The

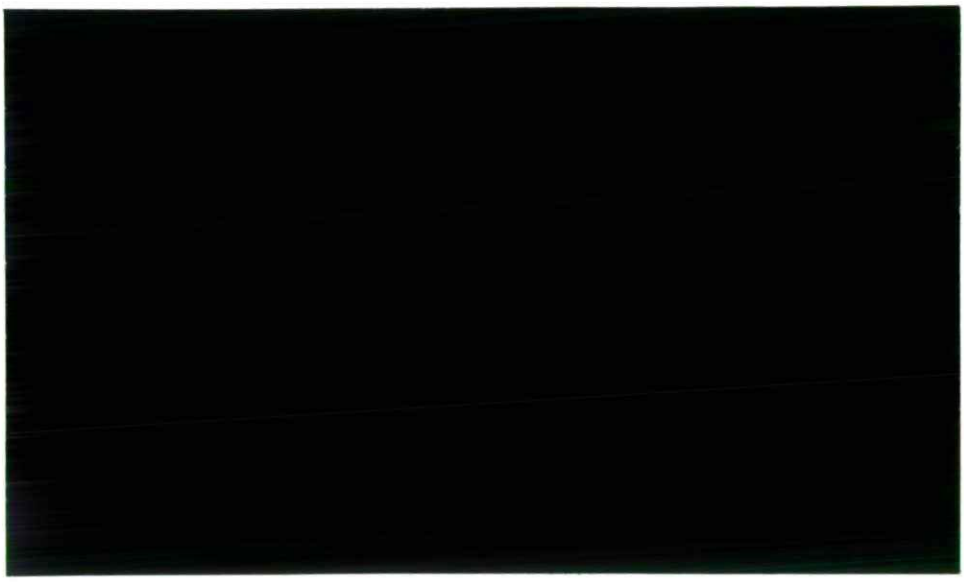
Centre for Ecology & Hydrology

Centre for
Ecology & Hydrology
NATURAL ENVIRONMENT RESEARCH COUNCIL

Formerly the Institutes of Hydrology,
Terrestrial Ecology, Freshwater Ecology,
and Virology and Environmental Microbiology

The **Centre for Ecology and Hydrology** is one of the Centres and Surveys of the Natural Environment Research Council (NERC). It was established in 1994 by the grouping together of four NERC Institutes, the Institute of Hydrology (IH), the Institute of Terrestrial Ecology (ITE), the Institute of Freshwater Ecology (IFE) and the Institute of Virology and Environmental Microbiology (IVEM). In 2000, the four component institutes were merged into a single research organisation.

Institute of Hydrology | Institute of Terrestrial Ecology | Institute of Freshwater Ecology | Institute of Virology and Environmental Microbiology



The CEH mission

- To advance the sciences of ecology, environmental microbiology (including virology) and hydrology through high-quality and internationally recognised research leading to a better understanding and quantification of the physical, chemical and biological processes relating to land and freshwater and living organisms within these environments.

- To investigate, through monitoring and modelling, natural changes in the ecological, microbiological and hydrological environments, to assess both past and future changes and to predict man's impact on these environments.

- To secure, expand and provide ecologically and hydrologically relevant data to further scientific research and provide the basis for advice on environmental conservation and sustainable development to governments and industry.

- To promote the use of the Centre's research facilities and data, to provide research training of the highest quality and to enhance the United Kingdom's research base, industrial competitiveness and quality of life.

Julien Brunet,
MST Chimie et Biologie Végétales.

Centre for Ecology & Hydrology
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RAPPORT DE STAGE
Stage execute du 22/04/02 au 28/08/02.

Effect of Chemical and Physical Environment on *Crassula helmsii* Spread

Maître de stage: H. Dawson

Pour: Université Claude Bernard, Lyon I

Le 28 Août 2002.

Index:

Introduction:

I) About the CEH

The **CEH** (**C**entre for **E**cology and **H**ydrology) is one of four Centre Surveys of the Natural Environment Research Council (NERC), the UK leading organisation researching national environmental issues of major importance. Thanks to the British Antarctic Survey, the Proudman Oceanographic Laboratory and the British Geological survey and the CEH, NERC provides long-term research in the environmental sciences gathering full range of atmospheric, earth, terrestrial and aquatic sciences to improve understanding of the natural environment behaviour and its resources (figure 1). The results of the researches are relevant to academia, government, industry and the public. Through communication they are intended to promote public awareness and understanding.

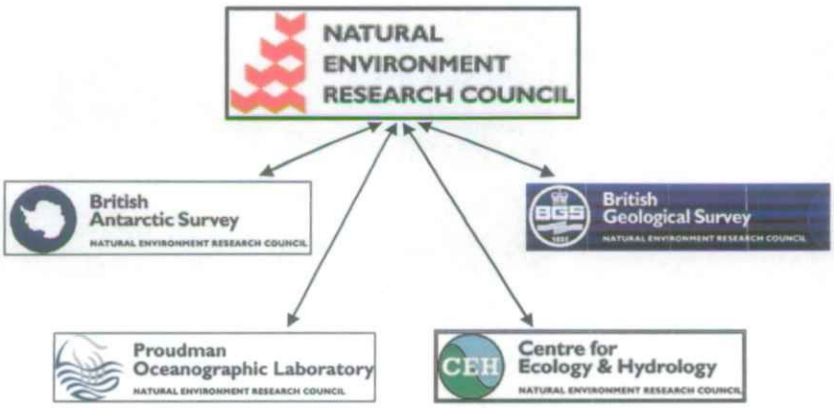

Figure 1 : organisation of the Natural Environment Research Concil, the main body in the UK for research, survey, and monitoring of the environment.

The CEH is the main UK body for research, survey and monitoring to describe and understand dynamics of terrestrial and freshwater environments. By carrying out long-term strategic and integrated research it predicts human impacts on the environment and generates potential solutions to improve quality of life.

The CEH is spread throughout the UK, in 9 sites:

- CEH of Banchory
- CEH of Edingbourg
- CEH of Windermere
- CEH of Merlwood
- CEH of Bangor
- CEH of Monks Wood
- CEH of Oxford
- CEH of Wallington
- **CEH of Dorset**

The CEH of Dorset comes from the merger of two former institutes :

♦ The Institute Freshwater Ecology's River Laboratory

Institute of Freshwater Ecology

♦ The Institute of Terrestrial Ecology's Furzebrook Research Station in.

Institute of Terrestrial Ecology

It carries out independent research in terrestrial, freshwater and estuarine environments and undertakes commissioned research.

All the investigations are ultimately designed to determine the processes dictating the distribution and dynamics of populations or communities of plants and animals.

In that aim, the CEH of Dorset is divide in three sections:

- Chemical and Molecular Ecology
- Vertebrate Population Ecology
- Biodiversity and Conservation Management

Each of these sections comprises several groups of research. The Biodiversity and management section has got four of groups :

- The Myrmecophily Research Group
- The River Communities Research Group
- The Conservation Management Research Group
- The Aquatic Botany Research Group, where I have carried out my investigation in the framework of this internship

A recent reorganisation has placed the Aquatic Botany group in the new River Ecology section.

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II) project

Biological invasions are large-scale phenomena of widespread importance, which represent one of the major threats to European biodiversity. In recent years many introduced plant species have caused problems for nature conservation.

Some of these species have a naturally vigorous growth strategy such as *Crassula helmsii* (T. Kirk), which show extreme competitiveness and are now predominant in many sites. This plant exhibits distinctly a fast-growing strategy invading the British wetland habitats such as ponds, lakes, gravel pits and other static water-bodies. Because of this, it has been the focus of much research regarding its impact.

Because of its abundance of water bodies, the New Forest (Hampshire) is a very sensitive area to *C. helmsii* invasion. Since the end of the 50's, numerous ponds and streams have been recorded as invaded. This area is a good model in the study of features which can be involved in its spread.

The aim of this investigation is to define the range of habitats suitable for this alien and consequently to estimate the limits of its invasion.

During this investigation, different wetlands with and without *Crassula helmsii* have been visited. Physical features, which may relate to a potential for invasion have been recorded.

Secondly, surveyed sites have been sampled and the water composition analysed. This is intended to demonstrate its chemical and trophic requirements.

After a brief introduction describing *C.helmsii* and the history of its invasion in the United Kingdom, the process and the results of this study will be displayed.

Bibliographic introduction:

I) What is a biological invasion?

Biological invasions represent one of the major threats to European biodiversity. They are generally the intended or unintended consequence of decisions involving the use of exotic species in production and consumption, changes to and fragmentation of habitat, or movement of goods and people (Mack et al. 2000).

An introduced species can change the look and make-up of an entire system, changing species composition, decreasing rare species, and even changing or degrading the normal functioning of the system.

In each habitat species have evolved together and have established a balance with their ability to live in this place. Hence introduction of new species into habitats where they are not native can result in biodiversity disruption.

Not every species that is introduced into a new environment survives and establishes self-maintaining populations. The terms "invasive" are generally used for those alien species that out-compete and displace native species. Because of the lack of natural controls such as predators, diseases, or competition with other organisms, they can become harmful outside their native range and affect the entire ecosystem.

Nevertheless, ecosystems vary in their natural susceptibility to invasion. Deserts, semi-deserts, tropical dry forests and woodlands, arctic systems, and pelagic marine systems appear to be least susceptible, while mixed island systems, and lake, river, and near-shore marine systems appear to be most susceptible (Heywood 1995).

Similarly, systems with low natural diversity (especially if they are without existing predators or competitors) appear to be more susceptible than systems with high natural diversity (Rejmanek 1989). But susceptibility also depends on land use. Habitat fragmentation, habitat conversion, and agricultural disturbance have all been blamed for increasing susceptibility to invasion (Williamson 1996, 1999).

Plants are the most commonly seen non-native organisms. Numerous exotic plants, many introduced in Europe little more than 200 years ago, have become successfully established over large areas. Furthermore, in many natural sites, invasive plants have smothered and crowded out native plants.

Most of these plants have been introduced by humans for agricultural and horticultural benefits or for garden trade. Hence non-native plants may also be transported unintentionally with other plants or in soil. Seeds (or sometimes roots or branches that can sprout) can also be the origin of an invasion. Many of the most problematic aliens are fast growing, vigorous competitors which are easily propagated (Starfinger *et al.* 1998). This plants can became real problem for environment in the case of out-competition with the indigenous species.

II) Crassula helmsii

a) Morphology and main features

Crassula helmsii (T. Kirk) Cockayne originated in New Zealand and Australian growing on damp soil near or in water.

It is identifiable by its short dense and mid-yellowish green succulent-like appearance. Pairs of unstalked opposite leaves (4-24mm) are borne on rigid stems often with a creeping habit and capable of rooting at the nodes. The leaves bases are joined giving a 1mm collar, which is a distinctive characteristic.

Figure 2: stem and flower of *Crassula helmsii*

The plant seems to be confined to areas, which have levels of precipitation of between 0.1 -0.55 m in summer (Nov -April) and 0.2 -3 m in winter (May -Oct). Its daily mean temperature requirements are restricted to a winter range of 0 – -15°C including extended periods under snow and a summer range of 20 – 25°C. It can tolerate drying for extended periods and has been shown to survive temperatures below freezing (Kirby, 1965). The climate in the British Isles falls well within these limits, providing the plant with optimal growing conditions. This plant is now invading aquatic ecosystems across the Great Britain and shows an extreme resistance to all attempts at eradication. (Leach and Dawson, 1999).

In Britain *C. helmsii* exhibits three growth forms (Dawson & Warman, 1987):

• When growing fully submerged in deep water (>0.5m), it is well rooted at the base, reaching a maximum length of 1.3 m. Plants growing in deep water have particularly thin, barely succulent leaves which are sparsely distributed along the upper stems. Inter-nodal lengths are often very long (20-25 mm) in the lower part of the plant but are much shorter (5-12mm) towards the apical end.

▪ In shallow water (<0.5m), growth becomes dense with much more branching. The stems become emergent during summer, either by the growth of the plant, or a decrease in water level. In these fluctuating conditions the plant forms a very dense mat of sprawling stems from which many side shoots with succulent leaves arise. This produces a turf, which is very effective in inhibiting the growth of other species. Culture trials in static water suggest that the change from production of the submerged to emergent leaves and *vice versa* is rapid. In addition a shallow-water growth form can occasionally be seen extending into deeper water where, although rooted along the bank, its sprawling stems grow out across the surface to form an anchored floating mass.

Figure 3: growth shape of *C. helmsii.* On the right, a submerged stem. On the left, an emergent shoot.

▪ In seasonal ponds and pond margins, *C. helmsii* is leafier and more compact than shallow water forms. Branching is more frequent as inter-nodal distance decreases, allowing the plant to form a well-anchored turf. Growth on damp mud at the margins of ponds and lakes is restricted to dense mats, which is considered more typical of its growth in its native range.

Toelken (1981) lists *Crassula helmsii* as an annual but there are indications from cultures and field observations that annual die-off and re-growth from seed is not the common strategy for this plant in Britain. Established plants flower annually. Nevertheless the viability of the seed in the British conditions is uncertain. Germination of this introduced alien has never been observed, although seed formation occurs (CEH Dorset, unpublished document).

Its method of growing means it can successfully propagate from very small fragments called "propagules". Just one centimetre of the weed is capable of rooting and growing, meaning there is no realistic way of mechanically removing it from the water. These fragments can be carried between waters, through water on birds or on wet angling equipment and clothing. In the region of "New forest" this spread may also be due to the ponies, which are living in the wild.

