EUROPEAN AND MEDITERRANEAN PLANT PROTECTION ORGANIZATION
ORGANISATION EUROPEENNE ET MEDITERRANEENNE
POUR LA PROTECTION DES PLANTES

09-15161

Report of a Pest Risk Analysis for *Hydrocotyle ranunculoides*

This summary presents the main features of a pest risk analysis which has been conducted on the pest, according to EPPO Decision support scheme for quarantine pests.

Pest:	*Hydrocotyle ranunculoides* L. f.
PRA area:	EPPO Region
Assessors:	The EWG was held on 2009-03-23-25, and was composed of the follc experts: - M. Guillaume Fried, LNPV Station de Montpellier, SupAgro (fried@supagro.inra.fr), - M. Andreas Hussner, Institut für Botanik, Universitaet Duesseldorf (andreas.Hussner@uni-duesseldorf.de), - M. Jonathan Newman, CEH Wallingford (jone@ceh.ac.uk), - Ms Gritta Schrader, Julius Kühn Institut (JKI) (gritta.schrader@jki.b - M. Ludwig Triest, Algemene Plantkunde en Natuurbeheer (APNA) (ltriest@vub.ac.be) - M. Johan van Valkenburg, Plant Protection Service (J.L.C.H.van.valkenburg@minlnv.nl)
Date:	2009-05

STAGE 1: INITIATION

Reason for doing PRA:	*Hydrocotyle ranunculoides* originates from the American continent and was introduced into the EPPO region as an ornamental plant for tropical aquaria and garden ponds, where it is still sold under its correct name, sometimes under other names (*H. vulgaris, H. leucocephala*, and *H. natans* which is a synonym of *H. ranunculoides*). The plant was first recorded as naturalised in the south-east of the UK in the 1980s (Newman, 2003). Naturalisation in the Netherlands and in Belgium was recorded in the last decade of the twentieth century (Baas & Duistermaat, 1999; Baas & Holverda, 1996; Krabben & Rotteveel, 2003; Verloove 2006, Invasive Species in Belgium Website). Deleterious impacts have been reported in these three countries. The species is also recorded in France, Ireland, Italy, Germany (see EPPO, 2009) but several EPPO countries are still free from *H. ranunculoides* and there are concerns that it may be able to enter and establish in further countries. This PRA assesses the risks of its further introduction into other EPPO countries and its current and predicted impact. An initial EPPO PRA was performed and approved in 2005. After the proposal of listing this species in the Directive 2000/29, the European Food Safety Authority reviewed the initial PRA and made some comments. The initial PRA is therefore revised in the view of the EFSA comments and of information having become available after the initial PRA (EFSA, 2007).
Taxonomic position of pest:	Kingdom: *Plantae*

Class: *Magnoliopsida* (Dicotyledons)
Family: *Apiaceae*

STAGE 2: PEST RISK ASSESSMENT

Probability of introduction
Entry

Geographical distribution:

Native range:

H. ranunculoides is considered to be native to North and South America (Everett 1981). Nevertheless, natural enemies are only reported from South America, but not from North America (Cordo *et al*., 1982). Some studies are in progress to determine with accuracy the native area of the plant (Newman, pers. comm., 2009).

North America: Canada (British Columbia, Quebec), Mexico, the USA (Alabama, Arizona, Arkansas, California, Delaware, Florida, Georgia, Illinois, Kansas, Louisiana, Maryland, Mississippi, New Jersey, New York, North Carolina, Ohio, Oklahoma, Oregon, Pennsylvania, South Carolina, Tennessee, Texas, Virginia, Washington, West Virginia). In some States (Illinois, New Jersey, New York) it is considered as an endangered species. Further details on American records can be found in USDA (2004).

Central America and Caribbean: Costa Rica, Cuba, Guatemala, Nicaragua, Panama. Martin & Hutchins (1981) indicate presence in Tropical America generally.

South America: Argentina, Bolivia, Brazil, Chile, Columbia (Holm *et al*., 1979), Ecuador, Paraguay, Peru, Uruguay (Mathias & Constance 1976).

Introduced range:

EPPO region: Belgium, France, Germany, Italy, the Netherlands, the United Kingdom, Ireland (Maguire *et al*., 2008; EPPO Datasheet, 2009).
According to Flora Iberica (ref), the mention of *H. ranunculoides* in Spain (Tutin *et al*., 1964-1980) could have resulted from confusions with small forms of *H. vulgaris* or *H. verticillata*.
See Appendix 2 for detailed maps.

Asia: Lebanon (Conroy, 2006), Iran (Naqinezhad *et al*., 2007), Israel (old record), Syria (Mouterde, 1966), Yemen (Wood, 1997).

Africa: Angola, Ethiopia, Kenya, Malawi, Tanzania, Uganda, Democratic Republic of Congo (Gonçalves, 1978), Madagascar, Rwanda (Troupin, 1978), Zimbabwe (Chikwenhere, 2001). Possibly also Sudan.

Oceania: Australia (Queensland, Western Australia) (Ruiz Avila & Klemm, 1996).

Note: the fact that it is endangered in its northern range of distribution in North America is considered to be due to sub-optimal climatic conditions.
Although mentioned as present in Austria in the previous PRA, the species does not occur in this country (F Essl, pers. comm., 2009). It is as well not recorded in Denmark (H E Svart, pers. comm., 2009) and Portugal (H Marchante, pers. comm., 2009).

Major host plants or habitats:	Freshwater bodies and ecosystems: ponds, ditches, marshes, waterways etc, more particularly, in static or slow-flowing waters (Newman & Dawson, 1999). In waters of high nutrient content the species thrives extremely well (EPPO, 2009).
Which pathway(s) is the pest likely to be introduced on:	Pathways are:

- Intentional import as an ornamental aquatic plant for use outdoors and in aquariums

From the isolated nature of the sites in which the plant has been observed, it can be suggested that they are almost all derived from human activity, whether by direct planting, by throwing away unwanted plants, or through cleaning of tropical aquaria or garden ponds where the plant fragments enter the water system (J. Newman, pers. comm., 2009). The plant is more likely to be introduced by aquarium trade through the Internet rather than direct retail (Newman, pers. comm., 2009).
The species has been imported into the EPPO region but is not considered to be imported anymore because local production is far more cost effective than importation (van Valkenburg, pers. comm., 2009). The species is known to be produced and traded within the EPPO region.

The actual sale of *H. ranunculoides* is difficult to ascertain because of the misapplied names. *H. ranunculoides* could be traded under the misapplied name *Hydrocotyle vulgaris* or the synonym *H. natans.* In Belgium, the species has also been sold as *H. leucocephala* (E. Branquart, pers. comm. 2009). Other *Hydrocotyle* species are in trade, which although being different species could be mislaballed (*H. umbellata, H. novae zeelandiae*, *H. verticillata*, *H. moschata*, *H. sibthorpioides*). *H. ranunculoides* is cited as *H. americana* L. in various catalogues (Brickell (ed), 1996).

See Q 1.33 on spread helped by human activities for data on trade within the EPPO countries.

As the plant is no longer imported, but is produced and traded, the entry pathway is not further considered. The volume of *H. ranunculoides* being produced and sold is considered to be very low.

- Intentional import for non ornamental uses

EFSA (2007) identified another pathway to be considered in the PRA which is the introduction of *H. ranunculoides* being used in phytoremediation (Bretsch, 2004) due to its ability to accumulate heavy metals and phosphorous (Poi de Neiff *et al*. 2003) and the general interest in the use of aquatic macrophytes for bioremediation (Vajpayee *et al*. 1995). Experts on phytoremediation were contacted to gather additional information.

Dr McCutheon, Hydrologist and Environmental Engineer for the University of Georgia was contacted, and reported that the community working on phytoremediation is concerned about the use of alien species and typically limit itself to screening and selecting suboptimal plant species from indigenous communities.
http://www.scientificjournals.com/sj/all/AutorenProfil/AutorenId/5118

Mr Marmiroli from the University of Parma was contacted, but no answer was received.
Marmiroli, N., & McCutcheon, S.C. (2003). Making phytoremediation a successful technology. In McCutcheon, S.C., & Schnoor, J.L. (Eds.), *Phytoremediation: Transformation and Control of Contaminants.* (pp. 85-119). Hoboken, NJ: Wiley-Interscience, Inc.

Prof. Dr. Peter Schroeder, working for the German Research Center for Environmental Health (http://www.scientificjournals.com/sj/all/AutorenAnzeigeESS/autorenId/1136) have been contacted but no answer was received.

