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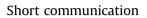
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1. Introduction

Excessive nitrogen input may change ecosystem composition and functioning (Galloway et al., 2003; Stevens et al., 2011). Most of the additional nitrogen that is received by ecosystems nowadays is due to atmospheric ammonia and nitrogen oxide, originating from agriculture, transport and industrial activities (Tilman, 1993; Thomas et al., 1999; Reich et al., 2001; Galloway et al., 2008). In general, atmospheric deposition of nitrogen leads to increased productivity and thereby to a decrease in species number by the exclusion of less competitive species (De Schrijver et al., 2011), and to an acceleration of the succession from grassland to woodland. As a result, species-rich dune grassland becomes increasingly rare in areas like The Netherlands that suffer from high nitrogen deposition (Provoost et al., 2011). Mitigating measures are applied to limit effects of nitrogen on the vegetation, both at the source of the pollutants (e.g. filters, catalysts) and at the polluted sites (e.g. sod cutting, mowing, grazing; Kelly et al., 2002; Tarason et al., 2003). Grazing is standard management in many semi-natural

ABSTRACT

Excessive nitrogen input in natural ecosystems is a major threat to biodiversity. A coastal dune area near Amsterdam in the Netherlands suffers from high atmospheric nitrogen deposition affecting sensitive habitats such as fixed coastal dunes with herbaceous vegetation ('grey dunes'). To mitigate its effect year round grazing was applied from 2007 until 2012. In winter, when natural food supply is low, the cattle received supplementary hay that caused additional inputs of nitrogen. Estimates based on nitrogen contents of hay, as well as of manure, showed the input through winter feeding (c. $3-14 \text{ kg N ha}^{-1}.\text{y}^{-1}$) is in the same order of magnitude as both the actual deposition (c. $17 \text{ kg N ha}^{-1}.\text{y}^{-1}$) and the critical load for a number of herbaceous habitat types ($10-15 \text{ kg N ha}^{-1}.\text{y}^{-1}$). Locally, the effect of winter feeding adds to the effect of nitrogen redistribution within the area caused by the cattle's terrain usage. We conclude that winter feeding may aggravate effects of atmospheric nitrogen deposition.

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grasslands to keep vegetation open and short. When grazing is applied year round, additional feeding may be necessary during winter. However, this results in additional input of nitrogen, which is not desired in natural areas that already suffer from nitrogen overload. To investigate the magnitude of this effect we set up a simple nitrogen budget for a grazed coastal dune area in the Netherlands, which has been shown to be extremely sensitive to atmospheric deposition of nitrogen (Kooijman et al., 1998; Van Dobben et al., 2014). We attempted to make this budget spatially explicit by tracing the redistribution of nitrogen via the cows' excrements.

2. Material and method

2.1. Study area

We selected the Amsterdam Water Supply Dunes (Amsterdamse Waterleidingduinen; 52°20′ N, 4°32′ E, West of Amsterdam; Fig. 1) for a pilot study. This area is an EU protected 'Natura 2000' site (Council of the European Communities, 1992) hosting several rare vegetation types, typical for nutrient-poor conditions (Fig. 2). In this paper we use the habitat typology (as in Annex I of Council of the European Communities, 1992) refined by subtypes defined for the Dutch situation (as in Annex I of Van Dobben et al., 2014). The area is grazed by cattle to prevent succession of open dune to shrub and forest. The area is also used for water infiltration for the production of drinking water for the Amsterdam area.