Figure 4: Propagule of *Crassula helmsii* giving rise to a new sprig

Where New *Crassula helmsii* appears it can rapidly spread over the shallow areas of water. It can cover and choke the entire surface to the exclusion of any other plants, leading to the loss of native aquatic flora and the natural habitats for the fauna. A previous survey undertaken by D. McMahon (Student in internship at the CEH) showed that the plant could have a suppressive effect on surrounding epiphytic and planktonic algae more than the effect of shade (CEH Dorset, unpublished results).

Figure 5: pond chocked by C.helsii (Holmsley Natural Reserve, le 16/5/02)

b) History of invasion

Living material of *Crassula helmsii* was originally sold under the name *Tillaea recurva* by Perry's Hardy Plant Farm, Enfield, Middlesex from 1927. It was sold as a submerged oxygenating aquatic (Laundon 1961). This specie was only available from this one supplier until the 1970's but it is now available from many aquatic suppliers (Dawson & Warman, 1987).

It seems likely however that there has been only one introduction. Examination of 11 enzymes expression using starch-gel electrophoresis from about 40 sites was not conclusive but indicate that there is no morphological variation throughout Britain when compared with the range in plants collected across Australia (F. H. Dawson, 1990).

Also DNA analysis of microsatellites indicates Australia as the origin of all British *C.helmsii* (CEH Dorset, unpublished).

The first collection of *Crassula helmsii* (T. Kirk) Cockayne in a naturalised situation was made in Greenstead, Essex (South East of England) in 1956 (Laundon, 1961) and had led to the designation of *C. helmsii* as an established alien in Britain, where it is currently known by the names of "New Zealand Pigmyweed" or "Australian Swamp Stonecrop" or simply "Crassula".

The plant is now widely distributed throughout the British Isles (Figure 1 distribution map), at a range of altitudes from sea level to 278 m.

Its invasion was monitored relatively early by the Institute of Freshwater Ecology (IFE now CEH), which have recorded its spread throughout the British Isles. After the initial introduction into a new area *C. helmsii* has many modes of dispersal including, transfer from pond to pond by anglers, wildfowl, ponies (e.g. in the New Forest), children pond dipping and even botanists survey equipment and their boots. These modes of distribution are possible because of the regeneration potential of even the smallest stem fragment.

By 1995 the number of recorded sites reached 600 and records were predicted to reach 1000 by the year 2000. Its actual distribution is probably far greater. Thus one recent estimate suggests that over 10,000 sites may have been colonised.

Crassula helmsii was first noticed in North Ireland in 1984, in a pool at Gosford and at the flooded Glastry clay-pits in Co.

Because the plant is now become to common, the new sites of invasion are not any more recorded.

Figure 6: Distribution of *Crassula helmsii* in the British Isles in 1998

Methods:

I) Study area : The New Forest

Historically, the "New Forest" is an English medieval deer hunting area created in 1079 by William the Conqueror and it is still largely in the possession of the Crown. This area is a nationally important environment of woodland, pasture, heaths and bogs and the remains of 17th, 18th & 19th century coppices and timber plantations. It is grazed by the ponies and cattle of the local people or 'Commoners' who breed them in semi-wild conditions. This area is also characterised by an abundance of water bodies such as ponds and streams.

Currently the Forest is recognised and controlled as an area of special conservation interest by main of the special conservation interest by bodies including English Nature, Forest Enterprise and Hampshire Wildlife Trust.

The previous surveys about *Crassula helmsii* in the New Forest undertake by the Hampshire Wildlife Trust

In March and April 2000, an assessment of the spread of *Crassula helmsii* in the New Forest was undertaken by the "Hampshire Wildlife Trust"(HWT), the leading wildlife charity concerned with conservation of wildlife in Hampshire. That survey is still the only source of data available for the CEH for the study of this invasion.

During that investigation, 191 individual sites were surveyed and entered into the database.

Of these 191 sites, 75 were labelled as invaded by *C. helmsii*; see the table below.

	Nb of site total	% of the sites recorded	Nb. of infected sites	% infected
Pond*	163	*85.3403*	59	*36.1963*
Ditch	9	*4.71204*	5	*55.5556*
Stream	7	*3.66492*	4	*57.1429*
Marsh	2	*1.04712*	1	*50*
Other	10	*5.2356*	6	*60*
Total	191	*100*	75	*39.267*

Table 1: Results of the survey undertaken by the HWT in 2000

II) Fieldwork

One part of the time of this internship has been devoted to fieldwork of collecting relevant data about the spread of *C.helmsii*. It consists of the observation and sampling of sites.

65 of the sites formerly surveyed by the HWT have been chosen for study as representatives of sites invaded and not by *C.helmsii*. The map references and description of sites in the 2000 database (HWT survey) were used for location of the sites. GPS system should enhance the ease and accuracy of location in the future.

Some of the area listed in the 2000 database includes water bodies or part of complexes of wetlands with different characteristics. A number of separate samples were taken at different points at such sites. In the present study, individual sites on the 2000 database were split into several sampling locations.

a) Observation

On arrival at a site, a sketch map of the site is drawn recording morphological features, the area where the plant occurs, the shaded zones and inputs or outputs of streams associated with the sites. This data may be useful in understanding the origin of the invasion and the preferred place of growth of *C.hemsii*. Other relevant data such as the turbidity of the water, type of surrounding vegetation or substratum can also be specified.

Walking along the banks is generally necessary to give a good view of the area and to establish extent of *Crassula helmsii* invasion.

b) Sampling

The main part of the time spent in the field is devoted to sampling the site. All the samples must be well referenced and dated.

• *Samples of water*: The water of each site must be sampled, whether they are invaded or not by *C.helmsii*. Water samples filtered through 45µm pores filters to remove the particles and microorganisms. Such particles have potential to damage analysis equipment as well as influence the amount of ions dissolved in the water by sorption or assimilation.

The bottles used to contain the water samples are previously steeped overnight in lightly acidified water (0.05M HNO_3), to remove the ions adsorbed. The water are then rinsed in deionised water.

These samples must be made with carefulness not to contaminate or dissolve the water of the site. All the equipment used (bottle, syringe, filter) has to be rinsed twice with distilled water and then by the water of the site.

These samples can be conserved few days in a cold room.

• *Samples of vegetal material*: In order to secure the identification of *Crassula helmsii* on the fieldwork and to improve its recognition for further surveys, all the specimens of plants, which are supposed to be *Crassula helmsii* or which look like it must be collected. These plants will be identified subsequently. By this way the mistakes due to confusion of indigenous species with *C.helmsii* will be avoided.

III) Water analysis

In order to characterise chemically the surveyed sites, samples of water will be analysed. Several features, which have the most influences in the vegetation growth such as conductivity, pH, alkalinity and the concentrations of phosphorus, nitrogen and potassium, will be measured.

a) Conductivity

Conductivity is directly measured in the water by a calibrated probe (Ciba-Corning Checkmate 90). Conductivity gives indication of the total ion concentration in the water, including mineral ions or organic ions.

b) pH

pH is measured at with an accurate potentiometric pH meter. This measurement is done after the pH of the sample is stabilised. CO_2 accumulation due to the organic matter degradation during the storage can be responsible for a lower pH. After about 2 minutes agitation, the CO_2 concentration can be regarded as in-balance with the atmosphere, and then the pH can be noted.

c) Alkalinity

Alkalinity refers to the capability of water to neutralize acid. This is really an expression of buffering capacity. In water bodies the main buffering power is due to the carbonate-bicarbonate system ($CO_2/HCO3^-/CO3^{2-}$). The presence of calcium carbonate or other compounds such as magnesium carbonate contribute carbonate ions to the buffering.

The alkalinity is expressed in mili-equivalent.l^{-1} of $CaCO_3$ (m.eq.l^{-1}); calcium carbonate is the main form in the water.

1) CO_2 (gas) ⇆ CO_2 (aqueous) ($pK_h = 3.5$)

2) CO_2 (aqueous) + H_2O ⇆ H_2CO_2 (aqueous)

only a small fraction (ca. 0.2%) is actually converted to carbonic acid. Most of the CO_2 remains as solvated molecular CO_2.

3) H_2CO_2 (aqueous) ⇆ HCO_3^- (aqueous) + H^+ ($pK_{a1}=6.37$)

4) HCO_3^- (aqueous) ⇆ CO_3^{2-} (aqueous) + H^+ (pK_{a2}= 10.32)

5) Ca^{2+}+ CO_3^{2-} ⇆ $CaCO_3$ (aqueous) (pKs= 8.32)

The alkalinity is measured by titration with sulphuric acid. It is taken to an end-point reading of pH 4.5 ($pKa_{(CO_2/HCO_3)}$).

$$\frac{[HCO_3^-][H^+]}{[H_2CO_3]} = 10^{-6.38} = K_{a1}$$

A pH = 4.5 , $[H^+] = 10^{-4.5}$

$$\frac{[H_2CO_3]}{[HCO_3^-]} = \frac{10^{-4.5}}{10^{-6.38}} \approx 10^2$$

Consequently at pH = 4.5, about 99% of the inorganic carbon dissolved into the water is on the form H_2CO_3.

Alkalinity is measured by titration of 10.0ml filtered water sample. The amount of acid required to reach pH 4.5 and consequently to neutralize all the carbonate ion is expressed in millilitres. A N/100 (M/200) sulphuric acid solution is used as well as 1ml of acid used correspond with $1 m.eq.l^{-1}$. (CEH; SOP 39/05.11.97).

To express as $mg.l^{-1}$ $CaCO_3$, alkalinity =vol. used {ml} x 50

(M_{CaCO_3} = $100 g.mol^{-1}$, $1 m.eq.l^{-1} = 0.5 mol. l^{-1}$).

A 2mM Na_2CO_3 solution is prepared as quality control. 10ml of this solution are titrated to the pH 4.5 endpoint. The volume used must be 4.00 ± 0.01 ml.

These titrations are undertaken by semi-automatic equipment, which titrates and records the volume of acid used to reach the specified endpoint (pH 4.5).

d) Phosphorus

Phosphorus typically functions as the "growth-limiting" factor because it is usually present in very low concentrations. The natural scarcity of phosphorus can be explained by its adsorption to organic matter and soil particles. Consequently amount in the solution could vary very quickly during the storage of the samples. Accordingly, the whole quantity of phosphorus present in the sample, as organic forms as well as free mineral forms, will be measured.
This assessment of phosphorus amount will be undertaken by UV-Visible spectrophotometry. This analyse is carried out in three steps.

- First, the sample, or a solution of this sample, must be digested in order to release all the phosphorus as soluble form.

Phosphates present in organic and condensed inorganic forms (meta-, pyro- or other polyphosphates) must be converted to reactive orthophosphate before analysis. Digestion of the sample with acid and heat provides the conditions for hydrolysis of the condensed inorganic forms. Organic phosphates are converted to orthophosphate by heating with acid and persulfate.

10 ml are deducted at each sample and diluted to half in a small (40ml) autoclave-proof bottles. Then, 1 ml ± 0.1 ml of 1.01N sulfuric acid and 0.15g ±0.1g of potassium persulfate are added to this solution before autoclaving it. The digestion is undertaken at 121°C during 45 min.

The same must be done with the standards for the calibration of the method.

- The second step of this analyse consist of the formation of phospho-molybdate complex obtained by interaction between ammonium molybdate and orthophosphate in the presence of antimony. This complex is then reduced with ascorbic acid to form a blue compound which has a maximum absorbency at 880 nm. This method given in Murphy and Riley (1962) remains one of the most convenient methods, in terms of rapidity and ease, for the determination of orthophosphate in seawater.

The reagent is prepared as follows (CEH; SOP 48/19.04.01):

Dissolve 0.57±0.01g of Antimony potassium (+) tartrate ($KSbO.C_4H_4O_6$) in approximately 500ml of water. When dissolved, carefully add 45 ± 1ml of concentrate sulphuric acid (*sp. gr 1.84*) while stirring and cooling the solution.

Dissolve 8.52 ± 0.01g of sodium molybdate ($NaMoO.2H_2O$)in approximately 400ml of water. Mix the two solutions together and dilute in a calibrated flask. (this solution is stable for several month in the dark)

1.0 ± 0.1 ml of this reagent is required to allow the colour to develop.

- Finally, the concentration of phosphorus in all samples and standards is determined by measuring the absorbance at 880nm. This measurement must undertaken after the same time interval following addition of reagent: about 12 min.

e) Potassium

Potassium concentration is assessed by atomic absorption spectroscopy. A lamp with hollow cathode Na/K (sodium-potassium) is used to produce radiations corresponding to Potassium and Sodium spectrum lines. The absorption is measured at 766.5nm, one of the wavelengths of the potassium spectrum.

The concentration of potassium in water bodies generally belongs falls in the range 0.1 to $20mmol.l^{-1}$. Nevertheless for the highest concentrations the Beer-Lambert law is not suitable; the relation between concentration and absorbance is not linear. Therefore, the spectrometer is calibrated with a 0.05 to 2mg/L set of standard and each sample is diluted to 1/5.

The absorbance of each sample atomised in the flame is noted. By calculating the absorption coefficient (E‰) from the standards, the concentration of potassium is then assessed in each sample.

The presence of other alkali elements in the sample can reduce ionisation of potassium and thereby enhance analytical results.