In the EPPO region, other species are usually used for phytoremediation including *Phragmites australis*, *Typha* spp., etc (Cooper, 2001). Trials have been made in Belgium, and the species was planted along watercourses in the Ghent area, from where it spread towards the border of the Netherlands (See Appendix 2). The species has also been tested for phytoremediation in Germany under controlled situation (Hussner, pers. comm., 2009).

If an EPPO country was willing to use *H. ranunculoides* for phytoremediation, the species is already available in the region.

- Unintentional introduction: hitch-hiking with other aquatic ornamental plants.
According to Maki & Galatowitsch (2004), *H. ranunculoides* has not been found as a contaminant of other traded aquarium plants in Minnesota (USA). In their study, a total of 681 individual plants (corresponding to 123 species) were ordered from vendors across the USA between May and September 2001, and were composed of the following types: 66 emergent plants, 16 submersed plants, 34 floating leaved plants and 6 free-floating plants.
Some *Hydrocotyle* spp. produced within the EPPO region have been found to be contaminated with *H. ranunculoides* (J van Valkenburg,

pers. comm., 2009). Such contamination is considered as a spread pathway (see Q. 1.33 and picture in Appendix 4)

Identified pathways are:
- trade for ornamental and aquarium purposes on the Internet
- trade for ornamental and aquarium purposes in direct retail
- use for phytoremediation.

The plant is more likely to be traded for ornamental and aquarium purposes through the Internet rather than direct retail.

Entry is not considered because the most important pathway is intentional import.

Establishment

Plants or habitats at risk in the PRA area:

Freshwater bodies and ecosystems: ponds, ditches, marshes, waterways etc, more particularly, in static or slow-flowing waters (Newman & Dawson, 1999).

Climatic similarity of present distribution with PRA area (or parts thereof):

largely similar

Uncertainty: Medium

H. ranunculoides is already established in several EPPO member countries (Belgium, France, Italy, the Netherlands, United Kingdom, Ireland, quite recently also Germany).
The climatic conditions experienced over winter result in a smaller suitable area restricted to the margins of waterbodies (Newman, 2003).

The species is endangered in parts (U.S. federal states of Illinois, New Jersey and New York (New York Environmental Regulations, 2000; USDA, 2004)) of its native range where it is vulnerable to low temperatures. However, in its introduced range, even if emergent leaves die at the first night frosts and floating leaves die when enclosed in ice, leaves of *H. ranunculoides* submerged below ice cover are reported to survive the winter months, and new plants can grow up in spring from these overwintering parts (Hussner & Lösch, 2007).

In Western Europe populations may be strongly reduced during cold winters, but recovery occurs quickly in the following season.

Optimum temperatures for gas exchange (linked with photosynthesis) at the leaves surface have been recorded to be comprised between 25°C and 32°C (Hussner & Lösch, 2007).
At 35°C, the gas exchanges dropped.
The species being aquatic, it is not considered to be susceptible to air drought or humidity as long at it rooted in water. The species prefers growing in full sun, and is limited by shade.
According to the Climex simulation, the Atlantic and Mediterranean areas of the EPPO region that are characterized by mild winters are the most at risk. (see Appendix 3).

The countries at risk are: Albania, Algeria, Austria, Azerbaijan, Belgium, Bosnia & Herzegovina, Bulgaria, Croatia, Cyprus, Denmark, France (including Corsica), Greece, Ireland, Israel, Italy (including Sardinia and Sicilia), Jordan, Germany (mostly western part), Hungary, Moldavia, Morocco, Portugal, Romania, Russia (Black Sea), Serbia, Slovenia, Spain, Switzerland, Tunisia, Ukraine Black Sea), the United Kingdom, Republic of Macedonia, Romania, Turkey.

Nevertheless, so far, the species has expressed invasiveness in North-Western EPPO countries (Belgium, the Netherlands, United Kingdom,), while the areas which seem to be the most suitable are the Mediterranean and the Atlantic areas. This may be due to other elements such as the use of the plant and the eutrophication of waters.

There is some uncertainty, how the plant would perform in Northern and Eastern Europe. However, severe continental winters and hot and dry summers (e.g. in continental conditions) are likely to limit distribution of the species.
Additional shortage of water during summer would also limit the success of the species.

Characteristics (other than climatic) of the PRA area that would favour establishment:

H. ranunculoides is found in static, slow-flowing and occasionally flowing water bodies, especially ditches, canals, lakes and ponds. In the Netherlands, the species is found over a broad range of water quality conditions: from mesotrophic pools to the eutrophic lake Ijsselmeer margins. The environmental conditions in such habitats are present in most if not all EPPO countries. It is also important to note that eutrophic conditions are preferred: *H. ranunculoides* shows a much higher growth rate in high nutrient conditions, while maintaining similar rates of growth to native species in low nutrient conditions (Newman, 2002).

Sediments nutrients
In controlled conditions, optimal growth was observed in water with 20 mg N l^{-1} with a N uptake rate of 41 mg N g-1 (dw) of plant tissue (Reddy & Tucker, 1985). In Germany, fields' measurements showed that monospecific stands could occur in water with 6.2 to 11.5 mg of NO_3-N / kg of sediment and 2.9 to 61.9 mg of P_2O_5 / 100 g sediment (Hussner & Lösch, 2007).

Water quality (see map in Appendix 1)
In an area in France, the species remained confined to a restricted pond, most probably due to acidic waters which limit the vigour of the species (E Tabacchi, pers. comm., 2009).
In the EPPO region, there are no macronutrients limitations.
Arocena & Mazzeo (1994) showed the importance of alkalinity, total phosphorus and total inorganic nitrogen in the development of several macrophytes. Optimal development of *H. ranunculoides* was recorded in waters with the following mean values (extrema between brackets): total suspended solids: 63 mg +/- 52 [21-213] pH=7.1 +/- 0.4 [6.5-7.9], alkalinity: 5.0 meq/l +/- 2.1 [1.3-8.5], phosphorus: 21 μM+/- 10 [7-45], nitrogen: 116 μM +/- 77 [11-241].
In Belgium, summer field measurements found *H. ranunculoides* on sites with the following ranges of (Nijs *et al.*, 2009):
O2: 6-11 mg/l

pH: 6.7 – 7.5
conductivity: 232-699 µSiemens/cm
Total Phosphate (PT): 0.066-0.82 mg/l
Soluble reactive phosphorus: 0.005-0.21 mg/l
Dissolved inorganic nitrogen: 0.018-4.14 mg/l
These data show no particular preference for specific water quality parameters.

Physical characteristics of waterbodies

Experiments show that under stable water level regimes, *H. ranunculoides* adopted different morphologies, with highest biomass occurring in fully aquatic conditions (Hussner & Meyer, accepted). Water level fluctuation limit or decrease the biomass accumulation (Hussner, pers. comm., 2009).
In its native range in Argentina, Gantes & Sánchez Caro (2001) studied the distribution of aquatic plant in streams and reported that emergent plants including *H. ranunculoides* were relatively ubiquitous with the independence of their distribution in relation to the hydrological variables: current velocity (from ~ 0 to 35cm/s), stream width (from 100 to 700cm), stream depths (from 7cm to 50cm).
The EWG concluded that in the EPPO region, the species grows in waterbodies with velocities up to 1 m/s and depth up to several metres.

Water flow velocity

In the UK, significant infestations were found in 4 locations (River Chelmer, River Wey, Pevensey Levels, Gwent Levels), all of which are slow-flowing rivers or wide channels, which could be an abiotic factor favouring infestations (Newman & Dawson, 1999).
Static and very slow flowing waters are considered to be optimal habitats (Newman, pers. comm., 2009).

Salinity

The salinity tolerance of *H. ranunculoides* has been tested in a study by the Centre for Aquatic Plant Management, UK. The results of the study show a decrease in leaf number and an increase in leaf death rate above 6.5 ppt salinity. The effect is sharply marked, with a 0.5 ppt increase causing a dramatic effect (Rothamsted Research, 2000). As a comparison for salinity levels, undiluted seawater has a salinity of 35 ppt, and eutrophic fresh water of 4 ppt. The salinity tolerance is possibly physiologically linked with a capacity to take up metals from water, *H. ranunculoides* has substantial metal absorption capacities (Pinochet *et al*., 2002).

These abiotic factors are very common and largely similar to the ones in the native range.

The EFSA opinion suggested that the levels of eutrophication in water bodies as monitored by the Water Information System for Europe (WISE) of the European Environment Agency should be taken into account. The species is not borne to euthrophic waters, and the level of eutrophication does therefore not influence the distribution of the species. These maps have been checked by the

EWG but are not considered to provide any accurate additional information.