Since the 1990s the species-rich, open and low-productive vegetation has been in a process of succession towards a species-poor and more productive tall grass vegetation. The succession is partly ascribed to nitrogen deposition and partly to the near extinction of the rabbit by myxomatosis (Sumption and Flowerdew, 1985) and later by rabbit haemorrhagic disease (Van de Bildt et al., 2006). Also the increasing



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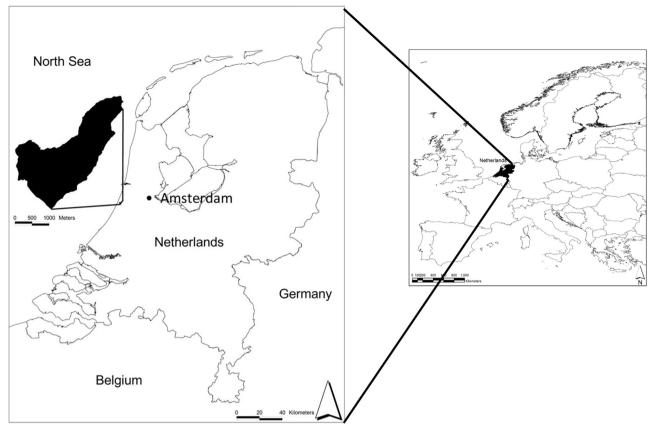


Fig. 1. Location of the study area.

dominance of the exotic invasive species *Prunus serotina* plays a role (Ehrenburg et al., 2008). To counteract these unwanted effects *P. serotina* was removed and year round grazing by cattle, including pregnant cows, was installed in 2007, in a relatively high density in order to tackle regrowth and rejuvenation of *P. serotina*. In winter the cattle received supplementary food in the form of hay harvested outside the area, to compensate for the temporary lack of food (see also Klimkowska et al., 2010).

Our study area is 439 ha, approximately half of which consists of fixed coastal dunes with herbaceous vegetation ('grey dune') which is highly sensitive to nitrogen input (critical load 10–15 kg N/ha/y) depending on the sub-type (Van Dobben et al., 2014) (Table 1, Fig. 2). Approximately 15% consists of oak-birch forest which is also highly sensitive. Less than one third is shrubland dominated by *Hippophae rhamnoides* but also containing *P. serotina*; this type is less sensitive to nitrogen deposition (note that H. *rhamnoides* is associated with the nitrogen-fixing actinomycete *Frankia*). Small parts of the area consist of relatively insensitive wet dune slack and wet forest.

2.2. Nitrogen deposition

We estimated nitrogen deposition on a 250*250 m grid by applying the regional model INITIATOR (De Vries et al., 2003). This model uses input from a geographical information system for agricultural companies (GIAB; Naeff, 2003), including location, animal type and number and farm details. Based on the emission, the deposition from local sources was calculated with the model OPS (Van Jaarsveld, 2004). The background deposition was taken from CPB/PBL (2006) and added to the local deposition. The total deposition in the area ranges between 15.7 and 20.9 kg N ha⁻¹j⁻¹ (Fig. 3).

2.3. Dung heap counts

We counted dung heaps in 60 150 m² areas to estimate the manure input per habitat type (Fig. 3). These areas were randomly selected, stratified over the habitat types indicated in Fig. 2. The input was extrapolated over the whole area, assuming our counts to be representative for each habitat type. We thus assume that cattle uses every habitat patch in the same manner as in the sub-areas where dung heaps were counted. We performed a check to ascertain that the quantity of dung received by each Habitat type was not strongly influenced by the proximity of each Habitat type to the feeding station (Fig. 3) (Klimkowska et al., 2010).

2.4. Calculation of nitrogen input due to supplementary winter feeding

We estimated the additional nitrogen input through supplementary winter food in two ways, namely through hay and through manure.

2.5. Estimation of nitrogen input through hay

For this estimation we hypothesize that the net storage of nitrogen in the cattle's biomass is negligible over the considered period (20 weeks). Also, we hypothezise that all nitrogen excreted by the cattle becomes plant-available i.e. no nitrogen in stored in non-degradable soil organic matter. The cattle were fed with two types of hay: from semi-natural grassland and from agricultural grassland. The hay was presented to the cattle in the form of silage, i.e. after partial fermentation. For both types of hay, the nitrogen content was determined in 5 samples and averaged. The cattle received seven bales per week of semi-natural hay for thirteen weeks, seven bales per week of agricultural hay for two weeks (total feeding period: 20 weeks). We assumed that the cattle consumed all the hay supplied. The N input was calculated as:

$$Ninp = Nbales^*Mbale^*(N/100)$$
(1)

in which: Ninp: N input through winter feeding (kg N y⁻¹), Nbales: total number of bales (91 for the semi-natural hay and 37 for the agricultural hay), Mbale: average mass of a bale (400 kg), %N: N-content of fresh 'hay' (i.e., wet grass silage) (3.6 ± 0.12 and $3.8 \pm 0.34\%$ for the semi-natural and agricultural hay, respectively).

