# Analysis of succession and the relation with hydrogeochemistry in a former tidal wetland in the Grevelingen, The Netherlands over a period of 43 years

What is the influence of soil development processes on the development of the vegetation in a former tidal wetland which is no longer inundated.



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Different types of vegetation at the Slikken van Flakkee<sup>1</sup>

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<sup>1</sup> (Foto: Ilona Noorlander; <u>https://boswachtersaanzee.wordpress.com/2016/05/31</u> /voorjaar-op-de-slikken-van-flakkee)



## Summary

Deltas and estuaries are widely recognized as areas with important high natural values that are under threat by human activities such as coastal defence. The Grevelingen estuary in the South West of The Netherlands was closed off from the North sea in 1971 to protect the area from disasters similar to the storm flood in 1953. The closing of the Grevelingen dam removed the tidal influence from coastal wetlands in the Grevelingen like the 'Slikken van Flakkee' leading to the development of a unique natural area. In the northern part of the area, natural succession has led to the emergence of a diverse landscape containing forests and different types of grasslands. The southern part of the area which has been influenced by nature management in the form of extensive grazing and regular mowing has led to a more uniform landscape containing Red list species with high natural values.

The aim of this research is to determine which hydrogeochemical processes occur at the 'Slikken van Flakkee' area and what their influence is on the different vegetation types. Soil samples were collected from nine representative locations in the field and analysed in the laboratory. Vegetation data gathered since 1972 were analysed to determine the major trends in vegetation succession. We found evidence for desalination, pyrite oxidation and cation exchange. Desalination was the only hydrogeochemical process that has a significant effect on species composition. The observed difference between the vegetation types at the northern and the southern part of the area is due to the different management strategies.

A modelling study is recommended to gain insight into the past and future development of the hydrogeochemical conditions at the 'Slikken van Flakkee' to investigate whether the current management choices suffice to maintain the high valued Red list species in the area.

# Preface

With my background as a marine biologist I have always been interested in the dynamics between human activity and ecosystems in coastal areas. Investigating the relationship between the effects of coastal defence measures was therefore a very interesting subject for my master thesis. During the research and the writing of my thesis I learned a lot about the dynamics between soil processes and ecosystem succession and I got fascinated by the unique species composition found in coastal wetlands. Even though data analysis using the statistical program R was not easy, I learned how gratifying it can be when you manage to visualize your data into insightful images. The whole thesis process also improved my planning and data management skills thanks to the useful advice from my daily supervisor Martin van der Weiden. All things considered I found that writing this thesis was a challenging activity which taught me a lot about fieldwork, labwork and scientific writing and confirmed my interest in the combination of scientific research and practical issues about how our activities as a society influence the world around us.

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## 1. Introduction

The intergovernmental Panel on Climate Change (IPCC) defines deltas as low lying coastal complexes that combine natural and human systems with a population density of 10 times the global average (Wong et al. 2014). Over the past 50 years, sediment reduction, relative sea level rise and changes in delta and river management have led to widespread degradation of deltas. This is expected to intensify as a result of increased coastal flooding, erosion and salt water intrusion because of global sea level rise (Wong et al. 2014).

After the stormflood of 1953 which killed 1800 people and inundated thousands of hectares of land, the delta in the south west of the Netherlands was closed from the sea by the Delta works (Bannink et al. 1984). One of the open sea arms which was closed is the Grevelingen estuary (Figure 1). The closing of the Brouwersdam in 1971 changed the Grevelingen from an open sea arm to a salt water lake with no tidal dynamics (Bannink et al. 1984; van der Pluijm & de Jong 2003). Relatively elevated areas of the coastal wetlands in the Grevelingen which used to be diurnally inundated with salt water are now permanently dry while the lower zones are still permanently below the water level mean water level and regularly flooded (van der Pluijm & de Jong 2003; Van de Haterd et al. 2010).

The largest of these wetlands is the 'Slikken van Flakkee' area. Located along the northern shore of lake Grevelingen. The 'Slikken van Flakkee' consists of an area of 1500ha of former tidal wetlands which are now permanently above the mean water line (van der Pluijm & de Jong 2003). Since 1971, the northern and the southern part of the 'Slikken van Flakkee' have developed into different ecosystems. In the northern part, natural succession has led to the development of thickets consisting mainly of Salix species, Hippophae rhamnoides, and Pteridium aquilinum. The succession in the southern part, that had been managed by grazing with livestock (since about 1980) and regularly mowed, has led to species-rich grasslands including plant species that represent a high natural value and which are on the European Red list for endangered species (Bilz et al. 2011). This includes species such as Parnassia palustris, Blackstiona perfoliatte ssp., Potentilla anserina and orchid species like Epipactis palustris and Dactilorhiza majalis spp. (van der Pluijm & de Jong 2003). Grasslands of this composition are normally found in CaCO<sub>3</sub> rich dune valleys where hydrological and geochemical conditions are comparable to the conditions in the former tidal wetlands (van Haperen 2011). While the northern part of the 'Slikken van Flakkee' has been allowed to develop without human interference, the southern part has been subjected to cattle grazing. According to Van der Pluijm en de Jong (2003) the difference between the northern and southern part of the 'Slikken van Flakkee' is due to these different management styles of both areas (van der Pluijm & de Jong 2003). However, differences in vegetation communities in saline wetlands have also been linked to the hydrogeochemical processes in the soil (Ratas et al. 2003; Wu et al. 2011). Up to now no scientific research had been carried out in the 'Slikken van Flakkee' that could conclude on the relative impact of management versus hydrogeochemistry.

Besides the Grevelingen there are many other areas in the Netherlands where tidal influence have been banned. An example of such an area is the Krammer-Volkerak lake. This former tidal salt water system was cut off from the sea in 1987 and since then flushing with fresh water has changed it into a fresh water lake with a fixed water level (Tosserams et al. 2000). Soil development in the former tidal zone has been largely determined by the composition and the average particle size of the sediment. The relatively high plates are characterized by coarse sandy sediment and relatively fast desalination due to easy penetration and flushing by fresh rain water of the soil. The lower areas consist of finer sandy material with layers of clay leading to less flushing by fresh water and slower desalination (Tosserams et al. 2000). The soil water development at the former tidal wetlands of the Krammer-Volkerak lake has been modelled using the geochemical modelling tool PHREEQC (Parkhurst & Appelo 1999) by Appelo et al. (1998). For this model a sediment core from the dried up wetlands at the Krammer-Volkerak lake was oxidised completely using hydrogen peroxide in order to determine the reactions occurring naturally in the soil when it is aerated (Appelo et al. 1998).

The increased oxidation of the soil of a former tidal wetland can lead to the oxidation of pyrite (FeS<sub>2</sub>), a common mineral in Dutch geological deposits (Roskam & Griffioen 2011). Pyrite oxidation has been found to cause severe acidification of pore water dyked wetlands in the Unites States (Portnoy 1999; Portnoy & Giblin 1997). The following reaction takes place when pyrite is oxidised:

$$FeS_{2}(s) + 15/4O_{2} + 7/2H_{2}O \rightarrow Fe(OH)_{3}(s) + 2SO_{4}^{2-} + 4H^{+}$$
(1)

When sufficient amounts of carbonate are available in the environment, the acid produced by reaction (1) is neutralized by reaction (2) (Nicholson et al. 1988; Appelo et al. 1998).

$$FeS_{2}(s) + 15/4O_{2} + 7/2H_{2}O + 4 CO_{3}^{2-} \rightarrow Fe(OH)_{3}(s) + 2SO_{4}^{2-} + 4HCO_{3}^{-}$$
(2)

After the carbonate is depleted, further acidification is buffered by the solution of calcium carbonate (3) leading to decalcification of the soil (Appelo et al. 1998).

$$CaCO_{3}(s)+H^{+} \rightarrow Ca^{2+} + HCO_{3}^{-}$$
(3)

A third hydrogeochemical process that can change pore water condition is cation exchange (Appelo et al. 1998). During this process cations that are sorbed to soil particles are exchanged between the particles and the pore water by reaction, for instance (4)

$$Ca^{2+} + Mg X_2 \rightarrow Ca X_2 + Mg^{2+}$$
(4)

Ecological, geochemical and hydrological data need to be combined into one ecohydrogeochemical model to quantify the relationship between succession and soil development in wetland soils that are no longer inundated. Creating such a model, which can be used to quantify the effects of different management strategies, is the goal of the PhD research by drs. Martin van der Weiden (Van der Weiden 2015). Figure 2 shows the processes that are considered to determine the development of the soil and the vegetation for wetland ecosystems which are no longer diurnally inundated (Van der Weiden 2015). The current master thesis is set up as a pilot study to determine the relationship between vegetation and soil development at the 'Slikken van Flakkee' in the framework of the envisaged PhD thesis of van der Weiden.

The current master thesis work will combine existing data with actual measurements in the field. Data on vegetation development has been recorded on a yearly basis since 1973 by 'Rijkswaterstaat' and Anton van Haperen (Van Haperen, n.d.). Hydrological and meteorological data are available in the following two 'Rijkswaterstaat' reports:

- Slager, H., Fluijt, D.J. & Rook G.J., 1986. *Waterhuishouding en zouthuishouding van de Slikken van Flakkee*. Rapport nr. 198624abw, Rijksdienst IJsselmeerpolders, Afdeling Delta, Lelystad.

