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1 **Point source ammonia emissions are having a detrimental impact on prairie vegetation**

2

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4

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21

1 **Abstract**

2 Prairie grasslands are very species rich, but have declined in their extent considerably due to
3 land-use change and exploitation. Many remaining prairie fragments are situated within an
4 agricultural matrix and can be subject to high levels of atmospheric ammonia deposition from
5 animal units. Three prairie fragments in Minnesota that were located in close proximity to
6 feedlots were selected and 500 m transects studied at increasing distance from the feedlot.
7 Changes in soil pH, soil nitrate concentration and soil ammonium concentration with
8 increasing distance from source were variable between the sites, possibly due to differences in
9 the processing of nitrogen in the soil and the degree of nitrogen limitation.
10 Species richness showed significant negative relationships with ammonia deposition and soil
11 nitrate concentration whereas above-ground biomass showed a positive relationship with
12 ammonia deposition. Both the richness and biomass of non-graminoid species declined with
13 increasing soil nitrate concentration whereas graminoid biomass was positively related to
14 ammonia deposition and richness negatively associated. *Bromus inermis*, a non-native
15 perennial grass, was the main species that increased at high deposition.
16 The results of this study have important implications for the conservation and restoration of
17 prairie grasslands.

18

19 **Keywords**

20 **Ammonia, soil biogeochemistry, Bromus inermis, nitrogen deposition, prairie, species**
21 **richness**

1 **Introduction**

2 Prairie grasslands support a diverse assemblage of plants, invertebrates, birds and mammals
3 but they are under considerable pressure from agriculture and development. The extent of the
4 decline in tallgrass prairie exceeds all other major ecosystems in the US. Prior to European
5 settlement prairies covered large portions of the state of Minnesota but as settlements grew,
6 many were converted to croplands. Now, less than 1% of the original native prairie remains.
7 This usually only survives in areas that are unsuitable for agricultural development and some
8 of this remaining prairie is subject to exploitation by recreation and overgrazing (Samson and
9 Knopf, 1994).

10

11 One of the consequences of such extensive habitat loss is that the remaining areas of prairie
12 grasslands are fragmented; scattered through an agricultural matrix. These are frequently small
13 and isolated, and can be found in close proximity to intensive agriculture and resultant
14 emissions. Intensive animal units, sometimes housing several hundreds of cows, or tens of
15 thousands of poultry provide a very large source of nitrogen (N) pollution in the form of
16 ammonia volatilised from animal waste. Ammonia concentrations close to animal housing can
17 be large but decline exponentially with increasing distance from the source (Pitcairn et al.,
18 1998). This nitrogen is deposited to the vegetation and soil with the potential to negatively
19 impact nearby ecosystems.

20

21 Numerous N addition experiments in different grassland types have shown the potential for N
22 deposition to impact species richness, soil chemistry and soil microbial processes (e.g.
23 Mountford et al. 1993; Phoenix et al. 2003, Wedin and Tilman 1996). Combined with
24 evidence from regional surveys (Stevens et al., 2004; 2006) there is clear support for a loss of
25 diversity in grasslands that can be linked to atmospheric deposition of N. Long-term studies at
26 Cedar Creek in Minnesota have shown declines in species richness over a period of 24 years in

1 response to low level N applications equivalent to levels of atmospheric N deposition (10 kg N
2 ha⁻¹ yr⁻¹) (Clark and Tilman, 2008). At higher levels of N application (20, 95 and 120 kg N ha⁻¹
3 yr⁻¹) species richness declined after two years of N addition with annuals accounting for an
4 increased proportion of the vegetation (Wilson and Tilman., 1991). At the same time the
5 root:shoot ratio of vegetation decreased, productivity increased and individual species changed
6 in their abundance (Wilson and Tilman, 1991; 1991a). Above and below-ground productivity
7 was elevated throughout the year in all N treatments and litter showed the same response. As a
8 consequence of the increased productivity, light penetration into the canopy was reduced
9 (Wilson and Tilman, 1993). Increases in productivity have been reported in response to N
10 addition have also been observed in European grasslands (e.g. Wilson et al., 1995) together
11 with increases in species typical of more fertile conditions (e.g. Kirkham & Kent, 1997). This
12 is not always the case and where N is not the limiting nutrient or where the effects of
13 acidification are dominant there may not be an increase in productivity (e.g. Horswill et al.,
14 2008).

