Rev. sci. tech. Off. int. Epiz., 2010, **29** (2), 299-310

Biosecurity measures to prevent the incursion of invasive alien species into Japan and to mitigate their impact

K. Goka

National Institute for Environmental Studies, 16-2 Onogawa, Tsukuba, Ibaraki 305-8506, Japan
E-mail: goka@nies.go.jp

Summary

The 2004 Japanese 'Invasive Alien Species Act' was enacted to control invasive alien species and prevent the damage that they cause to ecosystems. The Act defines invasive alien species as those recognised as, or suspected of, causing damage to ecosystems, human safety, agriculture, forestry and fisheries. Invasive alien species are carefully regulated: raising, planting, keeping or transporting them is prohibited without the express permission of the relevant minister. The Act represents a revolutionary advance for biological conservation in Japan. However, enforcing the Act is problematic. Dealing with the European bumblebee (*Bombus terrestris*), for example, involved resolving a bitter dilemma between biological conservation and agricultural productivity. The Act also has a serious loophole; it does not include alien micro-organisms. The incursion of amphibian chytridiomycosis into Japan caused confusion for scientists and the Japanese Government because control of such an alien micro-organism was not anticipated in the Act. Japan faces particular difficulties in attempting to control alien species because of its reliance on imports.

Keywords

Batrachochytrium dendrobatidis – *Bombus terrestris* – Bumblebee – Chytridiomycosis – Control – Hitchhikers – Invasive Alien Species Act – Invasive species – Japan – Legislation – Risk assessment.

Introduction

The history of alien species is considered to have begun when people first started to migrate between continents. Both the distances travelled and the numbers of alien species have increased rapidly since the days of the great voyages of discovery in the 15th Century. Of course, it was difficult to predict the risks posed by alien species in ancient times, due to lack of ecological knowledge, but the understanding of ecology has improved enormously since then. Thus, the risks caused by alien species today should be assessed and, as far as possible, prevented, based on modern methods of ecological analysis.

The concept of controlling alien species includes the following:

– conducting risk assessments of alien species before they are introduced

– taking appropriate control measures to deal with those alien species that have already been introduced

– eradicating harmful and invasive alien species and preventing their entry

– making economic use of native species instead of alien species, wherever possible.

Alien species can increase and migrate autonomously and can also evolve to adapt to new environments. Thus, it is very difficult to control the impacts caused by alien species once they are established in the environment, compared to, for example, the effects caused by chemical pollutants, which have a half-life. Consequently, preventing the introduction of alien species is one of the most economic and effective measures for avoiding environmental and economic costs. It has been pointed out internationally that such prevention is much more important than controlling alien species that are already established (23).

To prevent the introduction of alien species, effective checks and quarantine measures are needed 'at the water's edge', i.e. at all entry points where the transportation of humans and goods may also introduce alien species. On the other hand, alien species that aid high productivity or provide economic benefits, such as green plants and agricultural bio-materials, will probably be introduced intentionally. Where species are introduced intentionally, they must be controlled so that they do not establish themselves in local ecosystems with harmful effects. Consequently, effective risk assessments based on scientific data and a strict system of regulation are needed.

If an alien species has, unfortunately, already become established, the following measures must be considered:
– eradicating the alien species
– controlling the alien species
– doing nothing.

In the past, since problems caused by the introduction of alien species have only been superficially recognised, the establishment of these species has been largely ignored. In the present day, the impacts caused by alien species should be able to be predicted, based on sound ecological data. If any detrimental impacts are predicted, eradication or control measures must be adopted against the alien species. But eradication or control are effective only when the establishment area is quite restricted.

If an alien species has already expanded its distribution, it is necessary to calculate the costs of control measures and the probability of their success. If control measures look likely to be ineffective, the alien species should be ignored. Of course, resources (money, time and labour) to invest in controlling alien species are always limited, so it is important to decide priorities. To rank the importance of the invasive species, their impact must be assessed from not only the ecological but also the economic viewpoint.

One of the most effective ways of preventing the introduction of alien species is through the legal system. Regulations can reduce the entry of alien species if there are severe penalties for letting them through and consequent obligations to eradicate (or pay for eradicating) them. In this review, the author examines a Japanese law enacted to control alien species, explaining both the legislation as a whole and also looking at particular case studies of alien species.

In particular, the author focuses on the case of the European bumblebee, *Bombus terrestris*, which has caused much argument. The author also examines the measures taken against chytridiomycosis invasion, as an example of an alien parasite which has 'fallen through' a loophole in the law.

The Invasive Alien Species Act in Japan

Japan is a country of islands with unique ecosystems. In particular, the Ryukyu Islands to the south-west and the Ogasawara (Bonin) Islands to the south support several endemic species. However, their local ecosystems are so fragile that such endemism could easily be affected by invasive alien species (IAS).

As a nation, Japan depends on international trade, importing massive volumes of goods, including living organisms. Many unwanted species are unintentionally brought into the country, both with the imported goods and in the transporting containers.

The present Japanese quarantine system was set up within the framework of the International Plant Protection Convention and the recommendations of the World Organisation for Animal Health. It is designed to prevent adverse effects from IAS on agriculture, forestry and fisheries but does not apply to wild fauna and flora and ecosystems. In light of this situation, and to implement the provisions of Article 8(h) of the Convention on Biological Diversity (5), and the guiding principles for the implementation of Article 8(h), adopted as Conference of the Parties (COP) 6 decision VI/23 (6), the Japanese Government created a new Act to address IAS in 2004.

The Invasive Alien Species Act was approved in 2004 and came into force in July 2005 (37). It aims to control IAS and prevent them from causing damage to ecosystems. The Act defines IAS as alien species recognised as, or suspected of, causing damage to ecosystems, human safety, agriculture, forestry and/or fisheries. The full text of the Act and its basic policy are available on line (37).

Relevant ministers have the responsibility of assessing and deciding, on advice from scientific experts, which species should be designated as IAS (Fig. 1). Species are assessed on whether they will:

– prey on native species

– compete with native species for ecological niches

– disturb the reproduction of native species by interspecies crosses

– destroy native ecosystem bases (for example, weeds).

Species considered alien are limited to those that have been introduced into Japan since the Meiji era (*circa* 1868), when Japanese trade with the rest of the world markedly increased and there was a rapid increase in introduced alien species.

Invasive alien species are subject to various regulations. Raising, planting, keeping or transporting them is prohibited without the express permission of the relevant ministers. Permission is a prerequisite for importing IAS and releasing them to the wild is not allowed at any time.