Grevelingen. Flevobericht 312, 'Rijkswaterstaat' Directie Flevoland, Lelystad.

Slager, H. & Visser j., 1990. Abiotische kenmerken van de drooggevallen gebieden in de

Figure 1: Lake Grevelingen (red) in the south west of the Netherlands and the two research areas (black).

Geochemical data will be obtained by taking soil and water samples at representative locations.

Data SIO, NOAA, U.S., Navy, NGA, GEBCO



**Figure 2:** Conceptual diagram of interactions between hydrology, geochemistry and vegetation in dried up marine soils. Translated from Van der Weiden, 2015.

#### **Problem definition**

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Coastal zones are areas with important ecosystem functions and high natural values. Protective measures against sea level rise like the Deltaplan in the Netherlands have lead to changes in hydrology,

Google earth

geochemistry and ecology in former tidal ecosystems causing changes in ecosystem functioning. Areas that used to be diurnally inundated like the 'Slikken van Flakkee' are now permanently dryer and rarely flooded. Different geological conditions and management styles have led to the development of diverse ecosystems, some with high natural values. However, continuing hydrogeochemical processes like desalination and decalcification may lead to a loss of plant communities with high natural values and abundance of Red list species. In order to choose management measures that will contribute to the preservation of the highest natural values it is important to be able to predict the development of the hydrogeochemical conditions in the soil.

#### Aim

The aims of this research are:

- 1. to analyse the vegetation succession at the 'Slikken van Flakkee' since the closing of the Grevelingen from the North Sea which ended the diurnal flooding by the tides;
- 2. link vegetation succession to the hydrogeochemical development of the soil;
- 3. understand differences in succession and vegetation development between the northern and southern sub-area (the latter being mown and grazed and the former having no management).

#### **Research Question**

The main research question for this thesis is:

How is the ecological succession of vegetation related to the hydrogeochemistry of the soil in this former tidal wetland?

To answer the main research question the following sub questions were formulated:

- 1. How has the vegetation at the 'Slikken van Flakkee' developed since the closing of the Grevelingen in 1971?
- 2. What are the current hydrogeochemical conditions at the 'Slikken van Flakkee'?
- 3. Which relationships exist between the direction of succession and hydrogeochemical conditions?
- 4. How is vegetation succession influenced by the different nature management choices at the 'Slikken van Flakkee?
- 5. Which measures can add to the preservation of vegetation communities with high presence of Red list plant species?

#### Hypothesis

We hypothesise that succession at the 'Slikken van Flakkee' is influenced by ongoing soil development which will lead to increasing desalination and a decrease in the buffering capacity of the soil due to decalcification. These processes are expected to lead to a will decrease of abundance of Red list plant species in the 'Slikken van Flakkee'. Differences in hydrogeochemistry between both areas are expected to be an important driver for ecosystem development directions occurring at the 'Slikken van Flakkee' and need to be taken into account besides the different nature management styles (mowing, grazing) in order to understand the existing variation between the northern and southern part of the area.

#### Comments second reader proposal

The second reader had some comments on the proposal <sup>2</sup>. This comment is dealt with in the following chapters.

<sup>&</sup>lt;sup>2</sup> Letter of University Utrecht, Dated March 31, 2016, Subject: Approval Research Proposal, Reference: EXCIE/2016/16.355 HI, Filename: 16.355\_SD\_GCE\_Kanters\_TJ\_approval Research proposal.pdf.

#### 2. Material & Methods

#### 2.1 Ecological succession

The composition of the vegetation at the 'Slikken van Flakkee' has been monitored on a yearly basis at multiple permanent quadrants (PQ's) (appendix 1) using Braun-Blanquet recordings. Vegetation recordings started in 1972 and most of the monitored plots recorded annually. The Braun-Blanquet abundance scores of van Haperen were transformed into a Van der Maarel decimal cover scale (van der Maarel 1979). The difference in vegetation composition between the PQ's was determined by calculating the Bray-Curtis dissimilarity index (Formula 4) (Bray & Curtis 1957) using the *vegdist* function from the R package *vegan* (Oksanen et al. 2016).

$$BC_{(i,j)} = 1 - \frac{2C}{(A+B)}$$
 (4)

Where C is the sum of the minimum abundances of the various species defined as the abundance at the site where the species is rarest, A is the sum of the abundance of all species at PQ *i* and B is the sum of the abundance of all species at PQ *j* (Legendre & Legendre 1998).

The PQ's from the entire dataset of van Haperen (n=1799) were subsequently clustered with an average linkage cluster analysis using the *hclust* function from the R *vegan* package (Oksanen et al. 2016). The resulting dendrogram from cluster this analysis was then sorted manually to determine the dominant vegetation types and sub-types occurring in the area over the entire period.

#### 2.2 Hydrogeochemistry

General geophysical and hydrological data were obtained from Slager et al., (1986) and Slager & Visser (1990) (Slager et al. 1986; Slager & Visser 1990).

To study the hydrogeochemical processes that occur at the 'Slikken van Flakkee', sediment samples were collected at nine locations representative for the different hydrological conditions (Slager & Visser 1990) and management styles in the area (Table 1) (Van Haperen, personal communication). The samples were taken using an Akkerman sampler which takes sediment cores by hammering a stainless steel tube into the soil at the desired depth. An Edelman drill was used to reach the top of the sampling depth. The steel tube is then removed from the sampler and closed off from the air by a steel disc and plastic caps. The Akkerman sampler was rinsed with demineralised water after each sample. To minimize the amount of air coming into contact with the sample, aluminium tape was used to seal the plastic caps to the sediment core. Dissolved oxygen in groundwater was also planned to be measured in the field but eventually turned out not possible due to the unavailability of an oxygen meter and due to the applied sample technique.

Soil samples were taken at four different depths, 0-20 cm, 30-50 cm, 70-90 cm and 110-130 cm below ground level to gain insight into the hydrogeochemical processes occurring at different depths. Besides taking soil samples a soil profile (Figure 3) was created at every location using a gauge to determine thickness of the humus layer, the highest groundwater level and the lowest groundwater level. An indication of calcium carbonate content was determined using hydrochloric acid (HCl) in the field.



Figure 3: Photo of Soil profile at PQ 17 on June, 29, 2016.



Figure 4: Photo of content glovebag on June, 27, 2016.

The sediment cores were placed in a coolbox in the field and stored in a refrigerator at  $5^{\circ}$ C in the laboratory until sub samples were taken for further analysis. In order to maintain the redox situation of the sediments cores the sub-sampling was done is a glovebag filled with nitrogen gas at the 'Utrecht Castel' (laboratory of Utrecht University). The sediment cores, required materials and sub sample containers were placed in the glovebag which was flushed four times with N<sub>2</sub> gas until the oxygen level in the glovebag was reduced to  $\pm 5\%$  of the normal oxygen concentration in the air (Figure 4).

Table 1: Height above NAP in 1986 (Slager et al. 1986) and management style at the investigated PQ's. Two series of
samples were taken in the Southern part representing a series with a stable groundwater level (5-40cm under ground level)
and a more dynamic groundwater level (0-80cm under ground level) due to an impermeable clay layer in the underground
(Slager & Visser 1990).

Area	PQ	Height (cm + NAP)	Management
North	45	100	None
	49	35	None
	73	10	None
South (stable)	10	70	Grazing and Mowing
	58	35	Grazing and Mowing
	59	5	Grazing and Mowing
South (dynamic)	17	90	Grazing and Mowing
	22	10	Grazing and Mowing
	65	10	Grazing and Mowing

When the desired  $O_2$  concentration was reached, the Akkerman cores were opened in the glovebag and the first centimetre of sediment was discarded. First, 30 grams of sediment were taken from the Akkerman cores and placed into an airtight glass bottle for iron (Fe) and manganese (Mn) oxyhydroxide analysis. Another 30 gram was placed in a similar bottle for phosphorus (P) analysis.

Pore water samples were taken from the sediment core using a ceramic Rhizosphere CSS soil moisture sample. Because the sediment cores were too compact to insert the Rizon, a steel wire of the same diameter as the Rizon was inserted into the sediment core first. When the wire was removed, the Rhizon was inserted into the sediment and coupled to a 10ml syringe which was put under negative pressure. 1.5 ml of the pore water from the syringe was placed into a glass IC vial for anion analysis, 1.5 ml was placed in a 15 ml Greiner tube for sulphate (S<sup>2-</sup>) analysis, 3 ml was placed in a glass test tube for dissolved organic carbon (DOC) analysis and 5.5 ml in another 15 ml Greiner tube which was subsequently acidified using 5.5 μl 65% suprapure nitric acid (HNO<sub>3</sub>) for cation analysis. The S<sup>2-</sup> sample was conserved using a 2% zinc-acetate (ZnAc) solution. A third 15 ml Greiner tube was filled with 6.5 ml pore water for alkalinity, ammonium ( $NH_4^+$ ) and total nitrogen ( $N_{tot}$ ) measurements. All samples were placed in a refrigerator at 5°C until they were analyzed. After the pore water sampling, 70 g of soil was removed from the top part of the sediment core and placed into an aluminium dish to determine the dry matter, calcium carbonate (CaCo<sub>3</sub>), carbon (C), nitrogen (N) content and the grain size distribution of the sediment. In cases where it was impossible to obtain enough pore water from the Rhizon the sediment cores where transferred into a plastic centrifuge bottle with the air hole taped off to prevent oxidation and subsequently centrifuged for 30 minutes at 2.000 rpm in a Sorvall centrifuge. The centrifuge bottles were then taken back into the glovebag and the extracted pore water was divided over the remaining samples using a 10 ml syringe with a 45  $\mu$ m filter. Eventually, we were able to collect sufficient pore water for all analyses.