15

16 Changes were also seen in the soil with an increase in the availability of nitrate (Wilson and
17 Tilman, 1993). Increases in available N with increases addition or deposition of N have been
18 widely observed (e.g. Emmett et al., 1995; Buchmann et al., 1996; Magill et al., 1997; Stuanes
19 & Kjonaas, 1998) although many studies have reported increases in ammonium but not nitrate
20 possible due to rapid plant uptake combined with the mobility and lability of any excess nitrate
21 (Matson et al., 2002).

22

23 After six years of N addition to prairie grassland the decline in species richness with increasing
24 N addition was still apparent with colonization decreased and loss of species increased with
25 increasing N. Short perennial forbs were particularly sensitive to high inputs including
26 *Solidago nemoralis* and *Hieracium* sp. (Wilson and Tilman, 2002). Furthermore, in a larger, 12

1 year study of the impact of N on Minnesota grasslands, Wedin and Tilman (1996) showed a
2 loss of species with increasing N deposition with greatest losses occurring up to 50 kg N ha⁻¹
3 yr⁻¹. In these plots C₄ grasses were seen to decline and C₃ grasses increased.

4

5 This study will investigate the impact of atmospheric ammonia deposition from animal units on
6 prairie vegetation and soils at three sites in Minnesota using 500 m transects starting close to
7 the ammonia source. The following hypotheses will be tested: 1) Species composition will
8 change with increasing distance from the ammonia source, with a greater species richness
9 further away from the ammonia source; 2) Biomass will be higher close to the ammonia
10 source; 3) Soil pH, nitrate and ammonia concentrations in the soil will change along the
11 gradient of ammonia deposition; pH is expected to be lower as a result of acidification from N
12 deposition, and nitrate and ammonium concentration higher close to the source.

13

14 **Methods**

15

16 Three study sites in Minnesota were selected where prairie fragments, consisting of a large
17 proportion of native species and thus of high conservation value, were found in close proximity
18 to animal feedlots.

19

20 The first site was an area of prairie grassland between a railway line and a highway near
21 Fairfax, Minnesota. It was on the opposite side of a road, approximately 250 m from a poultry
22 unit housing 10,000 chickens and turkeys. The farm was established in 1990. The prairie
23 extended a kilometer beyond the feedlot, along the road. Dominant species in the prairie
24 grassland included *Rosa arkansana*, and *Bromus inermis*. The second site was located in
25 north-west Minnesota, near Brooten. The prairie covered approximately 80 hectares. Adjacent
26 to the site, on the other side of a small river approximately 800 m away was a feedlot with 150

1 dairy and beef cattle. This farm is thought to be over 100 years old. Dominant species at this
2 site included *Andropogon gerardii* and *Solidago gigantea*. The third site selected was located
3 near Arlington to the south-west of Minneapolis and was also adjacent to a railway. It was
4 approximately 500 m from a large beef unit established in 1997 with 820 cattle and
5 approximately 400 m from an older smallholding with 50 cattle which is over 100 years old.
6 The prairie grassland was limited in its extent to a length of 400 m. Dominant species included
7 *A. gerardii* and *Petalostemon purpureum*. Soils at all three sites were classified as black soils.

8

9 At each site three replicate transects were surveyed. Each transect began at a point where
10 prairie vegetation was close to the animal unit. Samples were collected at 50 m intervals up to
11 400 m from the start of the transect and a final sample at 500 m. The distance between the
12 animal unit and the start point varied. Vegetation was surveyed in 1 x 1 m quadrats, all higher
13 plants were identified to a species level and percentage cover recorded. Soils were collected
14 from each quadrat at a depth of 0-10 cm below the litter layer. Biomass samples were
15 collected from strips 10 cm wide and 3 m long. Strips were placed parallel to the transect
16 originating in the centre of each quadrat. Samples were sorted into functional groups in the
17 field and all vegetation was thoroughly dried and the mass determined.

18

19 Soils samples were air dried and ground to 2 mm. Soil pH was determined using a pH meter
20 with a soil solution of 5 g soil and 25 ml deionised water. Nitrate and ammonium
21 concentrations were determined using a Bran-Luebbe AA3 auto analyser following extraction
22 by shaking with 1M KCl.