Competition

The high Leaf Area Index of up to 5.47 +-0.2, is an indication that the species is able to outcompete submerged vegetation (Hussner & Lösch, 2007).

In Belgium, it has been observed to reduce by more than 50% the number of native aquatic plant species (up to 100% of the submerged species (Nijs *et al*., 2009).

H. ranunculoides may be able to produce allelopathic anti-algal compounds (Della Greca *et al*., 1994).

Natural enemies

In Germany, observations showed that coypus (*Myocastor coypus*) can eat *H. ranunculoides* (Hussner & Lösch, 2007). Some populations were partially grazed by this mammal, which exclusively eats the leaf lamina of these plants. However, grazing does not prevent the establishment of the species.

During summer, cattle will eat the plant when it grows at the water margins, but this again has not prevented the establishment of the species, and even encourages the spread of the plant due to fragmentation (Newman, pers. comm., 2009).

Managed environment

The optimal habitat of *H. ranunculoides* are static or slow-flowing waters, and the creation of the slowing down of waters by creating dams may favour the establishment of the plant.

Restoration of water bodies and the creation of new ponds would encourage the establishment of the species to new sites.

Two factors contribute to the establishment of *H. ranunculoides*:

- high nutrient levels through agricultural, urban and industrial run-offs favour the rapid growth
- and impoundment of waters by creating dams, altering hydrological regimes.

Which part of the PRA area is the endangered area:

Freshwater bodies and ecosystems: ponds, ditches, marshes, waterways etc, more particularly, in and static or slow-flowing waters (Newman & Dawson, 1999).

According to the Climex simulation, the atlantic and mediterranean areas of the EPPO region that are characterized by mild winter are the most at risk. The countries at risk are: Albania, Algeria, Austria, Azerbaijan, Belgium, Bosnia & Herzegovina, Bulgaria, Croatia, Cyprus, Denmark, France (including Corsica), Greece, Ireland, Israel, Italy (including Sardinia), Jordan, Germany (mostly western part), Hungary, Moldavia, Morocco, Portugal, Romania, Russia (Black Sea), Serbia, Slovenia, Spain, Switzerland, Tunisia, Ukraine, the United Kingdom, Republic of Macedonia, Romania, Turkey. There is some uncertainty, how the plant would perform in Northern and Eastern Europe. However, severe continental winters and hot and dry summers (e.g. in continental conditions) are likely to limit distribution of the species. (See Appendix 3). Additional shortage of water during summer would also limit the success of the species.

The species is considered to be limited by acidic waters, as shown by the map of acidity of soils in Appendix 1. Acidic soils are found in the Centre of France, in Toscana, in Corsica which may explain why the species is not as invasive yet as in other localities where it is present in the EPPO region. Acidity of soils (and therefore waters) may in future limit the species in some places like the northern Atlantic coast of Spain, West of Sardinia, Scandinavia, Western France, etc.

POTENTIAL ECONOMIC CONSEQUENCES

How much economic impact does the pest have in its present distribution:

Effect on crop yield and/or quality to cultivated plants or on control costs

In the Canning River in Western Australia *H. ranunculoides* became a serious problem in 1992. A program costing over AU$ 200,000 in the first year was implemented (Atkins, 1994, Ruiz Avila, Klemm, 1996; Newman & Dawson 1999), and the species is still present in Australia.

Control costs: In the Netherlands, some water boards faced a doubling of costs each year during the 1990s, and, in 2000, the total annual control costs were around 1 Million Euro (van der Krabben & Rotteveel, 2003). In 2007, in the Netherlands, 11 water boards out of 26 responded to an inquiry stating that they spent an additional 1.8 millions euros for the management of *H. ranunculoides* over and above normal operating costs for this plant (van Valkenburg, pers. comm., 2009).

In Flanders, the estimated cost for the management of *H. ranunculoides* is 1.5 million euros per year (needed during 3 years from 2009) (Triest, pers. comm., 2009).

In the UK, the estimate for control of the total area infested by *H. ranunculoides* by herbicides was between £250,000 and £300,000 per year (Harper, 2002). In 2008, £1.93 million were spent for the management and disposal of *H. ranunculoides* (Newman, pers. comm.., 2009). In 6 years, the costs were multiplied 7 times.

Flooding caused by the plant may also have an economic impact due to loss of crops (Newman, pers. comm., 2009).

Environmental impact

Since 2005 (date of the previous PRA), much more information on environmental impacts was made available.

Direct effects

The EWG concluded that in most sites, 100% cover is often observed over large distances (25 km), which is detrimental for the ecosystem (see pictures in Appendix 4). The plant is perennial and present all year long in the UK.

In Belgium, it has been observed to reduce by more than 50% the number of native aquatic plant species up to 100% of the submerged

species, and to reduce the native cover from 50% to 10 (Nijs *et al*., 2009).
In Sardinia, the species is considered invasive, and although no specific impacts have been studied, the thick coverage of the species at the surface of the water is considered to outcompete other species (G Brundu, pers. comm., 2009).

In the PRA area, where present, *H. ranunculoides* competes with many plant species due to its ability to establish in different habitats. Examples: different *Carex*/sedge and *Juncus* species, *Rorippa amphibia*, *Myosotis palustris* (syn. *M. scorpioides*), *Nasturtium officinale* (A. Hussner, pers. comm.., 2009). In Germany, the native *Myriophyllum spicatum*, *Callitriche* spec. and *Potamogeton crispus* were displaced (Hussner, 2008). Nevertheless, these species are not endangered.
Due to the high LAI of up to 5.57 +/- 0.2 it seems obvious, that the species is able to outcompete submerged vegetation (Hussner & Lösch, 2007). Many more species can be outcompeted due to *H. ranunculoides'* capability to build floating carpets that shade out other plants.

Data on impacts in dense infestation are rare because of dangerous surveillance conditions underneath dense floating mats.

Indirect effects
Indirect effects on other biota and food web (phytoplankton, zooplancton, fishes) is caused by its summer biomass and by moments of decay (lowering of oxygen) and alteration of detritus (impact on macroinvertebrates) (Alien impact report, 2009; L Triest, pers. comm., 2009).
The EWG considered that *H. ranunculoides* causes many significant changes of ecological processes and structures by :

- reduction in flow;
- increased sedimentation resulting in acceleration of ecological succession;
- changes in O2 concentration;
- loss of accessible open water at the margins for wildlife (e.g. birds);
- loss of light;
- increased flood risk.

Presence of *H. ranunculoides* prevents attainment of good ecological quality status under the Water framework Directive (http://ec.europa.eu/environment/water/water-framework/index_en.html).

Social impacts
Effects on tourism (swimming, water sports, fishing, navigation, leisure etc.) can locally be expected to be large.
As waterways covered with *H. ranunculoides* are not attractive for recreation and may hinder traffic, even the movements of boats, some profit losses have been observed in the Netherlands (van Valkenburg, pers. comm., 2009).

	Dense vegetation mats can present a direct safety risk to the public and livestock. Cattle have drowned in the UK (Newman, pers. comm., 2009). Loss of aesthetic value in nature reserves has been reported in Belgium (Triest, pers. comm., 2009). Increased costs for drainage and/or flood prevention will be borne by the users (agriculture and general society). The water boards tax the inhabitants and enterprises of their management area.
Describe damage to potential hosts in PRA area:	There are currently no impacts recorded in crops, but the EWG considered that flooding of low lying agricultural areas is possible due to blockage of water level control structures. Environmental impact is supposed to be the same wherever the species grows in suitable conditions. For instance, in Essonne (France) and in Italy, similar impacts can be expected as in the Netherlands, UK and Belgium. In France, the species is currently only present in 7 sites, but already exhibits up to 100% cover of water surface in some of them.
How much economic impact would the pest have in the PRA area:	Control costs could be similar to those already spent in infested parts of the PRA area. Social impact is supposed to be the same wherever the species grows in suitable conditions.