These other alkali metals release their external electron in the flame and therefore limit potassium ionisation.

The ionisation suppressive effect of sodium is small if the ratio of Na to K is under 10. For a more stringent control of ionisation, the addition of caesium (more electropositive and consequently with a highest ionisation suppressive effect) is required. 0.2% (weight) of caesium is added in each solution before analysis.

f) Nitrogen

Nitrate and nitrite concentration have been measured for most of the samples. This measurement was carried out by the chemical and molecular ecology section by following the SOP: 53/ 03.03.00,displayed in appendix 1.

Because of a lack of time, the results of nitrogen analysis cannot have been analysed. Nevertheless these data will be displayed as appendix.

Results and Discussion:

During this part of the report, the results of this project will be displayed and analysed. By looking at the distribution of *Crassula helmsii* against different parameters, its preferential habitat will be found out.

In this analysis the relationships between the frequency of invasion and chemical or physical parameters are not strictly representative of the natural situation because the sites of surveyed have not been chosen randomly (see *methods*). Nevertheless they are useful for a comparative point of view.

I) Occurrence of *Crassula helmsii*

65 sites were visited during these investigations in the "New Forest" and 79 samples have been collected from the wetlands at these sites.

The following table shows the occurrence of *Crassula helmsii* observed in these sites and compare them with the results of the WHT.

				PRESENCE OF CRASSULA HELMSII		
Ref	Name of the Pond	Grid reference	Type of site	Results of the survey	Results of the HWT	Agreement
1	Wootton Heath Farm	SZ236987	Pond	YES	YES	YES
4	Little Wooton inclosure.	SZ228990	Pond	NO	YES	NO
5	Little Wooton inclosure (south - west edge).Pond B	SZ223988	Pond	NO	NO	YES
5	Little Wooton inclosure (south - west edge).Pond A	SZ223987	Pond	YES		
5	Little Wooton inclosure (south - west edge).Pond C	SZ223987	Marl Pit	NO		
5	Little Wooton inclosure (south - west edge).PonD D	SZ219987	Pond	NO		
9	Little Wooton inclosure edge (south).	SZ224985	Pond	NO	YES	NO
11	Little Wooton Inclosure South-west edge	SZ225985	Marl Pit	NO	YES	NO
12	Little Wooton inclosure South-east edge.	SZ226985	Pond	NO	NO	YES
13	Little Wooton inclosure South edge / iron gate.	SZ228985	Pond	NO	NO	YES
16	Hinchesiea Bog.	SU277005	Pond	NO	YES	NO
16	Hinchesiea Bog.	SU277006	Marsh	NO		
17	Durns town.	SZ287987	Pond	NO	YES	NO
19	Mallpit Oak, Setley.	SZ286997	Marl Pit	NO	YES	NO

				PRESENCE OF *CRASSULA HELMSII*		
Ref	Name of the Pond	Grid reference	Type of site	Results of the survey	Results of the HWT	Agreement
20	Mallpit Oak, Setley.	SZ286997	Marl Pit	YES	YES	YES
21	Mallpit Oak, Setley.	SZ286997	Marl Pit	YES	YES	YES
23	Latchmoor pond.	SU293004	Pond	YES	YES	YES
24	Balmor lawn (near hotel).	SU304033	Pond	NO	YES	NO
25	Balmor lawn (near hotel).	SU306035	Lake	NO	YES	NO
27	Black Knowl/ Butts lawn.	SU289027	Pond	NO	YES	NO
29	Furzy cottage.	SU287018	Pond	NO	YES	NO
30	Clay hill (Burley).	SU233024	Pond	NO	YES	NO
30	Clay hill (Burley).	SU233024	Pond	NO		
31	Holman's Bottom (Burley).	SU229024	Pond	YES	YES	YES
32	Cot bottom (Burley).	SU224025	Pond	NO	YES	NO
33	Pigsty Hill (Burley).	SU224026	Pond	YES	YES	YES
34	Burley Golf Course.	SU218026	Pond	NO	YES	NO
34	Burley Golf Course.	SU218027	Pond	NO		
37	Whitten Pond (Holmsley).	SU204012	Pond	NO	YES	NO
38	Holmsley Walk pond.	SU206013	Pond	NO	NO	YES
39	Church Moor/ Cranes Moor (Brown Lane).	SU199022	Pond	NO	NO	YES
39	Church Moor/ Cranes Moor (Brown Lane).	SU198020	Pond	YES		
39	Church Moor/ Cranes Moor (Brown Lane).	SU19905	Pond	NO		
40	Cranes Moor.	SU199011	Pond	NO	YES	NO
40	Cranes Moor.	SU199012	Pond	NO		
43	Bagnum Rough - Kingston Great Common.	SU183026	Stream	NO	YES	NO
44	Trenley Lawn drain (Brockenhurst).	SU281010	Ditch	NO	NO	YES
45	Headwaters - South weirs (Fuzzy Hill).	SU284013	Stream	NO	YES	NO
46	Thorney Hill Holms (Cross Ways).	SU201002	Pond	YES	YES	YES
46	Thorney Hill Holms (Cross Ways).	SU201002	Pond	YES		
46	Thorney Hill Holms (Cross Ways).	SU200004	Pond	NO		
49	North Weirs.	SU291021	Stream	NO	YES	NO
52	Wooton.	SZ246984	Pond	NO	YES	NO
53	Eastley Wooton.	SZ241988	Pond	NO	YES	NO

				PRESENCE OF *CRASSULA HELMSII*		
Ref	**Name of the Pond**	**Grid reference**	**Type of site**	**Results of the survey**	**Results of the HWT**	**Agreement**
54	Wooton Woods.	SZ244987	Pond	NO	NO	YES
56	Old house,Backley Plain.	SU220052	Stream	YES	YES	YES
56	Old house,Backley Plain.	SU220053	Stream	YES		
60	Wilverly Drain.	SU257015	Pond	NO	NO	YES
61	Hatchet Pond.	SU367014	Pond & Marl Pit	NO	NO	YES
69	Pond opposite the Royal Oak pub.	SU161119	Stream	YES	YES	YES
70	Opposite Gorley Tea rooms.	SU161118	Pond	YES	YES	YES
71	Pond by Mandy's Cattery.	SU160117	Pond	YES	YES	YES
72*	Pond north of Buddle Hill lane.	SU161116	Pond	YES	YES	YES
73	North of Jones farm.	SU161116	Pond	YES	YES	YES
74	Ponds immediately west of telephone box.	SU161115	Pond	NO	NO	YES
75	Gorley Triangle.	SU162105	Pond	NO	NO	YES
77	South of plantation cottage.	SU163099	Pond	YES	YES	YES
78	Pond next to EH1 village hall.	SU164097	Pond	YES	YES	YES
78	Pond next to EH1 village hall.	SU164098	ditch	YES		
82	Unnamed Anti-Aircraft Emplacement.	SU405049	Pond & Marl Pit	YES	YES	YES
83	Unnamed AA emplacement next toward Beaulieu from site 3.	SU405047	Pond	YES	YES	YES
84	Unnamed AA emplacement east of the road.	SU405047	Pond	YES	YES	YES
85	Unnamed roadside depression west of road.	SU406051	Pond	YES	YES	YES
88	Hartford Heath Pond.	SU401043	Pond	YES	YES	YES
89	Hill top Pond.	SU402031	Pond	YES	YES	YES
90	Moonhills pond.	SU408026	Pond	YES	YES	YES
96	Unnamed pond.	SU236098	Pond	NO	YES	NO
98	Near Fritham.	SU228147	Lake	YES	YES	YES
121	Bignell Pond.	SU286135	Marl Pit	NO	NO	YES
122	Longcross Pond.	SU246152	Pond	NO	NO	YES
123	Jamesmoor Pond.	SU246134	Pond	NO	NO	YES
128	Unnamed Pool.	SU272079	Pond	NO	NO	YES
131	Unnamed Gravel Pit.	SU221101	Ditch	NO	NO	YES
133	Slufters Pond.	SU223095	Pond	NO	NO	YES

				PRESENCE OF *CRASSULA HELMSII*		
Ref	**Name of the Pond**	**Grid reference**	**Type of site**	**Results of the survey**	**Results of the HWT**	**Agreement**
134	Slufters Seepage Pond.	SU222096	Pond	NO	NO	YES
135	Broomy Shade Pond.	SU211106	Pond	NO	NO	YES
135	Broomy Shade Pond.	SU211107	Pond	NO		
137	Cadnam's Pool.	SU229122	Pond & Brackish	NO	NO	YES
138	Green Pond.	SU226135	Pond	NO	NO	YES
139	Small Pond near Green Pond.	SU227136	Pond	NO	NO	YES
New site	Milkham enclosure.	SU217105	Pond	NO		

Table 2: Presence of *Crassula helmsii* at the different sites of survey

Of the 79 wetlands surveyed during this study, 27 are invaded by *Crassula helmsii.*

It seems to be clear that ponds are the best type of wetland bodies for *Crassula helmsii* growth. Nevertheless this invasive plant can also be found in other habitats, such as seasonal ponds (if these latter remain damp most of the year), in streams with very low flow, or ditches with no flow at all.

The results collected during this survey show a number of mismatches with the data collected by the HWT. The disagreements occurring with the HWT survey are shown in the last column. It concerns 21 sites of the 65, or about 32% of the surveyed sites.

It is more than likely that *C. helmsii* has colonised new sites since 2000 and consequently that sites previously listed as without *C. helmsii* are now infected. Nevertheless, in view of its fitness and its potential for spread, the hypothesis that *C.helmsii* has been eliminated from the sites previously labelled by he HWT as invaded, seems unlikely. This is the case for most of the mismatches between these two surveys. These mismatches can only be explained by confusion with indigenous species of plants. Moreover, the identification of *C.helmsii* can be all the more difficult because of the polymorphism it shows in different habitats.

At any site where the HWT survey recorded *C. helmsii* but the current survey did not find it, any plant which looks like and could have been confused with *C.helmsii* as been sampled and identified.

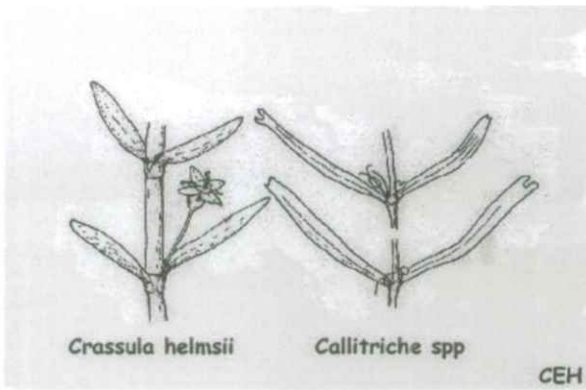
The following are examples of plants found in the sites mislabelled as invaded in the HWT surveys and which could at casual inspection be confused with *C. helmsii*:

⬩ First of all, ***Montia fontana*** is certainly the plant which looks most like *C.helmsii*. This plant grows in wet area in emerged conditions. It can be distinguished from *C. helmsii* by its curled stems and its more oval leaves.

figure 7: *Montia Fontana*

⬩ Several species of ***Callitriche*** could be confused with *C.helmsii*. Mismatches can be avoided by noting the obviously notched leaf tip of the *Callitrice sp*.

Figure 8: Two species of *Callitriche sp.* found in the sites: *C. stagnalis* and *C. autumnalis* (or *C.hermaphroditica*



Figures 9: Distinguishing Crassula helmsii from Callitriche. Callitiriche sp has notched leaf stips and no collar at the base of the leaves.

• ***Gallium sp*** can also be misidentified as *C. helmsii*. The plant shows a similar shape to the submerged form of *Crassula helmsii*. Nevertheless, *Gallium sp* bears four leaves in each nodes against only two for *C. helmsii*.

figure 10: *Galium saxatile*

• ***Sphagnum sp*** can also be mistaken for *C. helmsii* when viewed at a distance. Its shape and especially the colour of this marginal moss remind one of *C. helmsii* when this latter occurs on the banks.

Figure 11: Shagnum sp

• ***Polytrichum sp***, a terrestrial moss, bears long and massive erected leaves. Even if these two plants do not really look similar, they show the same growth habits in dense patches. Casual observation of stand of *Polytrichum sp.* at distance could result in confusion with patches of *C.helmsii*.

Figure 12: Polytrichum sp.

II) Study of the preferential habitat of Crassula helmsii

The water collected in each site has been analysed in order to assess several of its chemical characteristics. The water chemistry of wetland has a main influence in the plant growth. It is also a good reflection of the nature of the substrate in which the plants are rooted.

The pH, alkalinity and conductivity of each sample have been measured. The results are shown in the table below.

