

Supplementary Material: Appendix A

The header of the article: Effects of Long-term Exposure to Ammonia from Animal Farms on the Dry and Dune Heath Vegetation: Lessons from a Gradient Study

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Vegetation and climate data

As part of the study, we examined the vegetation of each site to compile new species lists and compare the species recorded at the sites. Table 1a shows that there is an overlap of only four species between the sites. However, since our study includes only plant species recorded within the plots, the actual species overlap between the sites is likely higher. Table 2a shows climate data from the two sites. There is a remarkable difference in precipitation between the sites, where the dry heath has significantly more annual precipitation (200 mm) compared to the dune heath. Table 3a shows the species list from a survey by Hansen (1976) at the dry heath in 1970. Our results show an overlap in species between the sites; however, there is an apparent discrepancy between the recorded species, which probably is related to the different sampling protocols, where the study by Hansen (1976) specifically targeted the rare species while our plots were placed along a transect.

Table 1a The species and genera that were recorded at the dune heath (H) and the dry heath (T). The number of pinpoint hits is the combined number of hits from the two sites

Species	Number of pinpoint hits	Sites
<i>Achillea millefolium</i>	1	T
<i>Acrocarp(Campylopus introflexus)</i>	2	T
<i>Acrocarp(Dicranum)</i>	2	H
<i>Ammophila arenaria</i>	2	H
<i>Calamagrostis epigejos</i>	1	T
<i>Calluna vulgaris</i>	34	T & H
<i>Carex arenaria</i>	15	H
<i>Carex canescens</i>	1	T
<i>Carex pilulifera</i>	2	T
<i>Cladonia spp.</i>	6	H
<i>Avenella flexuosa</i>	15	T & H
<i>Empetrum nigrum</i>	24	T & H
<i>Festuca ovina</i>	1	T

<i>Galium saxatile</i>	6	T
<i>Hieracium umbellatum</i>	1	T
<i>Hylocomium splendens</i>	1	H
<i>Hypochoeris radicata</i>	1	T
<i>Molinia caerulea</i>	15	T
<i>Phalaris arundinacea</i>	1	T
<i>Pilosella officinarum</i>	1	T
Pleurocarp(<i>Hypnum</i>)	2	T
<i>Pleurozium schreberi</i>	12	T & H
<i>Polypodium vulgare</i>	1	H
<i>Populus tremula</i>	2	T
<i>Potentilla erecta</i>	5	T
<i>Vaccinium myrtillus</i>	3	T
<i>Vaccinium uliginosum</i>	2	T
<i>Vaccinium vitis-idaea</i>	9	T

Table 2a Mean temperature and yearly precipitation from the dune heath (Holmsland) and the dry heath (Trehøje) from 2011 to 2020. Data from DMI (DMI, 2020)

Site	Temperature	Accumulated yearly precipitation
Dune heath (Holmsland)	9.66	789.82
Dry heath(Trehøje)	8.95	1026.11

Table 3a List of species of bryophytes, lichens, and vascular plants recorded in our study and by (Hansen, 1976) from the dry heath (Trehøje)

Species	Our study	The survey conducted (Hansen, 1976)
<i>Achillea millefolium</i>	X	
<i>Agrostis capillaris</i>		X
<i>Arctostaphylos uva-ursi</i>		X
<i>Arnica montana</i>		X
<i>Avenella flexuosa</i>	X	X
<i>Calamagrostis epigejos</i>	X	
<i>Calluna vulgaris</i>	X	X
<i>Campylopus introflexus</i>	X	
<i>Carex arenaria</i>		X
<i>Carex canescens</i>	X	
<i>Carex nigra</i>		X
<i>Carex panicea</i>		X
<i>Carex pilulifera</i>	X	X
<i>Cladonia sp.</i>		X
<i>Empetrum nigrum</i>	X	X
<i>Erica tetralix</i>		X
<i>Festuca ovina</i>	X	X
<i>Galium saxatile</i>	X	X
<i>Genista anglica</i>		X
<i>Genista pilosa</i>		X
<i>Hieracium umbellatum</i>	X	X
<i>Hypnum sp.</i>	X	
<i>Hypochoeris radicata</i>	X	
<i>Luzula multiflora</i>		X
<i>Maianthemum bifolium</i>		X
<i>Molinia caerulea</i>	X	X
<i>Phalaris arundinacea</i>	X	
<i>Pilosella officinarum</i>	X	
<i>Pleurozium schreberi</i>	X	
<i>Populus tremula</i>	X	
<i>Potentilla erecta</i>	X	X
<i>Salix repens</i>		X
<i>Scorzonera humilis</i>		X
<i>Solidago virgaurea</i>		X
<i>Trichophorum cespitosum</i>		X
<i>Trientalis europaea</i>		X
<i>Vaccinium myrtillus</i>	X	X
<i>Vaccinium uliginosum</i>	X	X
<i>Vaccinium vitis-idaea</i>	X	X
Total species richness	22	29

Table 4a Results from the cover of groups from the dry heath omitting data from grazed plots. Significant effects are in bold (here, there are none). The covariate “Nitrogen deposition” is the total nitrogen deposition. The covariate “Distance” is “Distance in meters from the farm”. The covariate “pH” is the soil pH in the bleached sand layer. The table shows the parameter’s average and credibility interval with the 0.025, 0.5 and 0.975 quantiles

	Mean	0.025	0.5	0.975
The dry heath (Trehøje): Dwarf shrubs				
Intercept	6.992	-2.244	7.002	16.168
Nitrogen deposition	-0.455	-1.281	-0.456	0.372
Distance	-0.002	-0.004	-0.002	0.005
pH	-2.49	-5.041	-2.498	0.11
The dry heath (Trehøje): Graminoids				
Intercept	-2.283	-9.527	-0.298	9.044
Nitrogen deposition	0.089	-0.77	0.09	0.942
Distance	-0.002	-0.01	-0.002	0.006
pH	-0.052	-2.769	-0.049	2.643
The dry heath (Trehøje): Forbs				
Intercept	-9.387	-21.724	-9.436	3.229
Nitrogen deposition	0.793	-0.402	0.794	1.976
Distance	0.003	-0.009	0.003	0.015
pH	1.295	-2.247	1.316	4.718

Fig. 1a View over the dry heath dominated by *C. vulgaris*, in the front with bog bilberry *Vaccinium uliginosum* L. and english oak *Quercus robur* L. The tall green grass is *M. caerulea*



For constructing the two meshes, we used the INLA package in R (Belmont, 2022; Lindgren and Rue, 2015; R Core Team, 2021).

