ANCIENT DNA IN PALEOSOLS, SW GREENLAND

A tool for retrospective paleoenvironmental studies?

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Abstract

Ancient DNA (aDNA) is a useful tool for retrospective paleoenvironmental studies. Paleosols formed in Arctic environments constitute a potential archive of aDNA from terrestrial organisms living in past environments, given that the cold and dry climate prevailing at high latitudes favors DNA preservation and hamper post-depositional mobility of deposited fragments. However, to what extent aDNA is preserved in old buried soil layers (paleosol layers) are not well known. This study assesses to what extent DNA older than 100 years is present in a paleosol profile from southwest Greenland. My main hypothesis was that aDNA from both plants and animals could be extracted from old buried soil layers. I found that oldest studied soil layers were more than 800 B.P. yr old. These old layers contained DNA from both plants and animals. The clean sampling protocol used showed no signs of contamination, suggesting that the DNA was from soil layers and not from modern contaminants. I conclude that my hypothesis seems valid and that a majority of the analyzed plant and animal DNA is ancient. Indeed, aDNA could be used to infer species presence in past paleoenvironments and widen our knowledge regarding how Arctic organisms coped with climatic perturbations and thus, improve our understanding how they will respond to future climatic change.

Key words: Ancient DNA, paleosol, paleoenvironment, Greenland, Arctic
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1 Introduction

The Intergovernmental Panel on Climate Change (IPCC) has projected an increase in temperature by 1.4-5.8°C around 2100 relative to 1990, with high latitudes and continental regions to be affected at a larger scale (Hansen and Lebedeff 1987; Lashof and Ahuja 1990; Houghton et al. 2001). This has led to a growing interest in research on climate-induced environmental change and impacts in the Arctic. Estimations show the potential effect from greenhouse gas emissions and carbon sequestration possibly raising the global temperature to a level exceeding any period within the past 1-40 Myr (Houghton et al. 2001). An increase in temperature could lead to a latitudinal decline of the Arctic habitats and thereby threatening the species depending on it and allowing boreal species currently established in the outlier regions of the Arctic, occupying a wide range of habitats, to expand north with the potential to outcompete the native species (Callaghan et al. 2004). A well-known example is the phenomenon called shrubification, with woody plants expanding to the Arctic, shown to alter numerous environmental factors such as the structure of tundra ecosystems, energy fluxes and the regional climate (Myers-Smith et al. 2011). Shrubification is expected to escalate due to the global warming and thereby increasing the local extinction risk (Mod and Luoto 2016). Recent and past effects on species due to climate changes (Parmesan and Yohe 2003; Root et al. 2003; Benton and Twitchett 2003; Overpeck, Whitlock and Huntley 2002) and the anthropogenic effect on the current climate could cause an extinction of species within the near future (Benton and Twitchett 2003; Brooks, Pimm and Oyugi 1999). Historical reconstructions are an important tool to understand previous extinction events. Under oxygen deficient conditions, preservation of DNA is possible due to the increased withstand of environmental changes such as humidity, pressure and temperature. This enables DNA to remain intact for thousands of years (Rosenberg and Rosenberg 2012) preserved as ancient DNA (aDNA). The Arctic climate is favorable for long-term preservation of DNA due to the constantly low temperature (Willemse et al. 2002) and aDNA originated from teeth, bones, plants and wood, categorized as macrofossils, within the studied area has widely been used to link past animals and plants in time and space (Willerslev and Cooper 2005). But this method is dependent on macrofossils, thus limiting its usage for historical reconstruction.

