Electronic Supplementary Information (ESI)

Nitrification in a Soil-Aquifer Treatment system:
Comparison of Potential Nitrification and Concentration Profiles in the Vadose Zone

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Section S1: Table of primary water quality parameters

Table S1: Primary water quality parameters before and after the SAT process, averaged for 2015 flood cycles. The removal percentage is depicted for the Yavne 2 SAT [1]

Parameter[†]	Units	Yearly average concentrations 2015		Removal %	Number of samples
		Before SAT (Station 6)	After SAT (Observation well T/282)		Before/After SAT
DOC	mg/L	10.6±1.0	1.6±0.3	88	26/13
DO	mg/L	2.5±0.4	<1	-	29/13
Ammonia, as N	mg/L	4.02±1.50	<0.02	>99	39/13
Nitrate, as N	mg/L	0.85±0.7	2.71±0.82	-	32/13
Nitrite, as N	mg/L	1.07±0.6	0.01±0.01	99	31/13
Calcium	mg/L	63.0±4.0	75.5±4.6	-	96/10
Magnesium	mg/L	14.8±2.4	19.1±2.3	-	96/10
Sodium	mg/L	150.5±13.6	162.5±11.8	-	96/10
Potassium	mg/L	20.1±1.0	22.1±1.9	-	96/10
pH	-	7.4±0.1	7.2±0.1	-	126/13
Temperature	ºC	18.1±6.4	26.0±1.7	-	22/13

[†] **Dissolved Organic Carbon (DOC); Dissolved Oxygen (DO).**

Table S2: Monthly averages of ammonia as N for the 2015 flood cycles [1].

Parameter	Units	Monthly averages[†]											
		1	2	3	4	5	6	7	8	9	10	11	12
Ammonia, as N	mg/L	5.51	4.73	2.99	3.3	2.34	2.67	3.17	3.47	3.12	6.13	5.08	5.77

[†] **The number of tests per month were 3-5. The relative standard deviation was 7-30%.**

Section S2: Methods: Experimental protocols. Potential nitrification protocol and ammonium monitoring

Potential Nitrification (P.N) determination

P.N experiments were carried out according to the nitrite oxidation inhibition method described by Schinner et al., 1996[2] for the soil and water samples: 7 gr of wet soil/water were placed in a 50 mL Erlenmeyer flask. 20 mL of 0.5 mM of NH_4^+ solution and 0.1 mL of $NaClO_3$ were added to the soil. The slurry was then incubated for 5 hr in a rotary shaker at a set temperature of 25.0 ± 0.1 °C. After incubation, 5 mL of 2M KCl were added. The sample was stirred and 5 mL of solution were filtered through a 0.22 μm PVDF membrane filter (Millex®-GV). To the filtrate, 3 mL of ammonium chloride buffer (0.19M, pH 8.5) were added, followed by 2 mL of a color reagent. The reagent was prepared by dissolving 2 gr of sulfanilamide and 0.1 gr of N-(1-naphtyl)-ethylenediamine hydrochloride in 20 mL of concentrated phosphoric acid and 180 mL of distilled water. After 15 min, a reading was taken at 520 nm using a spectrophotometer (UV-2600, Shimadzu). The concentrations were calculated from a 7 point calibration curve (R^2=0.9997). Results are portrayed in units of mg NH_4^+ as N per Kg of dry weight per day for solid samples or as mg NH_4^+-N $L^{-1} d^{-1}$ for the water samples.

Ammonium monitoring as a function of depth and time

The extraction and determination of ammonium was carried out according to ISO/TS 14256-1[3]. Briefly, 25 mL of 2M KCl were added to 5 mL of wet soil or water, shaken for half an hour and then filtered through a 0.22 μm PVDF membrane filter (Millex®-GV) to give a total of 5 mL of clear solution. An ammonium kit was used for colorimetric determination of ammonium concentration (Ammonium Test 1.14750, Merck-Millipore Co.).