The remaining sediment was then divided into two 50 ml Greiner tubes for pH and cation occupation analysis and a plastic bag for the rest material.

## 2.3 Laboratory analysis

pH and alkalinity where determined in the laboratory using an Orion 520A pH ion meter. After measuring the initial pH, a GRAN titration was performed to determine the alkalinity. The anion content of the pore water was determined by measuring fluoride (F<sup>-</sup>), chloride (Cl<sup>-</sup>), nitrite (NO<sub>2</sub><sup>-</sup>), bromide (Br<sup>-</sup>), nitrate (NO<sub>3</sub><sup>2-</sup>), phosphate (PO<sub>4</sub><sup>3-</sup>) and sulphate (SO<sub>4</sub><sup>2-</sup>) concentrations using Ion Chromatography (Dionex IC). The acidified subsample was used to determine the cation content by measuring sodium (Na<sup>+</sup>), potassium (K<sup>+</sup>), magnesium (Mg<sup>2</sup>), calcium (Ca<sup>2+</sup>), iron (Fe), manganese (Mn) and strontium (Sr) concentrations with inductively coupled plasma optical emission spectrometry (ICP-OES). NH<sup>4+</sup> and S<sup>2-</sup> concentrations where determined spectrophotometrically using a Shimadzu UU 1800 spectrophotometer and DOC with a Shimadzu TOC-5050A total organic carbon analyser. Total nitrogen (N<sub>total</sub>) concentration and electrical conductivity (EC-lab) were planned to be analysed but not measured due to time constraints.

The sediment samples were dried in an oven for 16 hours at 105°C to determine the dry matter content. CaCO<sub>3</sub>, C and N content of the sediment samples were determined by TGA and CN analysis respectively. The S content could not be analysed because the CS Analyser was not available. All analysis were performed at 'Utrecht Castel'. Grain size, iron minerals and P analysis were also planned but eventually not possible within the timeframe of this master thesis. Measurements will be carried out separately.

### 2.4 Representation of pore water composition

Major ion concentrations in the pore water were used to create a Piper plot using the "Excel for hydrology" macro by the USGS (Halford, Keith 2005)In the Piper plot, the chemistry of the pore water is visualized by plotting the Ca<sup>2+</sup>, Mg<sup>2+</sup> and Na<sup>+</sup> plus K<sup>+</sup> cations and SO<sub>4</sub><sup>2-</sup>, Cl<sup>-</sup> and HCO<sub>3</sub><sup>-</sup> plus CO<sub>3</sub><sup>2-</sup> anions in two ternary plots which are combined into a diamond plot. The percentage of each ion is based on the total concentration of their respective group (cations or anions).

The water type was determined using the classification system by Stuyfzand (Stuyfzand 2012) (Figure 5). This systems creates a code based on the salinity (mg/l Cl<sup>-</sup>), alkalinity (mg/l), cation and anion concentration (%) and Base Exchange Index.



**Figure 5:** Explanation of water type determination using Hydrochemcal (HGC 2.1) Microsoft Excel spreadsheet (Stuyfzand 2012).

#### 2.5 Relationship between vegetation composition and hydrogeochemistry

The relationship between species composition and the environmental factors at the selected PQ's was determined with a canonical correspondence analysis (CCA) (Braak & Verdonschot 1995) using the *CCA* function in the R *vegan* package (Oksanen et al. 2016).

CCA is a multivariate method which we used on abundance data of species in the respective plots in 2015 and data on the environmental variables measured in the top layer (0-20cm) at the plots in 2016. Management was added to the model as a factor with two nominal values, Grazing and mowing and NoManagement. CCA extracts synthetic gradients as ordination axes from the measured environmental variables that maximise the niche separation among the species (Braak & Verdonschot 1995). Because CCA uses linear combinations of environmental variables that explain the species composition, it can be interpreted as a linear regression method to explain the ordination of the species composition within the investigated PQ's (Braak & Verdonschot 1995).

The final model explaining the differences in vegetation composition was determined by stepwise selection using the *ordistep* function in R *vegan* (Oksanen et al. 2016). This method uses Akaike's information criterion (AIC) to select the important environmental variables in the data. The significance of the resulting model was tested by an ANOVA-like permutation test with the *anova.cca* function in R *vegan* (Oksanen et al. 2016).

### 3. Results

#### **3.1 Ecological succession**

Cluster analysis of the vegetation data revealed 4 vegetation types which can generally be described as: species poor halophyte vegetation (1), *Salix* forests (2), *Rubus* thickets (3) and species rich grasslands (4). Type 2 and 4 were subdivided into 2 and 4 subtypes respectively (Table 2).

Vegetation type 1 is relatively poor in species and contains mainly of *Puccinellia maritima*, Spergularia salina, Salicornia europaea and Suaeda maritima. The two subtypes of the Salix forest are type 2A1 which consists mainly of Salix caprea, Tussilago farfara and Pteridium aguilinium. Salix cinerea and Hippophae rhamnoides are also present in type 2A. In type 2B Salix cinerea is the dominant species, Salix caprea is not present is this subtype. Hippophae rhamnoides also occurs in high numbers. Different species of grass like Festuca rubra and Agrostis stolonifera occur in subtype 2A and are not present in type 2B. Type 3 consists mainly of thicket species like Rubus caesius, Cirsium arvense and Urtica dioica. Common grasses in type 3 are Poa trivialis and Elytrigia atherica. The species rich grasslands of type 4 are characterized by the presence of Agrostis stolonifera and Centaurium pulchellum. Type 4A1 and 4A1 differentiate form type 4B and 4C by the presence of Poa trivialis and Cirsium arvense. 4A1 and 4A2 are separated by the presence of Juncus gerardii and Trifolium fragiferum in 4A1. Festuca rubra, Festuca arenaria and Trifolium repens are present in 4A2 and absent in 4A1. Juncus gerardi is a common species in type 4B which is further characterized by a high diversity of grasses and herbs including Potentilla anserina, Leontodon autamnalis, Carex distans, Leontodon saxatalis and Lotus glaber. This is also the vegetation type that includes rare and Red list species like Parnassia pallustris, Epipactis pallustris, Dactylorhiza incarnata and Dactylorhiza majalis. Type 4C is sepreated from the other vegetation types of group 4 by the presence of more halophyte species like Puccinellia maritima and Spergularia salina. Other important species in type 4C include Juncus gerardii, Plantago coronopus, Aster tripolium, Parapholis strigosa and Glaux maritima.

Figure 6 shows the development in space and time of the different vegetation types. At the northern part of the area, species poor halophyte vegetation is the dominant vegetation type in the first years. *Rubus* thickets are found closest to the dyke and remain the dominant vegetation type in this zone during all the recorded years. The *Salix* forests start do develop from 1979 onwards and eventually dominates the elevated zone starting with type 2B. After 1985, type 2A begins to develop in the highest lying zone of the *Salix* forests and slowly replaces type 2B. Spatially this zone in confined to a transition zone between 2B and the grassy vegetation. Type 4A1 starts to develop in the more elevated part of the area in 1975 and succeeds type 1 as the years proceed, spatially confining type 1 to a few PQ's near the shore. The succession in the grasslands between the forest and the shore goes from type 4A1 to type 4C. After 2000, type 4B slowly becomes the dominant grassland vegetation in the northern part.

The succession in the southern part of the 'Slikken van Flakkee' begins with type 4A2 and 1. Type 1 is succeeded by type 4A1 except for a few PQ's at the shoreline which develop into type 4C. After 1985, type 4B is dominating. Eventually replacing type 4A1 and 4A2 entirely. In 2015, all the PQ's in the southern area have developed into 4B except for only one PQ with type 1 and only three PQ's with

type 4C. For a more detailed overview of the succession at the 'Slikken van Flakkee', see the analysis of the succession by van Haperen (van Haperen, n.d.).



**Figure 6:** Time slices of the vegetation types at the 'Slikken van Flakkee' from 1975 to 2015 in 10 year steps. Note: not all PQ's were recorded every year. For a yearly representation of the different vegetation types see appendix 3.

