Science Advances

AAAS

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Supplementary Materials for

DNA from lake sediments reveals long-term ecosystem changes after a biological invasion

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Published 9 May 2018, *Sci. Adv.* **4**, eaar4292 (2018)
DOI: 10.1126/sciadv.aar4292

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Other Supplementary Material for this manuscript includes the following:
(available at advances.sciencemag.org/cgi/content/full/4/5/eaar4292/DC1)

Data file (Microsoft Excel format)

Supplementary Materials

appendix S1. The Kerguelen Islands: Historical information on rabbit introduction

The sub-Antarctic islands are extremely isolated. These islands, previously inaccessible to land animals by natural colonization are characterized by the absence of native mammal herbivores. Following their discovery in 1772, the Kerguelen Islands were subject to numerous alien plant and animal introductions (both desired and accentual) that have resulted in substantial changes to ecosystem structure and functioning (*32*). Among these introductions of alien species, the introduction of rabbits (*Oryctolagus cuniculus*) likely had the strongest effect on plant communities. Rabbits were introduced in 1874 by the sailors of HMS *Volage* (captain H. Faifax) (*12*). This expedition, dedicated to the observation of the transit of Venus, has notably stayed in the “baie de l’observatoire” (at the bottom of the golf of the morbihan - 15 km of the studied area). Thirty years later, E. Werth of the German expedition of the Gauss (1901-1903) observed that a 1,000 m coastal strip was already heavily colonized by rabbits with the disappearance of Kerguelen cabbage (*Pringlea antiscorbutica*), the rarefaction of *Azorella selago* and on the contrary, the predominance of *Acaena magellanica.* This effect on native plant communities has since been observed multiple times (*12, 13, 36*). The colonization of the main island likely occurred trough a step by step, colonization, and not trough long range migration (*30*). If in 1932 "only the eastern part of the island was colonized by rabbit" (*31*), while Lesel in 1967 affirms that "it can be said that every sector of the main island is occupied by the rabbit"(*30*). This suggests that the Lac La Poule Drainage has likely been colonized after 1932 but before the 1960’s, in strong agreement with the dates obtained through sediment analyses. The effect of rabbits on the ecosystem having become major, a campaign of eradication by mixomatosis was launched in 1955-1956. The voluntary introduction of the mixomatosis virus initially caused a major reduction of rabbit populations (with a minimum described around 1960-1961). However, even though many rabbits were still mixomatous in 1994 (*32*), Lesel considers that the population was again stable in the late 1960's (*30*).

appendix S2. Supplementary results

Analyses of plant communities were repeated excluding *Agrostis magellanica* and *Taraxacum officinale* (plant species detected in 1.5% of controls). The multivariate regression tree (cross-validation error = 0.456) confirmed the abrupt change of plant communities after 1948, with significant differences between the communities observed until 1948, and the most recent ones (permutation test: $P < 0.0001$, $R = 0.71$). Also in this case, rabbit abundance was a strong predictor of changes of plant communities, and explained most of variation (RDA: $P < 0.0001$, $R^2 = 0.73$).

The relationship between erosion rate and rabbit occurrence was repeated using rabbit sedDNA as dependent variable, instead of the abundance of *Sporormiella*. Also in this case, the variation in erosion rate was significantly related to rabbit occurrence (autoregressive model: $t_{18} = 2.95$, $P = 0.0085$, likelihood-ratio $R^2 = 0.33$).

Analyses were also repeated using *Sporormiella* concentration instead of influx. *Sporormiella* concentration was strongly correlated to influx ($r = 0.99$, $P < 0.001$), and all results were nearly identical if *Sporormiella* concentration (Fig. S4) was used instead of influx. Specifically, the detection of rabbit sedDNA and the concentration of *Sporormiella* spores were strongly correlated ($r = 0.78$, $N = 20$, $P < 0.0001$). In redundancy analysis, changes of plant communities were strongly related to *Sporormiella* concentration ($P < 0.0001$, $R^2 = 0.65$). Finally, the relationship between the variation in erosion rate and *Sporormiella* concentration was very strong (autoregressive model: $t_{18} = 5.28$, $P < 0.0001$, likelihood-ratio $R^2 = 0.61$).

appendix S3. Amplification of plant and mammal taxa using the P007 and g-h primers

For plants, in silico analyses were possible for sequences from 30 individuals representing 28 plant species for which reference sequences are available (*13*) (table S4). The g-h primers are able to amplify all these species; the average number of mismatches was 0.43 for the forward primer, and 0.63 mismatches for the reverse primer; amplicon length ranged between 14 and 63 bp (table S4). If only these species of Kerguelen flora was used as reference database, the resolution capacity of the primer was 100%; if the whole GenBank is used, the resolution of the primers was 87%. For animals, in silico analyses performed on representative GenBank sequences confirmed that the primers are able to amplify the alien herbivores on Kerguelen, as there are no mismatches on the primer regions (table S4).

table S1. Detection of NPPs in the analyzed sediment samples.