A more recent method is the usage of sedimentary aDNA deposited from cells and tissues such as urine, hair and skin, faeces, and other genetic material that can be leaked from animals and plants (Thomsen and Willerslev 2014). The studied sedimentary aDNA from Greenland has been preserved in ancient soils known as paleosols. This old soil was once the ground surface but was rapidly buried by loess sediment during the formation of aeolian sand-sheet deposits due to climatic, latitudinal, geomorphological, and other environmental changes (Filion 1984; Bégin et al. 1995). Sedimentary aDNA has shown to be of local origin (Haile et al. 2007) and has the potential to record physical, biological and chemical information about past conditions near Earth’s surface (Willemse et al. 2002; Tabor and Myers 2015) without the presence of macrofossils (Hebsgaard et al. 2008; Willerslev and Cooper 2005). Furthermore, it could be usable as a tool for monitoring past and present biodiversity (Thomsen and Willerslev 2014) with paleosols acting as an important resource for terrestrial environmental and climatic reconstructions (Tabor and Myers 2015). For example, by using sedimentary aDNA it has been shown that the survival of mammoth and horses in Alaska were thousands of years longer than what was previously thought when relying solely on macrofossils (Haile et al. 2009) and aDNA obtained directly from 450-800 000-year-old silty ice, stored at the base of the Greenland ice sheet, has revealed the youngest evidence of conifer forest in Greenland (Willerslev et al. 2007). Research on aDNA and rates of extinction has mainly focused on the large-mammal fauna - meanwhile, a majority of the small-mammal taxa has survived into the present. But even if a species survives great climatic changes such as the transition from glacial to interglacial periods, a dramatic decline in genetic diversity can be noticed when studying the
collared lemming *Dicrostonyx torquatus* (Prost et al. 2010). On the contrary from large mammals, the small-mammal fauna was not as vulnerable to human predation, thereby enabling a differentiation between human activity and climatic changes as a factor for the decline in genetic diversity (Brace et al. 2012).

Many of the previous studies about aDNA have been forced to rely on lake sediments, ice cores, or samples from various permafrost layers as this has been the most common method. But this has a disadvantage when considering the possibility to encapsulate material from a greater distance (e.g. water upstream or air from the atmosphere), resulting in an indirect archive for the surrounding terrestrial environment. Paleosols with aDNA has been used in far less extent but has the advantage to represent the direct terrestrial environment which it’s surrounded by. Although, paleosols has its disadvantages such as degradation of organic matter inhibiting the polymerase chain reaction (PCR) with humic acids (Matheson et al. 2010). Another risk with paleosols is the increased migration of DNA through the soil due to precipitation and leaching of plant DNA (Gulden et al. 2005; Poté et al. 2007), potentially contaminating ambient soils with external DNA and thereby increasing the risk of the analyzed DNA not being representative for its soil and depth.

The objective with this study was to analyze an Arctic soil profile for aDNA and evaluate its usage for reconstructing of historic plant and animal species. My hypothesis was that sedimentary DNA from ancient (>100 years old) plants and animals is preserved in Greenland paleosols.

## 2 Materials and methods

### 2.1 Study site

The fieldwork was performed about 25km east from the settlement of Kangerlussuaq (67°05'26.2"N 50°14'29.4"W), in the vicinity of Russell Glacier, southwest Greenland (Fig. 1). The area is based in the Precambrian zone, dominated by gneiss, once covered by ice (Storms, 2012), with its surroundings being one of the most studied areas in the Arctic. Kangerlussuaq has about 500 residents and is located at an extensive marine delta at the inner part of a 190km long fjord with the Watson River entering it, running from the Greenland ice sheet in the east. The environment towards the studied site is constituted by numerous geomorphological formations such as floodplains with dunes, braided and meandering river systems, moraine ridges, lakes, and low mountains (Yde et al. 2018). The climate is categorized as sub-Arctic, with a mean annual temperature of -5 to -6°C and a low mean annual precipitation (<200mm). By observations on the site the vegetation was noticed to be sparse and dominated by low shrubs such as *Salix* and *Betula*. The beginning of the last deglaciation in central West Greenland began around 8300-7500 B.P. yr cal (Willemse et al. n.d.) with the inland ice-margin reaching its present position around 5500 B.P yr cal (van Tatenhove et al. 1996) and experienced a recession during the early and mid-1900’s but between 1968 and 1999 a mean growth of about 7m yr⁻¹ was recorded (Knight et al. 2000). The deposition of aeolian sand in the area is believed to have been continuous after the latest deglaciation, with a significant increase in 900 B.P. yr cal. (Eisner et al. 1995) with the bulk of aeolian sands deposited prior to 3400 B.P. yr cal. and after 550 B.P. yr cal. (Willemse et al. 2002).
2.2 **Analysis of general paleosol properties**

General descriptions of stratigraphic units were determined visually in the field. Soil texture of the studied paleosol has previously been studied in detail using x-ray fluorescence geochemical composition (XRF) by Hällberg (2017), while organic matter content determined from mass losses after heating (LOI) has been previously been quantified by Sundin (2017), and these two datasets was compiled for assessment of the paleosol properties.