Legend	
Vegetation Type	Code
SPH (species poor halophytes)	1
Forest	2B
Forest	2A
	3
SRG (species rich grassland)	4A1
SRG (species rich grassland)	4A2
SRG (species rich grassland)	4B
SRG (species rich grassland)	4C

**Table 2**: Synoptic table indicating the frequency of species in the different vegetation types. I = low frequency (Van der Maarel score 1-3), II = intermediate frequency (Van der Maaral score 4-6), III = high frequency (Van der Maarel score 7-9). Species with low presence are omitted. For a more detailed representation see appendix 2. Type 4B includes Red list species representing a high nature value like *Blackstonia perfoliata s. spinosa, Parnassia palustris, Epipactis palustris, Dactylorhiza incarnata, Dactylorhiza majalis* and *Euphrasia stricta*.

Vegetation type	1	2A	2B	3	4A1	4A2	4B	4C	_
Number of PQ's	265	111	282	283	188	118	397	209	
									-
Puccinellia distans s. distans	Ш								
Puccinellia maritima								Ш	
Spergularia salina								II	
Suaeda maritima	Ш							II	
Salicornia europaea	Ш							II	
Salix caprea			Ш				Ι		
Salix cinerea			П						
Pteriduim aquilinum			П						
Hippophae rhamnoides					II		Ι		
Calamagrostis epigejos			П		II		II		
Moss (div)		II	I.	II	II	Ш	II		
Salix repens		II	I.				Ι		
Eupatorium cannabinum		Ι	П		I				
Phragmites australis		II	I.				Ι		
Rubus ceasius		П	П	Ш					
Cirsium arvense				П					
Poa trivalis			П	П	П	П			
Holcus lanatus				II	II				
Urtica dioica				II					
Galium aparine				II					
Agrostis stolonifera		Ι	I.			Ш		III	
Centautium pulchellum					II	Ш	II	II	
Festuca rubra F. arenaria					II	Ш			
Poa pratensis				Ι	II	Ш	II		
Juncus gerardii					II	Ш		III	
Parapholis strigosa					Ι		Ι	II	
Aster tripolium					Ι		II	II	
Plantago coronopus	Ш				Ι	I	Ι	II	
Lolium perenne					Ι	П			
Trifolium repens						Ш	II		
Trifolium fragiferum						Ш			
Lotus glaber						I			
Juncus articulatus							II		
Potentilla anserina							П		
Leontodon autamnalis							II		
Carex distans							П		
Cares extensa							Ι	I	
Glaux maritima							Ι	II	
Elytrigia athenica								I	

#### 3.2 Hydrogeochemical conditions

The hydrogeochemical conditions at PQ 49, 45 and 73 in the northern part of the 'Slikken van Flakkee' and PQ 10, 58, 59, 17, 22 and 65 in the southern part were sampled in June 2016.

The concentrations of major ions in the pore water were used to create a Piper diagram (Figure 7) and to determine the water types (Table 3) in the investigated plots.



**Figure 7**: Piper diagram of the major ion composition of the pore water. A , A and P represent PQ 45, 49 and 73 at the northern part of the 'Slikken van Flakkee'. A , A and P represent PQ 10, 58 and 49 and A , A and P represent PQ 17, 22 and 65 at the stable and dynamic zone of the southern part respectively. The red dots are different ratio's of rainwater and water from lake Grevelingen. The two ternary plots show the cation and anion concentrations. The diamond a matrix transformation of the anions and cations.

Depth (cm -	Area					
groundlevel)	North		South (stable)		South (dynamic)	
	PQ	Watertype	PQ	Watertype	PQ	Watertype
10	45	F4CaHCO3+	10	F2CaHCO3	17	g3CaHCO3
40		F4CaHCO3+		g3CaHCO3+		F3CaHCO3
80		F4CaHCO3+		g3CaHCO3		F3CaMix+
120		f4CaSO4+		g3CaHCO3+		F3CaHCO3+
10	49	f4CaHCO3	58	B5NaHCO3+	22	b3NaCl
40		B4CaMix+		f5NaHCO3+		b4NaCl
80		B4CaMix+		f5NaHCO3+		b4NaCl
120		B4NaCl+		F5NaHCO3+		S4NaCl+
10	73	b4NaClo	59	b4NaCl-	65	b4NaCl
40		S4NaCl+		b5NaCl+		b5NaCl-
80		S4NaCl		b5NaCl+		b5NaCl
120		S4NaCl+		b5NaCl		b5NaCl

**Table 3**: Chemical water types in the various PQ's at different depths, classification according to Hydrochemcal 2.1(Stuyfzand 2012).

The chemical water type of the pore water differs substancially between the investigated PQ's. The main pattern shows the occurrence of NaCl water types at PQ 73, 59, 22 and 65 reflecting the influence of the saline Grevelingen water on the water quality. PQ 73 shows an increase in salinity after 40 cm (8.700 mg/l Cl<sup>-</sup> > 15.000 mg/l Cl<sup>-</sup>) while PQ's 59 and 65 have a similar salinity over the entire 120 centimetres (3.700 mg/l Cl<sup>-</sup> - 5.700 mg/l Cl<sup>-</sup>). The salinity at PQ 22 shows an increase between 80 en 120 cm (8.400 mg/l Cl<sup>-</sup> - 12.000 mg/l Cl<sup>-</sup>). PQ's 45, 49, 10 and 17 which are situated relatively far from the shore show a CaHCO<sub>3</sub> water type in the top layer. At PQ 45 water type stays the same till a depth of 120 cm where it changes to CaSO<sub>4</sub>. The water type at PQ 49 changes after the first 40 cm to CaMix and again to NaCl at a depth of 120 cm indicating that desalination and decalcification have progressed less far compared to PQ 45. The water type at PQ 10 remains stable with only a slight decrease in salinity after the first 40 cm. PQ 58 has a NaHCO<sub>3</sub> water type which decreases slightly in salinity with increasing depth.

The pH differs only slightly between 6,8 and 8,4 in all pore water samples (Figure 8). At PQ 49, 45 and 73 in the northern part of the area and 58 and 59 in the southern part the pH increases slightly with increasing depth. pH decreases with depth at PQ 10, 17, 22 and 65. The alkalinity of the pore water generally increases after the first 20 cm.

To determine whether pyrite (FeS) oxidation is taking place at the 'Slikken van Flakkee' the sulphide concentrations were compared to the amount of  $SO_4$  that is expected due to the fraction of sea (Grevelingen) water in the pore water samples. Figure 9 shows the amount of  $SO_4$  measured with the lon Chromatograph compared to the expected amount based on the ratio between  $SO_4$  and Cl<sup>-</sup> in fresh and seawater. The deepest point at PQ 45 and all points at PQ 45 and 73 have  $SO_4$  concentrations lying above the expected amount due to the sea water content (shown by the Ratio Fresh – Sea line). In the southern part of the area PQ 17 shows  $SO_4$  excess at 40 and 80 cm depth. PQ 22 shows a  $SO_4$  excess which increases with depth.



Figure 8: pH and alkalinity of the pore water at different depths for each PQ.



**Figure 9**: Scatterplot of the Cl and SO<sub>4</sub> content of the pore water samples compared to the expected ratio based on the mixing between sea and fresh water. The Ratio Fresh-Sea line shows the expected SO<sub>4</sub> concentration based on the amount of Cl. Sulphate values above the Ratio Fresh-Sea line are an indication for occurring pyrite oxidation. Cl and SO<sub>4</sub><sup>2-</sup> concentrations in fresh (30 mg/l Cl<sup>-</sup> and 6 mg/l SO<sub>4</sub><sup>2-</sup>) and seawater (19.100 mg/l Cl<sup>-</sup> and 2.640 mg/l SO<sub>4</sub><sup>2-</sup>) are based on literature values (van Wirdum 1991; Van Breukelen et al. 1998).

The alkalinity ( $HCO_3^{-}$ ),  $Ca^{2+}$ ,  $K^+$ ,  $Mg^{2+}$ ,  $Na^+$ ,  $SO_4^{2-}$  and Fe concentrations were normalised to the Cl<sup>-</sup> concentrations and plotted against the Cl<sup>-</sup> (meq/l) concentration on a logarithmic scale to provide insight into the macro-chemical processes that occur in the soil at the 'Slikken van Flakkee' when the salinity decreases (Figure 10). Trends were visualized by including the moving average in the plot.



**Figure 10**: Major chemical components of pore water normalised to the Cl<sup>-</sup> concentration (meq/l) plotted against the Cl<sup>-</sup> concentration (meq/l) with moving average of each component. Cl<sup>-</sup> is assumed stable so deviations from the Compontent(s)/Cl = 1 line on the y-axis by other components with decreasing Cl<sup>-</sup> concentrations on the x-axis indicate hydrogeochemical processes in the soil. Increasing alkalinity and Ca<sup>+</sup> show the dissolution of CaCO<sub>3</sub>, increasing Fe and SO<sub>4</sub> are an indication of pyrite oxidation and fluctuations in Na<sup>+</sup>, K<sup>+</sup>, Mg<sup>2+</sup> and Ca<sup>2+</sup> indicate cation exchange between the pore water and the sediment.