Date (Min-Max)	*Sporormiella* (spores)	Multi-cellular ascospores of *Sporormiella*	*Podospora* (spores)	*Lycopodium clavatum* (marker)	Sample mass (g)
1998-2013	8	0	0	721	0.59
1988-1998	16	0	0	1639	0.67
1980-1988	14	0	0	1646	0.72
1972-1980	40	0	0	1036	0.81
1964-1972	15	0	0	874	0.60
1962-1964	26	0	0	1168	0.67
1959-1962	16	0	0	1021	0.53
1956-1959	29	1	1	1160	0.72
1952-1956	19	0	2	386	0.59
1948-1952	14	1	1	1051	0.67
1941-1948	19	1	0	679	0.96
1932-1941	0	0	0	798	0.88
1919-1932	0	0	0	397	0.92
1905-1919	0	0	0	551	0.79
1889-1905	0	0	0	703	0.95
1863-1882	0	0	0	505	0.91
1818-1842	0	0	0	739	1.68
1580-1617	0	0	0	441	0.55
1478-1520	0	0	0	463	0.43
1387-1432	0	0	0	728	0.53

table S2. Detection of sedDNA in the analyzed sediment samples. For rabbit (*Oryctolagus cuniculus*) we report the number of PCRs in which we detected DNA; for plants, we report the relative abundance of sedDNA within samples.

Date						Species			
Min-Max	*Oryctolagus cuniculus*	*Azorella selago*	*Acaena magellanica*	*Ranunculus biternatus*	*Agrostis magellanica*	*Taraxacum officinale*	*Pringlea antiscorbutica*	*Festuca contracta*	*Deschampsia antarctica*
1387-1432	0/8	68.0%	28.8%	0.6%	0.0%	0.0%	0.0%	2.6%	0.0%
1478-1520	0/8	71.3%	18.2%	10.0%	0.0%	0.0%	0.0%	0.0%	0.5%
1580-1617	0/8	72.9%	21.3%	5.6%	0.2%	0.0%	0.0%	0.0%	0.0%
1818-1842	0/8	51.8%	41.6%	6.6%	0.0%	0.0%	0.0%	0.0%	0.0%
1863-1882	0/8	44.5%	46.9%	7.0%	0.0%	0.0%	1.7%	0.0%	0.0%
1889-1905	0/8	59.1%	35.7%	4.8%	0.5%	0.0%	0.0%	0.0%	0.0%
1905-1919	0/8	53.0%	43.9%	2.9%	0.0%	0.0%	0.2%	0.0%	0.0%
1919-1932	0/8	48.0%	47.2%	3.3%	0.0%	0.0%	0.0%	1.5%	0.0%
1932-1941	0/8	44.0%	40.4%	11.9%	0.0%	0.0%	0.0%	3.7%	0.0%
1941-1948	1/8	94.5%	4.9%	0.2%	0.1%	0.0%	0.0%	0.2%	0.0%
1948-1952	0/8	36.4%	48.1%	10.4%	1.4%	3.7%	0.0%	0.0%	0.0%
1952-1956	0/8	41.1%	51.7%	3.7%	3.2%	0.0%	0.0%	0.4%	0.0%
1956-1959	2/8	28.5%	64.7%	3.8%	2.8%	0.1%	0.0%	0.0%	0.0%
1959-1962	3/8	28.0%	67.9%	1.6%	1.3%	0.0%	0.0%	0.6%	0.5%
1962-1964	1/8	20.9%	70.9%	4.1%	3.7%	0.0%	0.0%	0.0%	0.4%
1964-1972	4/8	23.2%	67.6%	4.8%	3.4%	0.0%	0.0%	0.0%	1.0%
1972-1980	3/8	23.7%	71.3%	2.2%	2.8%	0.0%	0.0%	0.0%	0.0%
1980-1988	4/8	23.6%	71.5%	1.1%	3.5%	0.0%	0.0%	0.3%	0.0%
1988-1998	1/8	13.4%	79.5%	1.9%	2.0%	1.4%	0.0%	0.0%	1.9%
1998-2013	1/8	10.1%	70.2%	0.9%	13.9%	4.8%	0.0%	0.0%	0.0%

table S3. Estimation of breakpoints in the erosion rate performed using a dynamic programming algorithm (*59*). BIC: Bayesian Information Criterion. The lowest BIC value indicates the model with the most likely number of breakpoints.