2.3 **Sampling**

Sampling was made in the beginning of August 2017 by digging a pit measuring 250cm in length, 145cm in depth and 125cm in width. A cleansuit, shoe protectors, face mask and a double pair of rubber gloves used on to prevent contamination of the soil with DNA from the sampler (Fig. 2). The pit exposed a profile with numerous aeolian stratifications. Areas for sampling was cleaned with an acid washed plastic spoon, removing any potentially contaminated soil and horizontally pressing an acid washed falcon tube as far as possible into the soil profile. Sampling was initiated from 145cm depth in the organic layer at the bottom, collecting cores upwards to limit contamination from the soil above, starting with every 5cm, changing to 10cm interval when reaching the loess soil at about 115cm depth. A total of 18 samples in three soil types were collected and classified by their stratigraphic unit using soil color charts (Munsell Soil Color Charts) and 1 surface sample was collected by pressing a tube vertically down in the soil. To verify that the method was minimizing the risk of samples being contaminated, DNA from Arctic char (*S. alpinus*) were applied at a sampled area and at a reference area sampled after being cleaned from char-DNA by using an acid washed plastic spoon. DNA were applied at 63cm (positive control) and 67cm depth (negative control) in the profile and sampled as previously.
2.4 Polymerase chain reaction

All samples were analyzed for plant and mammal DNA in a lab at Umeå University through a PCR analysis. Analyses were combined with a negative control, blanks and a positive control to verify the method and to exclude any contamination of the PCR reagents. All DNA extractions were carried out in a clean lab far from modern DNA labs and no DNA work or amplification has been performed previously as suggested (Cooper and Poinar 2000). The work bench was cleaned with bleach every time before DNA extraction. For plant and vertebrate analyses 0.3–0.5 g soil sample was used for DNA extraction by MoBio PowerSoil DNA Isolation Kit, and two replicates were performed for each soil sample. Two extraction blanks (extractions without soil) were collected for both plant and mammal analyses to detect sporadic contaminations from extraction procedure. The extracted DNA was amplified with different primer pairs targeting chloroplast P6 loop (trnL), Arctic char mitochondria (S. alpinus) and vertebrate mitochondrial (16S). Each DNA amplification was carried out in a 25 μL reaction containing 1X Taq buffer, 0.5 mM MgCl2, 0.2 mM of each dNTP, 0.4 μM of each primer, 0.625 U Taq DNA polymerase (Thermo Scientific), 2.5 μg of bovine serum albumin (BSA), and 1.5 μL of extracted DNA. PCR underwent the enzyme activation at 95°C for 3 min followed by 35 cycles consisting of 95°C for 30 seconds, annealing temperature (55°C for trnL, S. alpinus, and 16S) for 30–60 seconds, and 72°C for 30 seconds, followed by final elongation at 72°C for 5 min.

Mammal DNA amplification was made a second time, with the attempt to exclude human DNA. It was carried out in a 25 μL reaction containing 1X Taq buffer, 0.5 mM MgCl2, 0.2 mM of each dNTP, 0.4 μM of 16Smam primers (Willerslev et al. 2003), 2 μM of 16Smam_blkhum3 (Boessenkool et al. 2012), 0.625 U HotStartTaq DNA polymerase (QIAGEN), 2.5 μg of bovine serum albumin (BSA), and 1.5 μL of extracted DNA. 16Smam_blkhum3 is the Homo sapiens DNA blocking primer that could partly inhibit human DNA from being over-amplified. PCR underwent the enzyme activation at 95°C for 15 min and then 45 cycles consisting of 95°C for
30 seconds, 60°C for 30 seconds, and 72°C for 30 seconds, followed by final elongation at 72°C for 10 min.

2.5 Radiocarbon dating
Soil samples were manually sieved with 2.0mm aperture for macrofossils suitable (>3.0mg) for accelerator mass spectrometry (AMS) dating, using ^14C. A total of four macrofossils (Fig. 3) from various depths (24-26, 55-60, 110-115, and 122-124cm) were sent to, and analyzed, by Beta Analytic (4985 S.W. 74th Court Miami, FL USA 33155).