The pore water in the entire 'Slikken van Flakkee' area consisted of a mixture of about 80 % seawater and about 20 % riverwater before the closing of the Grevelingen (Slager et al. 1986) so the assumption is made that the normalised ion concentrations were equal to the Cl<sup>-</sup> concentration. Because Cl<sup>-</sup> is a conservative ion which does not react with other elements as the soil is flushed with rainwater and desalinizes, changes in the relative concentrations of the pictured pore water components indicate several hydrochemical processes.

Pore water samples where checked for ionic balances which were generally within the accepted range (Stuyfzand 2012). However the ionic balance for the deepest sample (120cm) at PQ 49 and 22 and at the shallow sample (20cm) at PQ 17 show relatively high deviations. This is possibly due to the dilution of the deep samples which was necessary in order to decrease the Cl<sup>-</sup> content for IC analysis. The sample from PQ 17 was also diluted in order to obtain sufficient material for analysis. For all porewater results see appendix 4.

The increase in alkalinity and Ca<sup>+</sup> show the solution of CaCO<sub>3</sub> as the salinity decreases. The increase in Fe concentration is an indication of pyrite oxidation. The slight lowering in the relative concentrations of  $Mg^{2+}$ , Na<sup>+</sup> and K<sup>+</sup> ions between 1.000 and 100 meq/l Cl<sup>-</sup> is an indication of cation exchange between the pore water and the soil.

Results for the chemical features of the soil are shown in Table 4. The dry matter content of the top layer is around 50% in all PQ's. Deeper samples have a dry matter content around 80%. C and N show the highest values in the top layers. While CaCO<sub>3</sub> content is generally the lowest in the top layer. All results from the soil analysis are given in appendix 5.

		Depth				
Area	PQ	(cm -groundlevel)	Dry Matter	С	Ν	CaCO3
North	45	10	48	10,33	0,53	2,56
		40	78	2,26	0,07	8,03
		80	80	1,32	0,01	9,09
		120	82	1,19	0,00	7,75
	49	10	51	8,73	0,36	2,68
		40	80	1,18	0,01	7,17
		80	80	0,99	0,01	6,29
		120	81	1,25	0,01	8,63
	73	10	63	7,22	0,26	2,29
		40	84	0,82	0,01	3,68
		80	84	0,85	0,01	3,82
		120	82	1,06	0,01	4,69
South (stable)	10	10	42	18,39	1,12	1,75
		40	83	1,11	0,03	6,15
		80	81	1,00	0,01	7,00
		120	81	1,12	0,01	8,08
	58	10	46	12,71	0,70	2,48
		40	80	1,21	0,01	7,61
		80	80	1,22	0,01	7,59
		120	81	2,03	0,01	13,77
	59	10	45	11,95	0,57	1,41
		40	79	1,10	0,01	6,14
		80	82	1,32	0,01	8,31
		120	80	1,59	0,01	9,14
South (dynamic)	17	10	49	12,10	0,78	0,95
		40	83	1,05	0,02	6,16
		80	83	0,87	0,01	6,16
		120	80	0,95	0,01	6,46
	22	10	43	11,40	0,66	2,84
		40	79	1,46	0,03	8,08
		80	81	1,13	0,01	7,62
		120	80	1,20	0,01	7,06
	65	10	52	7,53	0,39	2,71
		40	79	1,41	0,02	7,51
		80	80	1,53	0,02	8,83
		120	81	0,95	0,01	5,49

Table 4: Dry matter content in % in 70 grams of fresh sample and C, N and CaCO<sub>3</sub> content (% of dry weight).

# **3.3 Relationship between vegetation type and hydrogeochemical conditions in 2015**

A selection of environmental variables from the top layer (0-20 cm) of the soil was used to determine the relationship between the vegetation type recorded in 2015 and the environmental conditions in 2016. Variables that were selected for analysis are pH,  $Ca^{2+}$ ,  $Cl^-$  and  $HCO_3^-$  concentrations,  $SO_4^{2-}$  excess,  $CaCO_3$  and N content, water level (GHG) and management.

Figure 11 shows four different environmental zones at the 'Slikken van Flakkee': the part of the area without any management, relatively high  $Ca^{2+}$  and  $Cl^-$  concentrations,  $SO_4^{2-}$  excess and high  $CaCO_3$  content (lower left side of the diagram), the northern part with a lower water table (upper left side of the diagram), the part with management by grazing and mowing and a high nitrogen content of the soil (upper right side of the diagram) and the southern part of the area with relatively high Cl concentrations and slightly higher pH values.

In 2015, PQ 45 consisted of *Salix* forest (type 2A) which occurs where the water level is slightly lower in the no-management zone. The vegetation type at PQ 73 was species rich grassland with a relatively high abundance of halophytes (type 4C). This vegetation occurs close to the shore where the Cl<sup>-</sup> and Ca<sup>2+</sup> concentrations are high and pyrite oxidation is occurring. The rest of the PQ's had species rich grassland vegetation (type 4B) with a high abundance of herb species like *Trifolium repens, Lotus glaber* and *Leontodon autamnalis*. In the southern part of the area, with management by grazing and mowing, type 4B occurs over a wide range of environmental conditions while it restricted to a zone with relatively fresh water and slightly higher ground water levels in the northern part of the area.

A stepwise selection analysis was performed to identify the most important environmental factors that determine the occurrence of the different vegetation types. In the resulting model only management and Cl<sup>-</sup> concentration were selected. A further CCA was carried out in which only these factors were taken into account as constraining factors. The result from this analysis (Figure 11 and Figure 12) confirms that the difference in vegetation composition at the 'Slikken van Flakkee' is more correlated to management style and salinity then to the other hydrogeochemical variables in the area.

#### CCA 2015 Environmental Data



**Figure 11**: Canonical Correspondence Analysis showing correlation between vegetation type and a selection of hydrogeochemical variables of the top 20 cm of the soil (indicated by arrows) and management type (nMN = no management, mGR = grazing and mowing) at the investigated PQ's displaying 100% of the inertia. Eigenvalue: axis 1 = 0.74, axis 2 = 0.45. sfn and sfz represent the northern and southern part respectively. The number corresponds to the PQ number. sfn 49 consists of vegetation type 2A, sfn 73 of type 4C and all other PQ's of type 4B (table 2). The location of the PQ's within the plot is determined by the weighted average of the species abundance within the PQ's.



CCA 2015 Vegetation types - Significant Environmental variables

**Figure 12**: CCA showing correlation between vegetation type and important environmental variables (Cl<sup>-</sup> in the top layer and management) determined by stepwise selection. Cl<sup>-</sup> is represented by the arrow and management type by mNM (No Management) and mGR (Grazing and mowing). Eigenvalue: axis 1 = 0,59; axis 2 = 0,36. Model explains 45% of the variance. Permutation test significant (P < 0,01).

#### 4. Discussion

The aim of this project was to analyze the succession of the vegetation that has taken place at the 'Slikken van Flakkee' since the tidal influence was removed by closing the Grevelingen estuary from the North Sea in 1971 and to find the hydrogeochemical processes that occur in the former tidal wetlands. Furthermore, the effects of the different nature management strategies on the succession in the two sub-areas at the 'Slikken van Flakkee' were analyzed.

#### Vegetation types and Succession

The trends found in the succession of the vegetation show gradual change from species poor halophyte vegetation to a *Salix* forest and species rich grassland vegetation at the northern sub area. The *Salix* forest ecosystem develops in the more elevated part of the area where the salt content of the pore water has decreased relatively fast. In the lower lying part of the area where the salt content is still high, succession leads to species rich grasslands. In the southern part of the 'Slikken van Flakkee' species poor halophyte vegetation develops into species rich grassland vegetation with an increase in species with time. Eventually this leads to a uniform ecosystem which includes several rare species on the European Red list. The different vegetation types that are present right after the removal of the tides in the southern sub-area are due to the sowing of rye and grass species (van Haperen n.d.). Our results are in keeping with general trends found in other former tidal wetlands in The Netherlands (Joenje & Verhoeven 1993).

#### Hydrogeochemistry

The hydrogeochemical processes that where identified at the research area confirm the hypothesis that desalination and pyrite oxidation (Figure 9) occur in the soil. The variation in different chemical water types (Table 3, Figure 7) indicate that desalination and pyrite oxidation proceed in different rates within the research area. The hypothesized difference between the northern and southern part of the area has not been observed and seems mainly determined by elevation and influence of saline Grevelingen water within the respective sub-areas. The predicted acidification due to pyrite oxidation that is found in other former tidal wetlands (Portnoy 1999; Portnoy & Giblin 1997) has not been observed however (Figure 8). This is due to the high CaCO<sub>3</sub> content of the soil. The relatively low CaCO<sub>3</sub> content in the top layer can be explained by the formations of a humus layer and by decalcification due to humic acids. We find that cation exchange takes place with increasing desalination.

#### Relationship between Vegetation and Hydrogeochemistry

We found no evidence for the hypothesis that the difference in vegetation composition between the northern and southern sub-areas is driven by environmental factors other than the different nature management styles being practiced. The variation within the northern part is influenced mainly by differences in desalination. Other processes like pyrite oxidation and decalcification are present within the area but did not have a significant influence on the vegetation type (Figure 11 and Figure 12).