N breakpoints	BIC	Most likely dates
0	-20.87	-
1	-29.27	1948
2	-38.10	1948, 1964

table S4. Average climatic variables during the period 1912–2013, calculated on the basis of the 2014 version of the University of Delaware time series of monthly global gridded high-resolution station data (*60*).

Period	Mean annual temperature (°C)	Mean monthly precipitation (mm)
1912-1919	2.6	80.8
1919-1932	2.7	71.0
1932-1941	2.9	80.6
1941-1948	2.7	69.6
1948-1952	2.3	69.3
1952-1956	2.7	67.1
1956-1959	2.5	72.7
1959-1962	2.5	70.5
1962-1964	2.3	71.2
1964-1972	2.5	72.4
1972-1980	2.9	66.8
1980-1988	3.2	65.7
1988-1998	3.2	57.2
1998-2013	3.1	56.2

table S5. Match between primers and sequences from plants and mammals living in Kerguelen. In-silico analyses of the match a) between the g-h primers and plants of the Kerguelen flora; b) between the MamP007 primers and representative GenBank sequences of alien herbivores present in Kerguelen.

a) plants

specimen	Species	family	forward errors	reverse error	Amplicone length
1	*Azorella selago*	Apiaceae	0	0	50
2	*Azorella selago*	Apiaceae	2	2	50
3	*Taraxacum officinale*	Asteraceae	0	1	51
4	*Leptinella plumosa*	Asteraceae	0	1	51
5	*Pringlea antiscorbutica*	Brassicaceae	0	0	40
6	*Colobanthus kerguelensis*	Caryophyllaceae	0	1	63
7	*Stellaria alsine*	Caryophyllaceae	3	1	57
8	*Cerastium fontanum*	Caryophyllaceae	0	2	53
9	*Crassula moschata*	Crassulaceae	0	1	55
10	*Crassula moschata*	Crassulaceae	0	1	55
11	*Cystopteris fragilis*	Cystopteridaceae	3	3	22
12	*Trifolium repens*	Fabaceae	0	1	53
13	*Hymenophyllum peltatum*	Hymenophyllaceae	3	2	19
14	*Lycopodium magellanicum*	Lycopodiaceae	2	1	22
15	*Montia fontana*	Montiaceae	0	2	14
16	*Callitriche antarctica*	Plantaginaceae	0	0	44
17	*Poa kerguelensis*	Poaceae	0	0	54
18	*Aira praecox*	Poaceae	0	0	54
19	*Deschampsia antarctica*	Poaceae	0	0	54
20	*Poa nemoralis*	Poaceae	0	0	54
21	*Dactylis glomerata*	Poaceae	0	0	58
22	*Festuca contracta*	Poaceae	0	0	54
23	*Holcus lanatus*	Poaceae	0	0	54
24	*Elymus repens*	Poaceae	0	0	53
25	*Poa cookii*	Poaceae	0	0	54
26	*Agrostis magellanica*	Poaceae	0	0	59
27	*Ranunculus repens*	Ranunculaceae	0	0	44
28	*Ranunculus biternatus*	Ranunculaceae	0	0	48
29	*Ranunculus pseudotrullifolius*	Ranunculaceae	0	0	48
30	*Acaena magellanica*	Rosaceae	0	0	53

b) mammals

Accession number	Species	family	forward errors	reverse error	Amplicone length
KJ870169.1	*Rangifer tarandus*	Cervidae	0	0	74
DQ334838.1	*Oryctolagus cuniculus*	Leporidae	0	0	79
HM236185.1	*Ovis aries musimon*	Bovidae	0	0	74