Fig. 2a A diagram of the mesh constructed at the dune heath (Holmsland). The transect points plots are the blue dots

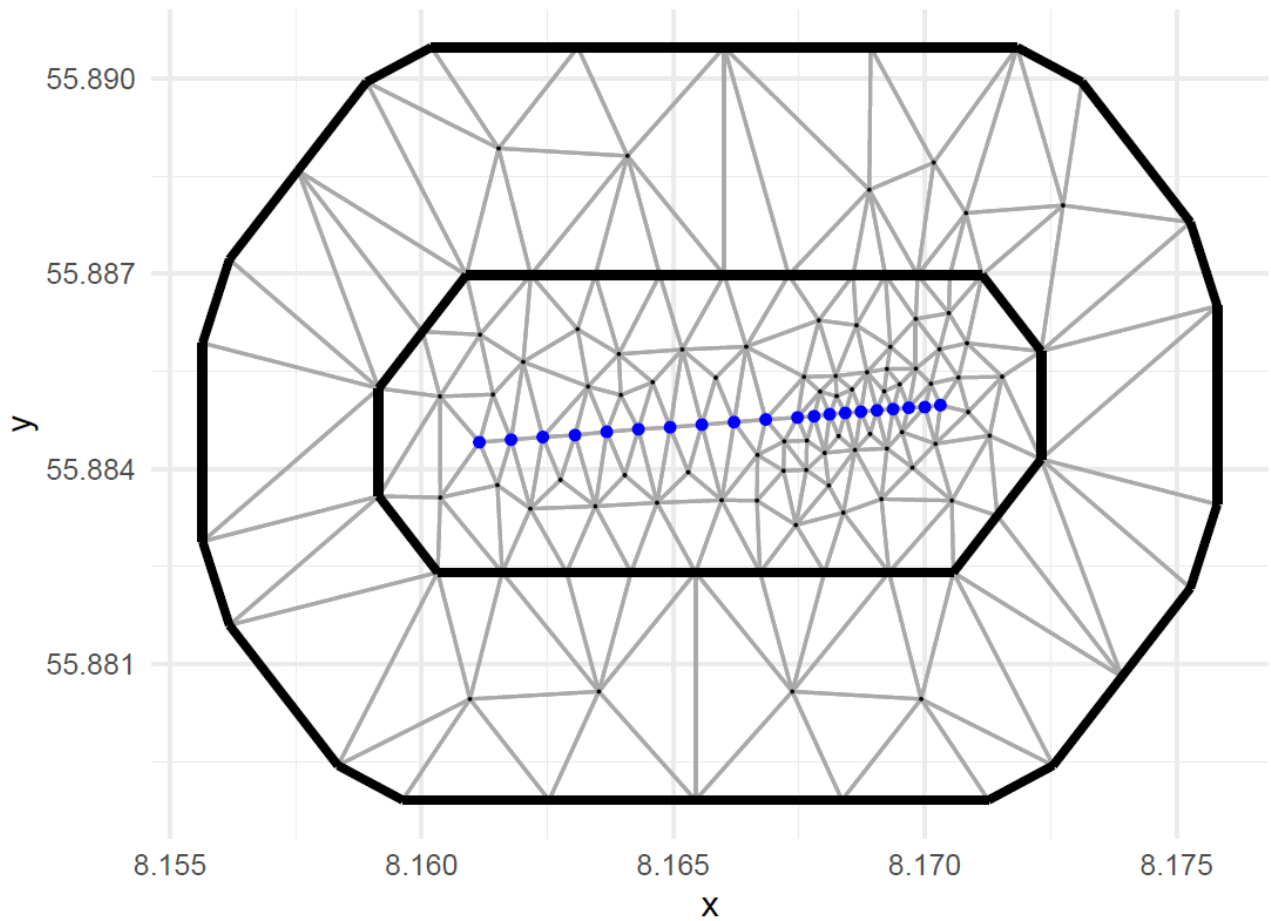


Fig. 3a A diagram of the mesh constructed at the dry heath (Trehøje). The transect plots are the blue dots. The disconnected points line was due to a farm field between the heathland remnants

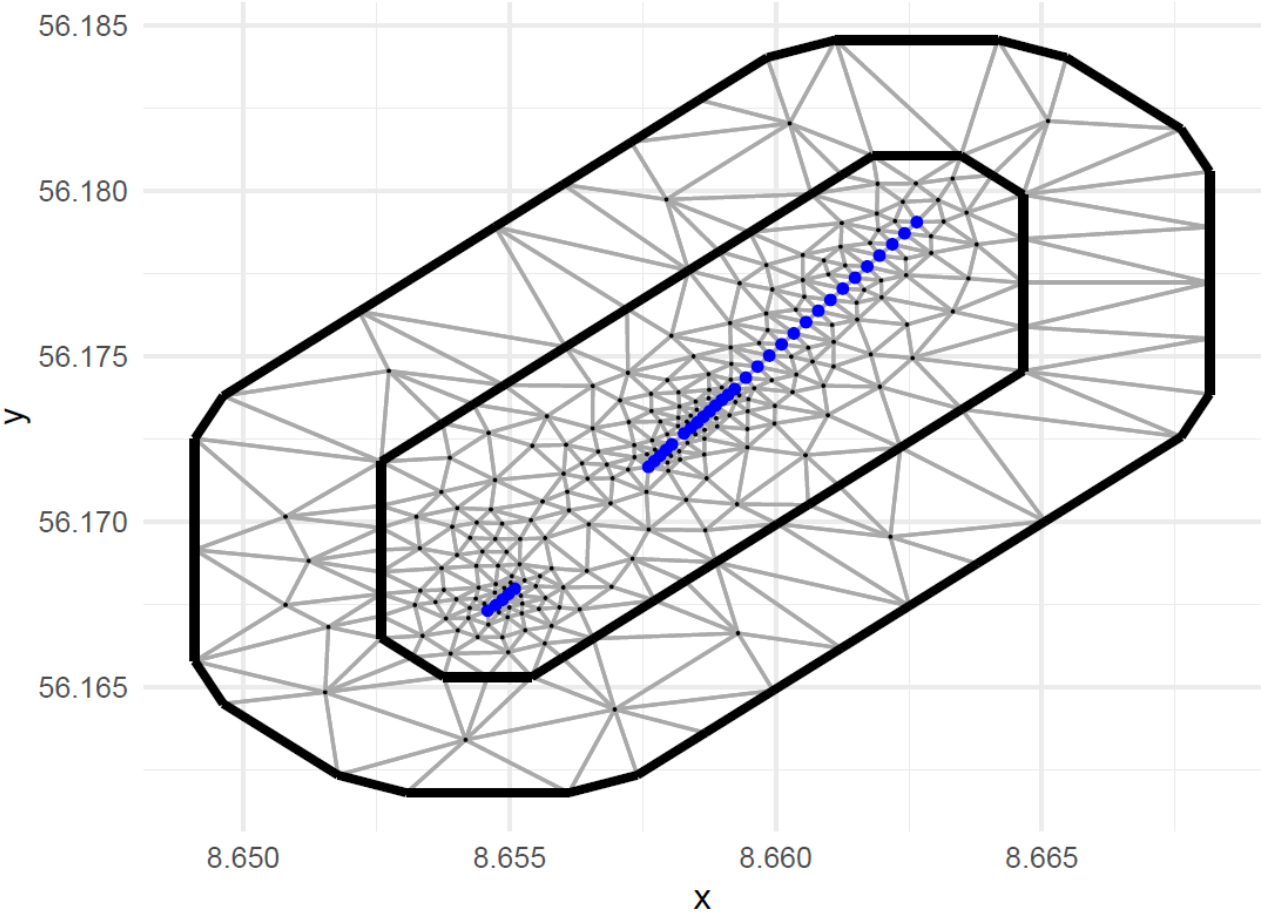


Fig. 4a Bearberry *Arctostaphylos uva-ursi* and *C. arenaria* at Trehøje heath. Unlike the results from Hansen (1976), where bearberry was reported to occur solely at south-facing slopes, this population of bearberry primarily occurred on hills facing west