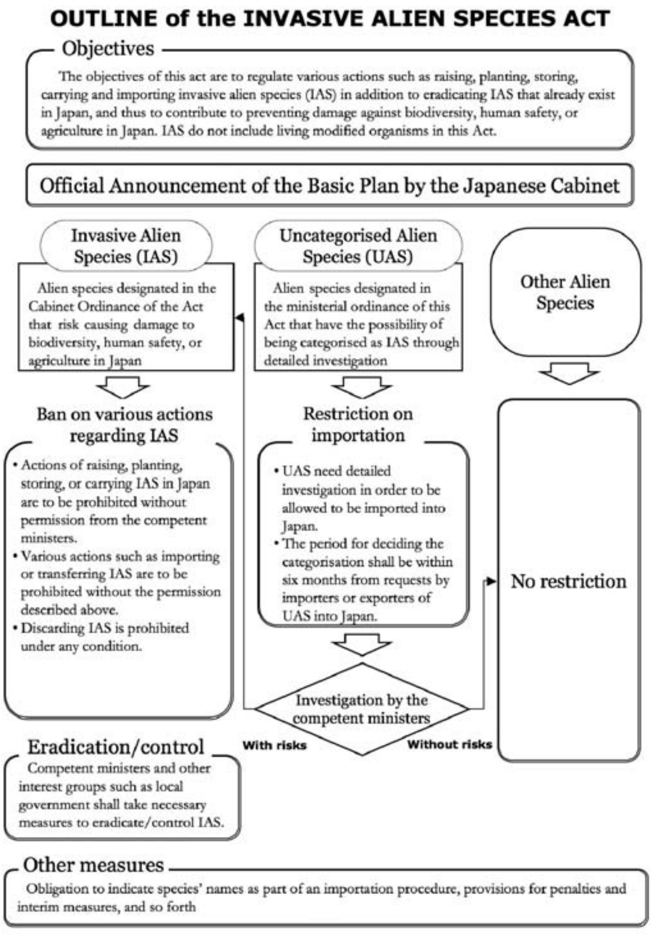


Fig. 1
An outline of the Invasive Alien Species Act 2004, Japan

The Act additionally defines species related to IAS as 'uncategorised alien species' (UAS). Species belonging to the same genus or family as an IAS can be defined as UAS because of the possibility of similar ecological impacts. For example, the Taiwan macaque (*Macaca cyclopis*) is designated as an IAS because the species competes with the Japanese native macaque (*M. fuscata*), so most other species in the genus *Macaca* are designated as UAS. Before importing UAS into Japan, importers must notify the relevant ministers of their intention and provide information on the ecological properties of the UAS. These species are evaluated by experts within six months of the application. Uncategorised alien species evaluated as posing a risk are immediately designated as IAS, whereas those posing no risk are permitted. By this process, six reptile and six amphibian UAS were judged to be IAS in 2008.

Although the IAS Act imposes controls on the importation of designated species, it would be more effective to prohibit all alien species. In Australia and New Zealand, the importation of all alien species is prohibited, except for species designated as safe. This quarantine system is known as a 'white-list system'. In contrast, the Japanese system is called a 'black-list system'. Although Japan would benefit from adopting a white list, Japan's economic situation would make this difficult as most of the natural resources on which Japan depends are imported from many countries. The introduction of a white-list system could impose obstacles on the supply of natural resources and the Japanese economy.

Selection of invasive alien species

The first selection of IAS was started in 2004. A general expert meeting and six working groups were established to deal with each group of alien species (mammals and birds, reptiles and amphibians, fish, insects, other invertebrates, and plants) and discuss which species should be designated as IAS. In addition, two special working groups were set up to discuss the largemouth bass (*Micropterus salmoides*) and the European bumblebee (*Bombus terrestris*). Largemouth bass were illegally released into lakes and ponds throughout Japan for game fishing and cause damage to fisheries and native fish species. European bumblebees have been widely used in greenhouses as pollinators. The sports fishing industry and farming industry were greatly concerned about the designation of these species as IAS because designation meant prohibition of their use. It was necessary to discuss the issue among stakeholders in special working groups to reach a conclusion.

Each working group convened and made draft recommendations, and the general expert meeting decided on the final recommendation to the appropriate minister. After public consultation, the ministers finalised their decision. More than 110,000 comments were received from the general public; most were against the designation of the largemouth bass as an IAS. Newspapers and other media reported the conflict widely, resulting in a rapid increase in public awareness of the IAS Act.

Species being proposed for designation were selected by identifying those species whose adverse effects had already been reported by international scientific publications. As a result, about 100 species were selected as IAS. The list of IAS is available on line (38).

On the other hand, alien species that were already widely established, such as the red swamp crawfish, *Procambarus clarkii*, and the red-eared slider, *Trachemys scripta elegans*, have not been listed as IAS because of the huge difficulties involved in control. In addition, the African lovegrass, *Eragrostis curvula*, which is a serious invading plant species for riverbed flora (34), has not been listed as an IAS because this species is widely used for planting on construction sites, and no alternative species have been found. Exotic stag beetles have been imported live into Japan as companion animals from many other countries, since 1999 (15). The annual number of imported stag beetles is more than one million. Although it is clear that some of the exotic species pose ecological risks for the native Japanese species, the Act has not controlled their entry. The number of exotic stag beetles bred in Japan alone is estimated at approximately half a billion, so, it is thought, if owners and breeders released them into the wild in anticipation of a ban, it would be impossible to control their numbers.

The case of the European bumblebee, *Bombus terrestris*

The introduction of an alien pollinator

The basic policy behind the IAS Act was decided by a Cabinet decision in 2005. Section 2.3 of the policy states that alien species causing damage or posing risks shall be considered, along with the social and economic consequences of their designation. The process for the designation of the European bumblebee provides an illuminating example.

The European bumblebee, *B. terrestris*, is one of the most successful biological agents used for commercial pollination. The bumblebee-breeding industry has flourished worldwide since the 1980s and has helped to increase rates of agricultural productivity (43). In Japan, the use of the bumblebee, mainly for pollinating tomatoes, has steadily increased since its introduction in 1991, and the number of commercial colonies used annually reached almost 70,000 by 2004 (29). The spread of commercial colonies of *B. terrestris* has increased tomato crop productivity, and has also led to a decline in the use of chemical pesticides, to protect the pollinating activities of the bee. This, in turn, has increased the quality and safety of tomato products.