CONCLUSIONS OF PEST RISK ASSESSMENT

Summarize the major factors that influence the acceptability of the risk from this pest:

Estimate the probability of entry:	High – Low uncertainty The plant has already entered the EPPO region. The plant is no longer imported, but is produced and traded. The volume of *H. ranunculoides* being produced and sold is considered to be very low.
Estimate the probability of establishment:	High – Low uncertainty The pest has already established in at least 6 countries of the EPPO region, the probability of establishment is therefore very high. According to the climatic prediction, additional countries are at risk (e.g.: Mediterranean countries, Black Sea area). Spread by human activities is very effective.
Estimate the potential economic impact:	High – Low uncertainty **Economic impacts:** medium to high risk. Economic impacts include management costs of the species and flooding of areas. Any economic benefit of the introduction of this plant as an ornamental aquatic plant is heavily outweighed by management costs. Flooding may also occur. It is very likely that these impacts would occur when the plant is introduced. **Environmental impacts**: medium to high risk. Invasion of slow flowing waters, degradation of aquatic ecosystem, loss of

biodiversity.
Social impact: low-medium risk. Where it occurs, it has an impact on navigation, recreation and fishing.

Degree of uncertainty	The degree of uncertainty is low. The areas of uncertainty identified are the following: - study on the varieties and forms, and the ones considered invasive. - the amount of internet trade - the amount of production in the EPPO region - the amount of exchange between gardeners and hobbyists, - Information of the situation in Italy - why did the plant disappeared from a river in Corsica (non found since 1968). - effects on water quality (e.g. O_2 content) and secondary effects on biota. Research needs identified: - data on outcompeted native species and their potential for recovery. - The effect of climatic change on the distribution and impacts of the plant - Biological control
OVERALL CONCLUSIONS	The risk of establishment of *Hydrocotyle ranunculoides* in waterways, and negative impacts on their vegetation and use, justifies measures to prevent its further spread in the EPPO region. **The pest qualifies as a quarantine pest.**

STAGE 3: PEST RISK MANAGEMENT

IDENTIFICATION OF THE PATHWAYS

Pathways studied in the pest risk management	**Intentional import as an ornamental aquatic plant for use outdoors and in aquariums**
Other pathways identified but not studied	/

IDENTIFICATION OF POSSIBLE MEASURES

Possible measures for pathways

- **Pathway 1: Intentional import as an ornamental aquatic plant for use outdoors and in aquariums**

Measures related to consignments:

Measures related to the crop or to places of production:
International measures
Prohibition of import and trade in the EPPO region and within the countries will effectively prevent further introduction into the EPPO region combined with accurate identification of species and synonyms.

National measures

Prohibition of the import, selling, planting, holding, movement, causing to grow in the wild, and possession of the plant may effectively prevent further establishment and spread within the EPPO region.

Other possible measures

Integrated management plan for the control of existing infestations

It is potentially highly effective if coupled with prohibition measures. Uncertainty concerns commitment to long-term implementation.

This would require:

- Accurate identification of the species
- Monitoring/surveillance in the countries where it is invasive or present (Belgium, France, Germany, the Netherlands, the United Kingdom, Italy), and surveillance in the countries at risk.
- Early warning consisting of exchanging information with other countries, and rapid response
- Control of existing populations.
- Publicity: aquatic plants producers and sellers and aquarium enthusiasts shall be informed of the problem and work should be undertaken with them to explain the prohibition of the species, and inform consumers. Administration should also be warned that the plant shall not be used as a phytoremediation species.

1 Monitoring and review

Performance of measure(s) should be monitored to ensure that the aim is being achieved. This is often carried out by inspection of the commodity on arrival, noting any detection in consignments or any entries of the pest to the PRA area.

EVALUATION OF THE MEASURES IDENTIFIED IN RELATION TO THE RISKS PRESENTED BY THE PATHWAYS

Degree of uncertainty Low

CONCLUSION:
Recommendation for possible measures:
PC= Phytosanitary certificate, RC=Phytosanitary certificate of re-export

Pathway 1:	Prohibition

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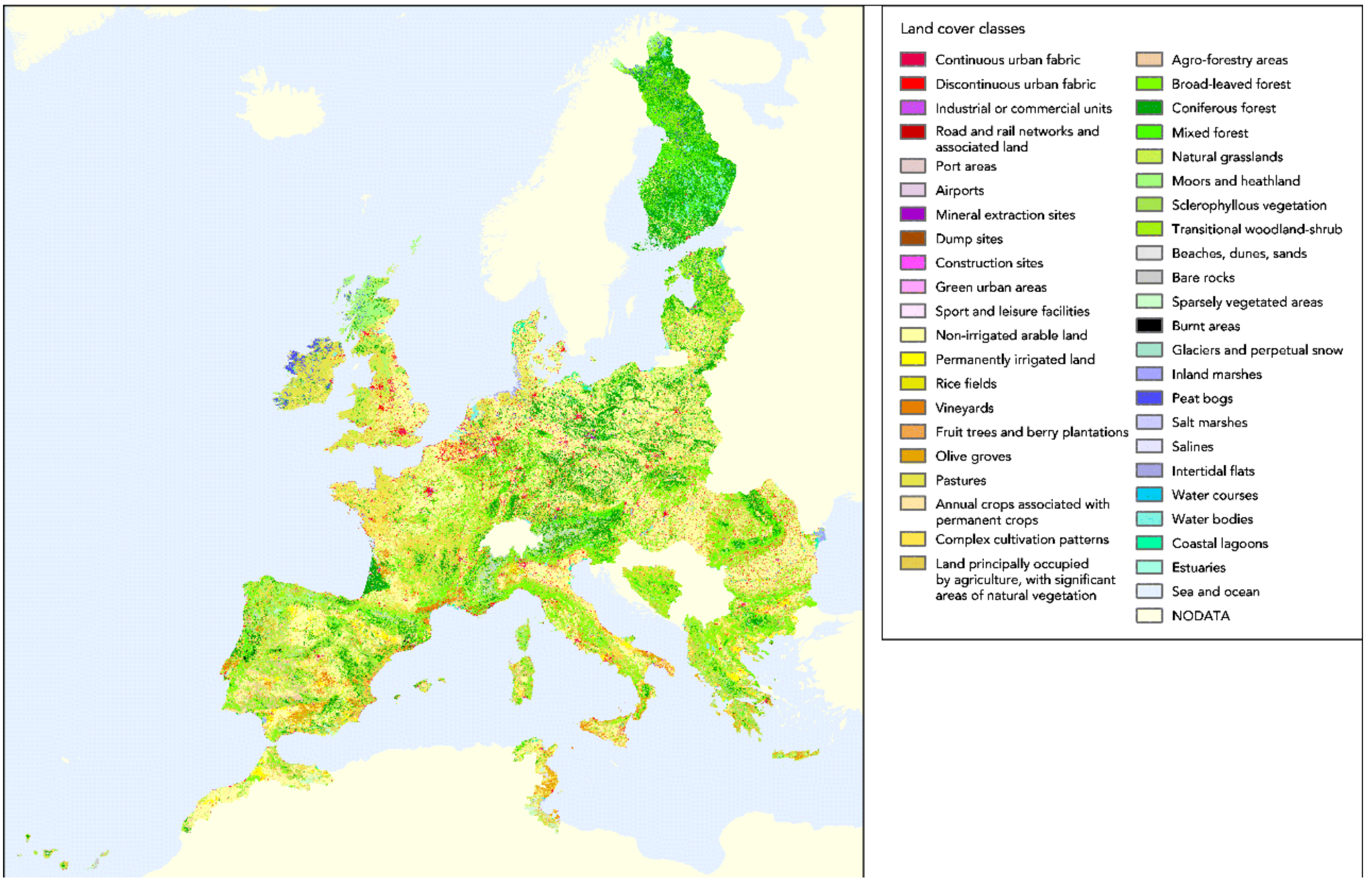
Appendix 1