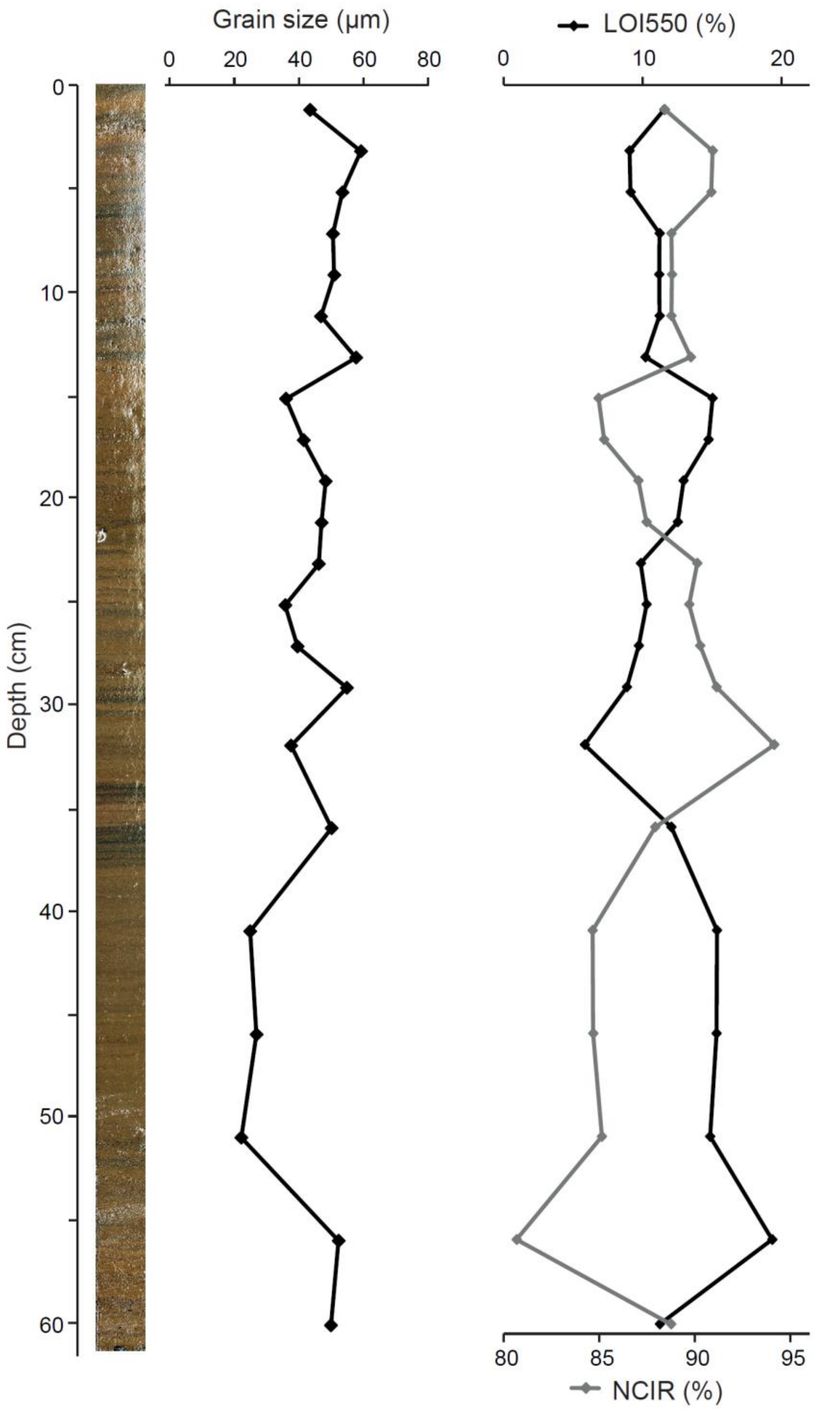

fig. S1. Data from core POU14P1 with photograph, grain size median, LOI550 (organic matter), and NCIR.