Figure 3. Macrofossils for radiocarbon dating using ^14C. A) Vascular plant. 24-26 cm depth. B) Moss, Dicranum. sp. 55-60cm depth. C) Wood fragment, likely Salix. 110-115cm depth. D) Wood fragment, likely Salix. 122-24cm depth.

3 Results

3.1 Stratigraphy
The paleosol was determined to have three main units based on the visual inspection in the field as well as the texture of the profile, with varying zirconium (Zr), silicon (Si), and lead (Pb) ratios along with a shift in sand (Fig. 4A), and LOI (Fig. 4B) concentrations. Section I: 0-65cm depth, consisting of mainly loess sediment, characterized by a high concentration of sand, and low carbon concentrations. Elevated Pb/Zr and Si/Zr ratios between 60-65cm depth. Stratigraphic unit at 10YR 6/2, with a lighter shade in the red and yellow spectrum. Section II: 65-120cm depth, consisting of mainly loess sediment but with increasing concentrations of organic matter when reaching 110cm depth, along with rapidly decreasing sand concentrations. LOI concentrations begins to vary. Elevated Pb/Zr and Si/Zr ratios between 65-85cm depth, with about 7x higher values at 75cm depth. Stratification layers becomes thicker and more distinct. Stratigraphic unit at 10YR 5/2, with a slightly darker shade than the sector above. Section III: 120-145cm depth, high in organic matter with LOI concentrations increasing drastically. Sand concentrations were at low. Pb/Zr and Si/Zr ratios returned to lower, stable values as seen in the first section. Stratigraphic unit at 10YR 3/1, with a considerably darker shade and a decreased purity in color.
3.2 Profile age
A higher abundance of woody fragments was noticed in the deeper part of the profile (>100 cm), with a higher frequency of mosses and roots towards the surface. The radiocarbon dating analysis (Fig. 4C) of the macrofossils dated the lower organic section at 122-124 cm depth to 1184-1275 +/- 30 cal. yr. A.D. with a 95.4% probability. Due to a large amount of iron replacement in the macrofossil at 110-115 cm depth and that it would dissolve completely away when using acid for the analysis, further analysis of this sample was therefore canceled. A high percent of modern carbon (pMC) in the two macrofossils in the top section (24-26 and 55-60 cm depth) inhibited a dating in calendar A.D. The amount was 101.38 +/- 0.38 pMC and 102.27 +/- 0.38 pMC respectively. Two possible ages could be inferred from these samples, including an age corresponding to the mid-1950 or from the last 10 years.

3.3 Ancient DNA analyses
3.3.1 Plants
No presence of the external tracer (char-DNA) were found before analyzing for plant and mammal DNA. Positive results from the PCR analysis for plant (chloroplast) DNA were found at all depths except in the organic layers below 120 cm (Fig. 4D). A clear positive plant DNA result of the organic extracts at 125 and 130 cm depth were visible after a 10x dilution. Chloroplast DNA is clearly shown at 200 bp, within 5-120 cm depth, with no DNA recorded in the negative control (Fig. 5A and 5B). Blanks tests and negative control analyses had no signs of contamination, with the positive control of pine indicating a strong hit (Fig. 5C).
Figure 5. Result from PCR analysis of plant DNA including duplicates. A) Result of chloroplast DNA at 5-65cm depth. N=Negative control. B) Result of chloroplast DNA at 65-120cm depth. N=Negative control. C) Blanks (B1-B3), positive control (Pine) and negative control (N) of 200bp, with duplicates.
3.3.2 Mammals
With the primer for human DNA blocker, positive hits of mammal DNA were found at 130 cm depth and towards the surface, with negative hits at 15, 25, 75 and 120 cm depth, along with no contamination in 2 out of 3 negative controls (Fig. 6). Specific targeting primers for muskox were unable to target any DNA, with negative results throughout the profile (Fig. 4D).

![Figure 6: Result from PCR analysis of mammal DNA, including duplicates. N=Negative control. A) Mammal DNA at 5-65 cm depth. B) Mammal DNA at 75-130 cm depth.]

