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# Nutrient Concentrations as Predictors of Nuisance *Hydrodictyon reticulatum* Populations in New Zealand

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## ABSTRACT

Hydrodictyon reticulatum (L.) Lagerh. has spread rapidly since its recent introduction into New Zealand, with persistent nuisance growths in several important recreational waterbodies causing economic problems for local communities. Samples of *H. reticulatum* and water were collected for nutrient analysis from a wide range of sites in the central North Island where *H. reticulatum* was present, had been present or where it was likely to have been introduced. The health of the *H. reticulatum* population was assessed at each site where present. No relationship between health of H. reticulatum populations and dissolved reactive phosphorus or tissue phosphorus concentrations was observed. There was a significant inverse relationship between population health and the tissue C:N ratio. C:N ratios below 18 supported growing populations. Dissolved inorganic nitrogen (DIN) concentrations above 30 mg m<sup>-3</sup> were shown to support growing populations, which suggests that nitrogen is likely to be the major limiting nutrient for H. reticulatum populations in this region of New Zealand. In conclusion, healthy natural populations of H. reticulatum in New Zealand are likely to be found where DIN concentrations are above 30 mg m<sup>3</sup> and tissue C:N ratios are below 18. The use of the logit link function analysis of the DIN concentrations and tissue C:N ratio in relation to health of H. reticulatum populations has provided a technique which allows the probability of active and inactive growth of *H. reticulatum* to be assessed for a given DIN or

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C:N ratio. Future work to increase the robustness of these curves will provide a useful tool for water managers.

*Key words:* nutrients, growth predictions, nuisance growths, water net.

## INTRODUCTION

Nuisance growths of *H. reticulatum* have been occurring in New Zealand since it was first reported in 1987 (Coffey and Miller 1988). Since then, *H. reticulatum* has invaded a wide range of sites from small spring fed farm ponds to large windswept lakes, spreading rapidly throughout the eastern Bay of Plenty and Central North Island region (Figure 1) over an area of approximately 300 km<sup>2</sup>. The main mechanisms for the spread of *H. reticulatum* are considered to be boats being moved between waterbodies and transfer by waterfowl (Hawes et al. 1991). *H. reticulatum* in New Zealand is occurring in habitats which are inconsistent with reports from overseas where it has primarily been found in small or sheltered nutrient-rich waterbodies (Prescott 1970, Round 1981). The only previous record of a nuisance occurrence in a large lake was Lake Zürich (Thomas 1963).

In many of the invaded sites in New Zealand, *H. reticulatum* has become a perennial problem with the formation of extensive surface mats occurring during the summer on a regular basis. At most sites a small population persists throughout the winter on the lake or pond bottom in shallow waters. The persistent occurrence of *H. reticulatum* in waterbodies in New Zealand is also contrary to overseas reports where it is reported to be a one off or irregular problem in invaded water bodies (Sorensen 1950, Corrilion and Geurlesquin 1977).

There is little information on the ecology and growth requirements of H. reticulatum and it has often been contradictory. For example Prescott (1970) suggested high pH and Ca were growth requirements, while Moss (1973) suggested that eutrophication, rather than high Ca, was the requirement for extensive growths. Fitzgerald (1981) suggested bloom conditions under low pH. There has also been debate over the levels of organic nutrients required with Sörensen (1950) suggesting low organic concentrations and Corillion and Geurlesquin (1977) recording growth only in organic rich waters. These data are not consistent with growth of H. reticulatum populations in New Zealand where extensive growth occurs in low Ca, neutral to acidic pH and in moderately enriched waters (Hawes et al. 1991, Hawes and Smith 1993). Dissolved inorganic carbon (DIC) is also unlikely to be a limiting factor as extensive growths of H. reticulatum have occurred regularly in Lake Rotorua where DIC values are low (0.28 n M) Schwarz and Howard-Williams (1993).

Recent comprehensive laboratory studies of a New Zealand isolate of *H. reticulatum* (Hawes and Smith, 1993) have shown that growth occurs over a temperature range of 5-40C with an optimum at 25C. These studies confirmed that *H. reticulatum* is shade adapted (Raven et al. 1979) with saturating photon flux densities for photosynthesis of 100 to 160  $\mu$ mol m<sup>2</sup>s<sup>-1</sup> at 12-20C respectively. Hawes and Smith (1993) also suggested that the isolate had a high affinity for NO<sub>3</sub> in comparison to other nuisance filamentous species and that concentrations of dissolved reactive phosphorus above 5mg m<sup>3</sup> and 20 mg m<sup>3</sup> NO<sub>3</sub> are likely to support rapid growth.