Fig. 5a The rare forb wolf's bane (*Arnica montana*) in the East-central part of the dry heath with *C. vulgaris* and pill sedge *Carex pilulifera*



Fig. 6a. The model syntax for the cover data

```
#####  
#####  
  
# Make models for Bryophytes  
  
# Start with loading the packages.  
  
library(RcmdrMisc)  
  
library(INLA)  
  
library(leaflet)  
  
library(viridis)  
  
library(geoR)  
  
library(rgdal)  
  
library(tibble)  
  
library(dplyr)
```

```

library(ggplot2)

library(raster)

library(inlatools)

# Load gradient data

library(readxl)

rdata <- read_excel("Specify our path")

rdata$Y=as.integer(rdata$Y)

summary(rdata)

# Non-spatial analysis of cover

nsmodel1=inla(Y~1, data=rdata, family="betabinomial", Ntrials=25)

summary(nsmodel1)

# Coordinate matrix

coords<-as.matrix(rdata[,2:1])

n <- nrow(rdata)

CRS("+init=epsg:4326")

utmproj <- "+proj=longlat +datum=WGS84 +no_defs"

spdat <- rdata

coordinates(spdat) <- ~ East+North

proj4string(spdat) <- "+init=epsg:4326"

new <- spTransform(spdat, CRS(utmproj))

plot(new)

loc1 <- SpatialPoints(as.matrix(coordinates(new)))

class(loc1)

# Create a mesh

mesh.loc <- SpatialPoints(as.matrix(coords))

mesh1 <- inla.mesh.2d(loc=loc1,

                      max.edge=c(0.005, 0.01),

                      min.angle=c(30, 21),

                      max.n=c(48000, 16000), ## Safeguard against large meshes.

```

```
max.n.strict=c(128000, 128000), ## Don't build a huge mesh!  
cutoff=0.00001, ## Filter away adjacent points.  
offset=c(0.002, 0.0035)) ## Offset for extra boundaries, if needed.
```

Plot the mesh:

```
plot(mesh1)  
  
plot(new, add = T, col = 'red', pch=21)  
  
mesh1$n  
  
#Variogram (geoR)  
  
v1=variog(coords=coords, data=rdata$Y, estimator.type='classical')  
  
plot(v1)
```

Create SPDE object

```
spde1<-inla.spde2.matern(mesh=mesh1,alpha=2)  
  
A.est<-inla.spde.make.A(mesh=mesh1,loc=coords)  
  
s.index<-inla.spde.make.index(name="spatial.field",  
                               n.spde=spde1$n.spde)
```

Stationary analysis

```
stationary.stack.est<-inla.stack(data=list(y=rdata$Y),  
                                A=list(A.est),  
                                effects=list(c(s.index,list(Intercept=1))  
                                ),  
                                tag="est")  
  
formula1<-y~1+f(spatial.field,model=spde1)  
  
stationary.output1<-inla(formula1,  
                          data=inla.stack.data(stationary.stack.est, spde=spde1),  
                          family="zeroinflatedbetabinomial1", Ntrials=25,  
                          control.predictor=list(A=inla.stack.A(stationary.stack.est),compute=TRUE),  
                          control.compute=list(waic=TRUE))  
  
summary(stationary.output1)
```

Non-stationary analysis pHsand.

```
non-stationary.stack.est<-inla.stack(data=list(y=rdata$Y),  
                                     A=list(A.est,1,1,1),  
                                     effects=list(c(s.index,list(Intercept=1)),  
                                                  list(ndep=rdata$Ndep),  
                                                  list(dist=rdata$Distance),  
                                                  list(ph=rdata$pHsand)  
                                     ),  
                                     tag= "est")
```

```
summary(nonstationary.stack.est)
```

Model 1 only with nitrogen deposition

```
formula1<-y~1~ndep+f(spatial.field,model=spde1)  
nonstationary.output1<-inla(formula1,  
                             data=inla.stack.data(nonstationary.stack.est, spde=spde1),  
                             family="zeroinflatedbetabinomial1", Ntrials=25,  
                             control.predictor=list(A=inla.stack.A(nonstationary.stack.est),compute=TRUE),  
                             control.compute=list(waic=TRUE),verbose=TRUE)
```

Check summary output

```
summary(nonstationary.output1)
```

#Model 2 with all terms

```
formula2<- y~ndep+dist+ph+f(spatial.field,model=spde1)  
nonstationary.output2<-inla(formula2,  
                             data=inla.stack.data(nonstationary.stack.est, spde=spde1),  
                             family="betabinomial", Ntrials=25,  
                             control.predictor=list(A=inla.stack.A(nonstationary.stack.est),compute=TRUE),  
                             control.compute=list(waic=TRUE))
```

Check summary output

```
summary(nonstationary.output2)
```

Fig. 7a The model syntax for the vertical density data

```
#####  
#####  
  
# Analyse graminoid vertical density from the dry heath  
  
library(RcmdrMisc)  
  
library(INLA)  
  
library(leaflet)  
  
library(viridis)  
  
library(geoR)  
  
library(tibble)  
  
library(dplyr)  
  
library(ggplot2)  
  
library(inlatools)  
  
# Load gradient data  
  
library(readxl)  
  
rdata <- read_excel("Specify your path")  
  
rdata$Y=as.integer(rdata$Y)  
  
summary(rdata)  
  
str(rdata)  
  
# Non-spatial analysis of cover  
  
nsmodel1=inla(Y~1, data=rdata, family="nbinomial", Ntrials=25)  
  
summary(nsmodel1)  
  
# Coordinate matrix  
  
coords<-as.matrix(rdata[,2:1])  
  
n <- nrow(rdata)  
  
CRS("+init=epsg:4326")  
  
utmproj <- "+proj=longlat +datum=WGS84 +no_defs"  
  
spdat <- rdata
```

```

coordinates(spdat) <- ~ East+North
proj4string(spdat) <- "+init=epsg:4326"
new <- spTransform(spdat, CRS(utmproj))
plot(new)

loc1 <- SpatialPoints(as.matrix(coordinates(new)))

# Mesh

mesh.loc <- SpatialPoints(as.matrix(coords))

mesh1 <- inla.mesh.2d(loc=loc1,

    max.edge=c(0.005, 0.01),

    min.angle=c(30, 21),

    max.n=c(48000, 16000), ## Safeguard against large meshes.

    max.n.strict=c(128000, 128000), ## Don't build a huge mesh!

    cutoff=0.00001, ## Filter away adjacent points.

    offset=c(0.002, 0.0035)) ## Offset for extra boundaries, if needed.