23

24 Levels of deposition of ammonia at each of the sampling points was determined using the
25 atmospheric dispersion model AERMOD (US EPA, 2007) with deposition to grassland using
26 a grid size of 25 x 15 m over the whole sampling areas and sampling points modeled

1 individually. Long term meteorological data from the nearest monitoring stations to each site
2 was used to parameterize the model. Emissions were estimated according to the number of
3 animals currently housed in the unit. Emission factors were based on Battye et al. (1994).
4 Within the model the cattle feedlots were treated as area sources and the poultry feedlot was
5 treated as two point sources due to ventilation fans located at one end of the two barns
6 comprising the feedlot. Ventilation rates were assumed to meet state recommended
7 requirements (Jani and Jacobson, 2003). In order to compare sites deposition estimated from
8 the feedlots was added to background deposition taken from the National Atmospheric
9 Deposition Program (2008).

10

11 Data were analysed using simple regression analysis and regression trees in R (R Core
12 Development Team, 2007).

13

14 **Results**

15 *Deposition*

16 Modeled ammonia deposition from the feedlots varied between the three sites depending on the
17 source size and distance between source and sink. Deposition ranged from 0.3 to 2.4 kg N ha⁻¹
18 yr⁻¹, 0.01 to 0.04 and at 0.08 to 0.27 kg N ha⁻¹ yr⁻¹ at sites one to three respectively. At sites one
19 and two deposition showed an exponential relationship with distance from the source. At site
20 three this relationship was complicated by the presence of two animal units (Figure 1).

21 Background deposition of ammonia at the three sites was the same (5.14 kg N ha⁻¹ yr⁻¹).

22

23 *Soil chemistry*

24 Examining all three sites together did not show any significant trends between ammonia
25 deposition and soil chemistry. When the sites were examined individually site one showed a
26 significant negative relationship between ammonia deposition and pH ($r^2=0.57$, $p<0.05$) and a

1 non-significant trend towards increasing soil nitrate concentration with increasing ammonium
2 deposition. There was no relationship between ammonia deposition and soil ammonia
3 concentration at site one. Site two showed very similar results with a significant negative
4 relationship between deposition and soil pH ($r^2=0.77$, $p<0.001$), a non-significant trend towards
5 increasing soil nitrate concentration with increasing ammonia deposition and no relationship
6 with soil ammonium concentration. Site three showed a slightly different response with a non-
7 significant trend towards a reduction in soil pH with increasing ammonia deposition and no
8 relationship with soil nitrate concentration. There was however, a significant relationship
9 between soil ammonium concentration and ammonia deposition ($r^2=0.39$, $p=0.05$).

10

11 Plant species composition

12 Looking at all three sites together, species richness was weakly negatively correlated with
13 ammonia deposition ($r^2=0.22$, $p<0.01$). At individual sites there was no consistent relationship
14 between ammonia deposition and species richness. Looking at all three sites there was also a
15 significant relationship between soil nitrate concentration and species richness ($r^2=0.29$,
16 $p<0.01$). The best fit for this decline in richness is not a linear relationship as it shows greater
17 losses at low nitrate levels (Figure 2).

18

19 Productivity (total above-ground biomass) showed a positive relationship with ammonia
20 deposition across the three sites ($r^2=0.50$, $p<0.001$) (Figure 3). If the outlier at the highest level
21 of deposition is removed the relationship is greatly improved ($r^2=0.76$, $p<0.001$). Productivity
22 and richness were negatively correlated ($r^2=0.35$, $p<0.001$).

23

24 Of the soil and deposition variables examined (ammonia deposition, soil nitrate concentration,
25 soil ammonium concentration and soil pH) graminoid productivity was most closely related to
26 ammonia deposition ($r^2=0.35$, $p<0.001$) showing a positive relationship. Graminoid richness

1 was also most closely correlated with ammonia deposition but with a negative relationship
2 ($r^2=0.36$, $p<0.001$) indicating an increased dominance of a few graminoid species. The
3 biomass of non-graminoid species (dominated by forbs) was most closely related to soil nitrate
4 concentration ($r^2=0.17$, $p<0.05$) as was the richness of non-graminoid species ($r^2=0.31$,
5 $p<0.01$). Both showed a negative relationship with nitrate concentration. Litter biomass
6 showed no consistent trend with increasing N deposition at the three sites.