It has been suggested that the success of *H. reticulatum* in New Zealand may be due to its ability to grow rapidly in relatively low N concentrations and low N:P ratios (Hawes and Smith 1992) and is able to fill a niche not previously filled by filamentous algae in the lakes of the Central North Island which are predominantly N-limited (White 1983). The proliferation of *H. reticulatum* may also be due to a lack of pathogens being introduced with its initial inoculum (Hawes et al. 1991).

In a number of the large lakes such as Rotorua, Rotoiti and Aniwhenua which are widely used for recreation and where large surface mats of *H. reticulatum* occur on a regular basis, the alga has become an economic nuisance resulting in pressure on water managers to control growths of the algae. No effective control method has been developed for use in New Zealand, although some control has been achieved by mechanical removal (Wells and Clayton 1993). To enable effective environmental manipulation as a potential control option the growth requirements of natural populations of *H. reticulatum* must be established.

### METHODS AND MATERIALS

During the period January 20 - February 18 1993, sixty four sites were sampled where *H. reticulatum* was present, had been previously reported or was considered likely to have been inoculated with *H. reticulatum* (Figure 1). Samples were collected for water and tissue nutrient analysis and water temperature was recorded. The health of the *H. reticulatum* population was assessed (Table 1).

Health group 1 populations were obviously unhealthy being yellow in colour and showing no signs of reproduction. In health groups 3 and 4, the populations were green in colour with a range of net sizes and in group 3 small numbers of daughter nets were present, in group 4 large numbers of daughter nets were present. Group 2 populations were in between the unhealthy group 1 populations and the obviously actively growing populations of groups 3 and 4. The group 2 populations were green in colour with some variability in net size but with no evidence of daughter net formation.

Water samples were filtered through acid soaked Whatman GF/F filters and stored frozen until analyzed for  $NO_3$ ,  $NH_4$  and dissolved reactive phosphorus (DRP) using a Technicon II autoanalyser (Downes 1988). Dissolved inorganic nitrogen (DIN), which is the sum of  $NO_3$  and  $NH_4$  concentrations, was calculated.

Tissue samples were oven dried at 65C and stored in a desiccator until analyzed. Carbon and nitrogen content were determined using a Perkin Elmer CHN Elemental Analyzer and phosphorus content was determined colorimetrically following wet digestion (Downes 1988). A generalized linear

TABLE 1. DEFINITION OF HEALTH GROUPS FOR H. RETICULATUM GROWTH.

Health group	Colour nets	Size of nets	No. daughter nets	
1	Yellow	one size	none	
2	Green	some variability	none	
3	Green	range sizes	some	
4	Green	range sizes	many	



Figure 1. Location of sample sites in North Island, New Zealand.

model (Nelder and Wedderburn 1972) with a logit link function was fitted to produce a logistic curve for the probability of nuisance growth against the logarithm of DIN concentration or C:N ratio. For this analysis health group 1 and 2 populations were grouped together as inactive populations as there was no evidence of daughter net production. Groups 3 and 4 were classified as active populations as daughter nets were present indicating actively growing populations.

#### **RESULTS AND DISCUSSION**

Temperatures ranged between 16.0 and 28.3C, well within the range known to support *H. reticulatum* growth. At 30 of the 64 sites sampled, *H. reticulatum* was not present. The presence/absence data showed no significant difference between the presence/absence groups on the basis of water nutrient concentrations at the sites. DIN values for the sites where *H. reticulatum* was absent ranged from 3.1 to 3509 mg m<sup>3</sup> and 0.5 to 1504 mg m<sup>3</sup> at the sites where it was present. DRP values ranged from 0.09 to 90 mg m<sup>3</sup> at sites where *H. reticulatum* was absent and from 0.09 to 96.80 mg m<sup>3</sup> at sites where it was present. This suggests that nutrient concentrations alone do not explain the distribution of *H. reticulatum* in the region.