Section S3: Calculation of the ammonium adsorbed fraction

CEC is expressed in meq per 100 gr of dry matter. It depends on soil surface area, which increases with clay and organic matter content[4]. The cations, which are commonly adsorbed on the soil are Ca^{2+}, Mg^{2+}, Na^+, K^+. The fraction of adsorbed cation in soil β_I is defined as the percentage of the exchangeable ion from the total CEC, where I is the exchangeable ion. Hence, the fractions follow eq. [S1]:

$$[S1] \quad \beta_{Ca^{2+}} + \beta_{Mg^{2+}} + \beta_{Na^+} + \beta_{K^+} = 1$$

However, in many cases, where additional cations are present, the ratios change accordingly. In our case, where NH_4^+ is present, then eq. [S2] applies[4]:

$$[S2] \quad \beta_{Ca^{2+}} + \beta_{Mg^{2+}} + \beta_{Na^+} + \beta_{K^+} + \beta_{NH_4^+} = 1$$

The fractions of different ions can be calculated based on repeated thermodynamic exchange coefficients[4]. These coefficients are the result of model fitting of a database of experimental results of the correlations between ion concentrations in the solute and soil[4–9]. For convenience, the exchange coefficients are represented relative to Na^+. As a result, the individual cation fraction can be written as eq. [S3]-[S6][4]:

$$[S3] \quad \beta_{Ca^{2+}} = \frac{\beta_{Na^+}^2 \cdot [Ca^{2+}]}{K_{Na^+ \backslash Ca^{2+}} \cdot [Na^+]^2}$$

$$[S4] \quad \beta_{Mg^{2+}} = \frac{\beta_{Na^+}^2 \cdot [Mg^{2+}]}{K_{Na^+ \backslash Mg^{2+}} \cdot [Na^+]^2}$$

$$[S5] \quad \beta_{K^+} = \frac{\beta_{Na^+} \cdot [K^+]}{K_{Na^+ \backslash K^+} \cdot [Na^+]}$$

$$[S6] \quad \beta_{NH_4^+} = \frac{\beta_{Na^+} \cdot [NH_4^+]}{K_{Na^+ \backslash NH_4^+} \cdot [Na^+]}$$

Where $K_{Na\backslash I}$ is the exchange coefficient between Na⁺ and the relevant cation. The exchange coefficients for the different cations, as presented in Appelo and Postma, 2005[4], are: $K_{Na\backslash Ca}$ = 0.4; $K_{Na\backslash Mg}$ = 0.5; $K_{Na\backslash K}$ = 0.2; $K_{Na\backslash NH4}$ = 0.25.

Substitution of eq. [S3]-[S6] in equation [S2], gives a quadratic equation (eq. [S7]):

$$[S7] \quad \beta_{Na^+}^2 \cdot \left(\frac{[Ca^{2+}]}{K_{Na^+\backslash Ca^{2+}}^2 \cdot [Na^+]^2} + \frac{[Mg^{2+}]}{K_{Na^+\backslash Mg^{2+}}^2 \cdot [Na^+]^2} \right) + \frac{\beta_{Na^+} \cdot [K^+]}{K_{Na^+\backslash K^+} \cdot [Na^+]} + $$

$$+ \frac{\beta_{Na^+} \cdot [NH_4^+]}{K_{Na^+\backslash NH_4^+} \cdot [Na^+]} + \beta_{Na^+} = 1$$

Using the concentration of cations, as were measured for the effluents (Table S1), we receive the following cation fractions (Table S2):

Table S3: Fraction of adsorbed cations (%)

$\beta_{Ca^{2+}}$	73.08 ± 2.32
$\beta_{Mg^{2+}}$	17.97 ± 1.97
β_{K^+}	2.22 ± 0.03
β_{Na^+}	5.63 ± 0.38
$\beta_{NH_4^+}$	1.10 ± 0.07

The adsorbed ammonium occupies 1.1% of the CEC of the soil.