Maps relevant for the distribution of *Hydrocotyle ranunculoides*

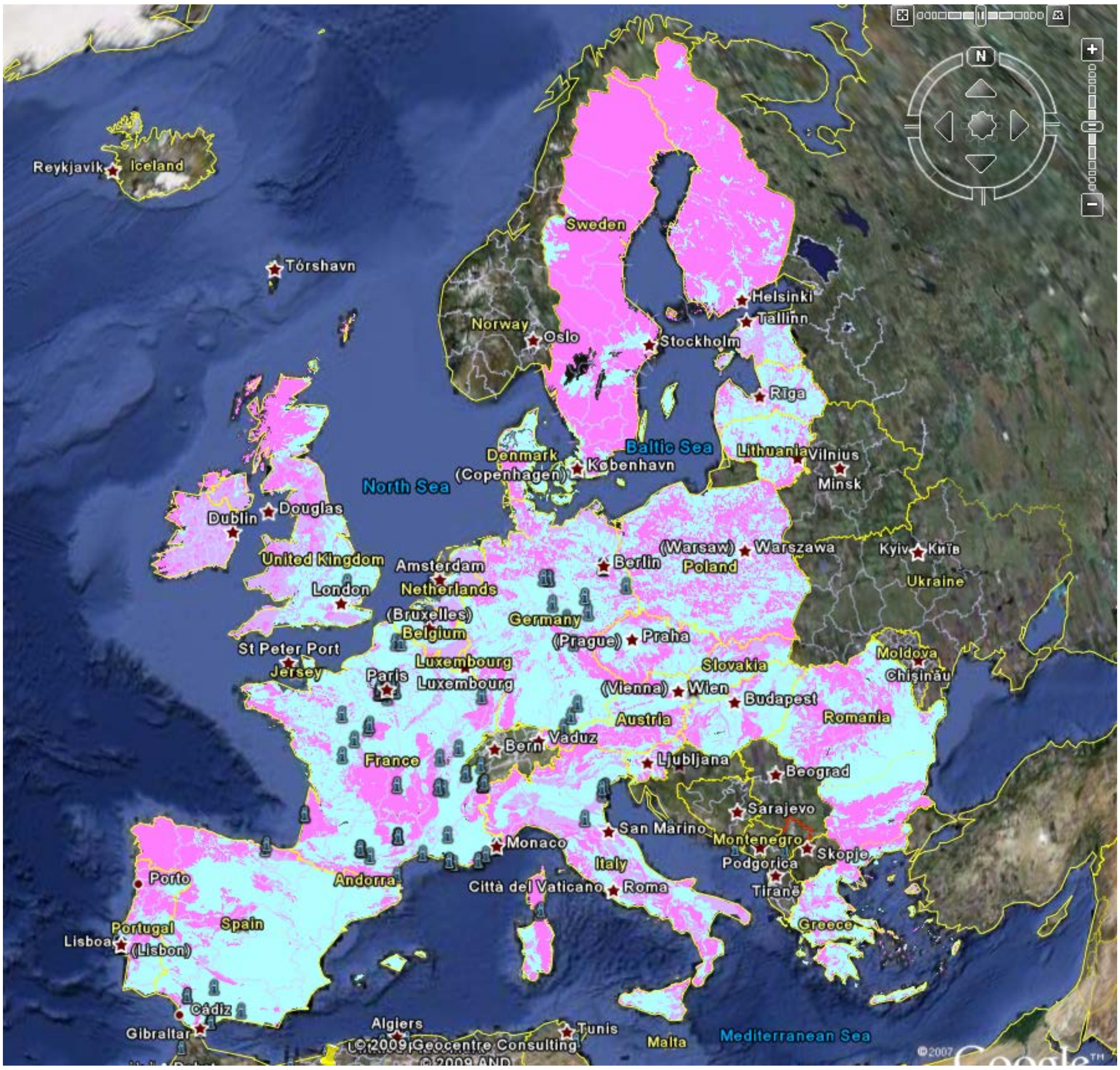
CORINE land cover classification

http://dataservice.eea.eu.int/download.asp?id=5859&type=gif.



pH maps

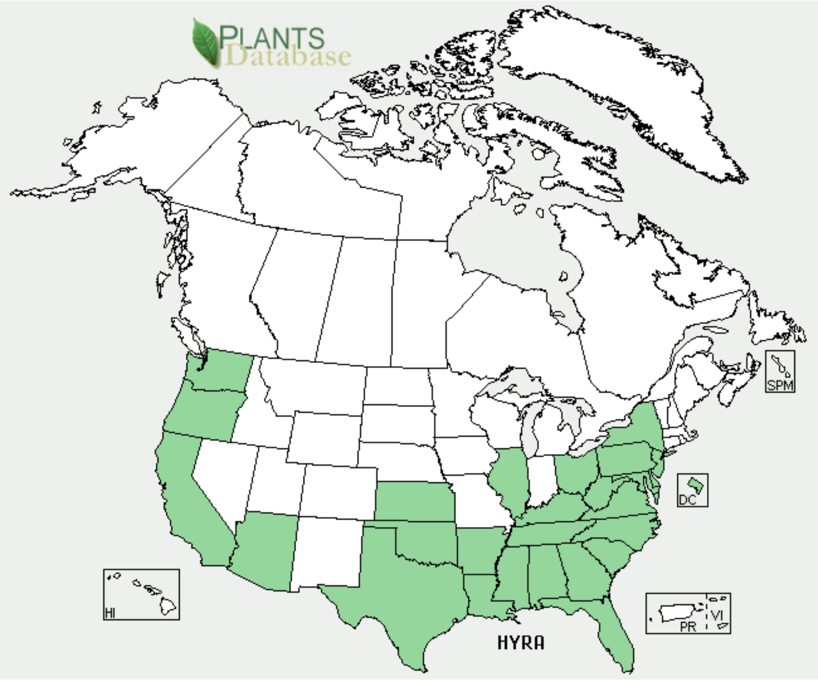
The following map can be found on the European Soil Portal maintained by the European Commission (http://eusoils.jrc.ec.europa.eu/, European soil data center > Data > European soil data base > Raster version or Google earth version> chemical properties > base saturation top soil (BS TOP)
The areas in pink (darker) represent acidic soils which are not suitable for *Hydrocotyle ranunculoides*.


Reykjavik
Iceland
Tórshavn
Sweden
Helsinki
Tallinn
Norway
Oslo
Stockholm
Riga
Baltic Sea
Denmark
(Copenhagen)
København
Lithuania
Vilnius
Minsk
North Sea
Dublin
Douglas
United Kingdom
Amsterdam
Netherlands
London
(Bruxelles)
Belgium
Berlin
(Warsaw)
Warszawa
Poland
Kyiv
Київ
Ukraine
Germany
(Prague)
Praha
St Peter Port
Jersey
Luxembourg
Paris
Luxembourg
Slovakia
Moldova
Chişinău
(Vienna)
Wien
Budapest
Austria
Romania
Vaduz
Bern
France
Ljubljana
Beograd
Sarajevo
San Marino
Monaco
Montenegro
Podgorica
Skopje
Italy
Porto
Andorra
Città del Vaticano
Roma
Tiranë
Greece
Portugal
Spain
Lisboa
(Lisbon)
Cádiz
Gibraltar
Algiers
Tunis
Malta
Mediterranean Sea
©2009 Geocentre Consulting

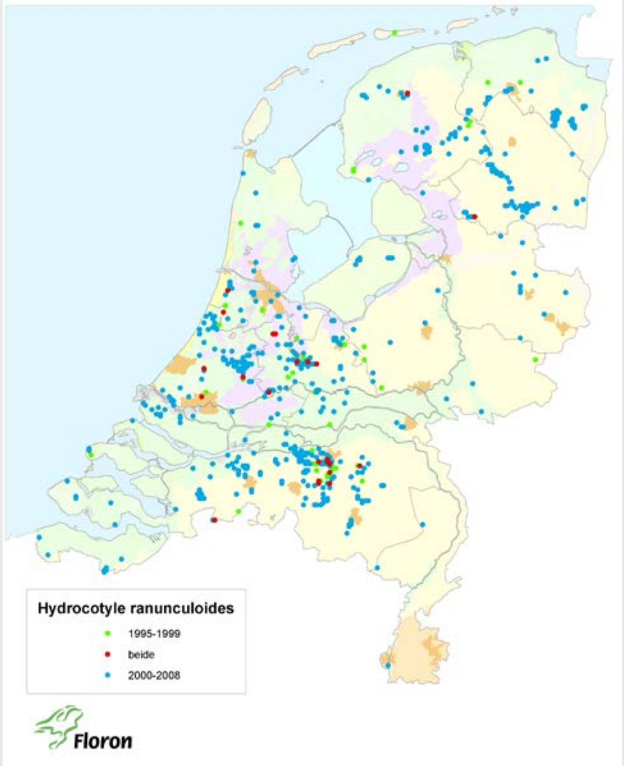
2 Appendix 2

Maps of occurrence and spread in countries of the EPPO region

North- America
Map available at http://plants.usda.gov/java/profile?symbol=HYRA
More detail data at the state scale are available on the website.



The Netherlands



Distribution of *Hydrocotyle ranunculoides* for the Netherlands for the period from 1995 till 2008.