```

Plot the mesh

```

plot(mesh1)

plot(new, add = T, col = 'red', pch=21)

mesh1$n

#Variogram (geoR)

v1=variog(coords=coords, data=rdata$Y, estimator.type='classical')

plot(v1)

```

Create SPDE object

```

spde1<-inla.spde2.matern(mesh=mesh1,alpha=2)

A.est<-inla.spde.make.A(mesh=mesh1,loc=coords)

s.index<-inla.spde.make.index(name="spatial.field",

    n.spde=spde1$n.spde)

```

Stationary analysis


```
control.predictor=list(A=inla.stack.A(nonstationary.stack.est),compute=TRUE),
control.compute=list(waic=TRUE))
```

Check summary output

```
summary(nonstationary.output1)
```

Model 2 fit with all the terms

```
formula2<-y~ndep+dist+ph+man+f(spatial.field,model=spde1)
```

```
nonstationary.output2<-inla(formula2,
```

```
data=inla.stack.data(nonstationary.stack.est, spde=spde1),
```

```
family="nbinomial", Ntrials=25,
```

```
control.predictor=list(A=inla.stack.A(nonstationary.stack.est),compute=TRUE),
```

```
control.compute=list(waic=TRUE))
```

Check summary output

```
summary(nonstationary.output2)
```

Fig. 8a. The model syntax for the ratio data

```
#####
#####
```

Analyses of ratio data

```
library(RcmdrMisc)
```

```
library(INLA)
```

```
library(leaflet)
```

```
library(viridis)
```

```
library(geoR)
```

```
library(tibble)
```

```
library(dplyr)
```

```
library(ggplot2)
```

```
library(inlatools)
```

Load gradient data

```
library(readxl)
```

```
rdata <- read_excel("Specify our path")
```

```

rdata$Y=as.numeric(rdata$Y)

summary(rdata)

names(inla.models())$likelihood

# Non-spatial analysis of cover

nsmodel1=inla(Y~1, data=rdata, family="beta", Ntrials=25)

summary(nsmodel1)

# Coordinate matrix

coords<-as.matrix(rdata[,2:1])

n <- nrow(rdata)

CRS("+init=epsg:4326")

utmproj <- "+proj=longlat +datum=WGS84 +no_defs"

spdat <- rdata

coordinates(spdat) <- ~ East+North

proj4string(spdat) <- "+init=epsg:4326"

new <- spTransform(spdat, CRS(utmproj))

plot(new)

loc1 <- SpatialPoints(as.matrix(coordinates(new)))

# Create a mesh

mesh.loc <- SpatialPoints(as.matrix(coords))

mesh1 <- inla.mesh.2d(loc=loc1,

  max.edge=c(0.005, 0.01),

  min.angle=c(30, 21),

  max.n=c(48000, 16000), ## Safeguard against large meshes.

  max.n.strict=c(128000, 128000), ## Don't build a huge mesh!

  cutoff=0.00001, ## Filter away adjacent points.

  offset=c(0.002, 0.0035)) ## Offset for extra boundaries, if needed.

# Plot the mesh

plot(mesh1)

```

```

plot(new, add = T, col = 'red', pch=21)

mesh1$N

#Coordinate matrix

coords<-as.matrix(rdata[,2:1])

# Plot of Y

pal <- colorBin("viridis", bins = c(6, 6.5, 7, 7.5))

leaflet(rdata) %>%

  addProviderTiles(providers$CartoDB.Positron) %>%

  addCircles(lng = ~East, lat = ~North, color = ~ pal(Y)) %>%

  addLegend("bottomright",

    pal = pal, values = ~Y,

    title = "Y"

  ) %>%

  addScaleBar(position = c("bottomleft"))

#Variogram (geoR)

v1=variog(coords=coords, data=rdata$Y, estimator.type='classical')

plot(v1)

# Create SPDE object

spde1<-inla.spde2.matern(mesh=mesh1,alpha=2)

A.est<-inla.spde.make.A(mesh=mesh1,loc=coords)

s.index<-inla.spde.make.index(name="spatial.field",

  n.spde=spde1$N.spde)

# Stationary analysis

stationary.stack.est<-inla.stack(data=list(y=rdata$Y),

  A=list(A.est),

  effects=list(c(s.index,list(Intercept=1))

),

  tag= "est")

```

```

formula1<-y~1+f(spatial.field,model=spde1)

stationary.output1<-inla(formula1,

  data=inla.stack.data(stationary.stack.est, spde=spde1),

  family="beta", Ntrials=25,

  control.predictor=list(A=inla.stack.A(stationary.stack.est),compute=TRUE),

  control.compute=list(waic=TRUE))

```

```
summary(stationary.output1)
```

Non-stationary analysis pHsand.

```

non-stationary.stack.est<-inla.stack(data=list(y=rdata$Y),

  A=list(A.est,1,1,1),

  effects=list(c(s.index,list(Intercept=1)),

    list(ndep=rdata$Ndep),

    list(dist=rdata$Distance),

    list(ph=rdata$pHsand)

  ),

  tag= "est")

```

```
summary(nonstationary.stack.est)
```

Model 1 considering only nitrogen deposition

```

formula1<-y~ndep+f(spatial.field,model=spde1)

nonstationary.output1<-inla(formula1,

  data=inla.stack.data(nonstationary.stack.est, spde=spde1),

  family="beta", Ntrials=25,

  control.predictor=list(A=inla.stack.A(nonstationary.stack.est),compute=TRUE),

  control.compute=list(waic=TRUE))

```

Check summary output

```
summary(nonstationary.output1)
```

Model 2 considering all terms

```
formula2<-y~ndep+dist+ph+f(spatial.field,model=spde1)
```

```
nonstationary.output2<-inla(formula2,
```

```
  data=inla.stack.data(nonstationary.stack.est, spde=spde1),
```

```
  family="beta", Ntrials=25,
```

```
  control.predictor=list(A=inla.stack.A(nonstationary.stack.est),compute=TRUE),
```

```
  control.compute=list(waic=TRUE))
```

Check summary output

```
summary(nonstationary.output2)
```

Fig. 9a Picture showing a pinpoint frame near the pig farm within the enclosure at the dry heath, entirely dominated by *C. vulgaris*. A pin was inserted in each cross (in total 25), and we noted the number of times the pin hit a specific plant species. A plant species, for instance, *C. vulgaris*, could have a cover of 1, meaning the pin had hit this species in all 25 cases. Vertical density was the number of times the pin hit a species in each cross within the pinpoint frame



Fig. 10a Showing cover of bryophytes from the dry heath against the distance in meters from the pig farm, nitrogen deposition in $\text{kg N ha}^{-1} \text{ year}^{-1}$, and pH in the mor - and the bleached sand layer. Plot (a) shows the cover of bryophytes against nitrogen deposition levels. Plot (b) and plot (c) show the bryophyte cover against soil pH in the mor or bleached sand layers, respectively. Plot (d) shows bryophyte cover against the distance to the farm. No statistical analyses were employed because of limited data