A) Basic chemistry

Ref	Name of the Pond	Presence of *C. helmsii*	Alkalinity (m.eq/L)	pH	Conductivity (µS)
1	Wootton Heath Farm	YES	1.58	7.59	286
4	Little Wooton inclosure.	NO	0.94	6.96	366
5	Little Wooton inclosure (south - west edge).	NO	3.21	8.31	368
5	Little Wooton inclosure edge (lower pond).	NO	4.13	7.98	508
5	Plain Heath (south)/ A35.	YES	2.27	7.92	282
5	Little Wooton inclosure edge (upper pond).	NO	2.87	7.86	363
9	Little Wooton inclosure edge (south)	NO	0.87	6.83	186.8
11	Little Wooton Inclosure South-west edge	NO	2.64	7.6	360
12	Little Wooton inclosure South-east edge.	NO	2.28	7.56	326
13	Little Wooton inclosure South edge / iron gate.	NO	0.2	6.03	168.7
16	Hincheslea Bog.	NO	0.22	6.43	119.1
16	Hincheslea Bog.	NO	0.43	6.58	178
17	Durns town.	NO	Dried out	Dried out	Dried out
19	Mallpit Oak, Setley.	NO	2.17	7.27	511
20	Mallpit Oak, Setley.	YES	1.3	6.8	244
21	Mallpit Oak, Setley.	YES	0.11	5.77	125.6
23	Latchmoor pond.	YES	0.12	6.3	117.1
24	Balmor lawn (near hotel).	NO	0.17	6.91	176.2

Ref	Name of the Pond	Presence of *C. helmsii*	Alkalinity (m.eq/L)	pH	Conductivity (µS)
25	Balmor lawn (near hotel).	NO	0.12	6.56	101.7
27	Black Knowl/ Butts lawn.	NO	2.77	7.67	385
29	Furzy cottage.	NO	0.82	7.2	134
30	Clay hill (Burley).	NO	0.04	5.12	45.8
31	Holman's Bottom (Burley).	YES	1.16	7.74	242
32	Cot bottom (Burley).	NO	0.93	7.68	200
33	Pigsty Hill (Burley).	YES	0.92	7.53	198
34	Burley Golf Course	NO	0.53	7.48	100.5
34	Burley Golf Course.	NO	0.09	6.22	56
37	Whitten Pond (Holmsley).	NO	0.05	5.07	127.5
38	Holmsley Walk pond	NO	0	4.1	136.4
39	Church Moor/ Cranes Moor (Brown Lane)	NO	0	4.36	77.6
39	Church Moor/ Cranes Moor (Brown Lane).	YES	0.08	5.91	152.3
39	Church Moor/ Cranes Moor (Brown Lane)	NO	0	4.32	73.7
40	Cranes Moor.	NO	0	4.1	93.8
40	Cranes Moor.	NO	0	4.03	70.5
43	Bagnum Rough - Kingston Great Common	NO	0.04	4.89	97.3
44	Trenley Lawn drain (Brockenhurst).	NO	1.13	7.8	284
45	Headwaters - South weirs (Fuzzy Hill)	NO	0.49	7.09	156
46	Thorney Hill Holms (Cross Ways).	YES	0.47	7.58	144.4
46	Thorney Hill Holms (Cross Ways).	YES	0.52	7.63	135.3
46	Thorney Hill Holms (Cross Ways).	NO	0.1	6.43	60
49	North Weirs	NO	0.57	6.68	190
52	Wooton.	NO	0.38	6.64	163.5
53	Eastley Wooton	NO	0.3	7.04	233
54	Wooton Woods	NO	0.08	5.35	353
56	Old house.Backley Plain	YES	0.35	6.85	177.2
56	Old house.Backley Plain.	YES	0	4.29	77.1

Ref	Name of the Pond	Presence of *C. helmsii*	Alkalinity (m.eq/L)	pH	Conductivity (μS)
60	Wilverly Drain.	NO	0.6	7.44	1202
61	Hatchet Pond.	NO	0.84	7.66	168
69	Pond opposite the Royal Oak pub.	YES	0.75	6.97	161
70	Opposite Gorley Tea rooms	YES	1.28	7.5	188.4
71	Pond by Mandy's Cattery.	YES	1.61	7.29	295
72*	Pond north of Buddle Hill lane	YES	0.09	5.99	29.5
73	South of pond no. North of Jones farm.	YES	0.46	6.81	91
74	Ponds immediately west of telephone box.	NO	0.55	7.14	92.7
75	Gorley Triangle.	NO	1.33	7.55	177.9
77	South of plantation cottage.	YES	0.93	6.82	177
78	Pond next to EH1 village hall.	YES	1.44	7.22	251
78	Pond next to EH1 village hall.	YES	0.7	7.45	256
82	Unnamed Anti-Aircraft Emplacement.	YES	0.17	6.48	52.3
83	Unnamed Anti-Aircraft Emplacement	YES	0.13	6.34	47
84	Unnamed AA emplacement east of the road.	YES	0.47	7.83	119
85	Unnamed roadside depression west of road.	YES	0.22	6.78	63.1
88	Hartford Heath Pond	YES	0.67	7.41	160.6
89	Hill top Pond.	YES	0.15	6.3	96.4
90	Moonhills pond.	YES	0	4.52	101.8
96	Unnamed pond	NO	0.32	6.81	123
98	Near Fritham.	YES	0	4.54	74.6
121	Bignell Pond.	NO	0.15	6.65	87.5
122	Longcross Pond.	NO	0.08	5.59	84.1
123	Jamesmoor Pond.	NO	0.35	6.88	124.3
128	Unnamed Pool.	NO	0.01	4.63	52
131	Unnamed Gravel Pit.	NO	0.04	5.7	64.4
133	Slufters Pond.	NO	0.03	5.27	66.5
134	Slufters Seepage Pond.	NO	0.16	6.62	82.9

Ref	Name of the Pond	Presence of *C. helmsii*	Alkalinity (m.eq/L)	pH	Conductivity (µS)
135	Broomy Shado Pond	NO	0.03	5.19	68.4
135	Broomy Shado Pond	NO	0.17	7.11	178.8
137	Cadnam's Pool	NO	0.04	4.3	51.5
138	Groon Pond	NO	0	6.58	123.4
139	Small Pond near Green Pond	NO	0.35	6.63	193.3
New site	Milkham enclosure	NO	0.05	5.43	68.5

Table 3: Alkalinity, pH and Condicuvity of the differents sites

*NB: The chemical results from the survey on 09/07/02 could have been distorted because of heavy rain over the previous few days. Dilution of pond water may have cause a drop of ionic strength and a decrease in pH especially at site 72. The standing water at the site appeared to be temporary accumulation of the recent rainwater. This opinion was supported by its low ionic concentration. In this site, *C. helmsii* occurs as a small patch. Normally the area remain damp due to infiltration of water from the adjacent pond, allowing *C. helmsii* to stay alive and proliferate when conditions are suitable.

The data of this site will not be taken into consideration during the basic and trophic analysis.

By analysing each chemical parameter, the range of habitats liable to permit the growth of *Crassula helmsii* will be defined.

In the following analysis, data from invaded sites will be compared with those from the whole population of ponds surveyed. The cumulated distribution of the two sets will be plotted for each chemical parameter.

NB: The non-cumulated distribution could be assessed by defining the derived curve of each of the cumulated distributions. The steeper the slope is, the more frequent are invasions.

To have a better view of these distributions, histograms will be drawn. Their classes have been chosen in order to contain a representative number of sites and to be the most relevant with the distributions of the sites.

a) pH

The charts below show the occurrence of the invasion by *Crassula helmsii* against the alkalinity of the sites.

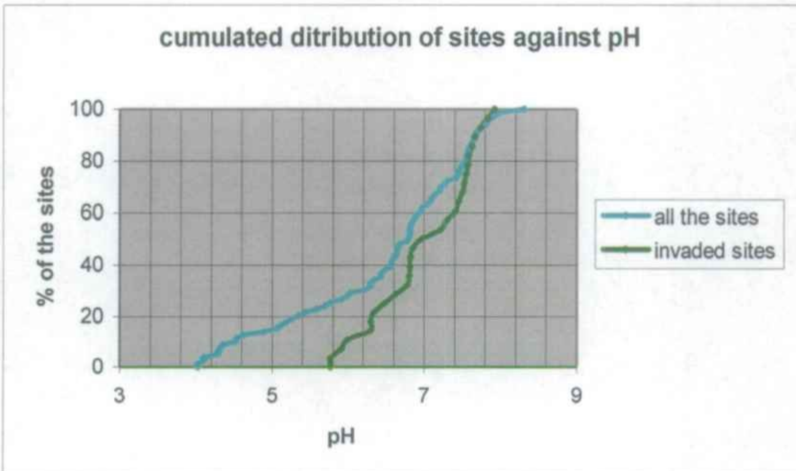


Figure 13: cumulated distribution of all the sites and the invaded sites against pH.

According to the chart, it seems to be clear that low pH could inhibit invasion by *Crassula helmsii*. The lowest pH noted on the 27 sites infected ponds is only 5.77 (site 21).

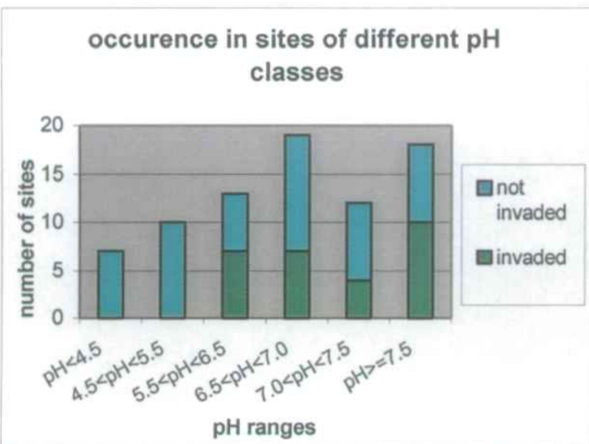


Figure 14: distribution of invaded and non-invaded sites against pH values.

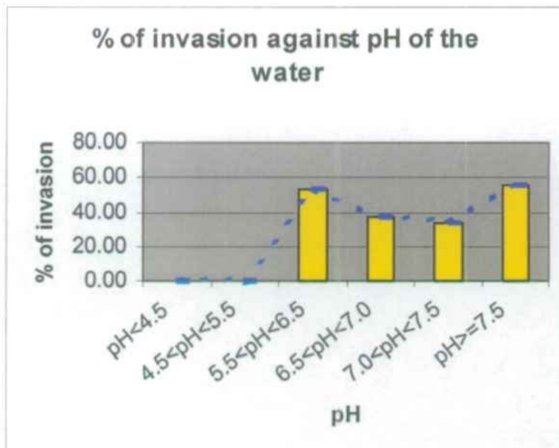


Figure 15: Percentage of occurrence of invasion against pH.

Above the pH of 5.5, no increase in invasion rate is evident with increasing pH.

b) Alkalinity

Similarly, the charts below, plot the distribution of the invasion by *Crassula helmsii* against the alkalinity of the water.

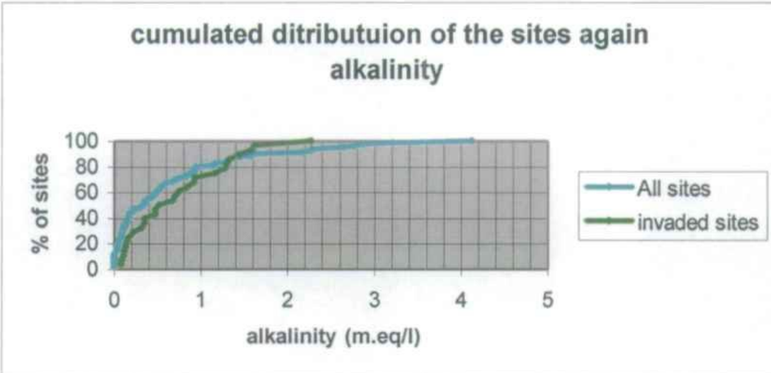


Figure 16: cumulated distribution of invaded sites and non-invaded one against alkalinity values.

The chart above appears to indicate that for both invaded and non invaded sites, there is a logarithmic distribution of number of sites with increasing alkalinity. Subsequently, to have best comparison of their distributions, the same curves will be re-plotted with a logarithmic scale.

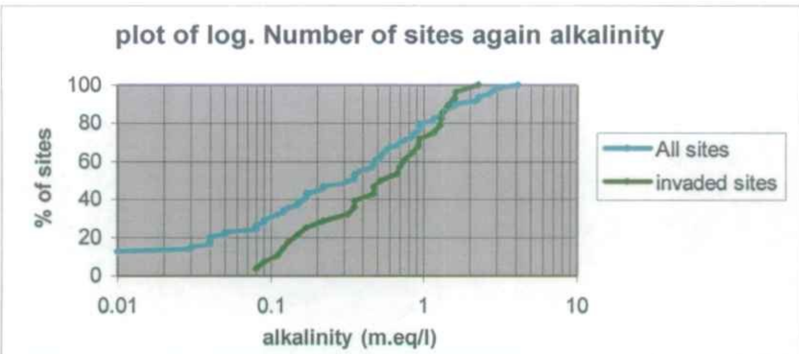


Figure 17: cumulated distribution of invaded sites and non-invaded one against alkalinity values, using a logarithmic scale

This results suggests that a minimum alkalinity may be required for *C.helmsii* to invade and establish a population. Nonetheless invasion can occur in low level of alkalinity. However in the site 39 (Pond 2), which is densely covered by *Crassula helmsii*, the alkalinity of the water is only 0.08 m.eq.l^{-1}.