7

8 The regression tree for species richness (Figure 4) showed the primary importance of ammonia
9 deposition for species richness. It also showed that at higher deposition levels soil nitrate
10 concentration becomes an important driver of richness indicating an additive effect of nitrate
11 concentration not correlated with ammonia deposition.

12

13 There were no significant effects of N deposition on the proportion of native species or the
14 proportion of C₄ grasses compared to C₃ grasses.

15

16

17 **Discussion**

18 Soil chemistry

19 The lack of a consistent response of the nitrate and ammonium concentrations in the soils at the
20 three sites suggests that N cycling and uptake differ at the three sites. This is most likely to be
21 due to different moisture regimes and different degrees of nutrient limitation at the sites. A
22 number of investigations have found that prairie grasslands in this part of the US are
23 commonly N limited (e.g. Wedin and Tilman, 1993). If N is the limiting resource, as it is
24 added to the soil it will be utilized quickly by plants and microbes and will not result in
25 elevated soil concentrations with increasing deposition. The relationship found between
26 ammonia deposition and total above-ground productivity supports this.

1

2 Sites one and two show non-significant trends towards increasing soil nitrate concentration
3 with increasing ammonia deposition, but no relationship between ammonia deposition and soil
4 ammonium concentration. This suggests that the ammonia not taken up by the vegetation is
5 nitrified, converting it to nitrate. This process produces protons acidifying the soil contributing
6 to the significant relationship between soil pH and ammonia deposition. Site three shows a
7 trend for increasing soil ammonium with increasing deposition but no trend with nitrate
8 suggesting that nitrification is not occurring to the same extent. If this site is more strongly N
9 limited than the other two sites plant N uptake and/or soil conditions may be limiting
10 nitrification. Less nitrification would also result in less soil acidification.

11

12 Soil chemical differences between the sites may also be related to amount of time that it has
13 received elevated deposition. N accumulates in the soil over time (Fowler et al., 2004) and
14 long term deposition may give a different response than deposition over a short period of time
15 (Skiba et al., 1999). All of the farms in this study had been established for many years but
16 information on animal numbers and exact times were not available.

17

18 Species composition

19 The general trend found across all three sites for reduced species with increasing levels of N
20 deposition and soil nitrate concentration is something that has frequently been observed in
21 prairie grasslands (e.g. Wilson and Tilman 2002; Wedin and Tilman, 1996). The increase in
22 productivity has also been observed in similar grasslands leading to reduced light penetration
23 of the canopy (Wilson and Tilman, 1993). The increase in available nutrients leads to vigorous
24 growth of those species able to take advantage of the increased availability of N. Species not
25 able to take advantage of these conditions, typically those adjusted to low soil N status, are
26 reduced in their abundance as a result of competition.

1

2 In this investigation the diversity of both grasses and forbs was negatively related to deposition
3 or soil nitrate but productivity of grasses increased, indicating an increased dominance of a
4 small number of grass species. Examining the percentage cover of individual grass species in
5 relation to ammonia deposition showed that most grass species, including the native grass
6 species, declined in their cover however, the invasive non-native grass *Bromus inermis*
7 increased in cover with increasing N deposition ($r^2=0.29$, $p<0.001$; Figure 5). If the outlier at
8 the highest level of deposition is removed the relationship is again considerably improved
9 ($r^2=0.68$, $p<0.001$). *B. inermis* is a fast growing species that can form a dense litter layer.
10 Another non-native perennial pasture grass, *Agropyron repens*, showed similar increases at
11 elevated N deposition in east central Minnesota (Wedin and Tilman 1996).

12

13 The increase in graminoid cover and a loss of non-graminoid species at higher levels of N
14 deposition is a phenomenon that has been observed in a number of habitats world-wide.
15 Stevens et al. (2006) report declines in forb richness and cover in calcifuge grasslands along a
16 national deposition gradient in the UK whilst similar trend have been reported in woodland,
17 heathland and bog communities (e.g. Pitcairn et al., 1998; Heil and Bobbink, 1993; Hogg et al.,
18 1995).