Grazing and mowing have a stronger influence on the development of the vegetation than the hydrogeochemical processes in the southern sub-area leading to a landscape with much less variation compared to the northern part of the area. However, the resulting vegetation type 4B (Table 2) contains the European Red list species which represent the highest natural value. It must be noted that at several PQ's in the southern sub area, vegetation type 1 and 4C exist at the edge of lake Grevelingen in 2015, indicating that water composition can have an influence on the vegetation.

#### Limitations

The results need to be interpreted in the light of several limitation. First, it has to be noted that differences in depth below the saturation level are not certain since the bore hole tended to collapse once the samples were taken from below the ground water level. Changes that occur above the saturated soil are more certain. Furthermore, since our date represents one point in time in the ongoing soil processes, it is not possible to be certain about how these processes have occurred since the tidal influence has been removed or how they will proceed in the future. To gain a more detailed insight in the evolution of these processes with time, hydrogeochemical modeling is required.

Because we were only able to investigate three PQ's in the northern sub-area there may be relationships between vegetation type and environmental conditions in the remaining PQ's that are now left undetected. To gain more insight in the environmental factors driving the variation within this sub-area, the hydrogeochemical conditions should be investigated at more PQ's. However, since the investigated PQ's are representative for the main vegetation types we expect that these analyses will yield similar results.

#### Management advice

In order to maintain the species with high natural values within the southern part research area the management by grazing and mowing should be continued in order to prevent the occurrence of natural succession. If the grazing and mowing is stopped the more elevated parts are predicted to develop into a *Salix* forest ecosystem. The vegetation type which contains the valuable species will probably still occur but at a much smaller zone where the water level is too high for the forests to develop and the salt concentration is low enough for the Red list species. Whether continuing pyrite oxidation and decalcification will eventually lead to a decrease in pH which might cause the Red list species to decline should be investigated by the aforementioned modeling using a geochemical model like PHREEQC. In the case that these processes occur, an optional choice might be to reintroduce the tides. However, this might decrease the zone where vegetation type 4B is able to persist due to an increase in salinity in the lower part of the area.

## 5. Conclusions

The main aim of this research was to study the differences in vegetation succession between the northern en southern sub-areas at the 'Slikken van Flakkee' by analyzing the vegetation data set compiled by Anton van Haperen and 'Rijkswaterstaat'. This data set was subsequently compared to current hydrogeochemical data obtained by analyzing soil and pore water from nine representative locations within the 'Slikken van Flakkee' area.

Our data suggests that the difference in the vegetation succession between the northern and southern area is mainly due to the different styles of nature management between the sub-areas. The variation within the northern sub-area is explained by differences in salt content of the pore water. Other chemical processes like pyrite oxidation and decalcification were observed. However, these processes did not have a significant influence on the development of the different vegetation types. Further research into the development of the hydrogeochemical conditions is required to determine whether the current nature management choices are sufficient to maintain the presence of valuable Red list species in the area.

## 6. Acknowledgements

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# 8. Appendices



# Appendix 1: Map of the PQ's at the Slikken van Flakkee.

**Figure 13:** Map of the PQ's at the Slikken van Flakkee where the vegetation has been recorded since 1972. Red dots represent the PQ's that were analyzed during this master thesis for hydrogeochemical compositions.

## Appendix 2. Heatmap.



**Figure 14**: Visual representation of all the vegetation recordings since 1972 at the Slikken van Flakkee with dendrogram (average clustering) and the important species. Vegetation types are indicated with colored rectangles. Blue = type 1, darkbrown = type 2B, lightbrown = type 2A, brown/green = type 3, darkgreen – blue/green = type 4A1, 4A2, 4B and 4C. Color key shows the cover percentage of the species in van der Maarel scale (1 = 0,1%, 2 = 1%, 3 = 3%, 4 = 5%, 5 = 12,5%, 6 = 28,5%, 7 = 37,5%, 8 = 62,5% and 9 = 87,5%).