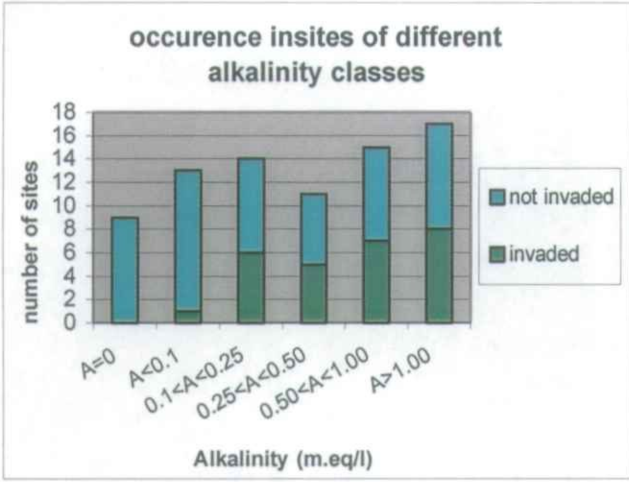


Figure 18: distribution of invaded and non-invaded sites against alkalinity values.

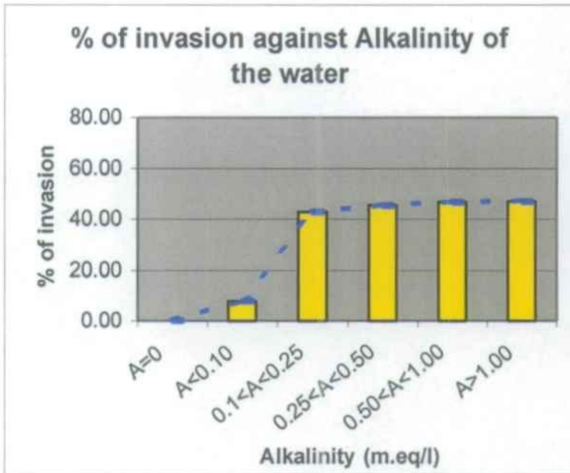


Figure 19: Percentage of occurrence of invasion against alkalinity.

Up to 1m.eq.l-1, the rate of invasion of the sites by *C.helmsii* remains stable. Alkalinity does not seem to have any influences on the rate of invasion above this value.

c) Conductivity

Conductivity is the ability of a substance to carry and exchange electrons. Consequently, a high conductivity in water is always associated with a high ionic concentration.

The charts below show the distribution of *Crassula helmsii* invasion against the conductivity of the water of the wetlands.

Figure 20: cumulated distribution of invaded and non-invaded sites against conductivity.

According to this chart, low conductivity does not seem to impose limitation to *C.helmsii* growth.

The charts below show the occurrence of *Crassula helmsii* invasion against the conductivity of the water of the wetlands.

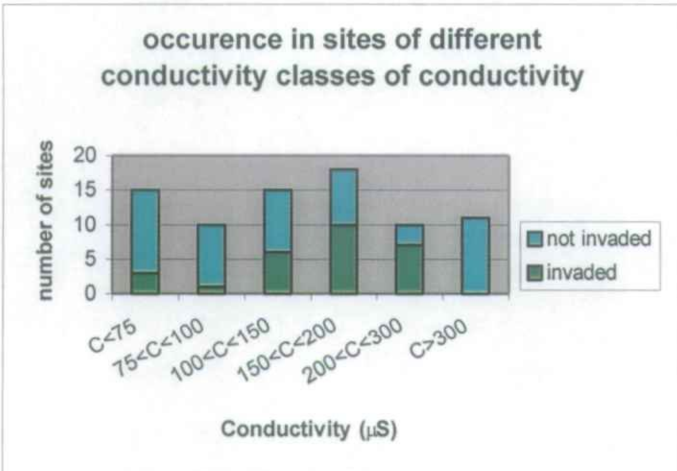


Figure 21: distribution of invaded and non-invaded sites against conductivity.

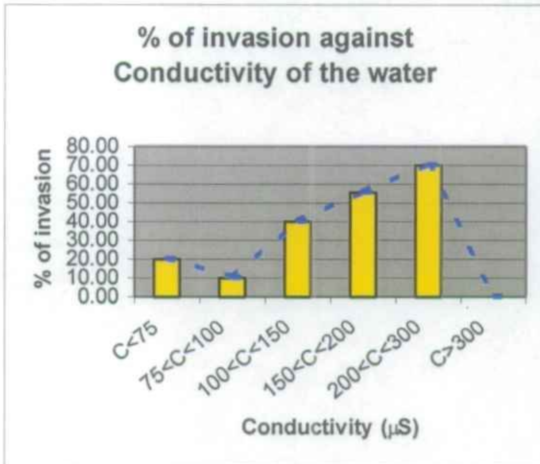


Figure 22: Percentage of occurrence of invasion against conductivity

It can be seen in both cumulative distribution chart and histograms that no *C. helmsii* has been observed at sites with the highest conductivity values (more than 300 μS). High conductivity may be responsible for exclusion of this invasive plant.

It was noted that, in a complex of 4 small ponds (site 5 A, B and C) grouped closely together and probably linked during flood conditionbs, only one was infected. Analysis of the water showed that the infected pond is the only one with conductivity lower than 300 μS.

Even if this hypothesis has to be confirmed by more data, several facts let think that a too high conductivity of the water could be responsible of its banishment.

d) discussion

During the previous chapters, involvements of the pH, the alkalinity and of the conductivity in the efficiency of invasion by *C.helmsii* have been exposed. Nevertheless, by examining the relation between these three parameters, it can be shown they may not be independent.

♦ By plotting the curve of pH against alkalinity, using a logarithmic scale, a tight relationship between these two parameters can be displayed.

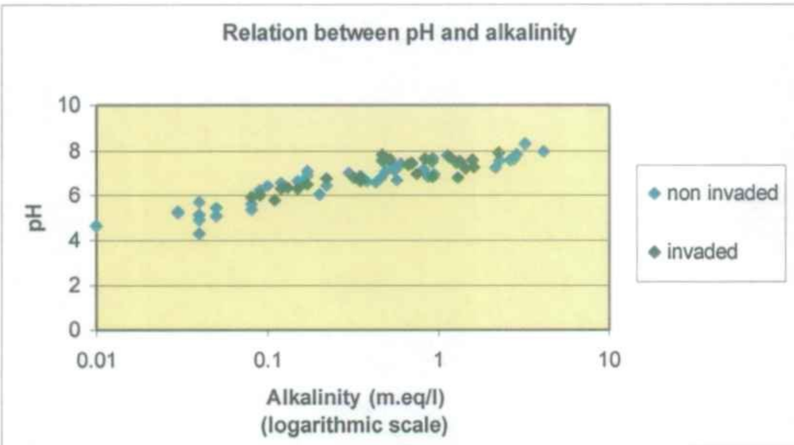


Figure 23: relation with pH and alkalinity for the surveyed sites.

This chart shows the predominance of the alkalinity in the pH setting. The more alkaline the water is, the more the pH is shift to the highest values by neutralisation of unbounded protons.

Consequently, because alkalinity is highly involved in pH establishment, these two parameters cannot be analysed independently. It cannot be said which of these two parameters is really involved in *C.helmsii*'s growth.

♦ In the same way the relation between pH and conductivity has been plotted below.

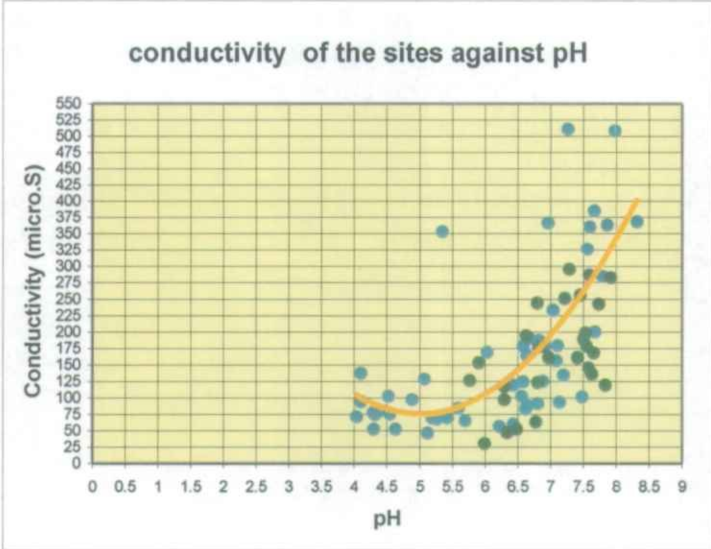


Figure 24: conductivity of the sites surveyed against their pH (blue: no invaded, green: invaded) and the trend line of all these points. One of the sites (site 61, not infected by *C.helmsii*) has been excluded because of its extremely high conductivity (1200µS, pH = 7.44); this point is out of the scale.

This charts shows that minimum conductivities observed for pHs are around 5.

- Above pH=6, and for more than 70% of the sites, the conductivity of the water increases proportionally with the pH. With high pH, the concentration of inorganic carbon ions ($HCO3^-/CO3^{2-}$), which are generally the main ions in freshwater, makes the conductivity increasing quickly.

-Below pH 6, an other phenomenon enters into consideration. At very low pH, the increase of conductivity is due to the accumulation of organic ions such as humic and fulvic acids, due to the slowdown of the microbial activity.

According to these previous relations, it cannot be said which of the parameters conductivity, alkalinity or conductivity is really involved in the limitation of the growth of *Crassula helmsii*. These three parameters must not be observed independently to determine the preferential habitat of this invasive plant.

Along with this first run of experiments, the main basic characteristics of the water bodies propitious to *Crassula helmsii* have been defined. These must have a high enough level of alkalinity in order to stabilise the pH of the water above the limiting value of 5.5. In so far as the water does not reach too high level of conductivity >300, the more conductive the water is, the more *Crassula helmsii* will fit in this site.

B) Trophic characteristics

The phosphorus, nitrogen and potassium concentrations of each sample have been noted. The results are shown in the table below.

Because of a lack of time, nitrogen analysis could not have been undertaken.

Ref	Name of the Pond	Presence of *C. helmsii*	Amount of Phosphorus	Amount of Potassium
1	Wootton Heath Farm	YES	529.396	8.56
4	Little Wooton inclosure.	NO	255.146	2.06
5	Little Wooton inclosure (south - west edge).	NO	37.479	2.87
5	Little Wooton inclosure edge (lower pond).	NO	6.301	0.32
5	Plain Heath (south)/ A35.	YES	32.282	3.53
5	Little Wooton inclosure edge (upper pond).	NO	0	2.6
9	Little Wooton inclosure edge (south).	NO	29.396	1.87
11	Little Wooton Inclosure South-west edge	NO	64.615	4.87
12	Little Wooton inclosure South- east edge.	NO	73.853	5.95
13	Little Wooton inclosure South edge / iron gate.	NO	95.215	3.16
16	Hincheslea Bog.	NO	26.509	1.47
16	Hincheslea Bog.	NO	20.735	2.63
17	Durns town.	NO		
19	Mallpit Oak, Setley.	NO	17.952	6.65
20	Mallpit Oak, Setley.	YES	35.169	1.91
21	Mallpit Oak, Setley.	YES	13.837	0.57
23	Latchmoor pond.	YES	5.146	0.16
24	Balmor lawn (near hotel).	NO	15.601	2.36
25	Balmor lawn (near hotel).	NO	22.656	0.16
27	Black Knowl/ Butts lawn.	NO		12.15
29	Furzy cottage.	NO	112.603	3.74
30	Clay hill (Burley).	NO	20.304	2.39
31	Holman's Bottom (Burley).	YES	232.629	2.39

Ref	Name of the Pond	Presence of *C. helmsii*	Amount of Phosphorus	Amount of Potassium
32	Cot bottom (Burley).	NO	29.973	4.93
33	Pigsty Hill (Burley).	YES	55.954	5.73
34	Burley Golf Course.	NO	19.128	2.24
34	Burley Golf Course.	NO	47.294	2.19
37	Whitten Pond (Holmsley).	NO	32.062	1.34
38	Holmsley Walk pond.	NO	15.601	1.15
39	Church Moor/ Cranes Moor (Brown Lane).	NO	26.771	1.99
39	Church Moor/ Cranes Moor (Brown Lane).	YES	36.177	3.2
39	Church Moor/ Cranes Moor (Brown Lane).	NO	6.195	1.81
40	Cranes Moor.	NO	196.255	0.79
40	Cranes Moor.	NO	7.958	0.55
43	Bagnum Rough - Kingston Great Common.	NO	8.61	1.46
44	Trenley Lawn drain (Brockenhurst).	NO	11.486	3.45
45	Headwaters - South weirs (Fuzzy Hill).	NO	11.486	1.32
46	Thorney Hill Holms (Cross Ways).	YES	22.068	2.19
46	Thorney Hill Holms (Cross Ways).	YES		
46	Thorney Hill Holms (Cross Ways).	NO	15.013	2.98
49	North Weirs.	NO	5.607	2.03
52	Wooton.	NO	91.174	3.12
53	Eastley Wooton.	NO	14.384	3.37
54	Wooton Woods.	NO	78.472	1.43
56	Old house,Backley Plain.	YES		1.95
56	Old house,Backley Plain.	YES		3.01
60	Wilverly Drain.	NO	11.486	2.05
61	Hatchet Pond.	NO	44.984	8.19
69	Pond opposite the Royal Oak pub.	YES	181.243	4.86
70	Opposite Gorley Tea rooms.	YES	159.881	4.67
71	Pond by Mandy's Cattery.	YES	51.336	2.09
72*	Pond north of Buddle Hill lane.	YES		

Ref	Name of the Pond	Presence of *C. helmsii*	Amount of Phosphorus	Amount of Potassium
73	South of pond no. North of Jones farm.	YES	69.811	1.78
74	Ponds immediately west of telephone box.	NO	32.282	3.71
75	Gorley Triangle.	NO	219.927	7.46
77	South of plantation cottage.	YES	83.091	3.02
78	Pond next to EH1 village hall.	YES	42.675	3.17
78	Pond next to EH1 village hall.	YES	100.412	3.58
82	Unnamed Anti-Aircraft Emplacement.	YES	51.336	2.23
83	Unnamed AA emplacement next toward Beaulieu from site 3.	YES	23.045	1.72
84	Unnamed AA emplacement east of the road.	YES	3.414	1.78
85	Unnamed roadside depression west of road.	YES	11.497	1.09
88	Hartford Heath Pond.	YES	29.973	2.26
89	Hill top Pond.	YES	75.008	2.53
90	Moonhills pond.	YES	19.003	0.77
96	Unnamed pond.	NO	20.304	1.64
98	Near Fritham.	YES	28.241	1.33
121	Bignell Pond.	NO	13.837	1.86
122	Longcross Pond.	NO	134.476	5.75
123	Jamesmoor Pond.	NO	31.705	4.69
128	Unnamed Pool.	NO	15.601	1.69
131	Unnamed Gravel Pit.	NO	8.546	1.32
133	Slufters Pond.	NO	30.298	0.32
134	Slufters Seepage Pond.	NO	12.661	0.57
135	Broomy Shade Pond.	NO	10.31	6.15
135	Broomy Shade Pond.	NO	53.226	1.33
137	Cadnam's Pool.	NO	28.241	1.6
138	Green Pond.	NO	109.65	6.94
139	Small Pond near Green Pond.	NO	9.722	2.74
New site	Milkham enclosure.	NO	1.492	0.07

Table 4: amount of Phosphorus and Potassium in the different sites

a) Phosphorus

The distribution of the sites against their phosphorus amount is displayed in the following charts.