19

20 This study suggests that a close proximity between prairie grasslands and large animal units is
21 linked to degradation in the quality of prairie grassland fragments. Through a loss of species
22 richness and an increase in non-native species, especially the non-native grasses *B. inermis*
23 (this study) and *A. repens* (Wedin and Tilman 1996), the conservation value of a prairie
24 fragment is reduced. Little of the original prairie of North America remains. For instance,
25 only about 2% of the native prairie of Minnesota is still intact. Our results suggest that
26 protection of these fragments may require practices that prevent locally-elevated deposition of

1 ammonium, such as occur from nearby feedlots, manure spreading or use of ammonia
2 fertilizers. Large, year-round ammonia sources, such as feedlots, may require sufficient
3 distance between the source and prairie fragments so that deposition levels return to close to
4 background levels. For the three sources we studied, distances of 0.5 to 1.0 km would seem
5 sufficient, but each case would merit explicit modeling. In total, our results suggest that even
6 small elevations in the soil N concentration can lead to changes in productivity and species
7 richness in prairie-grassland communities that should included in land use planning and
8 conservation.

9

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16 an earlier version of this manuscript.

17

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26 disturbance and nitrogen. *Ecology*, 83, 492-504.

1

2

1 **Figure legends**

2

3 **Figure 1.** Modeled ammonia deposition with distance along transect for sites one, two and
4 three. Size of source, distance of the transect from the source and position of the source
5 relative to the prevailing wind direction all influence the amount of ammonia deposition.

6

7 **Figure 2.** Mean species richness per 1 m² quadrat against mean soil nitrate concentration.

8

9 **Figure 3.** Mean above-ground biomass against total ammonia deposition (modeled feedlot
10 deposition plus background deposition).

11

12 **Figure 4.** Regression tree for species richness. Explanatory variables are given together with
13 threshold values separating the response. Values at the end of branches are mean species
14 richness with that category.

15

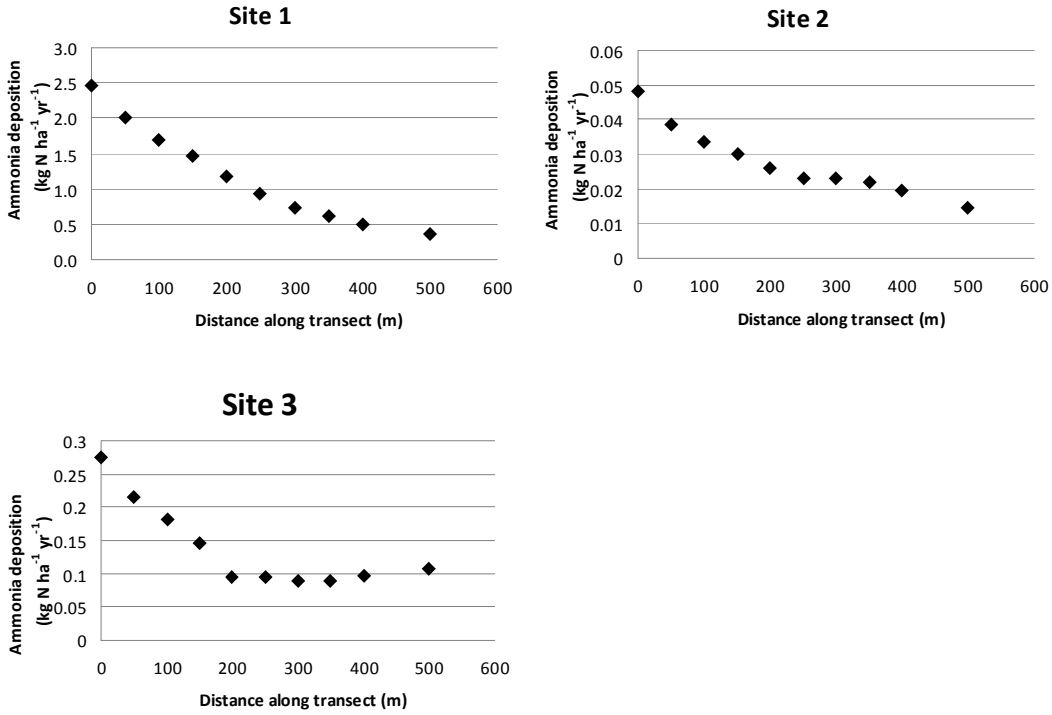
16 **Figure 5.** Mean percentage cover of *Bromus inermis* against total ammonia deposition
17 (modeled feedlot deposition plus background deposition).