4 Discussion

4.1 Profile origin
4.1.1 aDNA of plants and animals
The none-presence of the external tracer (char-DNA) in the samples suggest that contamination of modern DNA is very unlikely; hence, the DNA in my samples comes from the buried soil layers. In line with the main hypothesis my analysis revealed positive amplification of plant and vertebrate’s DNA from ancient soil layers, some being as old as 800 years. A high amount of organic matter in the organic layer (>120 cm depth) is most likely the reason for the negative plant DNA amplification because of PCR inhibition caused by a high concentration of humic acid in the extracts (Matheson et al. 2010) and is the reason that no soil below 130 cm depth were analyzed. This negative result itself does not necessarily imply that the DNA is not present, yet only that the analysis was not able to detect it. Positive results in the same layers were only visible with a 10x dilution, but simultaneously impairs the chance of detecting all DNA due to the increased amount of fluid compared to the amount of DNA.
Compared to the plants, the mammal DNA hits were overall positive, but with negative hits spread out at different depths, most likely due to the low abundance of mammal DNA and not being as concentrated as plant DNA. As the targeting for muskox DNA was negative, the origin of the analyzed DNA remains uncertain, thus increases the risk of the analyzed DNA to have been contaminated with contemporary DNA from layers above due to migration. Previous studies have detected sheep-DNA in 3300 years old sediment deposits in caves in New Zealand (Haile et al. 2007) and a recent study in the Kangerlussuaq area that simulated heavy rainfall on soil showed a migration of DNA to about 30cm depth within 24 hours (Karlsten 2017). Considering the thick, rapidly accumulated layer on top of the organic layer, acting as a protective barrier, combined with the preservative cold and dry climate of the Arctic, the chance of finding DNA at even larger depth seems likely. It also strengthens the possibility of the analyzed DNA, below 30cm depth in particular, to be ancient and not exposed to any contamination, but further analyses are required to verify this.

4.1.2 Accumulation
Average accumulation rate of the whole profile was 0.15cm yr$^{-1}$ based on the dated 125cm of soil, accumulated in a period of 800 +/- 30 years. But when considering the 60cm of soil with a high pMC, the accumulation rate becomes much higher, averaging 1cm yr$^{-1}$ over the last 60 years. This average aeolian sediment accumulation are in line with earlier studies in the area, estimating the overall aeolian sediment fluxes to 0.5-2.0cm yr$^{-1}$ (Willems et al. n.d). Because of the rapid accumulated modern layer, the average accumulation of the underlaying soil at 60-125cm depth becomes much lower, resulting in about 0.09cm yr$^{-1}$. A soil profile at a site within 300m of the studied area has previously been dated to a median age of 522-3423 B.P yr cal. with the top organic layer at 100cm depth dated at 549-653 B.P. yr cal. (Willems et al. 2002). With the 100cm depth and age acting as a reference, assuming an average accumulation rate of 0.09cm yr$^{-1}$, it would take about 278 years to accumulate another 25cm of soil, adding up to 827-931 yr B.P. This age falls within the radiocarbon dating result of 800 +/- 30 B.P. yr cal. at 125cm depth in the current studied profile. But spatial patterns are complex and greatly influenced by vegetation, irregular topography, grain sizes, and other factors that could affect the accumulation (Willems et al. n.d.). The rapid accumulation of aeolian sediment in the top 60cm layer could be due to a disturbance in the area during the current period, increasing the erosion and transforming the surrounding environment drastically.