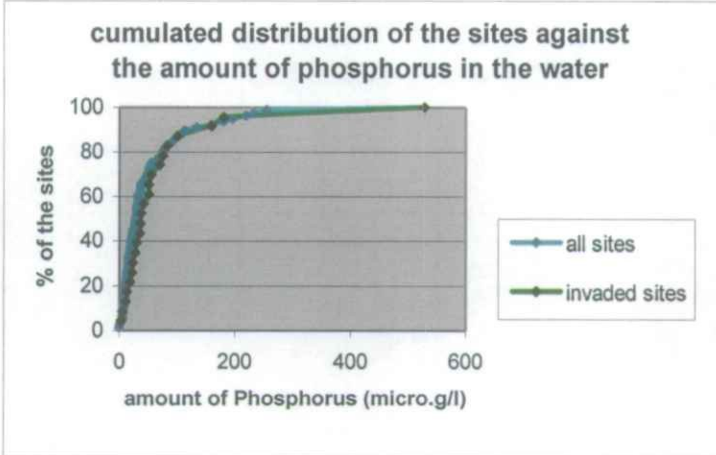


Figure 25: cumulated distribution of invaded sites and non-invaded one against phosphorus amount.

This chart indicates that both invade and uninvaded sites follow a logarithmic distribution. Subsequently, to have best comparison of their distributions, the same curves will be re-plotted with a logarithmic scale.

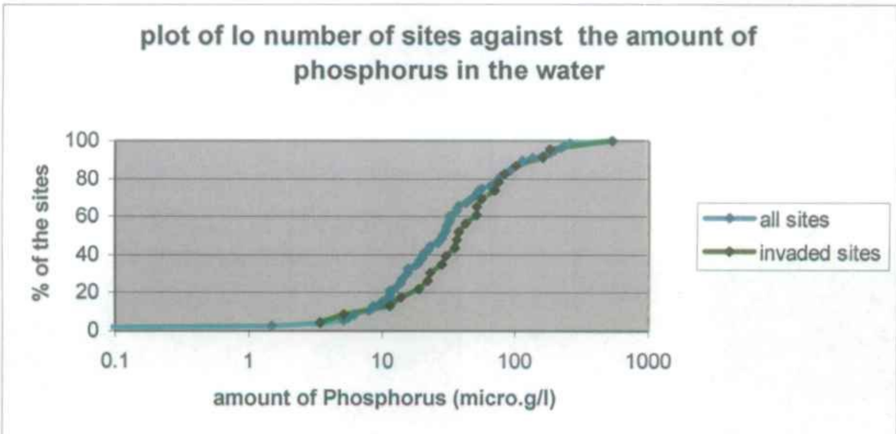


Figure 26: cumulated distribution of invaded sites and non-invaded one against conductivity (logarithmic scale).

Even if these two distributions present a similar shape, a shift occurs between them. The distribution of invaded sites seems to be more focussed in the highest values. The histogram will permit to have a better view of it.

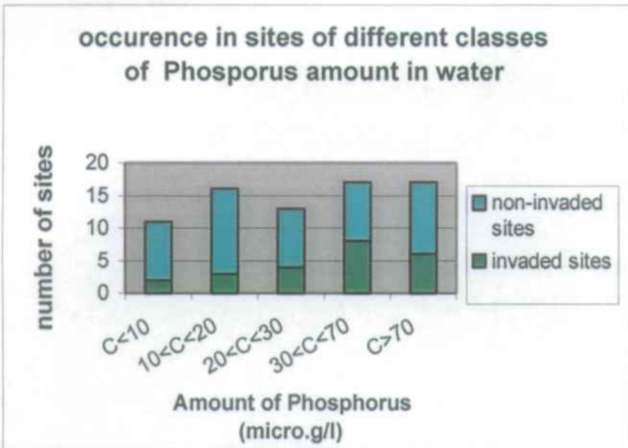


Fig 27: distribution of invaded and non-invaded sites against ranges of amount of phosphorus in water.

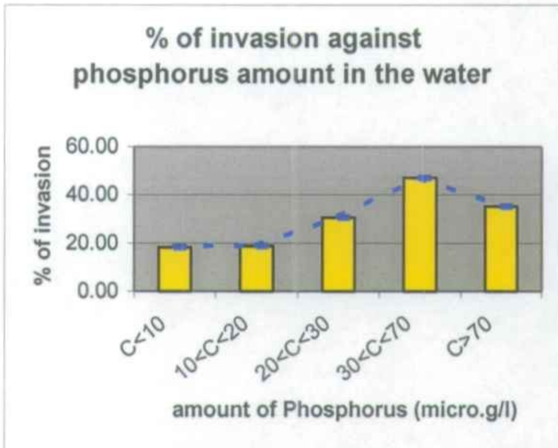


Fig 28: Percentage of occurrence of invaded sites against ranges of amount of phosphorus

Even if, *C.helmsii* can be observed in sites poor in phosphorus, such as the sites 82-B (3.4 $\mu g.l^{-1}$) or 23 (5.1 $\mu g.l^{-1}$), the potential for invasion seems to increase with the increasing of phosphorus content in the water. The most critical case of invasion is observed at the site 1, containing 529 $\mu g.l^{-1}$ of phosphorus, the highest value found during the surveys (figure 28).

b) Potassium

The distribution of the surveyed sites is shown in the chart below.

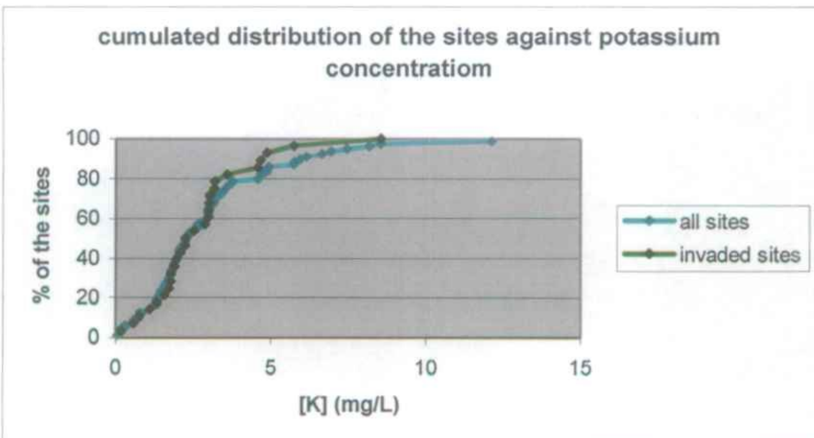


Figure 25: Cumulated distribution of the invaded sites and non-invaded sites again potassium concentration.

By this chart a large similarity can be noticed between the two curves, showing the same distribution between the two population.

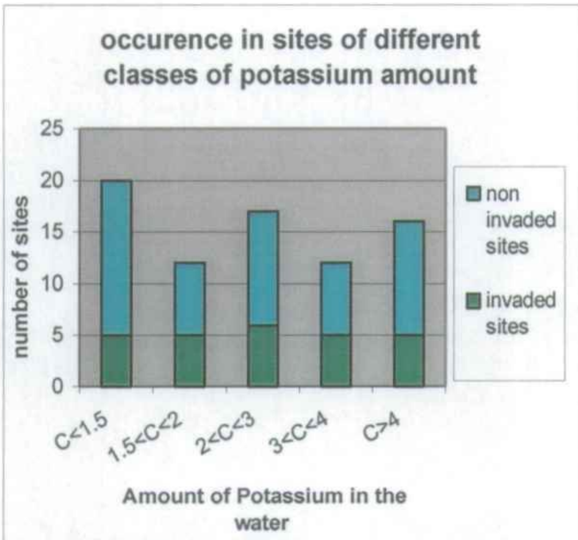


Figure 26: distribution of the invaded and non-invaded sites against potassium concentration.

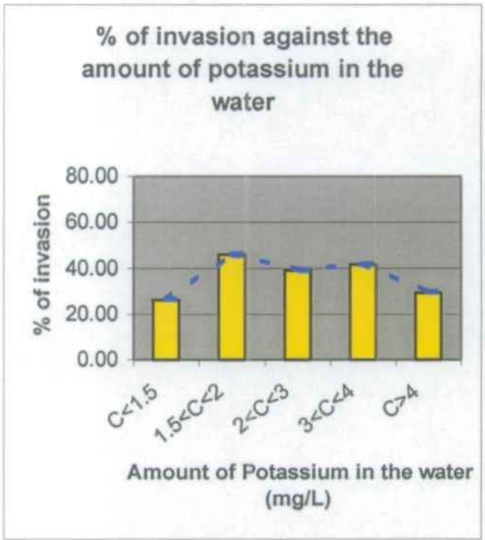


Figure 27: Percentage of occurrence of invasion against the concentration of potassium in the water

According to these previous charts, the amount of potassium in the water does not seem to have any influence on the invasion by *C.helmsii*. Indeed, can occur as well in sites with very low (site 23: 0.16 $mg.l^{-1}$) as in very high (site 1: 8.56 $mg.l^{-1}$) amount of potassium.

c) <u>Discussion</u>

During this investigation it has been shown that both low phosphorus and potassium levels in the water limit *C.helmsii* spread. Nevertheless, for sites with low nutrient amount and especially for Phosphorus (such as the site 23, 21 and 82-B) it was observed that the plant only occurs emerged on the banks. In these conditions, the plant is of short stature and is seems to be present as a component of the ecosystem rather than damaging the site by invading and choking the freshwater habitat (such as in the site 1).

C.helmsii seems to be characterized by an ability to up-take large amount of nutrients, giving it a competitive advantage over nature species. However this advantage seems to be lost in nutrient limited conditions (figures 28 and 29).

Thus, the amount of phosphorus present influences the growth of *C.helmsii* but not necessarily its potential for invasion.

Figure 28: site 1 (amount of P: 529.4 μg.l^{-1}, amount of K: 8.56 mg.l^{-1}), the pond is completely covered by *C. helmsii*, which is highly dominant in this ecosystem.

Figure 29: site 23 (amount of P: 5.1 μg.l^{-1}, amount of K: 0.16 mg.l^{-1}), *C. helmsii* is not present in the water, only little-sized specimen are colonizing the banks in association with others species.

Conclusion:

Through this investigation, limits in the potential of spread of *C.helmsii* have been found out.

Firstly, the range of physical habitat where it occurs is currently limited. Indeed the pant seems to be restricted to still or very slow flowing water such as pond or ditches. In addition, some aspects of the water chemistry seem to be related to its occurrence or absence. *C.helmsii* seems to be excluded from low pH and low alkalinity sites. Moreover it appears that the plant is absent from water with a very high ionic content.

Secondly, there appears to be situation where *C.helmsii* does not out-compete native species. The plant seems to be able to grow rapidly and dominate some ecosystems but not where nutrients – particularly Phosphorus – are low. Phosphorus can be the limiting nutrient in growth because of its scarcity. In this study sites with low levels phosphorus levels were found to have a lower incidence of invasion, and where present, the plant was less dominant.

C.helmsii is now wide spread in England. In the future it is possible that it will evolve and adapt to new habitats, even colonizing flowing water and invading waters with chemistry to which it is, at present, ill adapted. In this way, problems associated with this plant will increase.

Against this, there is a possibility that *C.helmsii* will become susceptible to natural control such as pathogens or invertebrates grazers to which it appears to be immune at present. Such pressures could reduce the dominance of the plant to the extent that it becomes a non-dominant addition to the flora of the country, as is the case of its natural range in Australia.

Additional activities:

Analysis of water samples for nitrate content is being undertaken but may not be complete before this report is finished. It is intended that results will be appended to the report when they are available.