On the other hand, there are fears that introduced biological agents can also become IAS. In Japan, many ecologists and entomologists have warned of the ecological risks posed by *B. terrestris* since its introduction (13, 47). In fact, a naturalised colony was found in Hokkaido in 1996 (47). Since then, the number of captive colonies of *B. terrestris* escaping into the wild from glasshouses or vinyl houses has continued to increase, suggesting that the rate of invasion by the alien bee is increasing (35). It is feared that the European bumblebee will eliminate native Japanese species of bumblebee through competition because of the similarity of their ecological niches (47).

Many ecologists and entomologists have argued that *B. terrestris* should be defined as an IAS under the IAS Act. Conversely, agriculturalists concerned about conserving agricultural productivity have objected to legal regulation of the use of *B. terrestris*. The alien bee has therefore become caught in a dilemma between two opposing agendas: biological conservation and agricultural productivity.

Although conservation of the Japanese ecosystem is the first principle of the Act, another policy is that the socio-economic background to the use of an introduced species should be considered in full before decisions are made to regulate that species. Thus, since the introduction of *B. terrestris* has both an economic aspect (through its contribution to agricultural productivity) and a social aspect (through improving the living standards of farming families), caution was required in deciding whether to declare *B. terrestris* an IAS.

Risk and impact assessment of *Bombus terrestris*

In light of this situation, the Ministry of the Environment set up a Bumblebee Specialist Group to discuss how *B. terrestris* would be managed. The group listed four ecological risks posed by *B. terrestris*:

– it excludes native pollinators through competition for food and nest sites

– it inhibits the reproduction of native plants by disturbing pollination ecosystems

– it disturbs the reproduction of native bumblebees through interspecies crosses

– it introduces alien parasites that could be pathogenic to native species.

The National Institute for Environmental Studies then set up a study project called: 'Development of control methods for ecological risks posed by introduced bumblebees'. The project was supported by the Research Project for Utilising Advanced Technologies in Agriculture, Forestry, and Fisheries, in collaboration with other institutes, universities, private companies and the government. This project has been dealing with the task of determining the four listed ecological impacts in the field since April 2005 and examining the accumulated scientific data.

On the basis of the scientific data confirming these four ecological impacts (9, 16, 17, 21, 22, 28), at the Specialist Group meeting held in December 2005, *B. terrestris* was declared to be an invasive threat to native Japanese flora and fauna and therefore to require regulation by law.

Controlled use of invasive alien bumblebees

The Specialist Group also considered a permission system for using *B. terrestris*. The species would be able to be used for agriculture but only in a secure facility that could prevent its escape.

In the study project, alongside ecological impact surveys, the Specialist Group developed methods for preventing the escape of *B. terrestris* from the glasshouses in which the species is used. One successful method is to cover the glasshouses with nets, making it possible to completely prevent the bumblebees from escaping (Fig. 2) (25). Furthermore, the Group developed a statistical method for estimating the number of nests of *B. terrestris* naturalised in the field; this method will be used to monitor the numbers of bees escaping into the wild (26, 27). On the basis of these new methods for controlling and monitoring the naturalisation of *B. terrestris*, the Ministry of the Environment decided to adopt a permission system for use of the species. Since March 2007, farmers have been required to completely cover their glasshouses with netting and obtain permission from the Ministry of the Environment before using *B. terrestris*.

Further issues associated with *Bombus terrestris*

In terms of controlling IAS, the decision to regulate *B. terrestris* under the Act in Japan is epoch-making from two perspectives. First, the Act may regulate even a beneficial species, such as a pollinator, when that species is declared invasive. Secondly, the Act can combine conservation ecology with agricultural productivity. The legal regulation of *B. terrestris* can, therefore, be considered

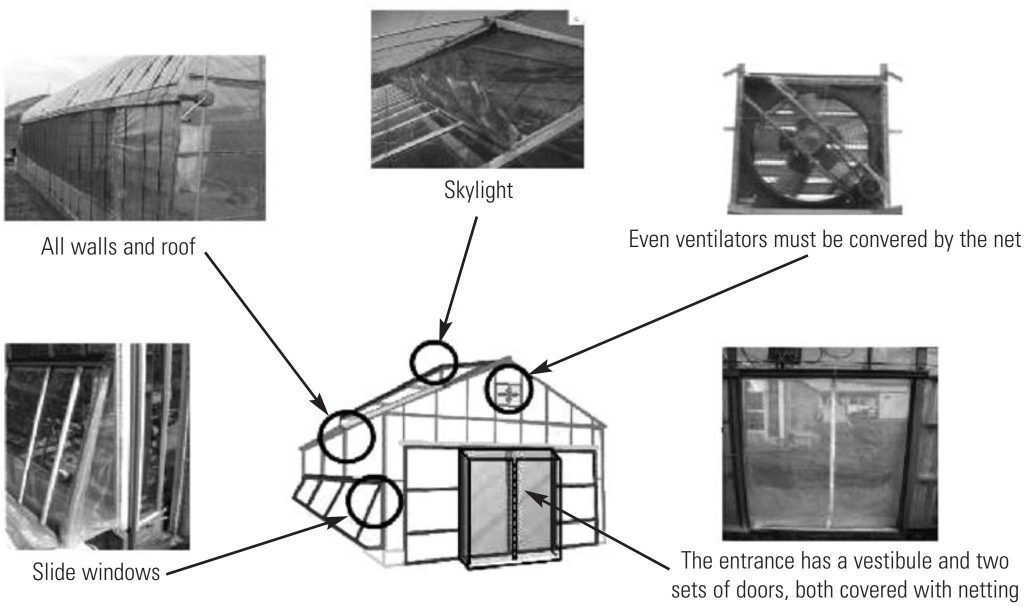


Fig. 2
Covering glasshouses with nets to prevent the escape of *Bombus terrestris*
Source: (25) (with some modification)

an innovative trial, aimed at reaching a state of mutualism between biodiversity and agriculture.

Nevertheless, many obstacles need to be overcome if this trial is to succeed. To secure the legal regulation of *B. terrestris*, the Group must educate farmers on the need to control this species. Monitoring systems that suit the control conditions in each glasshouse need to be constructed, and farmers must be helped to cover the costs of controlling *B. terrestris*.

As an alternative, the Ministry of Agriculture, Forestry and Fisheries is recommending the use of the native bumblebee species, *B. ignitus*, to pollinate tomato plants. As the native species is outside the scope of the Act, farmers cannot be penalised if the bee escapes from their glasshouses. However, even a native species poses a risk of acting as an IAS when it is transported artificially beyond its natural habitats. The first potent impact could be genetic introgression, caused by crosses between the natural and commercial colonies. Through DNA analysis, the Group has already found genetic diversity among local populations of *B. ignitus*. This observation indicates the need to account for the genetic endemism of *B. ignitus* before using commercialised colonies of the native bee (44).