Of the 34 sites where *H. reticulatum* populations occurred, 8 were classified in health group 1, 8 in group 2, 11 in group 3 and 7 in group 4. Neither DRP concentrations or temperature showed no significant difference between health groups with high variability in DRP and temperatures over all groups and no trend with increasing health status (Table 2). DIN however, showed a significant difference between health groups with a trend of increasing DIN with increasing health status (ANOVA, p < 0.05). Group 1 had a mean concentration of 65.4 and was significantly different from groups 3 and 4 with mean concentrations of 390 and 384 mg m<sup>3</sup> respectively (Table 2).

Tissue nutrient concentrations showed a similar trend to water nutrients with no apparent trend or significant difference between health groups on the basis of the C:P ratio with means for each group in the range of 52 - 79 (Table 2). The C:N ratio, however, showed a significant difference between health groups with a trend of decreasing C:N ratios with increasing population health (ANOVA p < 0.05). The mean of 23.8 for group 1 was significantly different from groups 2, 3 and 4 with means of 14.9, 11.6 and 9.4, respectively.

To determine the DIN concentration likely to be limiting growth and occurrence of natural populations the value which best splits the unhealthy (group 1) populations from the healthy populations (groups 3 and 4), needs to be determined. This line must minimize the misclassification of pop-

Table 2. Water nutrient concentrations (Mg  $\rm M^3)$  and tissue nutrient ratios for each health group (mean  $\pm$  standard error).

Parameter	Health group					
	0	1	2	3	4	
DIN	420±140	65±58	135±75	390±74	384±129	
DRP	21±5	21±10	26±12	23±8	35±13	
C:N ratio	_	$24\pm4$	$15\pm1$	$12\pm1$	9±1	
C:P ratio	_	79±10	$79\pm21$	75±18	$52\pm\!8$	

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Figure 2. Dissolved inorganic nitrogen concentrations of water and carbon: nitrogen ratio of *H. reticulatum* tissue with health groups plotted.

ulations. By definition, the group 2 sites should be split by this line as they fall between healthy and unhealthy populations. Figure 2 shows the line of best fit to be approximately 30 mg m<sup>3</sup>. This line results in one misclassification each side of the line. A similar approach can be applied to the C:N ratio data. The line of best fit was a C:N ratio of approximately 18 and results in one misclassification either side of the line (Figure 2).

The rapid spread of *H. reticulatum* has increased the need to be able to assess the risks of nuisance growths occurring in some areas. To enable assessment of the probability of nuisance growth occurring over a range of DIN concentrations, a logit link function was fitted to produce a logistic curve for the probability of nuisance growth (Figure 3). This allows the assessment of the probability of active or inactive population occurring at a given DIN concentration. For example, at a DIN value of 400 mg m<sup>-3</sup>, the probability of inactive populations is 0.2 and of vigorous growth 0.80, at a DIN of 4 mg m<sup>-3</sup> the probability of active populations growth 0.85 (Figure 3).

The same procedure using the C:N ratio data can also allow predictive assessments to be made. Figure 4 gives the probability curves for the active and inactive populations based on the C:N data. For example, a C:N ratio of 10 shows a probability of 0.80 of active populations and 0.2 for inactive populations, at a ratio of 25 the probabilities are 0.04 and 0.96 for active and inactive populations respectively.

The data collected in this study indicates that the health of field populations of *H. reticulatum* can be related to ambient DIN concentrations with mean DIN increasing with increasing health of the population (Table 2). DIN rather than  $NO_3$  or  $NH_4$  concentrations have been used as Stary et al. (1987) showed *H. reticulatum* is capable of taking up and utilizing both  $NO_3$  and  $NH_4$ .

Analysis of the data suggests that a DIN concentration of approximately 30 mg m<sup>3</sup> (Figure 2) required to support growth of field populations of *H. reticulatum*. This is close to the 20 mg m<sup>3</sup> identified by Hawes and Smith (1993) in labo-



Figure 3. Probability curve of active population of H. reticulatum occurring in relation to dissolved inorganic nitrogen concentration.

ratory studies as likely to support rapid growth. Nutrient requirements for growth determined under laboratory conditions are difficult to apply to field data and the close agreement of these values is surprising. Field concentrations of limiting nutrients and the ability of algae to take them up, vary over short-time scales (e.g., Gordon et al. 1981) and health of a population is a result of integration of this variability. Our field samples were a snapshot of this variation. There are a number of reasons why the two figures differ. The differences could be due to differences between strains of H. reticulatum. Wood and Leatham (1992) have shown considerable physiological differences can occur between strains of the same species. The initial inoculum of H. reticulatum into New Zealand is thought to have been small and potentially a single strain, but this has not been established. There may also be differences due to physiological or genetic adaptation by isolated field populations. The differences observed may also be due to potential seasonal variability in environmental factors contributing to variability in nutrient limitation as suggested by Fong et al. (1993).