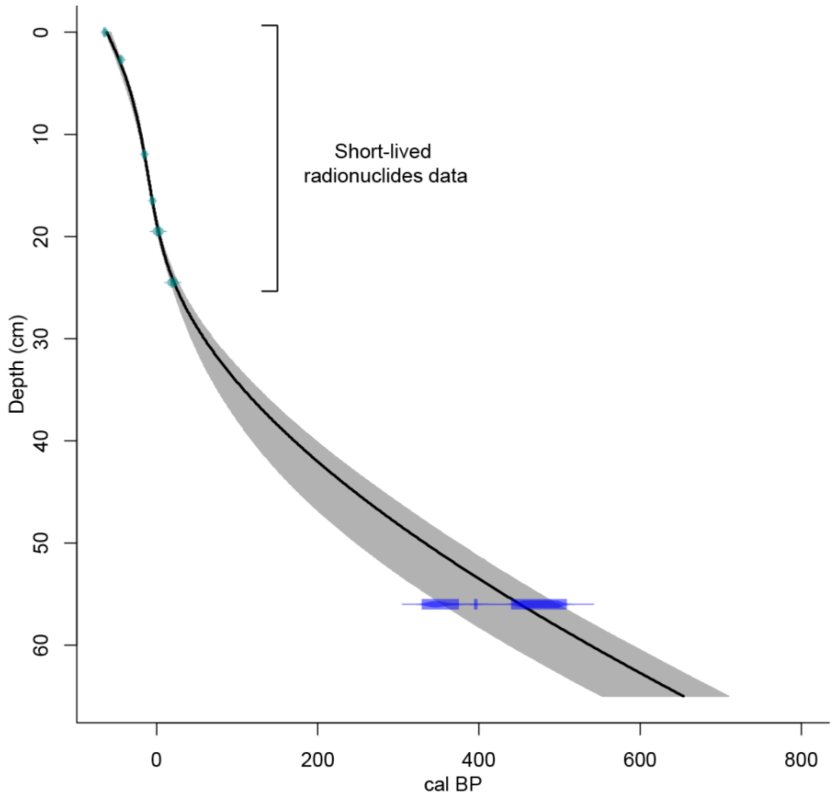

fig. S2. Age-depth model for the upper 65 cm of the sediment core POU14P1 based on radiocarbon (blue) and short-lived radionuclides derived ages (green).

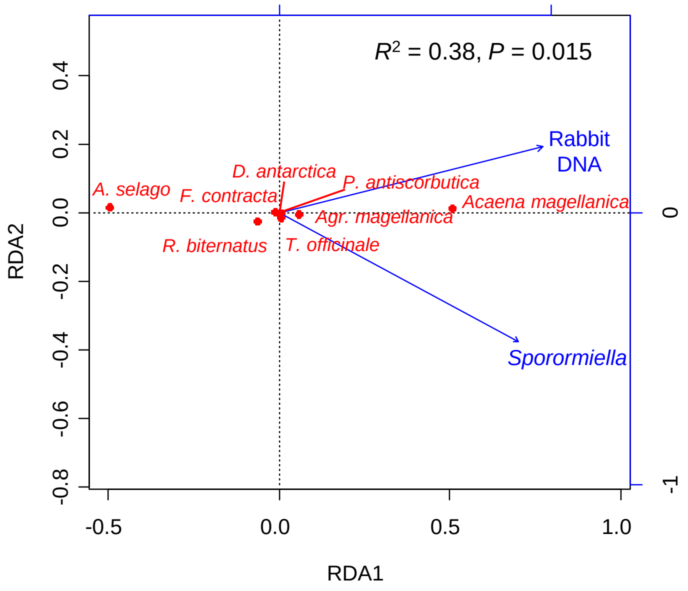

fig. S3. Constrained RDA, showing the relationship between the proxies of rabbit abundance (sedDNA and *Sporormiella*) and the abundance of different plant taxa. The analysis was repeated including the outlier sample (1941-1948). Taxa with negative scores along the first RDA axes have negative association with rabbit abundance.

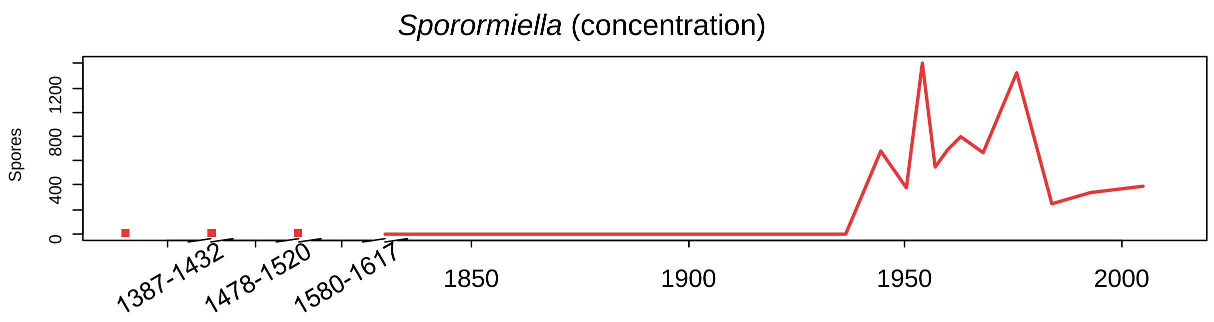

fig. S4. Temporal variation of the accumulation rate of spores of coprophilous fungi (*Sporormiella* sp.), measured as concentration (spores/cm^3).