Furthermore, recent shortages in the supply of the European honeybee, *Apis mellifera*, all over Japan are increasing the demand for bumblebees as alternative pollinators of various agricultural crops, in addition to tomatoes. Diversification of use could make the ecological risks posed by commercial bumblebees more difficult to control.

The case of the chytrid fungus, *Batrachochytrium dendrobatidis*

Invasion of an infectious disease of amphibians

There is a large loophole in the Invasive Alien Species Act. It does not cover alien micro-organisms. Under the present version of the Act, only alien species that can be visually identified must be inspected, and species too small to see (such as viruses, bacteria and fungi) are beyond the scope of the Act. Even though Japan has laws designed to control infectious diseases and parasites that affect humans, domestic animals and plants, these laws do not cover wildlife. Infectious diseases that affect wildlife populations are emerging at unusually high rates, and currently pose a great threat to the global conservation of biodiversity (20, 30, 46). Given that more than 500 million live animals are imported annually into Japan (36), the micro-organisms that accompany these animals are significant cause for concern.

This is the situation in which Japan faces a serious worldwide disease of amphibians caused by the chytrid fungus *Batrachochytrium dendrobatidis*. The chytrid fungus was first identified by Berger *et al.* (2) and described by Longcore *et al.* (32). Chytridiomycosis, caused by *B. dendrobatidis*, has been suggested as the infectious disease responsible for declines in wild frog populations in Australia, New Zealand, the United States, Central America, South America and Spain (2, 3, 4, 19, 31, 40, 42, 48). The fungus had never been reported in Asia until Une *et al.* (45) found it on a pet frog, imported from South America, in December 2006. This was the first report of amphibian chytridiomycosis in Japan.

Many Japanese herpetologists and ecologists were greatly disturbed by this first instance of the disease in Japan and the mass media described the news as a crisis in Japanese amphibians. To protect Japanese frog populations against this disease, it was vital first to understand the infection status of populations of both captive and free-ranging frogs, as well as the virulence of the fungus when it infects native Japanese species. Japanese citizens import many amphibians as scientific and medical materials, companion animals, food for companion reptiles, and for aquariums. These organisms come from many countries, including Central and South America (24), which are hotspots for *B. dendrobatidis* outbreaks. Furthermore, the African clawed frog (*Xenopus laevis*) and the American bullfrog (*Rana catesbeiana = Lithobates catesbeianus*) are alien species that have already become naturalised in Japan (1, 33). The amphibian chytrid fungus globally infects introduced populations of both species (12, 48). So, this situation was considered to pose a serious pandemic threat by *B. dendrobatidis* within Japan. The author and colleagues started to investigate, to determine the distribution of the fungus in Japan.

An urgent surveillance system for the amphibian chytrid fungus in Japan

At first, in order to identify amphibian chytrid fungal infections, the swab-sampling method was adopted, in which the skin of each collected amphibian was swabbed with a cotton swab and then DNA was extracted from each of the swab samples (Fig. 3). The author and colleagues developed a nested polymerase chain reaction (PCR) assay to obtain specific and highly concentrated PCR products of the internal transcribed spacer (ITS) gene in the fungus from the extracted DNA solution, which contained lots of contaminants (17).

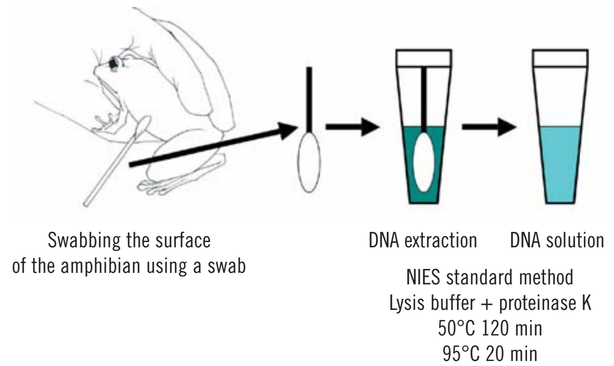
Then, they established a system for inspecting the infection status of amphibians in Japan, by collaborating with universities, non-governmental organisations, the Ministry of the Environment, local government, breeders and veterinarians (Fig. 4). Using this system, they collected swab samples from:

– 265 amphibians sold in pet shops

– 294 amphibians bred in institutes

– more than 5,000 amphibians collected at field sample points from northern to south-western Japan (14, 18).

The results of nested-PCR assays for these samples were surprisingly different from the author's expectations.



NIES: National Institute for Environmental Studies, Tsukuba, Japan

Fig. 2

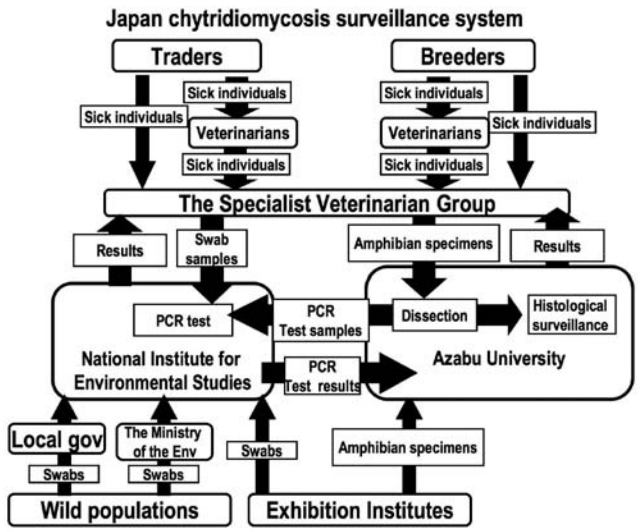
Taking swab samples from amphibians to detect *Batrachochytrium dendrobatidis* DNA

Source: (18)

Some infections were detected in both native and exotic amphibian species, both in captivity and in the wild (18). Sequencing of the PCR products revealed that Japanese amphibians carry more than 30 Japanese haplotypes of the *B. dendrobatidis* ITS region, including the haplotype already reported globally (called A type: Accession No. AY997031). Phylogenetic analysis of these haplotypes, combined with 48 *B. dendrobatidis* sequences already detected in other countries, such as the United States (J.P. Geartner, unpublished data) (41), Ecuador (J.P. Geartner *et al.*, unpublished data) and Italy (10), and inscribed in the DNA Data Bank of Japan/European Molecular Biology Laboratory/GenBank International DNA Database, showed that the genetic diversity of *B. dendrobatidis* was higher in Japan than in other countries (Fig. 5) (18). Furthermore, it was suggested that three of the haplotypes detected in Japan (B, J and K) were specific to the Japanese giant salamander (*Andrias japonicus*) and appeared to have established a commensal relationship with this native amphibian. In fact, although the incidence of infection was high in the giant salamander (> 40%), no disease signs were detected (18).