In the Netherlands, since 1995 when it was first recorded as invasive for the Netherlands, it is now present in all Provinces, and only absent from the Wadden Islands, separated by salt water from the mainland

<u>Germany</u>

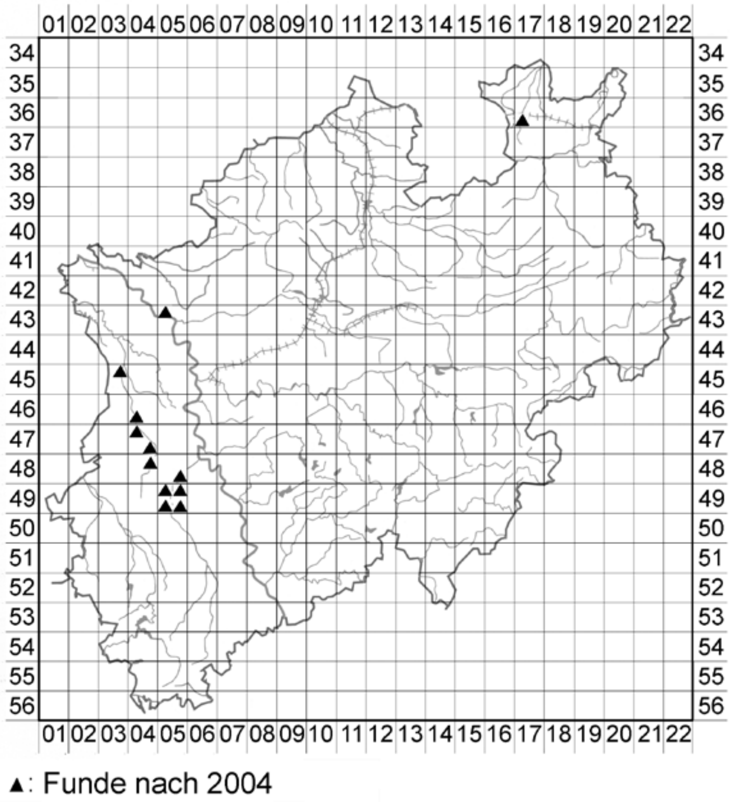
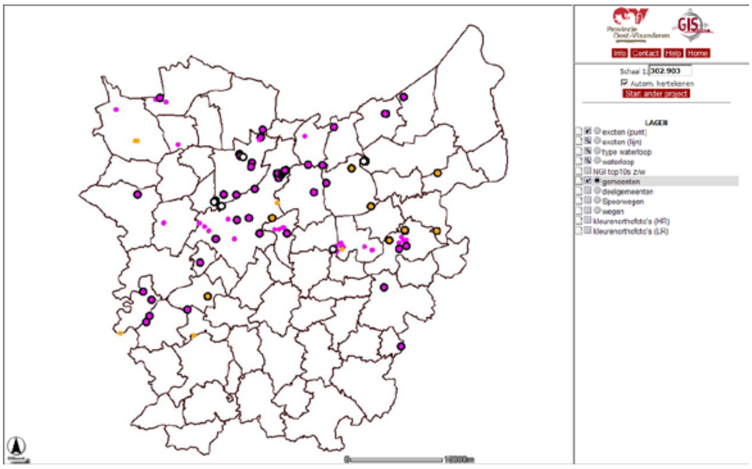


Figure: Known occurrences of *Hydrocotyle ranunculoides* in North Rhine-Westphalia (Germany) in 2008, species was present in all lower parts of the rivers Erft (a tributary of the River Rhine) and Niers (Hussner 2008).

Belgium

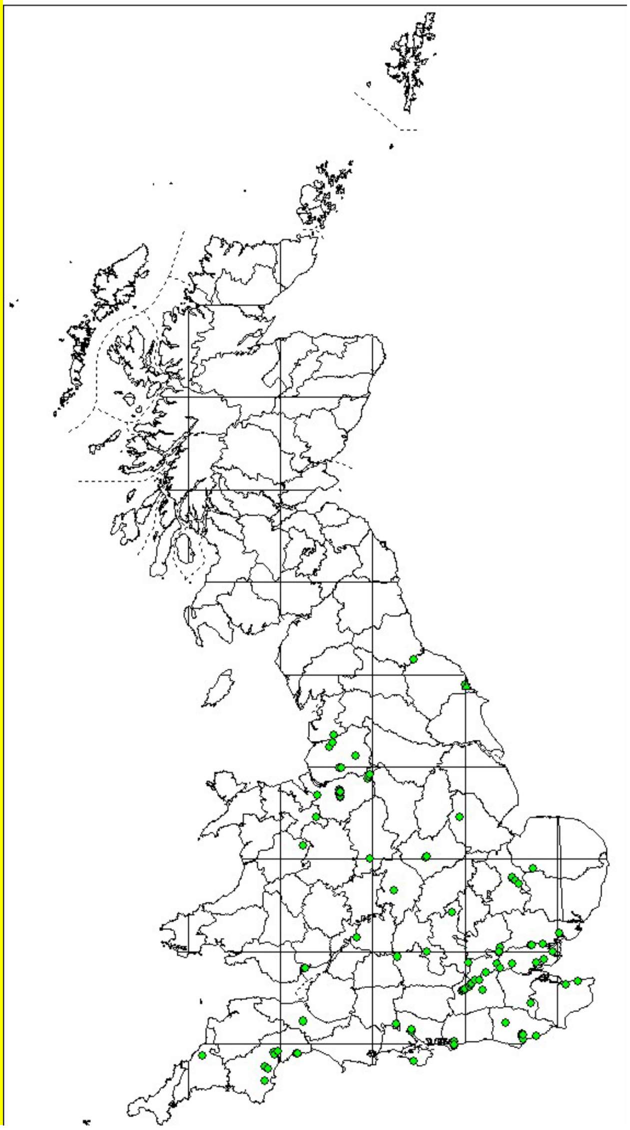
For the whole Belgium
Map available at http://ias.biodiversity.be/ias/species/show/63

Atlas Flora of Flanders, available at http://www.gisoost.be/exoten/ (go to “volledig gebied”)



Pink points represent localities where *Hydrocotyle ranunculoides* is present. Orange points represent localities where *Myriophyllum aquaticum* is present. Points circled in black represent unmanaged localities.

<u>The UK</u>



Dots correspond to sites where *H. ranunculoides* is present.

5 **Appendix 3**

6

7 Climatic prediction on *Hydrocotyle ranunculoides*

The CLIMEX model is a computer programme aiming at predicting the potential geographical distribution of an organism considering its climatic requirements. It is based on the hypothesis that climate is an essential factor for the establishment of a species in a country.

For *Hydrocotyle ranunculoides*, a compare location analysis has been undertaken.

1. Geographical distribution of the species and parameters

The distribution of *Hydrocotyle ranunculoides* was assembled from several sources: Global Biodiversity Information Facility (GBIF): http://www.gbif.org/, USDA http://plants.usda.gov, ForaWeb (ttp://www.floraweb.de/), etc. Distribution data in the EPPO region have been taken from question 7 and from distribution maps provided by individual countries (see Appendix 2).

Hydrocotyle ranunculoides is native from the American continent. Its northern boundary is reached in the USA and Canada (British Columbia, Quebec) where it becomes very rare. In the USA, the plant is only present in a belt including the southern states (except New Mexico), and north, the plant is mainly found along the east and west coasts. Its southern range is more obscure but it seems present in the whole tropical America (Martin & Hutchins, 1981), in almost all south American countries (Argentina, Bolivia, Brazil, Chile, Columbia, Ecuador, Paraguay, Peru, Uruguay). In the south, the species was recorded at latitude 35.34.030 and longitude 058.03.512 in the province of Buenos Aires (Newman, unpublished) but is known to go 200 km further south (J Newman, pers. comm., 2009).