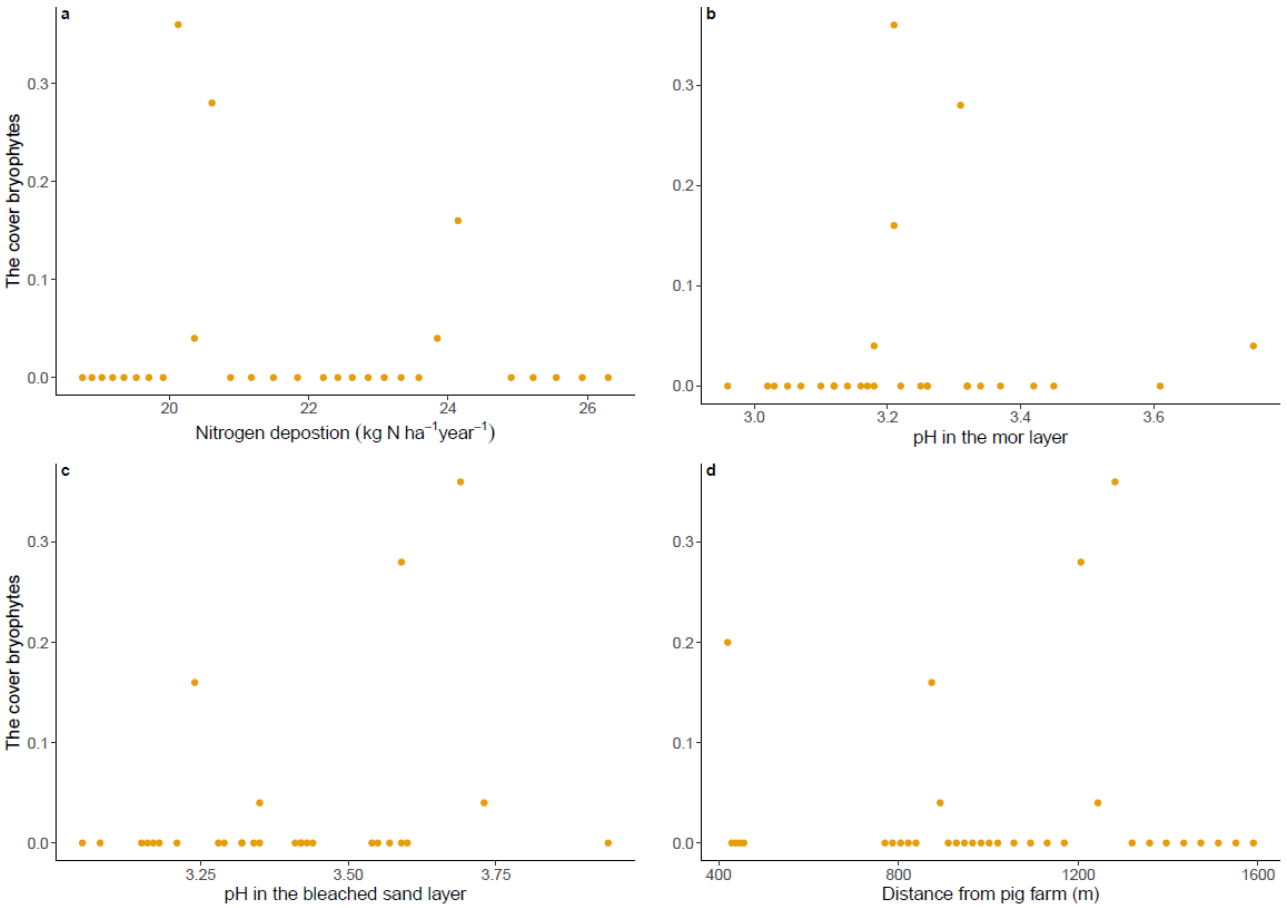


Fig. 11a Showing cover of graminoids from the dune heath. Plot (a) shows a higher graminoid cover with higher nitrogen deposition. Plots (b) and (c) show graminoid cover versus soil pH in the mor - and bleached sand layer. Plot (d) shows graminoid cover versus distance from the farm. The empirical analyses showed just a significant effect of nitrogen deposition

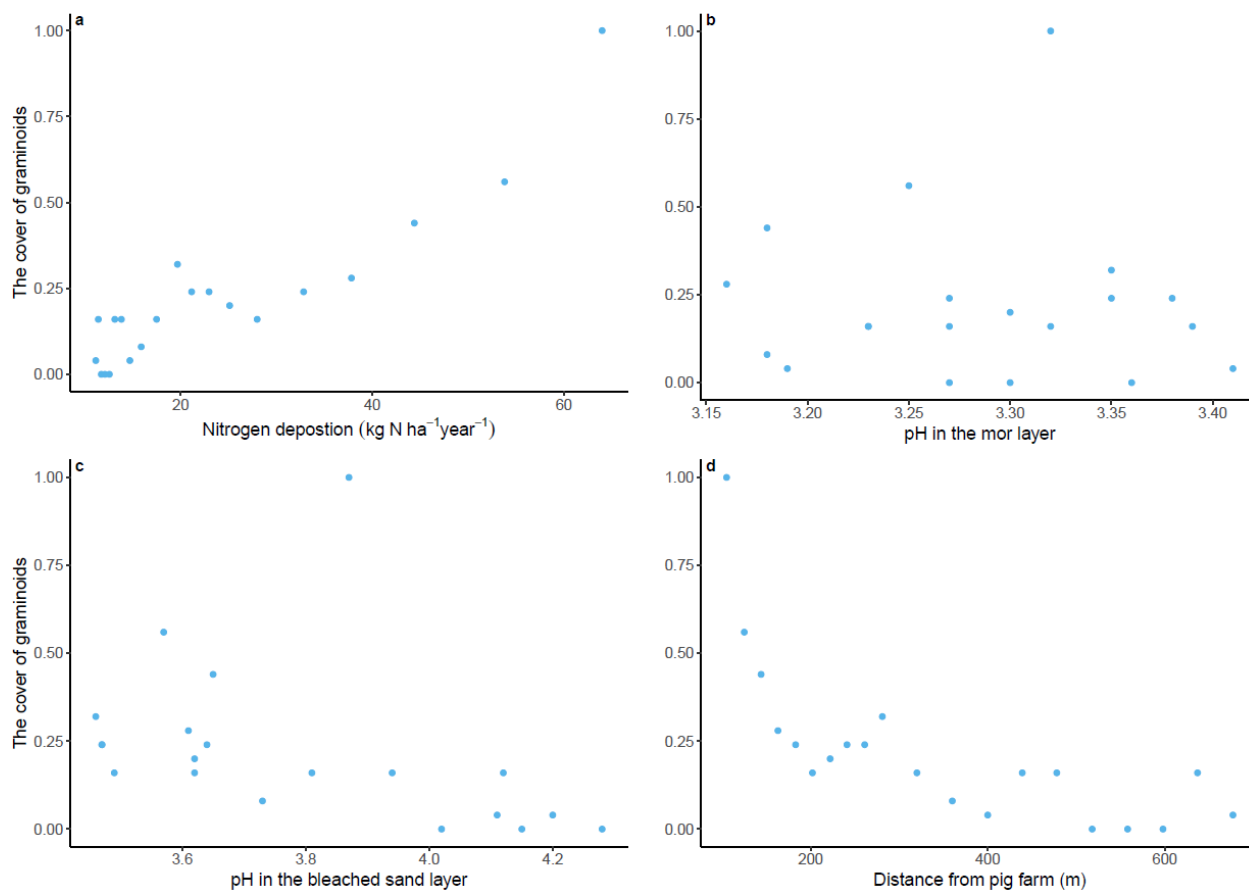


Fig. 12a Showing vertical density (VD) of graminoids from the dune heath. Plot (a) shows the graminoid VD versus nitrogen deposition. Plots (b) and (c) show graminoid VD versus soil pH in the mor - and bleached sand layer. Plot (d) shows the dwarf graminoid VD versus the distance from the pig farm. The empirical analyses showed a significant effect of nitrogen deposition