The highest infection rate of *B. dendrobatidis* (> 60%) and the highest genetic diversity were found in the sword-tail newt (*Cynops ensicauda popei*), which is endemic to Okinawa Island, in the south-western islands of Japan. The American bullfrog (*R. catesbeiana* = *L. catesbeianus*) showed the next highest genetic diversity of *B. dendrobatidis*, and also a relatively high incidence of infection (about 20%). The other alien species, the African clawed frog (*X. laevis*), which is believed to be the original host of the chytrid fungus, showed a high incidence of infection but the genetic variation of the fungus was relatively low (18).

On the other hand, the overall incidence of *B. dendrobatidis* infections in native amphibian species other than *C. ensicauda* and *A. japonicus* was extremely low (< 1%) (18). It has been suggested that *R. catesbeiana* is one of the key species responsible for expanding the global distribution of *B. dendrobatidis*, by acting as a carrier of the fungus (8, 11, 12, 39). However, the author and colleagues found no trend suggesting that the incidence of the fungus in amphibians increases around habitats where infected individuals of *R. catesbeiana* were found. On the contrary, they found that sites with infected native species were not adjacent to sites with infected *R. catesbeiana*. For example, the Okinawa sword-tail newt (*C. ensicauda*) population was infected with a relatively high incidence of *B. dendrobatidis* in a natural forest on Okinawa Island, as described above, but no alien amphibians were found in that area (18). These results suggest that horizontal infection between *R. catesbeiana* and native species is not the primary source of infection.



PCR: polymerase chain reaction
Gov: government
Env: environment

Fig. 4

Structure of the chytrid fungus surveillance system for Japanese amphibians

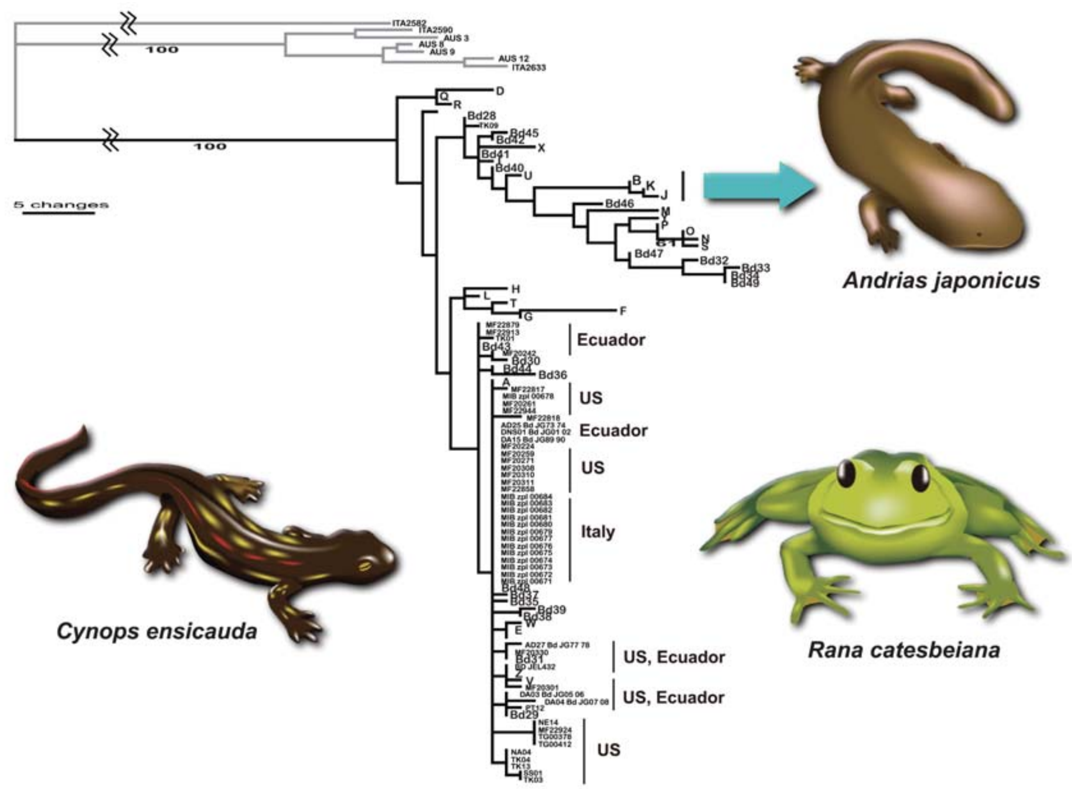


Fig. 5
Phylogenetic tree for the internal transcribed spacer haplotypes of *Batrachochytrium dendrobatidis*, using maximum parsimony analysis

Grey branches mean sister groups or out-groups (closely related fungi species). The code on the tip of each branch means the haplotype of the internal transcribed spacer gene of *B. dendrobatidis*. Large, bold letters indicate haplotypes of *B. dendrobatidis* detected in Japan. Small, slight characters indicate sequences identified in the United States (US), Ecuador and Italy

Source: (18) (with new data)

The low incidence of *B. dendrobatidis* in most of the native amphibian species in Japan suggests that the fungus is endemic to Japan and that many of the native species can tolerate infection by it. In fact, no signs of disease have been reported in indigenous Japanese amphibians infected with the fungus, in captivity or in the wild. These observations of diversity and endemism in *B. dendrobatidis* in Japan could lead to a new hypothesis for the source of the fungus: an 'Asian or Japanese origin hypothesis'. If so, the various haplotypes of chytrid fungus on imported and/or naturalised alien amphibian species in Japan might be propagated from native Japanese amphibians which are cryptically infected.

Implications from the hypothesis of an Asian origin for the chytrid fungus

If the amphibian chytrid fungus originated in Japan or Asia, how did it expand its distribution worldwide? The past trade in amphibians as food resources or companion animals between Asian and other countries might have caused the expansion. However, in Latin America and Australia, the remarkable damage caused to wild amphibians by the fungus has occurred mainly in tropical rainforests in the highlands. How, then, and why did the fungus arrive in such unexplored and undeveloped areas? The fungus would presumably be spread in human society and urban areas by artificial transport. But, instead, it has invaded undeveloped rainforest. The rapid development, alteration of landscape and flourishing of eco-tourism may have affected its expansion in these countries.