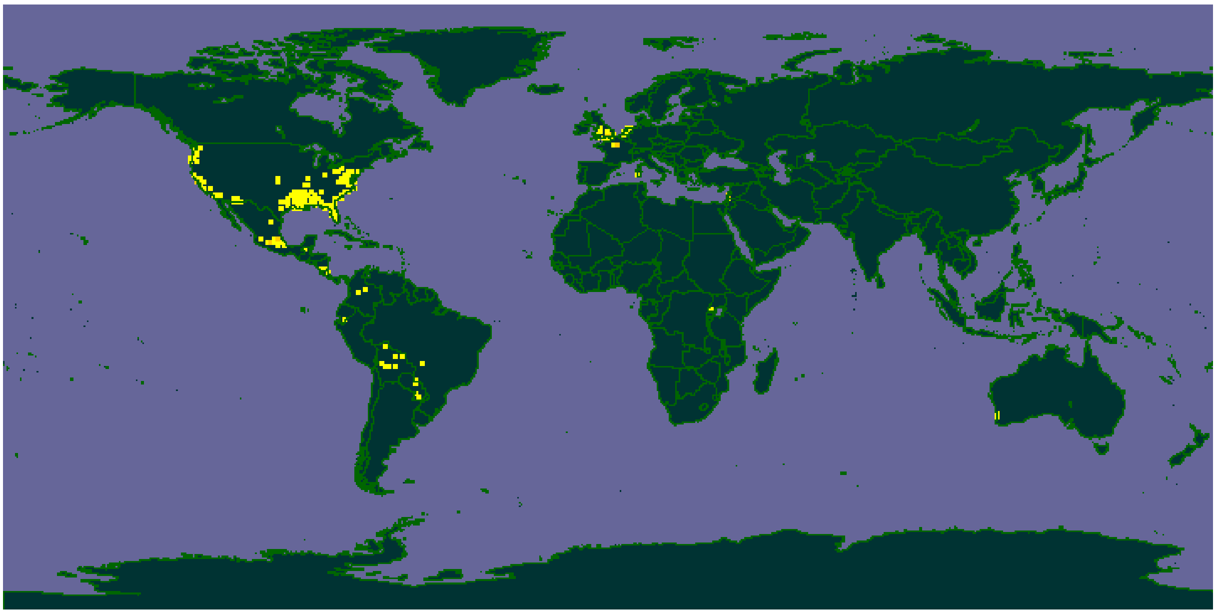


Fig. 1. World distribution of *Hydrocotyle ranunculoides* according to the GBIF.
This map is incomplete for data in Africa, in South America and in Europe.

Phenology

In Illinois (USA), the blooming period of *H. ranunculoides* occurs during the summer or early fall. In Australia, *H. ranunculoides* flowers in spring (September, October, November).

In Europe, plants grow slowly in spring and form small, up to 10 cm² large leaves. The plants flower and produce fruits between May and October. The maximal growth rate is reached during June and July (Hussner & Lösch, 2007)

Influence of climatic factors on distribution

Temperature

The species is reported to tolerate a wide range of temperatures, from 0°C up to 30°C of water temperatures (Kasselmann, 1995).

According to the climate calculations of Ackerly lab California Flora Climate Database (http://loarie.stanford.edu/calflora/index.php) which are based on mean climatic data where the species is recorded, the following information are available for temperatures:

- mean daily air temperature (Annual based on 18-year mean) = 14.31 °C
- minimum daily air temperature (Annual based on 18-year mean) = 1.58 °C
- maximum daily air temperature (Annual based on 18-year mean) = 30.82 °C

According to Hussner & Lösch (2007), optimal CO_2 exchange is between 25 and 32°C, meaning that optimal growth would occur at these temperatures; at 35°C, the gas exchanges dropped. Its presence in tropical America, in Africa and western Asia (Lebanon, Syria) shows however that *H. ranunculoides* could be present at higher temperatures.

Rainfall

According to the same Ackerly lab California Flora Climate Database, *H. ranunculoides* occurs in sites with 779.85 mm precipitation per year.

Fitting parameters

The parameters used in the CLIMEX model for *H. ranunculoides* are summarized in Fig 2. The role and meaning of these parameters are fully described in Sutherst *et al*. (2004), and their values are discussed below. It should be noted that the meteorological data used in this model represent long-term monthly averages, not daily values. This means that it is not possible to compare directly values derived using the model with instantaneous values derived through direct observations. This applies mostly to parameters relating to maximum and minimum temperatures.

The climatic requirements of *H. ranunculoides* were derived by fitting the predicted distribution to the known native distribution in America

		Edit Comments...		Copy to Clipboard	
☐ Moisture Index					
☑ Temperature Index					
DV0	DV1	DV2	DV3		
1	24	33	35		
☐ Light Index					
☐ Diapause Index					
☑ Cold Stress					
TTCS	THCS	DTCS	DHCS	TTCSA	THCSA
1	-0.001	9	-0.001	0	0
☑ Heat Stress					
TTHS	THHS	DTHS	DHHS		
35	0.001	0	0		
☐ Dry Stress					
☐ Wet Stress					
☐ Cold-Dry Stress					
☐ Cold-Wet Stress					
☐ Hot-Dry Stress					
☐ Hot-Wet Stress					
Day-degree accumulation above DV0					
DV0	DV3	MTS			
1	35	7			
Day-degree accumulation above DVCS					
DVCS	*DV4	MTS			
1	100	7			
Day-degree accumulation above DVHS					
DVHS	*DV4	MTS			
35	100	7			
Degree-days per Generation					
PDD					
0					

Fig. 2. Parameters used for *Hydrocotyle ranunculoides*

Moisture index

Moisture index is not considered since the plant is aquatic.

Temperature index
Based on the data described above, the minimum lethal temperature is set at DV0= 1°C, the maximum lethal temperature is set at DV3=35°C and optimal growth are set between DV1=24 and DV2=33°C. We then modify the parameters to better fit the potential distribution to the known distribution in America.

Stresses
Wet stress is not considered since the species is aquatic. The main stresses may be the *cold stress* which seems to limit the species in its northern range and to a lesser extent the *dry stress* which might limit the presence of its preferred habitats (for example in New Mexico).

Cold stress TTCS
As the plant is known to survive to 51 consecutive days of frost (Ackerly lab California Flora Climate Database), and to tolerate temperatures from 0 to 35°C, we set TTCS at 1°C and we supposed that the cold stress accumulates moderately slowly so the rate (THCS) was set at -0.001 (compared to *Eichhornia crassipes* for which it has been set at –0.01).

Cold stress DTCS
Additionally to be sensitive to a cold stress, the species might be sensitive to the fact that temperatures are not high enough to allow it to photosynthesise enough to offset minimum respiration demands. The parameters are therefore set (separately from the cold stress index) to 9 for DTCS. This parameter is set upon with an accumulation rate of -0.001 (DHCS) since the species is supposed to accumulate this stress slowly.

Heat stress
The plant is tolerant to temperatures of at least 30°C (Kasselmann, 1995). The plant is present in Lebanon, Syria or Yemen where temperatures are very high, the heat stress threshold was therefore set to 35°C. It is assumed that the stress accumulates quite moderately and the rate was set to 0.001 (THHS).

Dry stress
Dry stress is not considered as the species is aquatic.

2. **Climatic prediction in the native range**

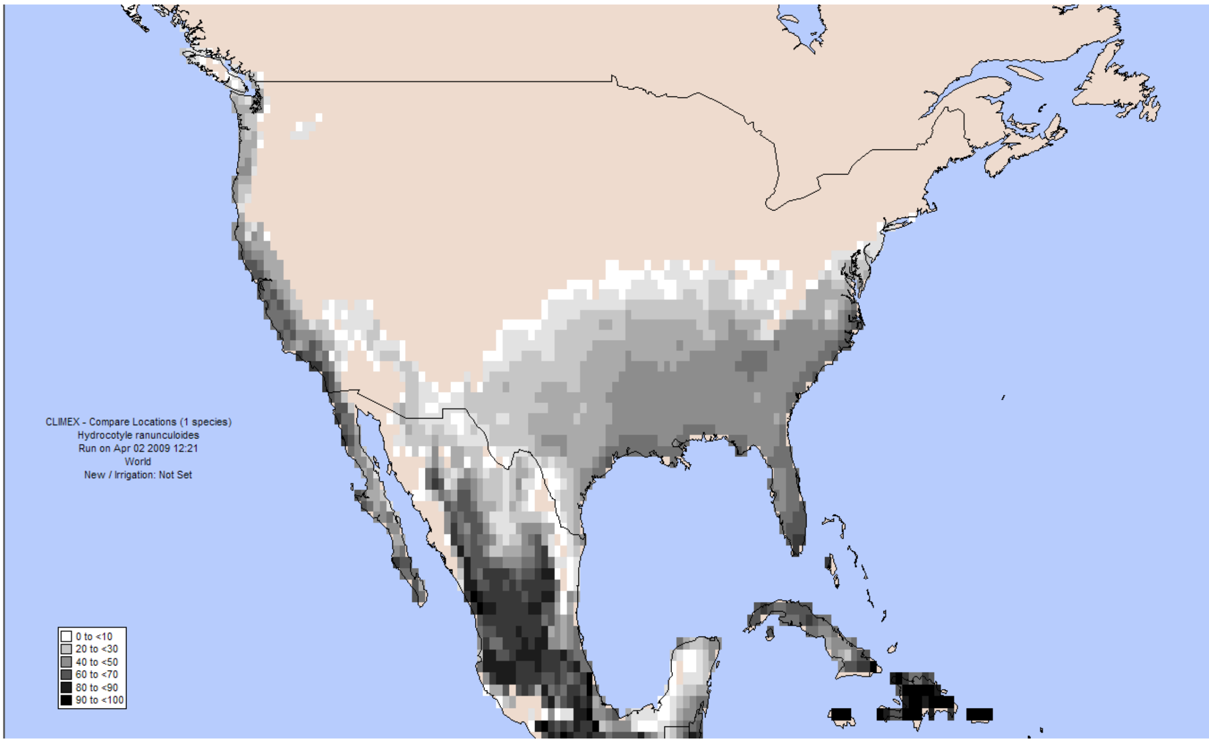


Fig. 3. Potential distribution of *Hydrocotyle ranunculoides* in North America

The fitting parameters provide a distribution into North-America very close to the current distribution of the species (see appendix 2 for the distribution of the species in North America). The West and east coasts are suitable for the species, as well as the southern part of the State.