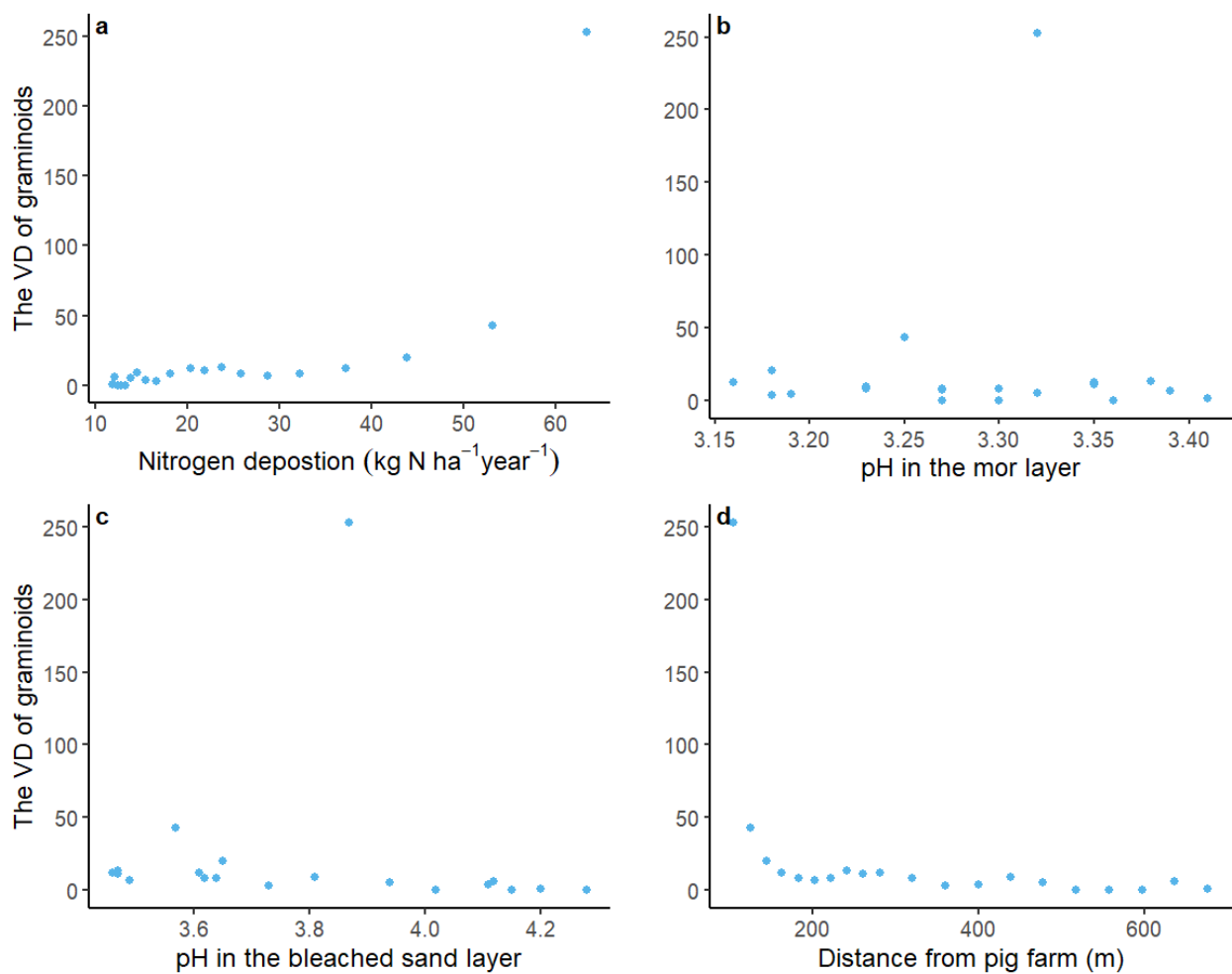


Fig. 13a Showing cover of graminoids from the dry heath. All data from the grazed site are omitted. Plot (a) shows the graminoid cover versus nitrogen deposition. Plots (b) and (c) show graminoid cover versus soil pH in the mor - and bleached sand layer. Plot (d) shows the graminoid cover versus the distance from the farm. The empirical analyses showed no significant effects