The genetic diversity and endemic nature of *B. dendrobatidis* detected in this study suggest a host–parasite co-speciation between amphibians and *B. dendrobatidis*, in which each of the fungal strains possesses a specific natural host. Anthropogenic disturbances of the environment and artificial transportation of amphibians into new habitats undoubtedly carry the chytrid fungus from its native habitats into non-native ecosystems. Under these conditions, the alien fungus must switch to a new host amphibian if it is to survive, and unnatural combinations of amphibians and fungal strains that have not co-evolved may explain the resulting pandemic. This scenario fits the case studies of emerging diseases that threaten human health, such as acquired immune deficiency syndrome and severe acute respiratory syndrome (7, 30). *Batrachochytrium dendrobatidis* thus provides significant evidence of the importance of conserving biodiversity, from the epidemiological viewpoint.

Finally, the argument that the entry of chytridiomycosis would cause a crisis in Japanese amphibians is now considered too presumptive. The conclusion of the author and others is that firm scientific data and evidence are needed to realistically assess the risks of alien species and that effective systems must be constructed to support risk assessments. Otherwise, there is a real danger that the true risks of IAS will be missed, minimised or overlooked.

Conclusion

Between 2003 and 2007, the number of live animals imported into Japan declined (Table I). One reason for this decline was the increased rigour of animal quarantine restrictions, especially against avian influenza. Another reason seems to have been the enforcement of the Invasive Animal Species Act. Raising public awareness of the risks of importation has brought about this change.

Table I
The decline in the numbers of live animals imported into Japan between 2003 and 2007

Category of animal	2003	2007	Change
Mammals	636,337	335,417	– 47.3%
Birds	121,114	35,346	– 70.8%
Reptiles	713,415	442,550	– 38.0%
Amphibians	28,912	4,571	– 84.2%
Ornamental fish	71,073,056	62,928,391	– 11.5%

Source: (36)

A number of challenges are involved in preventing the adverse effects of IAS. Preparing effective measures against the unintentional introduction of such species is one essential challenge. In addition, the Act does not cover the risk of infectious diseases to wild animals. To address these problems, it will be necessary to strengthen the existing legislation, but monitoring the many types of living organisms and imported goods entering the country will be difficult with limited staffing and resources. To mitigate the risks posed by IAS, it is vital that the Japanese public realise the precarious nature of Japan's strong economic dependence on international trade (15).

Acknowledgements

The author is grateful to all those who collaborated in the studies presented in this paper, including: Jun Yokoyama of Yamagata University; Yumi Une of Azabu University; Toshiro Kuroki of Kanagawa Prefectural Institute of Public Health; Kazutaka Suzuki, Maki N. Inoue, Atsushi Tominaga and Miri Nakahara of the National Institute for Environmental Studies; Shigeki Inaba of the National Institute of Technology and Evaluation; Tomoo Mizutani of the Ministry of the Environment; Alex D. Hyatt of the Commonwealth Scientific and Industrial Research Organisation (CSIRO), and Jean-Marc Hero of Griffith University. This study was supported by the Global Environment Research Fund (F-3 and F-081, leader: K. Goka) of the Ministry of the Environment, Japan, and the Research Project for Utilising Advanced Technologies in Agriculture, Forestry and Fisheries (No. 1701; leader: K. Goka) of the Ministry of Agriculture, Forestry and Fisheries, Japan.

■

Mesures de biosécurité destinées à prévenir l'introduction d'espèces allochtones envahissantes au Japon, ou d'en atténuer l'impact

K. Goka

Résumé
Au Japon, la loi de 2004 sur les espèces allochtones envahissantes a encadré la lutte contre ces espèces afin de prévenir leurs effets néfastes sur les écosystèmes. Cette loi définit le caractère envahissant d'une espèce allochtone en fonction de sa capacité avérée ou suspectée d'avoir des effets négatifs sur les écosystèmes, la sécurité des populations humaines, l'agriculture, la sylviculture ou la pisciculture. Les espèces envahissantes font l'objet d'une réglementation rigoureuse : la culture, la plantation, la conservation et le transport des espèces envahissantes végétales sont interdits sauf autorisation

expresse du ministère concerné ; il en va de même pour les espèces animales exotiques envahissantes, qui ne peuvent être élevées, gardées ni transportées sans autorisation. Cette loi représente une avancée révolutionnaire en faveur de la protection des ressources biologiques au Japon. Sa mise en œuvre pose toutefois quelques problèmes. Par exemple, dans le cas du bourdon terrestre (*Bombus terrestris*) et de sa gestion, il a fallu résoudre le cruel dilemme entre l'impératif de la conservation biologique, d'une part, et les exigences de la productivité agricole, de l'autre. Par ailleurs, cette loi comporte une grave lacune : elle ne couvre pas les micro-organismes exotiques. L'incursion de la chytridiomycose au Japon, maladie infectieuse affectant les amphibiens, a posé de grands problèmes aux scientifiques et au gouvernement japonais, car la loi ne prévoyait aucune mesure de contrôle à l'égard de ce type de micro-organisme allochtone. Le Japon dépend fortement des importations, ce qui entraîne des difficultés particulières en matière de gestion des espèces exotiques.

Mots-clés
Animal envahissant « autostoppeur » – *Batrachochytrium dendrobatidis* – *Bombus terrestris* – Bourdon terrestre – Chytridiomycose – Espèce envahissante – Évaluation du risque – Japon – Législation – Loi sur les espèces allochtones envahissantes – Lutte.

■

Medidas de seguridad biológica para prevenir la penetración en el Japón de especies foráneas invasoras y atenuar sus consecuencias

K. Goka

Resumen

En 2004 se promulgó en el Japón la "Ley sobre especies foráneas invasoras" con el objetivo de controlar a esas especies y prevenir los daños que ocasionan a los ecosistemas. Dicha ley define a las especies foráneas invasoras como aquellas que, a ciencia cierta o presumiblemente, resultan nocivas para los ecosistemas, la seguridad humana y la actividad agrícola, forestal o pesquera. Las especies foráneas invasoras son objeto de una minuciosa reglamentación: salvo autorización expresa del ministerio competente, está prohibido criarlas, plantarlas, mantenerlas o transportarlas. La ley representa un avance revolucionario en materia de conservación biológica en el Japón. Sin embargo, su aplicación práctica resulta problemática. El caso del abejorro común (*Bombus terrestris*), por ejemplo, planteó un amargo dilema entre conservación biológica y productividad agrícola. La ley adolece además de una importante laguna: no incluye a los microorganismos foráneos. La penetración en el país de la quitridiomicosis de los anfibios generó confusión en los científicos y el Gobierno japonés, toda vez que en la ley no había previsión alguna para combatir a este tipo de microorganismo foráneo. Dada la gran dependencia de las importaciones que tiene el país, el control de las especies foráneas plantea especiales dificultades en el Japón.