18

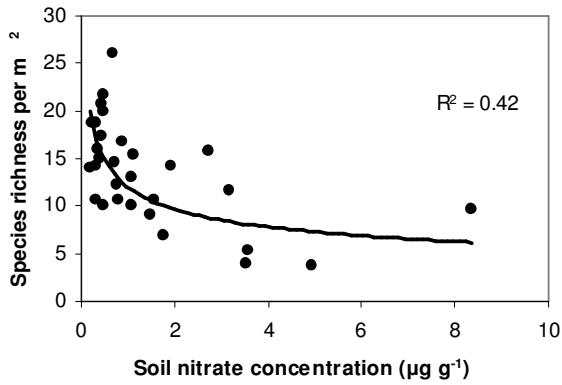
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1 **Figures**

4 Figure 1

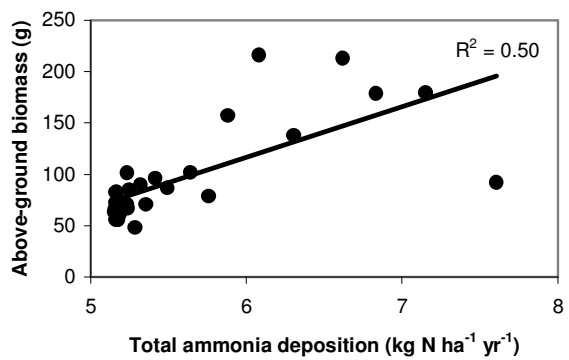


29 Figure 2



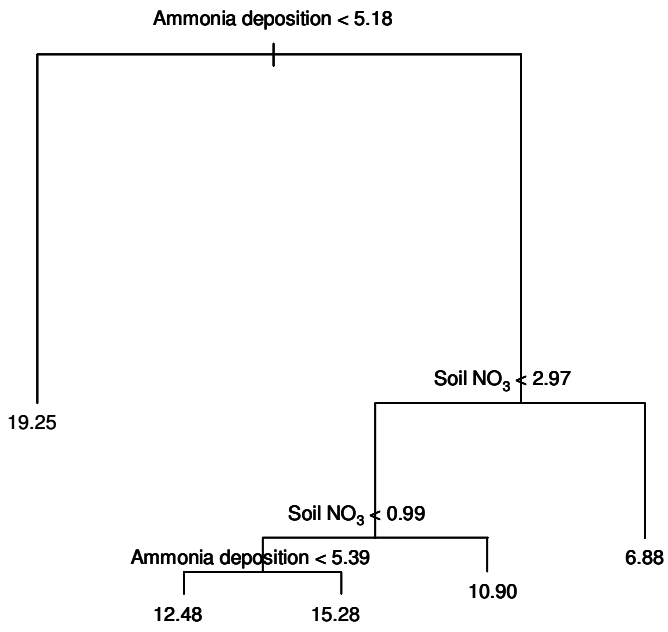
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1 Figure 3
2

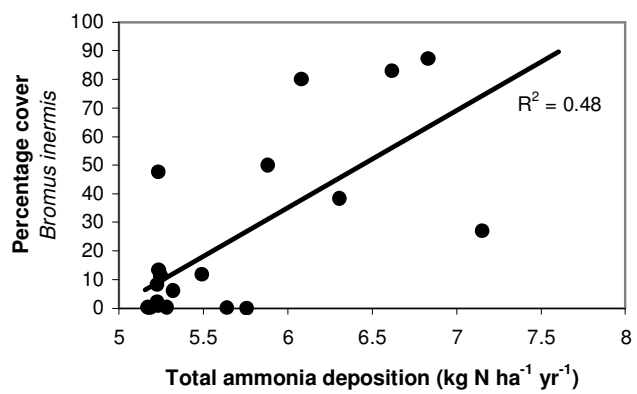


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4
5

1 Figure 4.



1 Figure 5.
2



3