A DIN requirement of approximately 30 mg m<sup>3</sup> to support healthy growth in field populations however confirms that relatively low concentrations of DIN will support growth of the alga and is consistent with the suggestion of Hawes and Smith (1993) that a major factor for *H. reticulatum*'s significant success in New Zealand may be due to its ability to grow at low ambient N concentrations.

Using a DIN of 30 mg m<sup>3</sup> and applying it to the presence/ absence data in this study, the absence of H. reticulatum from 13 out of the 30 sites can be explained on the basis of insufficient N to support a population growth. Using the DRP concentration of 5 mg m<sup>-3</sup> suggested by Hawes and Smith (1993) to support rapid growth of *H. reticulatum*, could explain the absence of the alga at a further 3 sites. The absence of H. reticulatum at the other 14 sites is unlikely to be related to nutrient concentrations. Similarly, light is unlikely to be limiting growth as alga is able to grow well at 10% surface irradiance (Hawes and Smith 1993). Factors which may be limiting the establishment of *H. reticulatum* populations are flow rate, salinity and lack of inoculum, although the latter is unlikely given the large number of waterfowl moving between waterbodies. H. reticulatum has been reported elsewhere as appearing only intermittently in some waterbodies (Sörensen 1950, Corrilion and Guerlesquin 1977). This phenomenon may



Figure 4. Probability curve of active population of *H. reticulatum* occurring in relation to tissue C:N ratio.

also be occurring in New Zealand as four sites where *H. reticulatum* was absent in this study had previously had *H. reticulatum* present.

The ability to assess the likelihood of H. reticulatum growth in a particular waterbody is an important management tool. The use of the curves in Figure 3 enables the probability of a healthy or unhealthy population of H. reticulatum to be assessed for a given DIN concentration. Caution must be exercised at this stage as the curves are formed from a relatively small number of data points from one-off samplings. There is a need to increase the robustness of the curves by increasing the number of sites and undertaking repeat samplings at the sites.

The tissue C:N ratio was a good predictor of population health with C:N ratios decreasing significantly with increasing health (Table 2). The C:N ratio shown to be associated with growth, is approximately 18 or below. This upper limit is higher than the C:N ratio of 10 suggested by Hawes and Smith (1992), from laboratory studies. Differences between laboratory and field data were also reported by Rosemarin (1982) where C:N ratios supporting growth in field populations were lower than those established using laboratory studies for *Cladophora*. A tissue C:N ratio of 18 is very close to

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the critical tissue N concentration for growth of 21 mg g-1 dry weight established for an estuarine *Cladophora* in laboratory culture (Gordon et al. 1981). Hanisak (1979) showed result in field and culture studies with a critical C:N ratio 13.5 for *Codium fragile*.

Once *H. reticulatum* has invaded a waterbody being able to predict whether nuisance growths are likely to occur can be important for waterbody management. The use of a logit link function analysis of the tissue C:N ratio data from this study can be used. Use of the curves in Figure 4 enables the probability of active and inactive populations for a given tissue C:N ratio. These curves are likely to give a better prediction of growth than those for DIN as it is well established that internal cellular concentrations of nutrients are better predictors of growth than external nutrient concentrations (e.g. Hecky and Kilham 1988).

The C:N ratio probability curves also have potential as a research tool. For example, in Figure 2 there is one site which has a health group of 1 and a C:N ratio of 8.3. The probability curves suggest that at that C:N ratio the chances of an inactive population is approximately 3% indicating that some factor other than nitrogen concentration is likely to be restricting growth. Using the probability curves in this

way will enable researchers to identify sites for further investigation of controlling factors. This may assist in developing potential control methods for nuisance populations of *H. reticulatum*.

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