Palabras clave
Abejorro común – *Batrachochytrium dendrobatidis* – *Bombus terrestris* – Control – Determinación del riesgo – Especie invasora – Japón – Legislación – Ley sobre especies foráneas invasoras – Polizones – Quitridiomicosis.

■

References

1. Arao K. & Kitano T. (2006). – *Xenopus laevis* from Hamamatsu City, Shizuoka Prefecture, Japan [in Japanese]. *Bull. Herpetol. Soc. Jpn*, **1**, 17-19.

2. Berger L., Speare R., Daszak P., Green D.E., Cunningham A.A., Goggin C.L., Slocombe R., Ragan M.A., Hyatt A.D., McDonald K.R., Hines H.B., Lips K.R., Marantelli G. & Parkes H. (1998). – Chytridiomycosis causes amphibian mortality associated with population declines in the rain forests of Australia and Central America. *Proc. natl Acad. Sci. USA*, **95** (15), 9031-9036.

3. Bosch J., Martinez-Solano I. & Garcia-Paris M. (2001). – Evidence of chytrid fungus infection involved in the decline of the common midwife toad (*Alytes obstetricans*) in protected areas of central Spain. *Biol. Conserv.*, **97**, 331-337.

4. Bradley G.A., Rosen P.C., Sredl M.J., Jones T.R. & Longcore J.E. (2002). – Chytridiomycosis in native Arizona frogs. *J. Wildl. Dis.*, **38** (1), 206-212.

5. Convention on Biological Diversity (1992). – Multilateral Convention on Biological Diversity (with annexes). Concluded at Rio de Janeiro on 5 June 1992. Available at: www.cbd.int/doc/legal/cbd-un-en.pdf (accessed on 23 May 2010).

6. Convention on Biological Diversity (2002). – COP 6 Decision VI/23: Alien species that threaten ecosystems, habitats or species. *In* COP 6 – 6th Ordinary Meeting of the Conference of the Parties to the Convention on Biological Diversity, 7-19 April, the Hague, the Netherlands. Available at: www.cbd.int/decision/cop/?id=7197 (accessed on 23 May 2010).

7. Daszak P., Cunningham A.A. & Hyatt A.D. (2000). – Emerging infectious diseases of wildlife – threats to biodiversity and human health. *Science*, **287** (5452), 443-449. Erratum: *Science*, **287** (5459), 1756.

8. Daszak P., Strieby A., Cunningham A.A., Longcore J.E., Brown C.C. & Porter D. (2004). – Experimental evidence that the bullfrog (*Rana catesbeiana*) is a potential carrier of chytridiomycosis, an emerging fungal disease of amphibians. *Herpetol. J.*, **14**, 201-207.

9. Dohzono I., Kunitake Y.K., Yokoyama J. & Goka K. (2008). – Effects of an alien bumblebee on native plant reproduction through competitive interactions with native bumblebees. *Ecology*, **89**, 3082-3092.

10. Federici S., Clemenzi S., Favelli M., Tessa G., Andreone F., Casiraghi M. & Crottini A. (2009). – Identification of the pathogen *Batrachochytrium dendrobatidis* in amphibian populations of a plain area in the northwest of Italy. *Herpetol. Notes*, **1**, 33-37.

11. Fisher M.C. & Garner T.W.J. (2007). – The relationship between the emergence of *Batrachochytrium dendrobatidis*, the international trade in amphibians and introduced amphibian species. *Fungal Biol. Rev.*, **21**, 2-9.

12. Garner T.W.J., Perkins M.W., Govindarajule P., Seglie D., Walker S.J., Cunningham A.A. & Fisher M.C. (2006). – The emerging amphibian pathogen *Batrachochytrium dendrobatidis* globally infects introduced populations of the North American bullfrog, *Rana catesbeiana*. *Biol. Lett.*, **2** (3), 455-459.

13. Goka K. (1998). – Influence of invasive species on native species – will the European bumblebee, *Bombus terrestris*, bring genetic pollution into the Japanese native species? *Bull. Biograph. Soc. Jpn*, **53**, 91-101.

14. Goka K., Hero J.M., Kriger K.M., Yokoyama J., Une Y., Suzuki K., Tominaga A., Mizutani T. & Hyatt A.D. (2010). – An inference of the origin of amphibian chytridiomycosis based on ITS-DNA variation: did it come from Japan? *Dis. Aquat. Organisms* (in press).

15. Goka K., Kojima H. & Okabe K. (2004). – Biological invasion caused by commercialization of stag beetles in Japan. *Global Environ. Res.*, **8**, 67-74.

16. Goka K., Okabe K. & Yoneda M. (2006). – Worldwide migration of parasitic mites as a result of bumblebee commercialization. *Popul. Ecol.*, **48**, 285-291.

17. Goka K., Okabe K. & Yoneda M. (2007). – Bumblebee commercialization has caused worldwide migration of parasitic mites. *In* Acarology XI (J.B. Morales-Malacara, V. Behan-Pelletier, E. Ueckermann, T.M. Perez, E.G. Estrada-Venegas & M. Badii, eds). Universidad Nacional Autonoma de Mexico, Mexico City, 25-32.

18. Goka K., Yokoyama J., Une Y., Kuroki T., Suzuki K., Nakahara M., Kobayashi A., Inaba S., Mizutani T. & Hyatt A.D. (2009). – Amphibian chytridiomycosis in Japan: distribution, haplotypes and possible route of entry into Japan. *Molec. Ecol.*, **18** (23), 4757-4774. E-pub.: 13 October 2009.

19. Green D.E., Converse K.A. & Schrader A.K. (2002). – Epizootiology of sixty-four amphibian morbidity and mortality events in the USA, 1996-2001. *Ann. N.Y. Acad. Sci.*, **969**, 323-339.

20. Harvell C.D., Kim K., Burkholder J.M., Colwell R.R., Epstein P.R., Grimes D.J., Hofmann E.E., Lipp E.K., Osterhaus A.D., Overstreet R.M., Porter J.W., Smith G.W. & Vasta G.R. (1999). – Emerging marine diseases – climate links and anthropogenic factors. *Science*, **285** (5433), 1505-1510.

21. Inoue M.N. & Yokoyama J. (2010). – Competition for flower resources and nest sites between *Bombus terrestris* (L.) and Japanese native bumblebees. *Appl. Entomol. Zool.*, **1**, 29-35.