3. Climatic prediction for the world

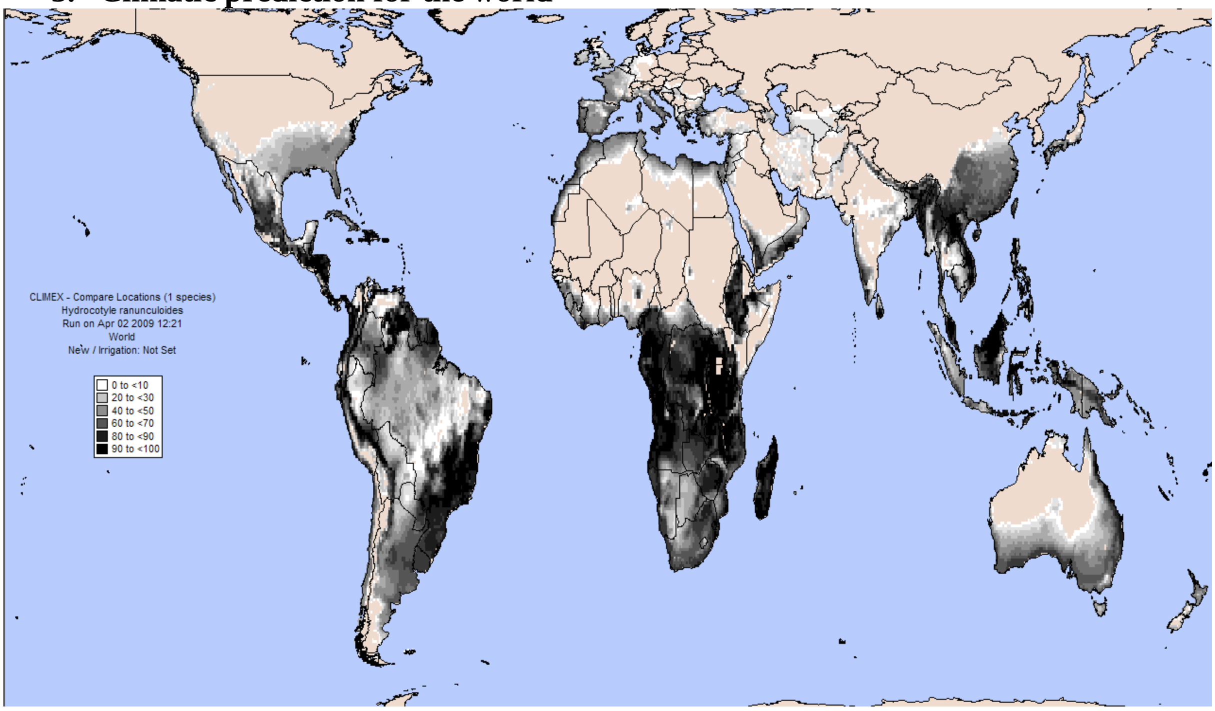


Fig. 4. Potential distribution of *Hydrocotyle ranunculoides* in the world.

The world distribution fits with known occurrences of the species.

4. Climatic prediction for the EPPO region

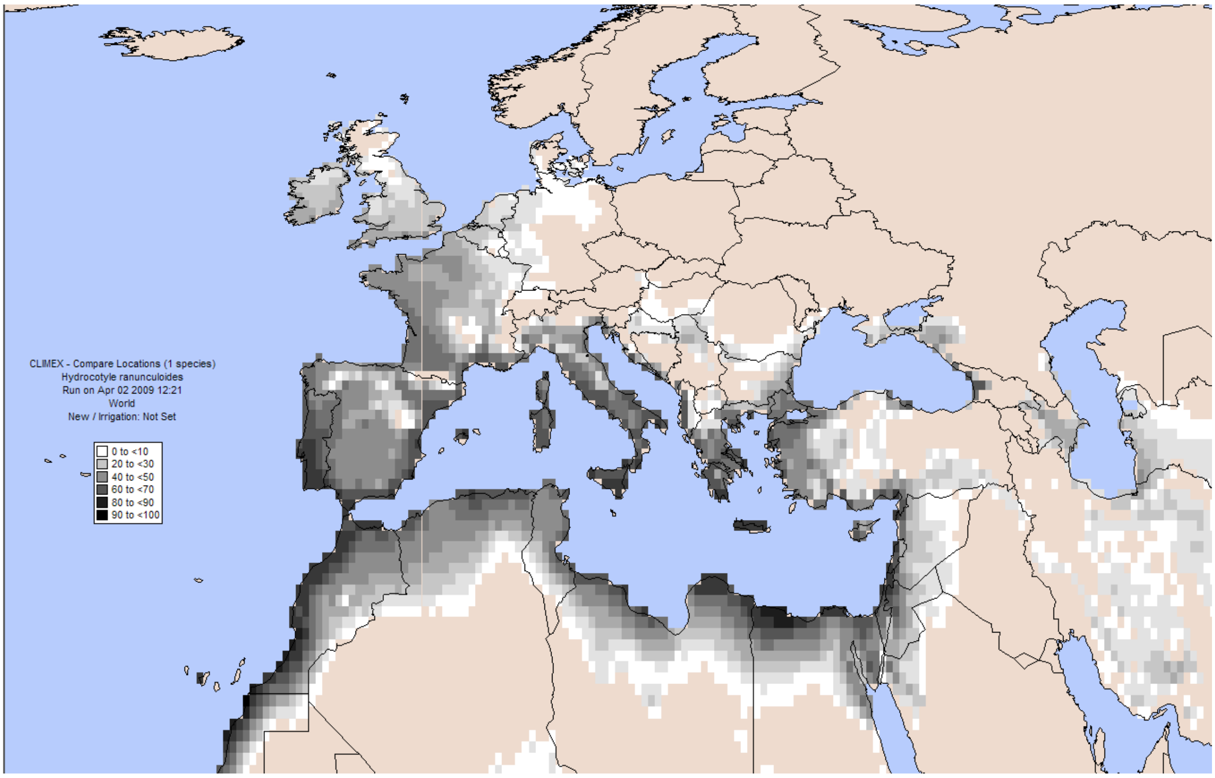


Fig. 5. Potential distribution of *Hydrocotyle ranunculoides* in Europe.

According to the Climex simulation, the Atlantic and Mediterranean areas of the EPPO region that are characterized by mild winters are the most at risk.

The countries at risk are: Albania, Algeria, Austria, Azerbaijan, Belgium, Bosnia & Herzegovina, Bulgaria, Croatia, Cyprus, Denmark, France (including Corsica), Greece, Ireland, Israel, Italy (including Sardinia), Jordan, Germany (mostly western part), Hungary, Moldavia, Morocco, Portugal, Romania, Russia (Black Sea), Serbia, Slovenia, Spain, Switzerland, Tunisia, Ukraine, the United Kingdom, Republic of Macedonia, Romania, Turkey.

Nevertheless, so far, the species has expressed invasiveness in North-Western EPPO countries (Belgium, the Netherlands, United Kingdom,), while the areas which seem to be the most suitable are the Mediterranean and the Atlantic areas. This may be due to other elements such as the use of the plant and the eutrophication of waters.

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Kasselmann C (1995) Aquarienpflanzen. Egen Ulmer GMBH & Co., Stuttgart. 472 pp. (In German)

Appendix 4

Pictures of invasion

Hydrocotyle ranunculoides as a contaminant on ornamental plants of *H. vulgaris* produced the Netherlands. Picture: J van Valkenburg

Invasion of a stream by *H. ranunculoides* in the UK. Picture: J Newman

Removal of *H. ranunculoides* in the UK. Picture: J Newman

Mechanical removal of *H. ranunculoides* in the U. Picture: J. ewman