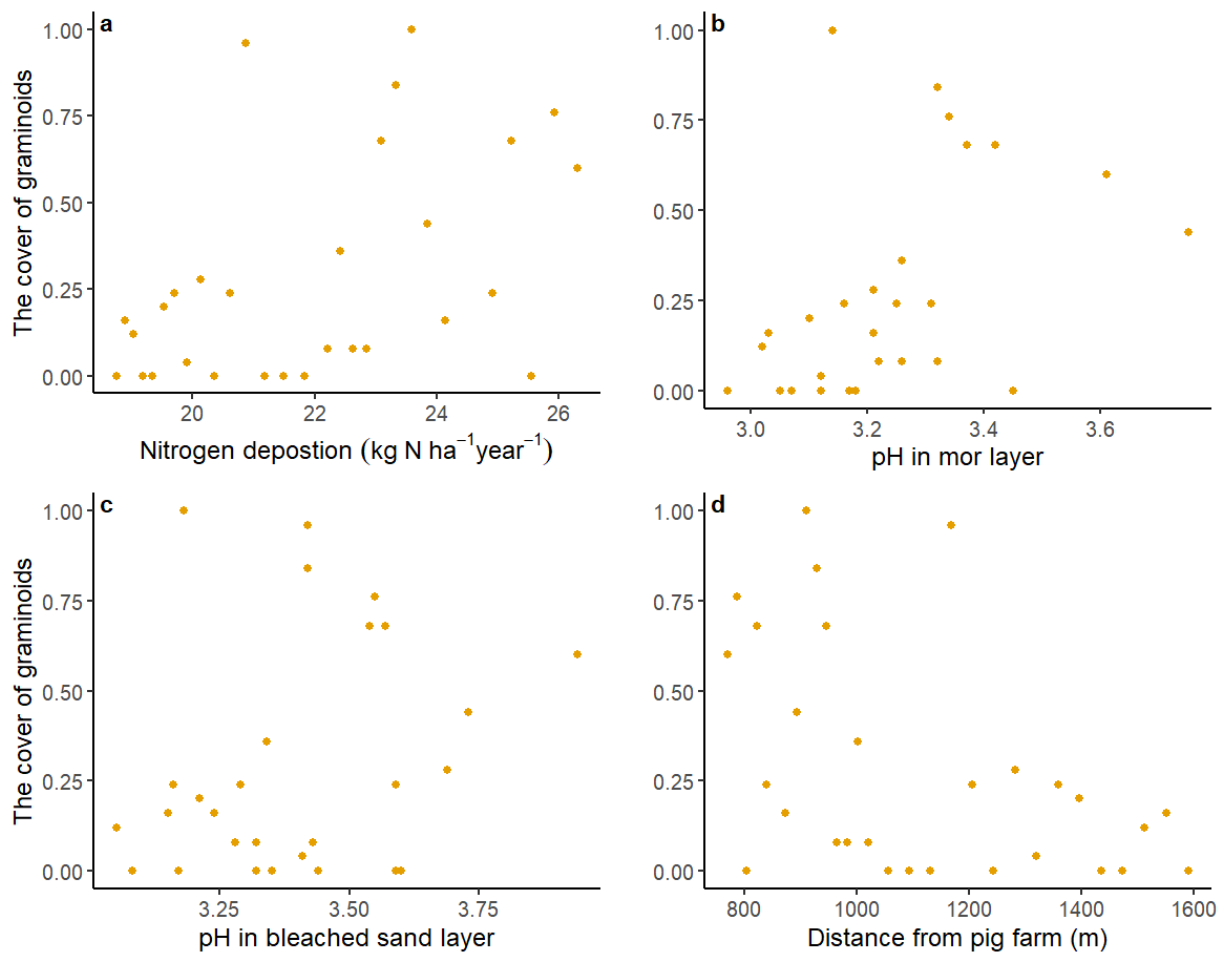


Fig. 14a Showing the graminoid/dwarf shrub ratio data from the dry heath. Data from the grazed site are omitted. Plot (a) shows the graminoid/dwarf shrub ratio versus nitrogen deposition. Plots (b) and (c) show the ratio versus soil pH in the mor - and bleached sand layer. Plot (d) shows the ratio versus distance from the pig farm. The empirical analyses showed no significant effects

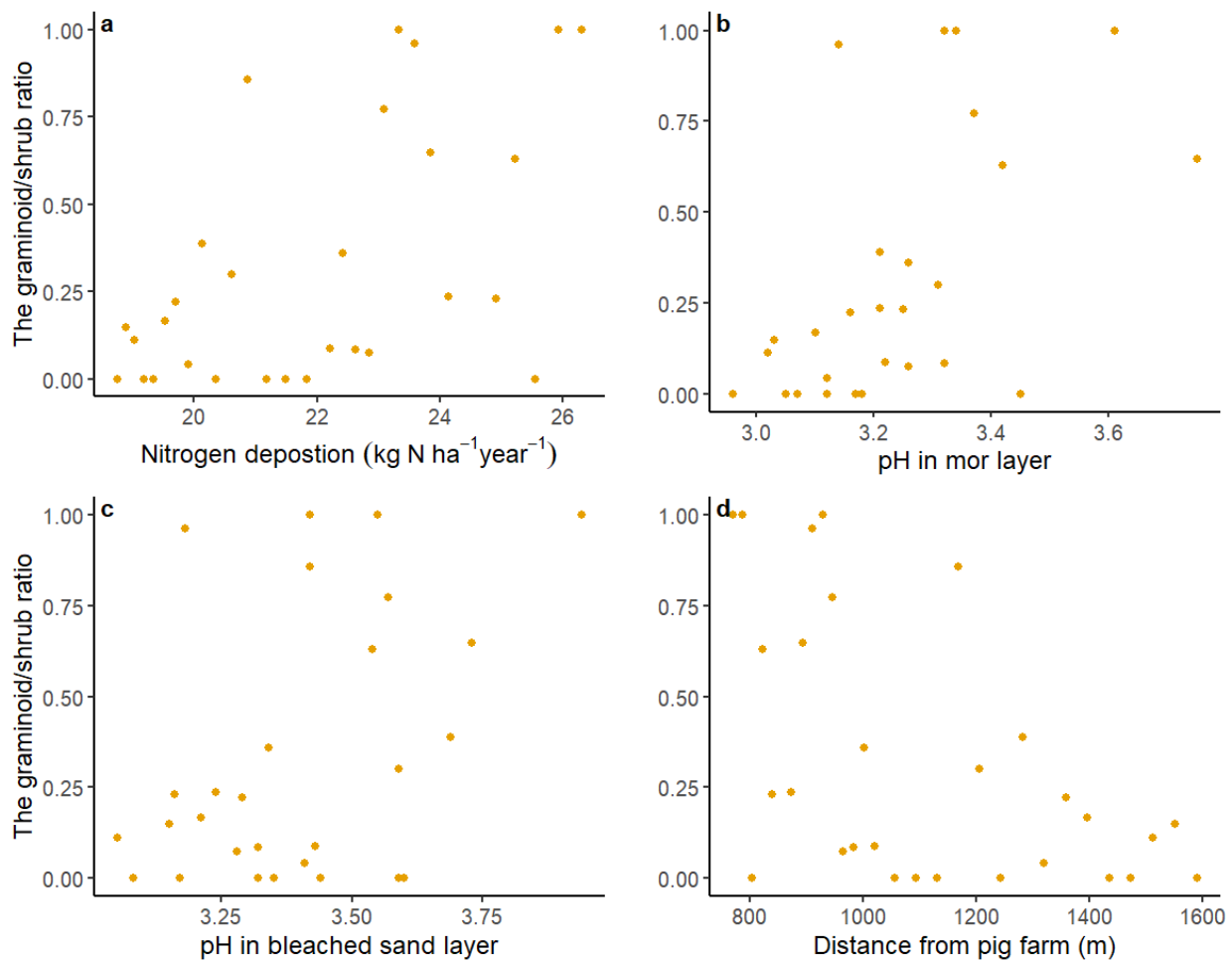


Fig. 15a Showing the vertical density of dwarf shrubs at the dry heath where data from the grazed plots is omitted. Plot (a) shows the dwarf shrub's vertical density versus nitrogen deposition. Plots (b) and (c) shows the dwarf shrub's vertical density versus soil pH in the mor - and bleached sand layer. Plot (d) show the dwarf shrub's vertical density versus the distance from the pig farm. The empirical analyses showed no significant effects