# Appendix 3. Development vegetation 1972-2015.







# Appendix 4. Results Water.

Research:	Ecohydrogeochemistry	of Former Ti	dal Areas		Back to Conte	ent .																				
Area:	Grevelingen, Slikken van	n Flakkee																								
Author: Date:	M. van der Weiden, T. Ka August 12, 2016	anters													_											
File:	Slikken van Flakkee Da	ata Processir	ng ??.xlsx																							
Sheet:	Results Water																									
Description:	Results of Water Analyse	es													_											
Instruction:	NA														_											
Source.	Authors														_											
Nr	Area	PQ High	/ Locatio	n	Samplecode	E. Conductivity	pH Na*	K*	Mg <sup>++</sup>	Ca <sup>++</sup> Sr	NH,* F Cľ	Bri	HCO <sub>1</sub>	CO <sub>3</sub> <sup>2</sup> SO <sub>4</sub> <sup>2</sup>		S	NO <sub>1</sub> *	NO <sub>2</sub> <sup>+</sup> P <sub>tot</sub>	PO <sub>4</sub> <sup>3-</sup> SiO <sub>2</sub> Fe <sub>rot</sub> Mn <sub>tot</sub> Al	DOC	Ionbalance 1)		Total Ratio's			
																					Sum	((SC-SA) /				
												(ICP-			(ICP-						Sum Cations Anions	(SC+SA)) *	Dissolved			
		Low /	X	Y	Z (cm below	Field	Lab				(IC)	OES)		(IC)	OES)	Measured					(SC) (SA)	100	Solids Ca*25/CI Mg*	5/CI Na^1,15/CI	K*55/CI CI SO4*10/CI	HCO3*200/CI Fe*250000/CI
		Salt			aroundlevel)	(mS/cm)	- ma/l	ma/l	ma/l	ma/I ma/I	ma/I ma/I ma/I	ma/l ma/l	mMol/I ma/I	ma/l ma/l	ma/l	ma/l	ma/l	ma N/I ma/I	ma/l ma/l ma/l ma/l ma/	/I maC/	'l mea/l mea/l	(%)	ma/l mea/l mea/	I mea/I r	nea/l mea/l mea/l	mea/l mea/l
1	Slikken Noord	45 High	600	71 42425	i6 -10 SvF-45-0,10		7,29 50,9	1,0	8,	,6 178,4 0,	,6 1,70 0,3 71	1 0,4	4 9,60 586	0	6,6 8	3 <mark>,7</mark> 0,00	5 C	0,5 0,2 0,17	23,1 0,6 0,85 0,1	12 35,	4 12,00 11,77	1	0,9 1.019 110,56	1,77 1,26	0,73 2,01 0,68	954,02 2.847,36
2		45			-40 SvF-45-0,40		7,28 55,4	1,8	13,	,9 203,4 0,	,8 0,73 1,0 77	2 0,1	7 11,21 684	0 1	,3 9	0,00	5 C	0,5 0,14	19,2 0,6 1,44 0,4	46 16,	0 13,87 13,68		0,7 1.111 116,71	2,63 1,28	1,15 2,17 1,08	1.030,97 2.388,06
3		45	-	-	-80 SVF-45-0,80	3.5	7,37 63,8	3,3	15,	4 205,1 0, 5 708.5 2	9 0,24 1,5 85 8 1.36 1.1 271	1 0,	7 11,38 694	0 2	7 26 8 1 306	0,00	6	0,15		33 12, 56 28	2 14,39 14,43 6 46.90 46.21	-	0,2 1.147 106,59	2,64 1,33	1,92 2,40 2,40	947,99 186,66 258,55 13,002,67
5	Slikken Noord	49 Low	597	44 42409	-10 SvF-49-0,10	3,3	6,89 97,1	8,3	18,	5 185,3 0,	6 0,12 0,3 174	48 0,6	6 9,96 608	0 1.57	4 7	.2 0,00	0 0	0,5 0,17	25,9 19,5 1 0,4	43 75,	.8 15,94 14,91		3,3 1.329 47,21	1,55 0,99	2,38 4,90 0,06	406,72 35.631,02
6		49			-40 SvF-49-0,40		7,03 232,9	22,9	58,	,4 270,1 1,	,5 0,63 1,0 420	136 1,1	1 9,56 583	0 31	,7 324	,9 0,00	3 0	0,5 0,14	16,3 2,3 1,14 0,5	57 36,	7 29,16 28,10		1,9 2.021 28,40	2,03 0,98	2,71 11,86 5,58	161,20 1.728,66
7		49	_	_	-80 SvF-49-0,80		7,47 342,5	58,3	87,	,8 201,0 1,	,7 0,56 1,2 507	214 1,6	6 11,84 722	0 33	3,1 350	0,7 0,00	4 C	0,5 0,16	17,5 6,5 0,6 0,6	61 34,	0 33,93 33,25		1,0 2.371 17,54	2,53 1,20	5,73 14,30 4,92	165,60 4.057,50
8	Slikkon Noord	49 72 Solt	604	20 42250	-120 SVF-49-1,20	5,6	7,46 761,1	99,2	91,	7 151,0 1,	<u>,5 4,19 1,0 635</u>	1.061 2,2	2 12,75 778	0 35	3,6 389	,3 0,00	6 0	0,7 0,43	32,0 0,2 0,06 0,4	43 31,	1 50,96 35,80	1	7,5 2.877 10,52 0.2 17,224 2.84	2,11 2,13	7,79 17,90 4,17	142,42 80,56
10	Silkken Noord	73 Jan 73	004	20 42230	-40 SvF-73-0,40		7,41 9,282.6	347.0	1.392.	.0 842.6 9.	0 0.12 1.9 15.073	17.194 50.3	3 10.70 653	0 4.22	4.584	0,00	3 21	1,1 14,3 0,12	NR 9.4 0.0 0 1.5	53 52.	7 569.21 524.34		4.1 31.996 2.47	1.35 1.09	1.15 425.14 2.07	5.03 0.00
11		73			-80 SvF-73-0,80		7,60 9.573,7	323,8	1.457,	6 723,0 8,	,2 3,78 0,6 18.856	17.743 67,4	4 12,19 744	0 4.03	4.188	,2 0,07	8 21	1,1 4,4 0,01	NR 21,2 0,3 0,3 1,4	43 42,	9 580,93 628,49	-	3,9 35.883 1,70	1,13 0,90	0,86 531,85 1,58	4,58 5,05
12	_	73	_		-120 SvF-73-1,20	50,6	7,87 10.125,2	337,6	1.528,	,0 719,3 8,	,4 6,33 0,6 17.334	18.630 64,6	6 13,51 824	0 3.57	4.324	,2 0,00	4 21	1,0 4,5 0,03	1,1 21,0 0,2 0,1 1,3	37 34,	,1 611,01 577,20		2,8 34.589 1,84	1,29 1,04	0,97 488,92 1,52	5,53 3,66
13	Slikken Zuid (dynamic)	17 High	624	94 41750	10 SVF-17-0,10		7,67 36,3	0,9	9,	8 121,6 0,	3 1,35 0,6 20	8 0,0	0 6,39 390	0	0,3 1	,4 0,00	3 0	0,6 0,4 0,56		31 20	6 8,56 7,09 7 10.14 0.11		9,4 795 274,20 5 3 787 116 61	7,30 3,28	2,40 0,55 2,00	2.310,73 2.017,08
14		17	-	-	-40 SVF-17-0,40 -80 SVF-17-0,80		7,00 40,4	1,2	12,	8 146.8 0	9 1.49 1.7 101	12 0.3	3 5.07 309	0 16	32 171	2 0.00	6 1	1.4 0.5 0.18	5.0 0.016 0.0046 0.2	21 20.	2 11.55 11.43		0.5 869 64.24	1.99 1.10	5.45 2.85 11.91	355.58 49.77
16		17			-120 SvF-17-1,20	1,6	7,45 64,6	21,5	18,	,6 164,4 1,	,1 1,75 0,7 97	13 0,3	3 6,87 419	0 14	8,3 161	,9 0,01	1 C	0,6 0,16	10,4 0,047 0,17 0,3	36 20,	6 13,20 12,74		1,8 1.000 74,90	2,80 1,18	11,04 2,74 11,27	501,66 152,67
17	Slikken Zuid (dynamic)	22 Low	618	06 41742	-10 SvF-22-0,10		7,85 1.742,8	112,2	154,	,8 367,0 2,	,6 0,23 0,8 2.244	3.115 4,2	2 7,63 466	0 34	9 <mark>,2</mark> 526	i,0 0,01	3	0,20	12,6 0,6 2,58 0	0,8 20,	4 109,85 78,24	1	6,8 5.506 7,23	1,01 1,38	2,49 63,29 1,15	24,11 89,12
18		22	-	-	-40 SvF-22-0,40		7,58 3.127,7	139,9	364,	6 425,7 3,	7 1,66 1,4 5.724	5.875 16,9	9 12,04 735	0 1.07	0,6 1.068	,5 0,00	5	0,12	10,9 0,2 1,6 0,7	79 13,	5 191,02 195,86	-	1,3 11.641 3,29	0,93 0,97	1,22 161,46 1,38	14,91 11,09
20		22			-80 SVF-22-0,80 -120 SVF-22-1,20	22.25	7,20 4.594,5	257.0	967.	7 516.2 6	3 7,54 0,8 8.399	12,659 36,4	6 12,44 759 4 11.90 726	0 2.60	2,4 1.922	9 0.46	4	0,14		26 21	4 401.95 403.63	-	0.2 23.788 1.91	1,12 0,97	1.07 337.50 1.61	7.05 15.92
21	Slikken Zuid (dynamic)	65 Salt	622	06 41675	4 -10 SvF-65-0,10		7,69 1.892,2	76,8	230,	6 326,1 2,	,3 1,43 1,4 3.707	3.544 9,3	3 11,68 713	0 48	3,5 311	,9 0,00	0	0,14	27,2 20,2 1,3 0,5	54 67,	6 120,36 126,48	-	2,5 7.657 3,89	0,91 0,91	1,03 104,56 0,97	22,34 1.726,33
22		65			-40 SvF-65-0,40		7,85 1.817,5	71,0	219,	,3 438,1 2,	,6 1,11 1,1 3.453	3.668 9,	1 19,83 1.210	0 30	, <mark>0</mark> 483	<mark>,9</mark> 0,00	2	0,15	26,5 3,4 1,3 0,7	72 62,	7 121,00 123,55	-	1,0 7.703 5,61	0,93 0,93	1,03 97,40 0,64	40,72 312,56
23		65			-80 SvF-65-0,80	110	7,81 2.342,1	118,5	272,	1 269,8 2,	3 2,57 0,8 4.320	4.390 11,4	4 18,68 1.140	0 51	,4 518	0,00	4	0,07	32,3 1,6 0,4 0,5	59 34,	4 140,97 151,36	-	3,6 9.106 2,76	0,92 0,96	1,37 121,86 0,88	30,66 117,55
24	Slikken Zuid (stable)	10 High	618	08 41921	-120 SVF-65-1,20 4 -10 SVF-10-0.10	14,6	7,10 2.372,8	127,2	2/5,	3 85 9 0	2 0.04 0.2 36	4.331 11,	5 17,38 1.060 0 3.97 242	0 1	2,8 600	0,00		0.8 0.2 0.69		32 5	0 5 91 5 24	-	6.0 436 105.95	2 17 0 90	20 72 1 01 2 30	28,79 244,76
26	Gilkken Zuid (Stable)	10 1191	010	41521	-40 SvF-10-0,40		7,62 14,9	7,2	7,	.8 134,7 0,	4 0,04 0,2 22	11 NR	6,94 423	0 1	3,4 16	.4 0,00	5 2	2,0 NR 0,14	NR 9,7 0,029 0,0281 0,2	26 13,	9 8,20 7,99		1,3 675 268,97	5,12 1,19	16,14 0,62 6,12	2.220,90 409,03
27		10			-80 SvF-10-0,80		6,86 13,5	1,7	6,	,9 132,8 0,	,5 0,17 0,2 19	9 0,1	1 6,40 391	0	,3 0,6233	37 0,00	6 0	0,4 0,0 0,15	NR 10,6 0,7 0,53 0,3	33 14,	,4 7,88 6,99		6,0 615 307,13	5,28 1,25	4,33 0,54 0,48	2.372,31 10.786,55
28		10			-120 SvF-10-1,20	1,05	6,92 16,4	2,9	4,	,6 84,0 0,	,3 0,25 0,2 20	14 0,1	1 4,74 289	0	2,8 0,4269	97 0,00	9 0	0,4 0,0 0,21	NR 7,6 0,03 0,06 0,1	12 15,	,1 5,38 5,38		0,0 466 186,41	3,38 1,46	7,26 0,56 1,05	1.687,13 473,27
29	Slikken Zuid (stable)	58 LOW	613	01 41880	15 -10 SVF-58-0,10		8,01 514,1	16,9	18,	5 89,2 0,	6 0.12 2.6 275	1/2 0,	5 17,37 1.060	0	3,6 9	0,00	6 2	2,2 0,1 0,19	NR 25,2 1,3 0,23 0,1	13 142,	3 28,82 28,26		1,0 2.468 10,40 0.7 2.420 12.07	0,71 2,40	2,22 10,70 0,07	324,77 1.046,32
31		58			-40 SVF-58-0,40		8,40 394,5	83,1	38,	2 50.6 0.	5 0,08 1,8 160	75 0,5	5 19,18 1.170	0 1	7,5 33	0,00	3 2	2,3 0,1 0,45		24 25,	4 24,96 24,18		1,6 2.006 14,00	3,49 4,38	25,92 4,51 0,81	850,85 83,61
32		58			-120 SvF-58-1,20	2,6	8,38 317,7	93,1	67,	,9 47,7 0,	,6 0,13 1,3 133	61 0,4	4 18,50 1.129	0	1,0 2	2,5 0,00	4 2	2,2 0,0 0,52	0,3 33,6 0,2 0,0091 0,0	09 21,	4 24,18 22,38		3,9 1.882 15,81	7,42 4,22	34,81 3,76 0,06	983,34 523,52
33	Slikken Zuid (stable)	59 Salt	612	08 41861	4 -10 SvF-59-0,10		7,50 2.666,5	78,7	304,	7 513,3 3,	3 0,29 0,6 5.759	5.507 14,8	8 8,34 509	0 70	(,1 676	,9 0,00	1 11	1,3 0,14	NR 45,1 9,5 3,3 0	0,8 39,	2 169,15 185,71	-	4,7 10.710 3,94	0,77 0,82	0,68 162,43 0,91	10,27 523,09
34		59			-40 SVF-59-0,40		7,94 1.688,1	/4,4	132,	8 203,6 1,	6 0,14 1,2 2.542	2.612 5,	7 24,41 1.489	0 13	5,3 146	0,04	8 2	2,2 0,5 0,09	NR 37,2 5,1 0,7 0,2	24 142,	8 96,63 99,05	-	2.4 7.101 2.10	0,76 1,18	1,46 /1,/1 0,40	68,08 636,79
36		59			-120 SvF-59-1.20	14.1	8.03 1.874.8	99.7	190,	.6 185.0 1.	9 0.35 1.1 3.250	3.151 7.6	6 19.91 1.215	0 17	3.1 160	.3 0.01	1 21	1.3 0.6 0.60	2.3 48.1 0.9 0.3 0	0.2 105.	7 109.07 115.68	-	2.9 7.332 2.52	0.86 1.02	1.53 91.66 0.40	43.44 87.91
101	Seawater					19.800	8,2 11000	478,47 400	132	20 420	19.800		145	27	75		1	1,3 0,06	4,4 0,002 0,005		618 619	-	0,1 35.866 0,94	0,97 0,99	1,01 558,49 1,03	0,85 0,03
102	Riverwater Rhine/Meuse					124	7,7 69,18	3,0091 5,89	10,	4 74,05	124		157		67			73 1,11	5,8 0,24 0,07		4 8 9	-	5,9 588 26,41	1,22 0,99	2,37 3,50 3,99	147,13 614,37
103	Grevelingenwater (G)		-	_		16.000	8.888,9	386,64 323,9	1.067,	1 353,2	16.000	451	147,3	2252	01		15,14	4.2 0.02	4,67 0,048 0,0176	1	5		29.053 0,98	0,97 0,99	1,01 451,30 1,04	1,07 0,95
10-1							<u> </u>	0,2	. 0,		3,9		1 1		<u> </u>	-	14	.,0,03	0,0 0,11 0,01		·~		20 0,00	1,00	2,00 0,11 12,11	0,00 0.302,30
Explanation																										
1) Calculated	with HGC		_						-						_					_						
INR	NO Result Detectionlimit / 2								-						_		-									
	Minimum value																									
	Not planned																									
	IC IC														_		_									
	Large deviation														_		-									
	30 0010001																1									
Legenda (Cor	nditional formatting for h	highlighting a	analytical e	rrors or lac	cking data)																					
		- I							-						_					_						
Conditional	tormatting with $\Sigma C = s_0$	um cations,	, ΣA = s ur	namions, i	n worksheets 3-6										_		-									
Cell colour	no colour	ta	in or	ange 🚥	colour tan oran	nge											-			-						
<b>社</b> XC-XA-10		B (%)	0	20	AEC-meas (%)	>0																				
2C+2A >6 m 5C+5A = 2.8	eqn. ⊲i meal ⊲6	1	2	>12	≪n ≪ö ≪5 <12 >>	>12																				
ΣC+ΣA <2 m	eq/L <10	<	0	≥20	<10 <20 >	≥20																				
IB (%) = 100	(ΣC-ΣΑ) / (ΣC+ΣΑ)		ΔEC-m	eas(%)=1	00 (EC <sub>MEAS</sub> - EC <sub>CALC</sub> ) / EC <sub>MEAS</sub>																					