We calculated the total N input as the sum of the values for both types of hay.

2.6. Estimation of nitrogen input through manure

We assumed that cows and calves produce 40 kg and 10 kg of fresh manure per day, respectively (Klimkowska et al., 2010). We collected 15 dung heaps (11 from cows and 4 from calves) and determined their nitrogen content. Most of the N is returned to the system though urine and we also accounted for this by estimating that 30% of the total input is through manure, based on Bokdam (2003). We estimated the N input though manure for both cows and calves as:

$$Ninp = Lp*Nanim*MP*(\%N/100)*(100/30)$$
(2)

in which: Ninp: N input through winter feeding (kg N y⁻¹), Lp: length of winter feeding period (day) (20 weeks = 140 days), Nanim: number animals (40 cows and

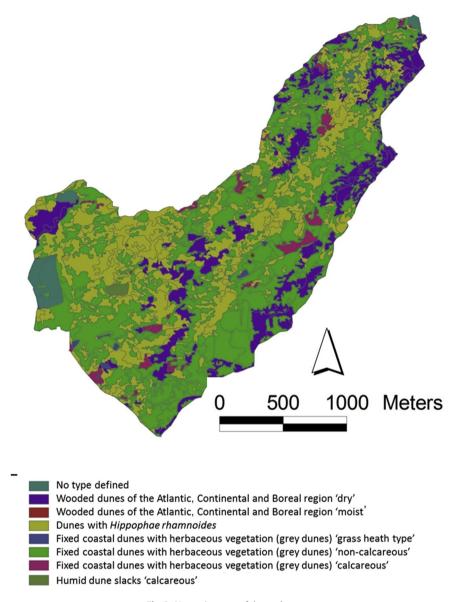


Fig. 2. Vegetation map of the study area.

26 calves), MP: manure production per animal per day (kg.day⁻¹, see above), %N: average N content fresh dung (0.311 \pm 0.011% and 0.400 \pm 0.042% for cows and calves, respectively), 100/30: correction factor for N in urine.

We calculated the total N input as the sum of the values for cows and calves.

We estimated the additional N input per habitat type in two ways. First we used the most straightforward method, which is evenly spreading the total amounts of nitrogen provided by hay or by manure and urine over the whole area. However, these estimates will be biased according to the intensity of the cow's terrain usage. Therefore, we also divided the additional N input over the habitat types in proportion to the quantity of dung found in each habitat type, as follows:

The fraction of the dung that lands in each habitat type can be calculated as:

$$Fract_{t} = (HpCnt_{t}*A_{t}) / \sum_{t} (HpCnt_{t}*A_{t})$$
(3)

in which: Fract_t: fraction of dung landing in habitat type t (-), HpCnt_t: heap count in habitat type t (recalculated to kg.ha⁻¹), A_t: surface areas of habitat type t (ha)

Therefore the amount of nitrogen that lands in each habitat type can be estimated as:

$$Ndep_{t} = (Ninp*Fract_{t}) / A_{t} = HpCnt_{t}*Ninp / \sum_{t} (HpCnt_{t}*A_{t})$$
(4)

in which: Ndept: additional N deposition in habitat type t (kg H ha^{-1} .y⁻¹), Ninp = estimated total additional N input (kg N y⁻¹, from Eqs (1) and (2)).

The estimates of N input through feeding were compared to the actual atmospheric N deposition and the critical loads for N deposition (CL, Van Dobben et al., 2014) of the habitat types in the area, and we investigated to what extent feeding contributed to exceedance of the CL.