Section S4: MATLAB interpolation of field data

For fig. 3 in the main article, the results of ammonium concentration, as were measured at different depths and time points, were interpolated into a 2D representation of the upper 80 cm of the unsaturated zone for 82 hr.

For the interpolation, MATLAB® (version 9.1.0.441655 (R2016b), release September 7, 2016) using the following script:

```
%% time is the matrix of time, depth is the matrix of depth and fullmatrix
are the concentrations.
```

```
[xx,yy]= meshgrid(time,depth);
New_Time= interp(time,5);New_Depth=interp(depth,5);
[xx2,yy2]= meshgrid(New_Time,New_Depth);
New_Matrix2= interp2(xx,yy,fullmatrix,xx2,yy2);
figure
surf(New_Time,New_Depth,New_Matrix2,'LineStyle','none','FaceColor','interp','
CDataMapping', 'scaled');
colormap('jet')
colorbar
view([0 90])
axis('ij','tight')
xlabel('Time [hr]','FontSize',20);ylabel('Depth [cm]','FontSize',20);
```
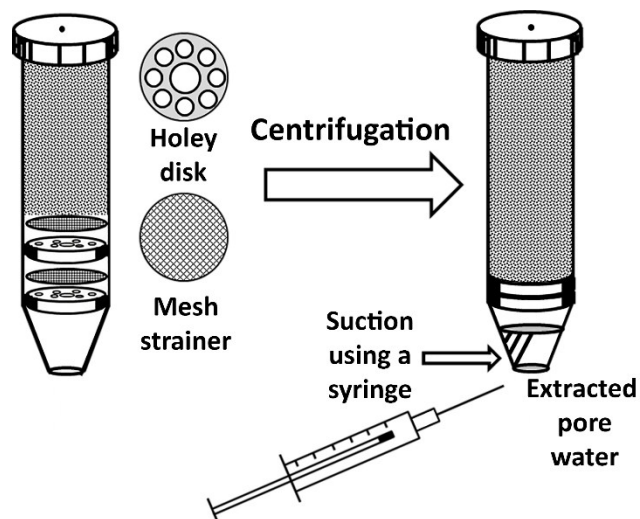
Section S5: Supporting figures

Figure S1: Homemade soil centrifugation system, equipped with Delrin^{TM} holey disks and stainless steel mesh strainers. The pieces are supported by the bottom cone of a Falcon tube. Soil is filled into the Falcon tube and centrifuged. The pore water is collected from the bottom cone by a syringe.
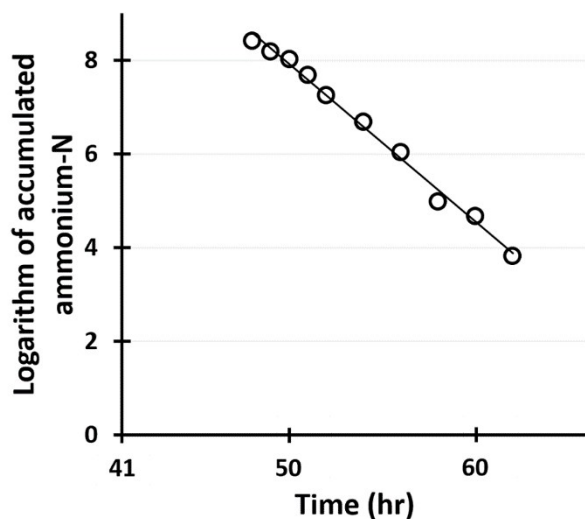
**Figure S2: Logarithm of total ammonium-N mass per m²
as a function of time, displaying a linear (R² = 0.9930)
first order kinetics.**

References for the ESI

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9   G. L. Gaines and H. C. Thomas, *J. Chem. Phys.*, 1953, **21**, 714–718.