It has been suggested that C.helmsii may take up and store nutrients in excess of immediate metabolic requirements. One aspect of the chemical investigation was intended to be measurement of the nutrient content of C.helmsii at sites where water has been analysed. Samples of C.helmsii have been taken from a selection of the invaded sites. These samples have been dried and some ground as a first stage in nutrient analysis. Time has not allowed completion of this aspect of the study.

As part of the current study, a photographic archive of the current state of the sites visited has been produced. Digital images of site and plants investigated have been transferred to CD for future reference.

A second archive of sketch maps of sites showing the extent and possible sources and routes of further invasion has been produced. These sketch maps include notes on orientation and physical features which may be of relevance to invasion by C.helmsii, such as substratum and shading. Time did not permit transfer of these to digital format but this may be done in the future.

As part of the general work of the section, a little time was spent assisting in the collection of bulk samples of sediments and waters. Samples and associated environmental measurements were taken under defined, quality-controlled conditions. Samples were sent, under contract, to commercial organisations for use in tests of environmental fate and toxicology of new compounds. A report on sampling conditions and environmental measurements was produced as part of the contract (Farr & Brunet, 2002).

Acknowledgement:

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I cannot thank I. Farr enough for all the time he devoted to me, for his advise and the confidence he had in me during this last four months.

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APPENDIX 1: SOP 55/03.03.00, Nitrate + Nitrite measurement

SOP: 53/03/03/2000 UPDATE: NONE

TITLE: THE DETERMINATION OF NITRATE IN WATER BY FLOW INJECTION ANALYSIS

AUTHOR: D.V.LEACH

EQUIPMENT

1. FIAstar 5012 Spectrophotometer and analyser
2. Chemifold Type II with Cd reduction column

PRINCIPLE

The aqueous sample is injected into a carrier stream of Ammonium Chloride. The nitrate is reduced to nitrite in a cadmium reductor. On the addition of acidic sulphanilamide a diazo compound is formed which then reacts with N-(1-Napthyl)-ethylenediamine dihydrochloride. A purple azo dye is formed, and measured at 540 nm.

Range: 0-10 mg/l NO_3 –N
Lod 0.01mg/l NO_3 –N

REAGENTS

All reagents should be of reagent grade purity. Use deionised water (>10 MΩ) to prepare reagents.

1. Standard Stock Solution, 1000 mg/l NO_3-N
   Dissolve 3.61 g of Sodium Nitrate, $NaNO_3$ in 500 ml of deionised water.

2. Running Standard Solutions, 0 - 10 mg/l NO_3-N

Standard number	Flask size (ml)	Volume added (ml)	Final Conc. (mg/l)
0	100	0	0
1	500	1000	2
2	250	1000	4
3	100	600	6
4	100	800	8
5	100	1000	10

3. Carrier Solution (Ammonium Chloride)
   Stock: 5 M NH_4Cl.
   Dissolve 133.75 g of Ammonium Chloride in 500 ml of deionised water

   Working Carrier: 0.1 M NH_4Cl.
   Dilute 10 ml of the Stock Solution to 500 ml with deionised water.

4. R1: Sulphanilamide Solution - Stable for several months

   Dissolve 5 g of Sulphanilamide in a mixture of 25 ml Hydrochloric acid and 300 ml of deionised water. Dilute to 500 ml.

5. R2: N-(1-Napthyl)-ethylenediamine dihydrochloride (NED) - Stable 1 week

   Dissolve 0.5 g of NED in 500 ml deionised water. Store in amber bottle.

REFERENCE

Perstorp Analytical Ltd. Application Note (ASN 62-02/83)

Hazards

Cadmium.

TOXIC, SUSPECTED CARCINOGEN and DANGEROUS TO THE ENVIRONMENT.

Avoid skin contact.

Wear gloves when handling and changing cadmium columns.

Wear gloves when clearing up any spillage.

Waste (reagents and samples after passing through column, old columns etc.) must be collected, clearly marked and disposed of by a recognised company

Sodium Nitrate.

Oxidising when in contact with combustable material.

Ammonium Chloride.

Harmful if swallowed and irritating to eyes.

Sulphanilamide.

Harmful if swallowed and irritating to eyes.

Hydrochloric acid.

Corrosive. Causes burns, irritating to lungs and eyes.

Wash with plenty of water if in contact with skin or eyes. If in contact with eyes or ingested then seek medical attention immediately.

N-(1-Napthyl)-ethylenediamine dihydrochloride (NED).

Harmful if swallowed and irritating to eyes.

APPENDIX 2 : Nitrate + Nitrite concentrations observed in the sites

Ref	Name of the Pond	Grid reference	Type of site	Nitrate + Nitrite concentration ($mg.l^{-1}$)
New site	Milkham enclosure.	SU217105	Pond	0.212
1	Wootton Heath Farm	SZ236987	Pond	0.293
4	Little Wooton inclosure.	SZ228990	Pond	0.225
5	Little Wooton inclosure (south - west edge).Pond B	SZ223988	Pond	0.196
5	Little Wooton inclosure (south - west edge).Pond A	SZ223987	Pond	0.254
5	Little Wooton inclosure (south - west edge) Pond C	SZ219987	Pond	0.226
9	Little Wooton inclosure edge (south).	SZ224985	Pond	0.208
11	Little Wooton Inclosure South-west edge	SZ225985	Marl Pit	0.246
12	Little Wooton inclosure South- east edge	SZ226985	Pond	0.214
13	Little Wooton inclosure South edge / iron gate.	SZ228985	Pond	0.266
16	Hincheslea Bog.	SU277005	Pond	0.250
16	Hincheslea Bog.	SU277006	Marsh	0.274
19	Mallpit Oak, Setley.	SZ286997	Marl Pit	0.268
20	Mallpit Oak, Setley.	SZ286997	Marl Pit	0.215
21	Mallpit Oak, Setley.	SZ286997	Marl Pit	0.208
23	Latchmoor pond	SU293004	Pond	0.174
24	Balmor lawn (near hotel).	SU304033	Pond	0.248
25	Balmor lawn (near hotel).	SU306035	Lake	0.229
27	Black Knowl/ Butts lawn.	SU289027	Pond	0.227
29	Furzy cottage.	SU287018	Pond	0.260
30	Clay hill (Burley).	SU233024	Pond	0.208
31	Holman's Bottom (Burley).	SU229024	Pond	1.829
32	Cot bottom (Burley).	SU224025	Pond	0.267
33	Pigsty Hill (Burley).	SU224026	Pond	0.645
34	Burley Golf Course.	SU218026	Pond	0.233
34	Burley Golf Course.	SU218027	Pond	0.233
38	Holmsley Walk pond	SU206013	Pond	0.247
39	Church Moor/ Cranes Moor (Brown Lane).	SU198020	Pond	0.277

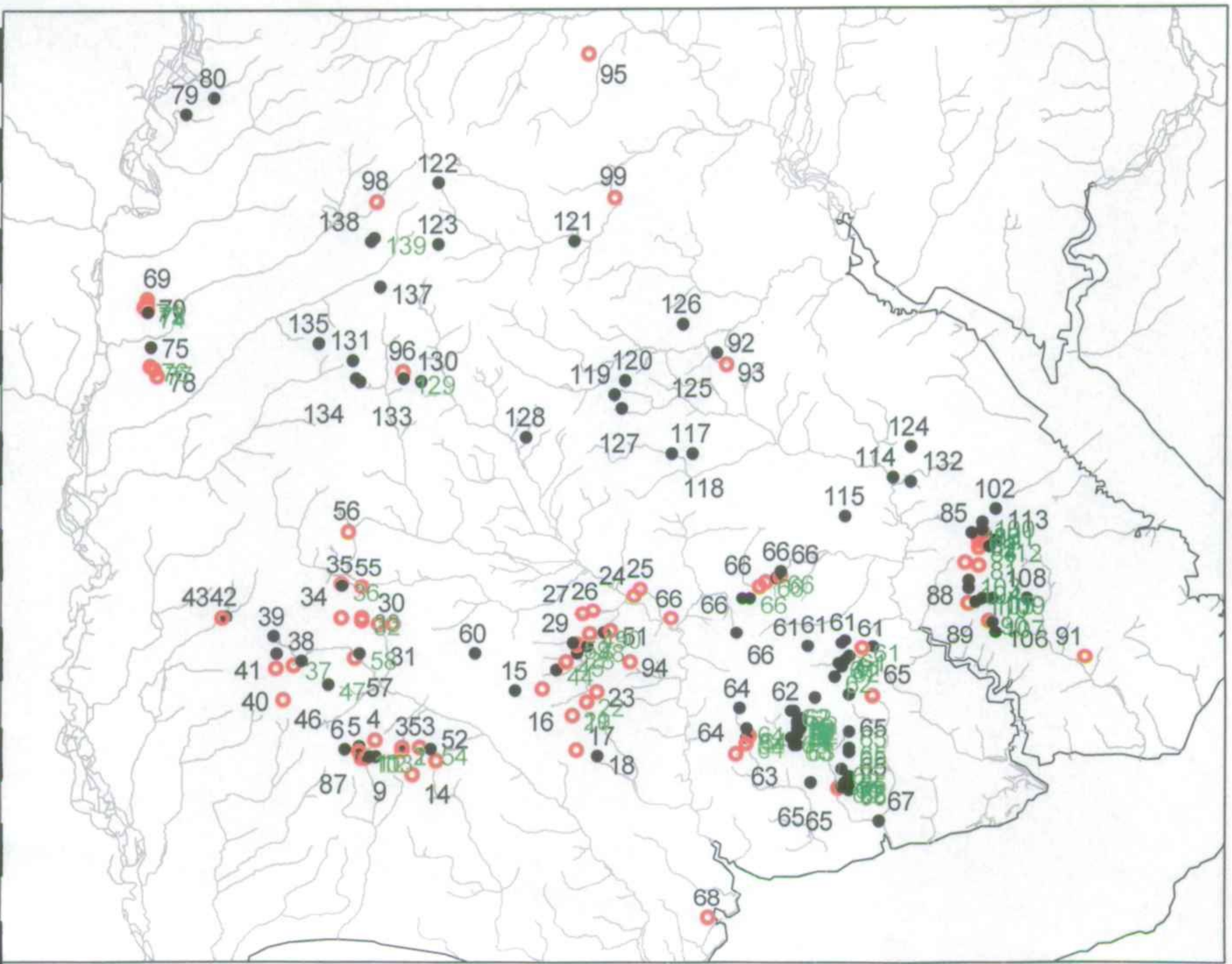
Ref	Name of the Pond	Grid reference	Type of site	Nitrate + Nitrite concentration $(mg.l^{-1})$
39	Church Moor/ Cranes Moor (Brown Lane).	SU19905	Pond	0.289
40	Cranes Moor.	SU199011	Pond	0.248
40	Cranes Moor.	SU199012	Pond	0.274
44	Trenley Lawn drain (Brockenhurst).	SU281010	Ditch	0.233
45	Headwaters - South weirs (Fuzzy Hill).	SU284013	Stream	0.243
46	Thorney Hill Holms (Cross Ways).	SU200004	Pond	0.264
49	North Weirs.	SU291021	Stream	0.246
52	Wooton.	SZ246984	Pond	0.234
53	Eastley Wooton.	SZ241988	Pond	0.240
54	Wooton Woods.	SZ244987	Pond	0.282
60	Wilverly Drain.	SU257015	Pond	0.269
61	Hatchet Pond.	SU367014	Pond & Marl Pit	2.537
69	Pond opposite the Royal Oak pub.	SU161119	Stream	0.815
70	Opposite Gorley Tea rooms.	SU161118	Pond	0.242
71	Pond by Mandy's Cattery	SU160117	Pond	0.334
73	North of Jones farm.	SU161116	Pond	0.290
74	Ponds immediately west of telephone box.	SU161115	Pond	0.267
75	Gorley Triangle.	SU162105	Pond	0.553
77	South of plantation cottage.	SU163099	Pond	0.250
78	Pond next to EH1 village hall.	SU164097	Pond	0.321
78	Pond next to EH1 village hall.	SU164098	ditch	0.651
82	Unnamed Anti-Aircraft Emplacement.	SU405049	Pond & Marl Pit	0.334
82	Unnamed AA emplacement next toward Beaulieu from site 3.	SU405047	Pond	0.248
82	Unnamed AA emplacement east of the road	SU405047	Pond	0.244
82	Unnamed Anti-Aircraft Emplacement.	SU405049	Pond & Marl Pit	0.253
88	Hartford Heath Pond.	SU401043	Pond	0.273
89	Hill top Pond.	SU402031	Pond	0.288
90	Moonhills pond.	SU408026	Pond	0.329
96	Unnamed pond.	SU236098	Pond	0.293
98	Near Fritham	SU228147	Lake	0.346
121	Bignell Pond	SU286135	Marl Pit	0.278
122	Longcross Pond.	SU246152	Pond	0.314