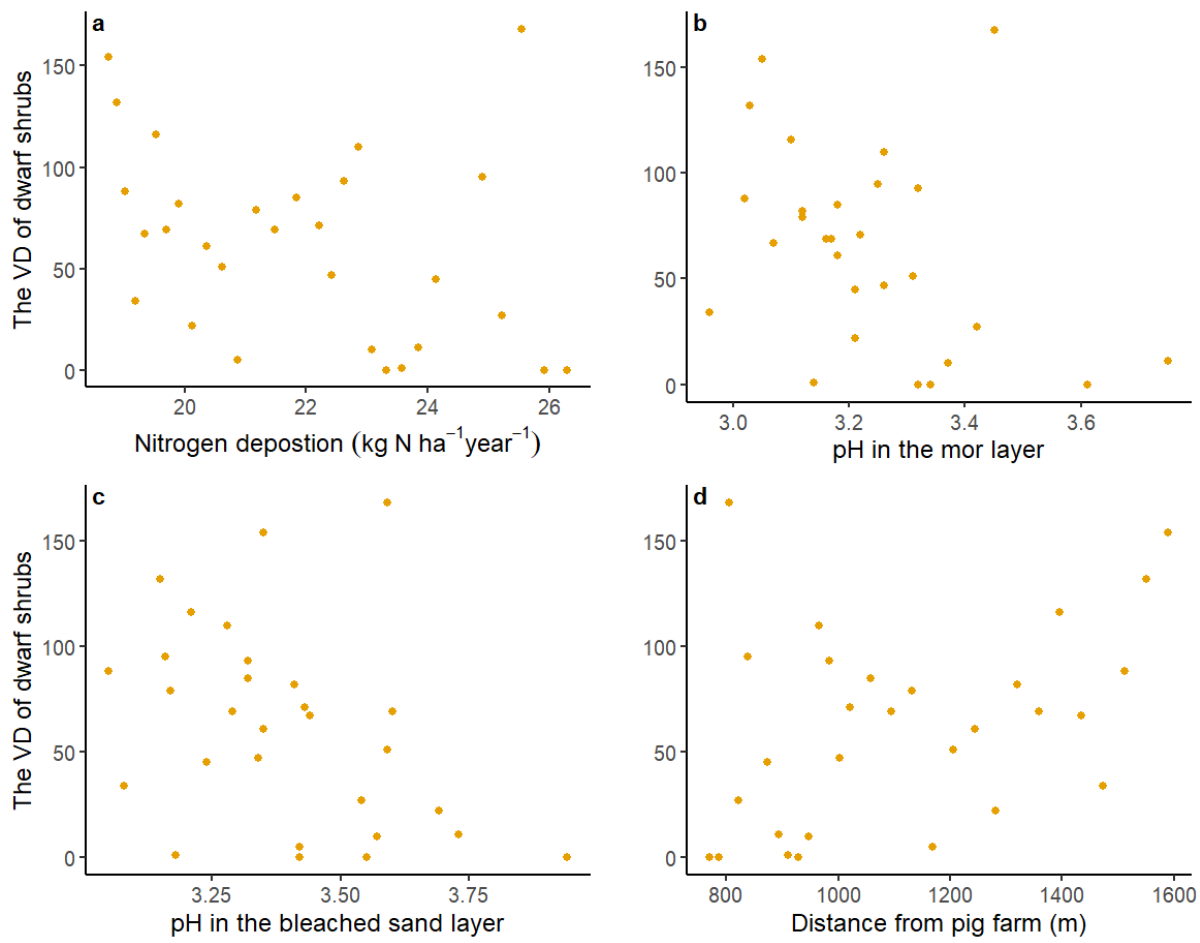
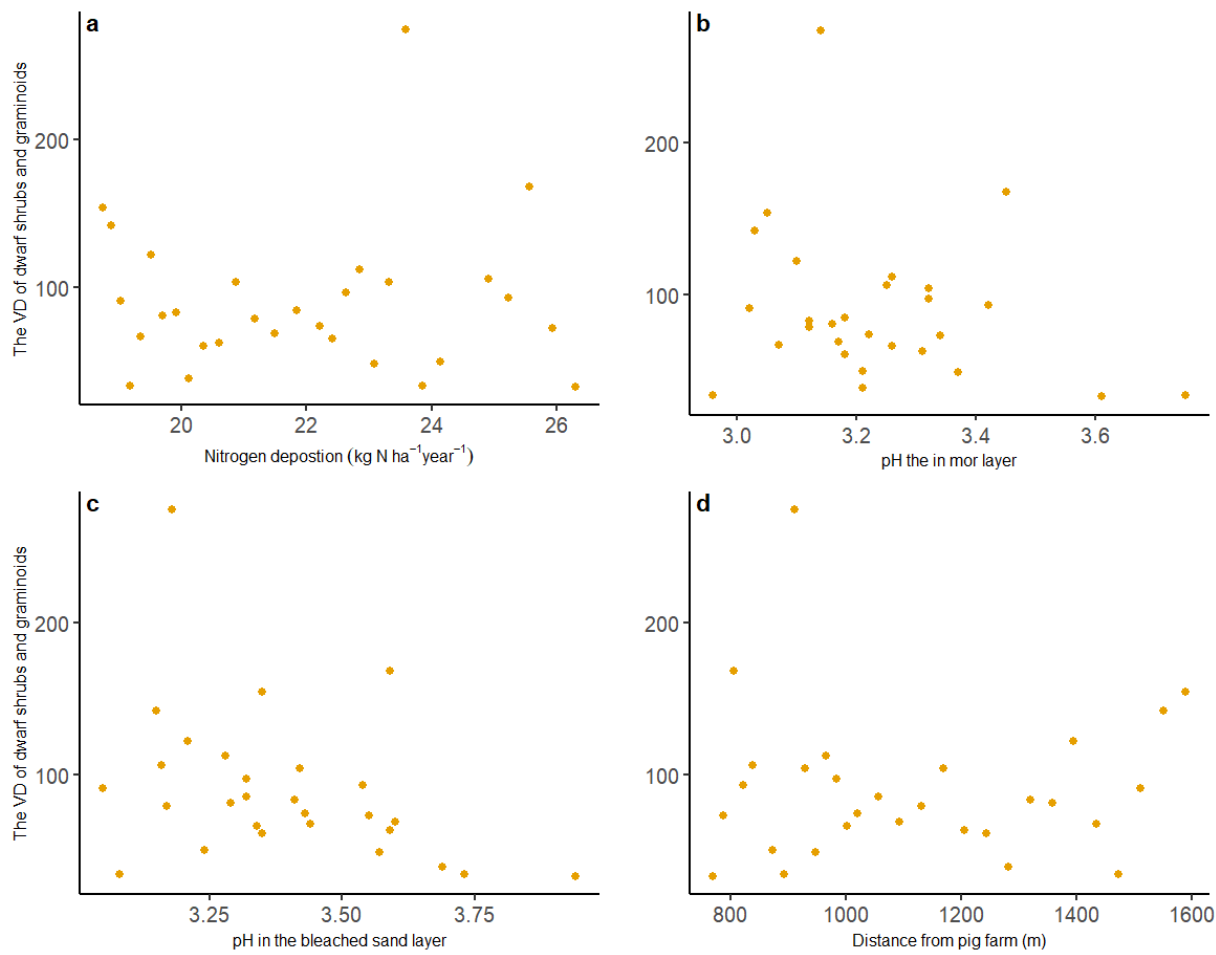


Fig. 16a Showing the vertical density of dwarf shrubs and graminoids at the dry heath where data from the grazed plots is omitted. Plot (a) shows the vertical density versus nitrogen deposition. Plots (b) and (c) shows the vertical density versus soil pH in the mor - and bleached sand layer. Plot (d) show the vertical density versus the distance from the pig farm. The empirical analyses showed a significant effect of pH in the bleached sand layer



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