3. Results and discussion

The total nitrogen input due to winter feeding was 1869 kg $N.y^{-1}$ estimated on the basis of hay supply and 2808 kg $N.y^{-1}$ estimated on the basis of manure production. If the nitrogen would be distributed equally over the area this would lead to an additional N input of at least 4.3 kg N ha⁻¹.y⁻¹, which is up to 43% of the critical load of its habitat types (Table 1). Thus its contribution to the total N input of the area is not negligible. The total N input exceeds the critical load for grey dunes (all subtypes) and dry wooded dunes. For these sensitive habitat types, the atmospheric deposition alone already causes critical load exceedance, which is aggravated by the N input from supplementary feeding. However, note that the critical load is not exceeded for all vegetation types, even if the N input though feeding is included.

Table 1

N-input as a result of supplementary winter feeding per habitat type, assuming an equal distribution of the extra N over the whole grazed area (ne: not estimated). Habitat types are given with their critical loads (CL).

Habitat type	Atmospheric deposition	Critical load	Input with hay as % of the CL	Sum deposition + input with hay	Deposition + input with hay vs. CL	
	kg N ha ⁻¹ y ⁻¹	kg N ha ⁻¹ y ⁻¹		kg N ha ⁻¹ y ⁻¹		
Dunes with Hippophae rhamnoides	16.4	28	15%	20.7	<cl< td=""></cl<>	
Fixed coastal dunes with herbaceous vegetation (grey dunes) 'calcareous'	16.6	15	28%	20.9	>CL	
Fixed coastal dunes with herbaceous vegetation (grey dunes) 'non-calcareous'	16.4	10	43%	20.7	>CL	
Fixed coastal dunes with herbaceous vegetation (grey dunes) 'grass heath type'	16.6	10	43%	20.9	>CL	
Wooded dunes of the Atlantic, Continental and Boreal region 'dry'	16.7	15	28%	21.0	>CL	
Wooded dunes of the Atlantic, Continental and Boreal region 'moist'	16.3	31	14%	20.6	<cl< td=""></cl<>	
Humid dune slacks 'calcareous'	15.9	20	21%	20.2	≈CL	
Humid dune slacks 'tall reed and sedge vegetation'	16.1	>34	<13%	20.4	<cl< td=""></cl<>	
no habitat type	16.6	ne	ne	20.9	ne	

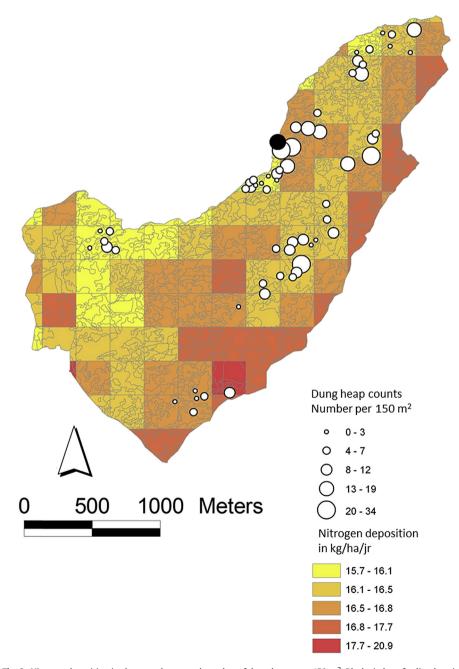


Fig. 3. Nitrogen deposition in the grazed area, and number of dung heaps per 150 m^2 . Black circle = feeding location.

Table 2

N-input as a result of supplementary winter feeding per habitat type, estimated on the basis of dung heap counts according to Eq. (4).