APPENDIX 3 : Substrate observed in the sites

Ref	Name of the Pond	Grid reference	Type of site	Substrate
1	Woolton Heath Farm	SZ236987	Pond	Clay
4	Little Wooton inclosure.	SZ228990	Pond	Mud/silt
5	Lillle Wooton inclosure (south - west edge).Pond B	SZ223988	Pond	Clay
5	Little Wooton inclosure (south - west edge) Pond A	SZ223987	Pond	Clay
5	Little Wooton inclosure (south - west edge).Pond C	SZ219987	Pond	Clay
5	Little Wooton inclosure (south - west edge) Pond D	SZ219987	Pond	Clay
9	Little Wooton inclosure edge (south).	SZ224985	Pond	Clay
11	Little Wooton Inclosure South-west edge	SZ225985	Marl Pit	Clay
12	Little Wooton inclosure South- east edge.	SZ226985	Pond	Clay
13	Little Wooton inclosure South edge / iron gate.	SZ228985	Pond	Clay
16	Hinchoslea Bog.	SU277005	Pond	Gravel
16	Hincheslea Bog.	SU277006	Marsh	Gravel
17	Durns town	SZ287987	Pond	Sand
19	Mallpit Oak, Setley.	SZ286997	Marl Pit	Clay
20	Mallpit Oak, Setley.	SZ286997	Marl Pit	Clay
21	Mallpit Oak, Setley.	SZ286997	Marl Pit	Clay
23	Latchmoor pond.	SU293004	Pond	Mud/silt
24	Balmor lawn (near hotel).	SU304033	Pond	Sand
25	Balmor lawn (near hotel).	SU306035	Lake	Clay
27	Black Knowl/ Butts lawn.	SU289027	Pond	Clay
29	Furzy cottage.	SU287018	Pond	Clay
30	Clay hill (Burley).	SU233024	Pond	Clay
31	Holman's Bottom (Burley).	SU229024	Pond	Mud/silt
32	Cot bottom (Burley).	SU224025	Pond	Clay
33	Pigsty Hill (Burley).	SU224026	Pond	Clay
34	Burley Golf Course.	SU218026	Pond	Clay
34	Burley Golf Course.	SU218027	Pond	Sand
37	Whitton pond (Holmsley)	SU204012	Pond	Sand
38	Holmsley Walk pond.	SU206013	Pond	Sand
39	Church Moor/ Cranes Moor (Brown Lane).	SU198020	Pond	Sand

Ref	Name of the Pond	Grid reference	Type of site	Substrate
39	Church Moor/ Cranes Moor (Brown Lane).	SU199022	Pond	Sand
39	Church Moor/ Cranes Moor (Brown Lane).	SU199025	Pond	Sand
40	Cranes Moor.	SU199011	Pond	Sand
40	Cranes Moor.	SU199012	Pond	Gravel
43	Bagnum Rough- Kingston Great Common	SU183026	Stream	Gravel & mud/silt
44	Trenley Lawn drain (Brockenhurst).	SU281010	Ditch	Gravel
45	Headwaters - South weirs (Fuzzy Hill).	SU284013	Stream	Gravel & mud/silt
46	Thorney Hill Holms (Cross Ways).	SU201002	Pond	Gravel & mud/silt
46	Thorney Hill Holms (Cross Ways).	SU201002	Pond	Gravel & mud/silt
46	Thorney Hill Holms (Cross Ways).	SU200004	Pond	Mud/silt
49	North Weirs.	SU291021	Stream	Clay
52	Wooton.	SZ246984	Pond	Clay
53	Eastley Wooton.	SZ241988	Pond	Clay/gravel
54	Wooton Woods.	SZ244987	Pond	Clay
56	Old house, Backley Plain	SU220052	Stream	Clay
56	Old house, Backley Plain	SU220053	Stream	Sand
60	Wilverly Drain	SU257015	Pond	Sand, gravel & mud/silt
61	Hatchet Pond.	SU367014	Pond & Marl Pit	Sand & gravel
69	Pond opposite the Royal Oak pub.	SU161119	Pond/Stream	Sand, gravel & mud/silt
70	Opposite Gorley Tea rooms	SU161118	Pond	Sand, gravel & mud/silt
71	Pond by Mandy's Cattery	SU160117	Pond	?
72*	Pond North of Buddle hill lane	SU161116	Pond	?
73	North of Jones farm.	SU161116	Pond	Mud/silt
74	Ponds immediately west of telephone box.	SU161115	Pond	Sand & Gravel
75	Gorley Triangle.	SU162105	Pond	Gravel
77	South of plantation cottage.	SU163099	Pond	Gravel
78	Pond next to EH1 village hall.	SU164097	Pond	Gravel
78	Pond next to EH1 village hall.	SU164098	ditch	Gravel
82	Unnamed Anti-Aircraft Emplacement	SU405049	Pond & Marl Pit	Gravel
83	Unnamed AA emplacement next toward Beaulieu from site 3.	SU405047	Pond	?
84	Unnamed AA emplacement east of the road	SU406051	Pond	?
85	Unnamed roadside depression west of road	SU405049	Pond	Gravel
88	Hartford Heath Pond.	SU401043	Pond	Gravel

Ref	Name of the Pond	Grid reference	Type of site	Substrate
89	Hill top Pond.	SU402031	Pond	Gravel
90	Moonhills pond.	SU408026	Pond	Mud/silt
96	Unnamed pond.	SU236098	Pond	Mud/silt
98	Near Fritham.	SU228147	Lake	Mud/silt
121	Bignoll Pond.	SU286135	Marl Pit	Mud/silt
122	Longcross Pond.	SU246152	Pond	Gravel & Mud/silt
123	Jamesmoor Pond	SU246134	Pond	Gravel & Mud/silt
128	Unnamed Pool	SU272079	Pond	Mud/silt & leaves
131	Unnamed Gravel Pit	SU221101	Ditch	Gravel & mud/silt
133	Slufters Pond	SU222095	Pond	Mud/silt
134	Slufters Seepage Pond	SU222096	Pond	Clay & mud/silt
135	Broomy Shado Pond	SU211106	Pond	Gravel & mud/silt
136	Broomy Shade Pond	SU211107	Pond	Gravel & mud/silt
137	Cadnam's Pool	SU229122	Pond & Brackish	Mud/silt
138	Green Pond	SU226135	Pond	Gravel & mud/silt
139	Small Pond near Green Pond	SU227136	Pond	Gravel
New site	Milkham enclosuro	SU217105	Pond	Clay, gravel &mud/silt



Colour code for *Crassula* sites:

Sites surveyed in 2000

grey = absent
red = present

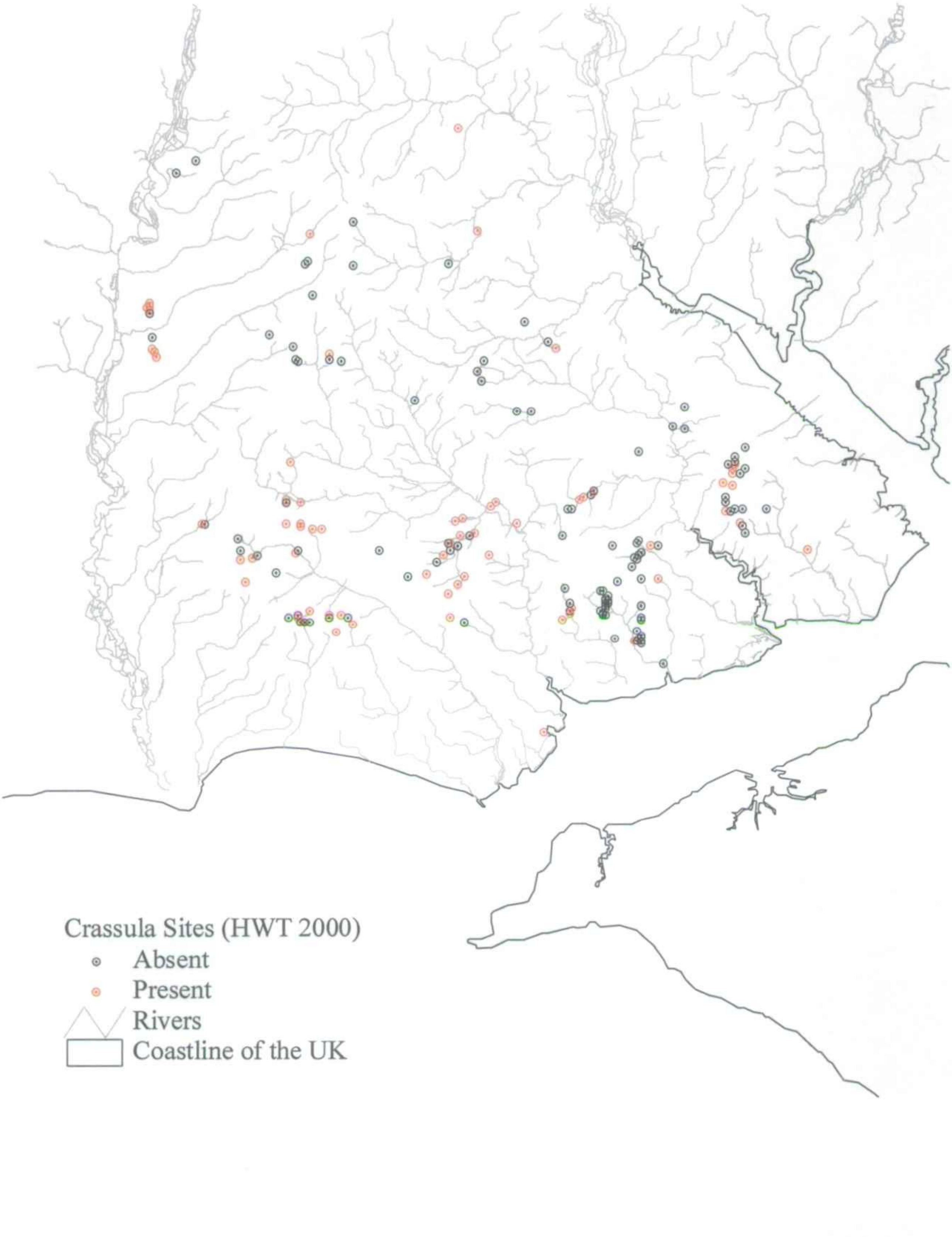
Historical *Crassula* sites

black = 1957-1969
light green = 1970-1979
dark green = 1980-1989
magenta = 1990 – 1996

To run Arcview *Crassula* project copy from t: drive the hampshire directory into your n:\av\ directory

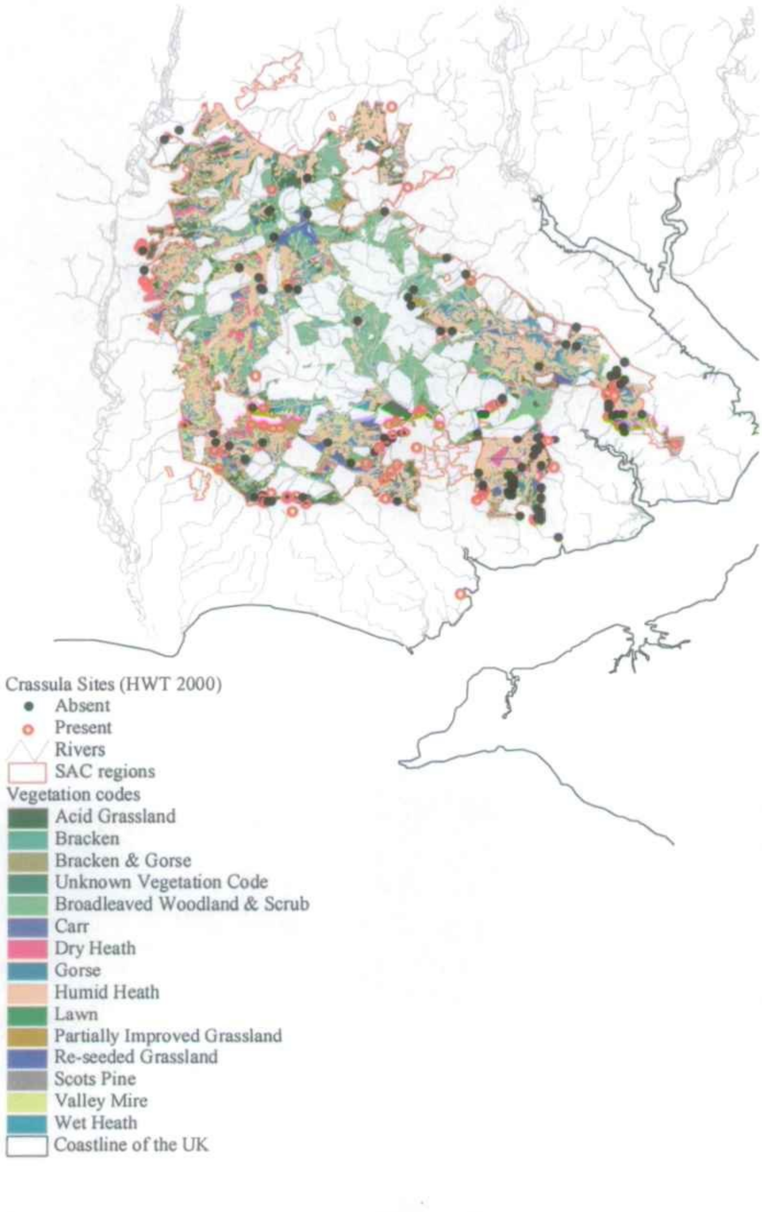
The Project file will be **n:\av\hamsphire\projectfile\crassula.apr**

I have generated the various views you asked for and few other variations for comparison. They are store in the **N:\av\hampshire\wmf files** directory



Crassula Sites (HWT 2000)
Absent
Present
Rivers
Coastline of the UK

APPENDIX 6: New Forest map and results of the Wildlife Hampshire Trust surveys



APPENDIX 7: History of Crassila helmsii invasion (about of HWT surveys)