22. Inoue M.N., Yokoyama J. & Washitani I. (2008). – Displacement of Japanese native bumblebees by the recently introduced *Bombus terrestris* (L.) (Hymenoptera: Apidae). *J. Insect Conserv.*, **12**, 135-146.

23. International Union for Conservation of Nature (IUCN) (2000). – IUCN guidelines for the prevention of biodiversity loss caused by alien invasive species. IUCN, Gland, Switzerland. Available at: http://cmsdata.iucn.org/downloads/2000_feb_prevention_of_biodiv_loss_invasive_species.pdf (accessed on 29 May 2010).

24. Japan Wildlife Research Center (JWRC) (2008). – Report of a study to grasp the situation of chytridiomycosis in Japan in 2007-2008 [in Japanese, with English summary]. JWRC Report. JWRC, Tokyo, 99 pp.

25. Koide T., Yamada Y., Yabe K. & Yamashita F. (2008). – Methods of netting greenhouses to prevent the escape of bumblebees [in Japanese, with English summary]. *Jpn J. Appl. Entomol. Zool.*, **52**, 19-26.

26. Kokuvo N., Toquenaga Y. & Goka K. (2007). – A simple visualization method to reconstruct nest-mate patterns among bumble bees (Hymenoptera, Apidae) using genetic data. *Appl. Entomol. Zool.*, **42**, 137-141.

27. Kokuvo N., Toquenaga Y. & Goka K. (2008). – Estimating colony number of *Bombus terrestris* (Hymenoptera, Apidae) queens foraging in Biratori, Hokkaido, Japan. *Appl. Entomol. Zool.*, **43** (1), 19-23.

28. Kondo I.N., Yamanaka D., Kanbe Y., Kunitake K.Y., Yoneda M., Tsuchida K. & Goka K. (2009). – Reproductive disturbance of Japanese bumblebees by the introduced European bumblebee *Bombus terrestris*. *Naturwissenschaften*, **96** (4), 467-475. E-pub.: 13 December 2008.

29. Kunitake K.Y. & Goka K. (2006). – A new aspect of the alien species problem: ecological risks caused by commercialization of the native bumblebee, *Bombus ignitus* [in Japanese]. *Nature & Insects*, **41** (4), 37-40.

30. Lebarbenchon C., Brown S.P., Poulin R., Gauthier-Clerc M. & Thomas F. (2008). – Evolution of pathogens in a man-made world. *Molec. Ecol.*, **17** (1), 475-484.

31. Lips K.R. (1999). – Mass mortality and population declines of anurans at an upland site in western Panama. *Conserv. Biol.*, **13**, 117-125.

32. Longcore J.E., Pessier A.P. & Nichols D.K. (1999). – *Batrachochytrium dendrobatidis* gen. et sp. nov., a chytrid pathogenic to amphibians. *Mycologia*, **91**, 219-227.

33. Maeda N. & Matsui M. (1989). – Frogs and toads of Japan [in Japanese]. Bun-Ichi Sogo Shuppan Publishers, Tokyo, 100-107.

34. Matsumoto J., Muraoka H. & Washitani I. (2000). – Whole plant carbon gain of an endangered herbaceous species *Aster kantoensis* and the influence of shading by an alien grass *Eragrostis curvula* in its gravelly floodplain habitat. *Ann. Bot.*, **86**, 787-797.

35. Matsumura C., Yokoyama J. & Washitani I. (2004). – Invasion status and potential ecological impacts of an invasive alien bumblebee, *Bombus terrestris* L. (Hymenoptera: Apidae) naturalized in Southern Hokkaido, Japan. *Global Environ. Res.*, **8**, 51-66.

36. Ministry of Finance of Japan (2008). – Trade statistics of Japan. Available at: www.customs.go.jp/toukei/info/index_e.htm (accessed in 2009).

37. Ministry of the Environment, Government of Japan (2004). – The Invasive Alien Species Act (Law No. 78 (June 2 2004)). Available at: www.env.go.jp/en/nature/as.html (accessed on 24 May 2010).

38. Ministry of the Environment, Government of Japan (2010). – List of regulated living organisms under the Invasive Alien Species Act. Available at: www.env.go.jp/nature/intro/1outline/files/siteisyu_list_e.pdf (accessed on 24 May 2010).

39. Ouellet M., Mikaelian I., Paul B.D., Rodrigue J. & Green D.M. (2005). – Historical evidence of widespread chytrid infection in North American amphibian populations. *Conserv. Biol.*, **19**, 1431-1440.

40. Pessier A.P., Nichols D.K., Longcore J.E. & Fuller M.S. (1999). – Cutaneous chytridiomycosis in poison dart frogs (*Dendrobates* spp.) and White's tree frogs (*Litoria caerulea*). *J. vet. diagn. Invest.*, **11** (2), 194-199.

41. Rodriguez E.M., Gamble T., Hirt M.V. & Cotner S. (2009). – Presence of *Batrachochytrium dendrobatidis* at the headwaters of the Mississippi River, Itasca State Park, Minnesota. *Herpetol. Rev.*, **40**, 48-50.

42. Ron S.R., Duellman W.E., Coloma L.A. & Bustamante M.R. (2003). – Population decline of the Jambato toad *Atelopus ignescens* (Anura: Bufonidae) in the Andes of Ecuador. *J. Herpetol.*, **37**, 116-126.

43. Ruijter A. (1996). – Commercial bumblebee rearing and its implications. *Acta horticult.*, **437**, 261-269.

44. Tokoro S., Yoneda M., Kunitake K.K. & Goka K. (2010). – Geographic variation of mitochondrial DNA in *Bombus ignitus*. *Appl. Entomol. Zool.*, **45**, 77-87.

45. Une Y., Kadekaru S., Tamukai K., Goka K. & Kuroki T. (2009). – First Asian report of spontaneous chytridiomycosis in frogs. *Dis. aquat. Organisms*, **82**, 157-160.

46. Ward J.R. & Lafferty K.D. (2004). – The elusive baseline of marine disease: are diseases in ocean ecosystems increasing? *PLoS Biol.*, **2** (4), e120. E-pub.: 13 April 2004.

47. Washitani I. (1998). – Conservation: ecological issues of the recent invasion of *Bombus terrestris* into Japan [in Japanese]. *Jpn J. Ecol.*, **48**, 73-78.

48. Weldon C., du Preez L.H., Hyatt A.D., Muller R. & Spears R. (2004). – Origin of the amphibian chytrid fungus. *Emerg. infect. Dis.*, **10** (12), 2100-2105.