Habitat type	Heap count Number. 150 m ⁻²	Surface area	Additional N by winter feeding		Atmospheric Critical N deposition load		N input through feeding as % of atmospheric input	
			kg N ha ⁻¹ y ⁻¹					
			Through hay	Through manure			Through hay	Through manure
Dunes with Hippophae rhamnoides	4.42	137.02	3.39	5.10	16.4	28	21%	31%
Fixed coastal dunes with herbaceous vegetation (grey dunes) 'non-calcareous'	8.19	207.11	6.29	9.45	16.6	10	38%	57%
Fixed coastal dunes with herbaceous vegetation (grey dunes) 'calcareous'	11.75	11.24	9.02	13.56	16.4	15	55%	83%
Fixed coastal dunes with herbaceous vegetation (grey dunes) 'grass heath type'	0.00	1.29	0.00	0.00	16.6	10	0%	0%
Wooded dunes of the Atlantic, Continental and Boreal region 'dry'	0.00	68.89	0.00	0.00	16.7	15	0%	0%
Wooded dunes of the Atlantic, Continental and Boreal region 'moist'	0.00	0.30	0.00	0.00	16.3	31	0%	0%
Humid dune slacks 'calcareous'	0.00	1.35	0.00	0.00	15.9	20	0%	0%
Humid dune slacks 'tall reed and sedge vegetation'	0.00	0.04	0.00	0.00	16.1	>34	0%	0%
no Habitat type	0.00	11.85	0.00	0.00	16.6	ne	ne	ne

If the additional N input is divided over the habitat types according to the dung heap counts the input through winter feeding may amount to 83% of the atmospheric nitrogen input (Table 2). However, this may be an over-estimate of the net input as the cattle also feeds on natural vegetation during winter and the N in manure therefore partly originates from the terrain itself and not from the hay. Based on the input through hay the contribution of winter feeding to N input is up to 55% of the atmospheric input which is still considerable.

The dung heap counts indicate that cows most often defecate in areas with a relatively low atmospheric nitrogen deposition (Fig. 3). Thus the effect of the manure input on the total nitrogen input is relatively high there. Moreover, the cows relatively often defecate in habitat types that are vulnerable for nitrogen deposition, especially the grey dunes (Table 2). Even without supplementary winter feeding this could lead to a net nitrogen addition in this habitat. We conclude that grazing, which is often used as a measure to counteract effects of atmospheric nitrogen deposition (Bakker, 1989; Smits, 2010), may unintendedly aggravate deposition effects if applied in combination with winter feeding.

This pilot study is based on a very simple nitrogen budget. The sources of nitrogen (dung and atmosphere) are supposed to be exchangeable. Dung and urine alkalise the soil and thus partly counteract the acidification caused by atmospheric deposition but this effect is not taken into account. Volatilisation of nitrogen is neglected and processes like mineralisation, nitrification, denitrification and nitrogen fixation are not included. Note, however, that the critical load concept does not take these processes into account either, and solely concentrates on the external input of nitrogen to the system (Stevens et al., 2011). In our study two estimates of additional nitrogen input were derived independently and they appeared to be in the same order of magnitude (they differ by a factor 1.5), which in our view adds to the credibility of our figures. The higher estimate based on dung heap count compared to input by hay may be due to nitrogen redistribution within the terrain (that is only included in the dung heap approach) but it may also be due to sampling error or to over-simplification in our budget approach.

It should be noted that in general, grazing does not remove substantial amounts of nitrogen, unless animals (e.g. calves) or animal products (e.g. milk) are taken from the system (Bokdam and Gleichman, 2000). However, the advantage of grazing is that it maintains an open vegetation with bare spots where new succession can start, and thus it may enhance the effect of measures that actively remove nitrogen such as mowing or cutting of trees or shrubs (De Bonte et al., 1999). If active redistribute of nitrogen within the area is required herded cattle may be used (Bokdam and Gleichman, 2000). In general, due to the low productivity, grazing of natural areas has a low or negative economic return and there may be animal welfare issues which is the reason for winter feeding.

In response to our figures the manager removed the cattle from our study area during the winter period after 2012. However, as grazing is an essential part of the dune ecosystem (Provoost et al., 2011) that is under pressure after the collapse of the rabbit population, some management intervention is required to maintain the typical grey dune vegetation. In our area herded sheep are now used to counteract regrowth of *Prunus serotina* after its nearly complete removal.

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