4.2 Paleosol formation
A large proportion of the upper profile was young and deposited after the 1950s. The high proportion of modern carbon (pMC) in two of the macrofossils (24-26 and 55-60cm depth) is most likely due to the atmospheric atomic bomb tests in the 1950’s and 60’s, vastly increasing the atmospheric radiocarbon values (Hua et al. 2013). Consequently, samples younger than about 1850 A.D. is hard to date correctly due to the increase of $^{14}$C in the specimen, and only the raw radiocarbon measurement as percent of the standard is retrieved (Hua et al. 2013). Using XRF to analyze the current profile has shown various ratios of Si, Zr and Pb, with stable Si/Zr and Pb/Zr ratios but with high peaks at 65-85cm depth (Hällberg 2017), indicating a change of the chemistry in the soil. Historical events with deposition of Pb on the Northern Hemisphere has spread as long-range air pollution (Rosman et al. 1997), and accumulates in the ground surface (Klaminder et al. 2008). Studies of the Greenland ice-sheet has previously been used to shown atmospheric pollution history at remote points at high latitudes (Wolff and Peel 1985). After the Second World War, the deposition of Pb increased considerably with a peak about 1970 (Renberg et al 2006), and considering the elevated Pb concentrations in the profile at 65-85cm depth, it seems most likely that this layer was the ground surface during the enrichment of Pb in the soil, exposed to the atmospheric deposition and became buried during the 1950’s.
But what could have generated such a dramatic accumulation of 60cm sand in just a few decades? Human and animal presence, alone or combined, could be a possible contributor for the increased accumulation of aeolian transported sediments in the area. A U.S. military air base named Bluie West Eight (BW-8), currently known as Sondrestrom Air Base, were established in 1941 during the Second World War and is still operating, although in smaller scale, under Danish control. The air base and the expansion of Kangerlussuaq has enabled large aircrafts to land in the area, allowing the tourism in West Greenland to grow and explore the surrounding nature, such as the area around Russell Glacier. Furthermore, in 1962 and 1965, 27 muskoxen were translocated from East Greenland to the area around Kangerlussuaq, with the objective to stabilize the meat resource for the Inuit hunters, and at the same time safeguarding the muskox against extinction in Greenland, should the indigenous muskoxen of Northeast and North Greenland perish (Olesen 1991). The muskox population increased rapidly with increment averages at 30%, highest ever recorded for this species, reaching 2600 individuals in 1990 (Olesen 1991). During the same period, the first signs of environmental effects were noticed, such as overgrazing and the destruction of willow thickets, shortly followed by an increased erosion of certain habitats (Olesen 1991). This phenomenon of an abrupt increase in erosion rate has been previously been studied on a sub-Antarctic island, where plant communities remained stable for centuries, until the detection of rabbit DNA in sediments, followed by the erosion rate to increase by double (Ficetola et al. 2018). It is difficult to specify any climatic event during the period that could have affected the profile since it is young, combined with the possible anthropogenic effects. But studies have detected a change in aeolian activity in Greenland and across the Arctic due to climate changes (Heindel et al. 2017). A change in the ice margin position that could affect the aeolian sediment flux in the area could be possible, with a withdrawal of the ice sheet leading to more land to be exposed to erosion, thus increasing the accumulation. This possibility remains yet to be evaluated. More recently, with a period of warming between 1981-2011 (Hanna et al. 2012), an enhancement of the rapid accumulation would be possible as well.

4.3 Historical reconstructions
The age of the analyzed sedimentary aDNA was within Holocene and enables the study of paleoecological development during a small part of this epoch. The global mean temperature has experienced an increase by 0.5°C over the past 150 years (Trenberth et al. 2007), but the Arctic has experienced a raise more than 2-3 times as high and even exceeds the century-scale warming during the period at the Pleistocene-Holocene transition about 11kyr ago (Overpeck et al. 1996). With the predicted climate changes, it has become more important to learn about the past to try to predict the future and what we might expect. A warmer climate, limiting the habitats, and pushing the latitudinal border north, will most likely influence the species currently living in Arctic environments. With the limited dating, along with the difficulty in resolving the history between the younger layer and its transition to the organic zone, as no radiocarbon dating was made at this depth, a more precise dating is crucial. Carbon extracted from the soil could be a tool for dating, but would result in an average age of the depth sampled, with an increased uncertainty of any event that could have affected the environment. Further studies would have to extend the area and study more than only one pit, how it may vary between different Arctic environments and not just merely in West Greenland. It would be in my wish to analyze the DNA further by sequencing it to clarify which species the DNA originated from, date more macrofossils and hopefully enable a link between species during specific time periods and climates. Previous paleoenvironmental studies and historical reconstructions has relied much on other sources for data, such as macrofossils or sediments. Being able to use aDNA from paleosols, the possibilities of historical reconstructions is broadened and might be a tool to anticipate on how different terrestrial ecosystems will be affected in the future, with species declining or becoming extinct, along with the migration of southern species to the Arctic.
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6 References


Hanna, E., Mernhild, S.H., Cappelen, J. and Steffen, K. 2012. Recent warming in Greenland


Hällberg, P. 2017. A proglacial paleosol sequence in West Greenland – Paleoenvironmental implications of magnetic and geochemical properties. *(Non-published).*

Karlsten, A. 2017. Rain: A possible transporter of animal DNA to old soil layers. *(Non-published)*