Remark: Results are not fully final due to some minor dilution corrections yet to be performed.

# Appendix 5. Results Soil.

Research:	Ecohydrogeochemistry	of Fo	ormer Tida											
Area:	Grevelingen, Slikken van		kee											
Author:	M. van der Weiden, T. H		S											
Date:	August 12, 2016													
File:	Slikken_van_Flakkee_Da		rocessina	??.xlsx										
Sheet:	Result Soil													
Description:	Results of Soil Analyses													
Instruction:	NA													
Source:	Authors													
	, lations													
Nr	Area	PQ	High /	Location			Samplecode	Dry matter content	CaCO₃	Organic matter (Humus)	Organic Carbon	N <sub>total</sub>	Ratios	
			Low /	x	Y	Z		1)	(TGA)	(TGA)	(CN Analyzer)	(CN Analyzer)		
			Salt			(cm below		0/_	% <b>DS</b>	% <b>DS</b>	% <b>DS</b>	% <b>DS</b>	C/N	
			Jan			groundlevel)		78	/1 03	<i>™</i> D3	~ 55	<i>№</i> <b>В</b> 3	O/N	
1	Slikken Noord	45	High	60071	424256	-10	SvF-45-0,10	47,68	2,56	18,0	10,33	0,53	19	
2		45				-40	SvF-45-0,40	78,18	8,03	2,5	2,26	0,07	33	
3		45				-80	SvF-45-0,80	80,28	9,09	0,5	1,32	0,01	100	
4		45				-120	SvF-45-1,20	81,76	7,75	0,4	1,19	0,00	247	
5	Slikken Noord	49	Low	59744	424098	-10	SvF-49-0,10	51,44	2,68	15,7	8,73	0,36	24	
6		49				-40	SvF-49-0,40	80,28	7,17	0,6	1,18	0,01	100	
7		49				-80	SvF-49-0,80	79,61	6,29	0,5	0,99	0,01	83	
8		49				-120	SvF-49-1,20	81,18	8,63	0,5	1,25	0,01	115	
9	Slikken Noord	73	Salt	60428	422584	-10	SvF-73-0,10	63,13	2,29	12,9	7,22	0,26	28	
10		73				-40	SvF-73-0,40	83,98	3,68	0,7	0,82	0,01	99	
11		73				-80	SvF-73-0,80	84,31	3,82	0,7	0,85	0,01	101	
12		73				-120	SvF-73-1,20	81,68	4,69	0,9	1,06	0,01	94	
13	Slikken Zuid (dynamic)	17	High	62494	417501	-10	SvF-17-0,10	42,32	1,75	35,0	18,39	1,12	16	
14		17				-40	SvF-17-0,40	82,57	6,15	0,8	1,11	0,03	44	
15		17				-80	SvF-17-0,80	81,09	7,00	0,4	1,00	0,01	101	
16		17				-120	SvF-17-1,20	81,43	8,08	0,4	1,12	0,01	109	
17	Slikken Zuid (dynamic)	22	Low	61806	417423	-10	SvF-22-0,10	45,96	2,48	22,4	12,71	0,70	18	
18		22				-40	SvF-22-0,40	80,50	7,61	0,6	1,21	0,01	142	
19		22				-80	SvF-22-0,80	80,19	7,59	0,6	1,22	0,01	163	
20		22				-120	SvF-22-1,20	80,99	13,77	0,7	2,03	0,01	223	
21	Slikken Zuid (dynamic)	65	Salt	62206	416754	-10	SvF-65-0,10	45,31	1,41	22,1	11,95	0,57	21	
22		65				-40	SvF-65-0,40	79,20	6,14	0,7	1,10	0,01	81	
23		65				-80	SvF-65-0,80	81,58	8,31	0,6	1,32	0,01	117	
24		65				-120	SvF-65-1,20	79,86	9,14	0,8	1,59	0,01	155	
25	Slikken Zuid (stable)	10	High	61808	419214	-10	SvF-10-0,10	49,17	0,95	23,6	12,10	0,78	15	
26		10				-40	SvF-10-0,40	82,89	6,16	0,7	1,05	0,02	46	
27		10				-80	SvF-10-0,80	82,65	6,16	0,4	0,87	0,01	92	
28		10				-120	SvF-10-1,20	80,39	6,46	0,5	0,95	0,01	75	
29	Slikken Zuid (stable)	58	Low	61301	418805	-10	SvF-58-0,10	43,06	2,84	21,3	11,40	0,66	17	
30		58				-40	SvF-58-0,40	79,41	8,08	1,0	1,46	0,03	52	
31		58				-80	SvF-58-0,80	80,54	7,62	0,4	1,13	0,01	88	
32		58				-120	SvF-58-1,20	80,06	7,06	0,7	1,20	0,01	86	
33	Slikken Zuid (stable)	59	Salt	61208	418614	-10	SvF-59-0,10	52,21	2,71	13,9	7,53	0,39	20	
34		59				-40	SvF-59-0,40	79,48	7,51	0,9	1,41	0,02	63	
35		59				-80	SvF-59-0,80	80,40	8,83	0,8	1,53	0,02	74	
36		59				-120	SvF-59-1,20	80,81	5,49	0,5	0,95	0,01	102	
Explanation														
1) Soil sample n	ot saturated due to lower v	vaterle	evel and du	ue to porew	ater sampli	ng.								
	Analytical result													
	Average of Duplo													
	Calculated													

<u>Remark</u>: Results of Dry Matter content are not fully final due to some minor weight corrections yet to be performed.