# Supplemental Online Material (SOM)

# An automated apparatus for the simulation of prebiotic wet–dry cycles under strictly anaerobic conditions

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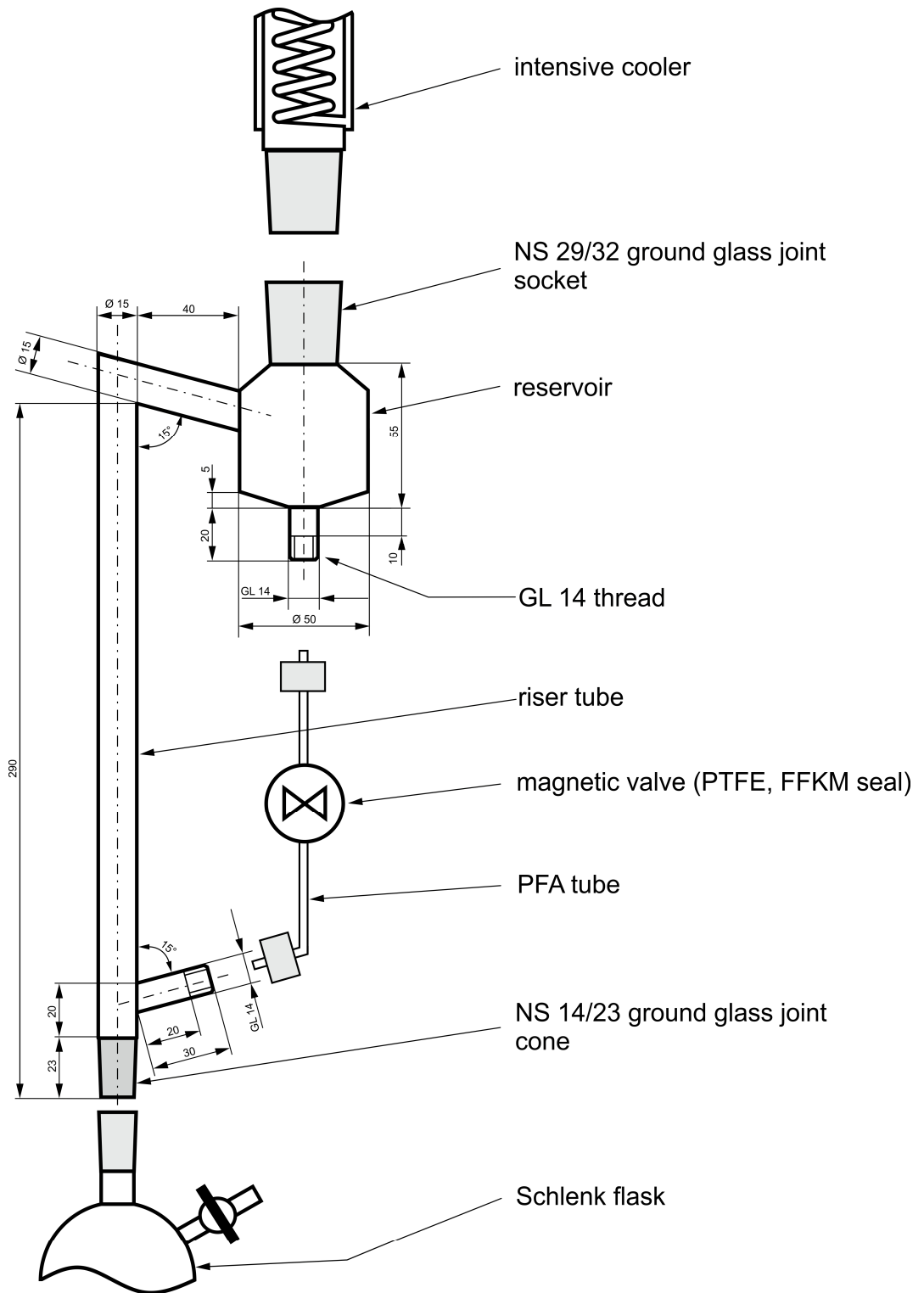
**SOM-Fig. 1.** Technical drawing of the wet–dry apparatus (WDA). Dimensions are given in millimeters.

**Details of the hardware and software setup of the WDA controller**

*Hardware*

Eight GPIO (general purpose input/output) ports are used to control an eight-channel 5 VDC relay module (SainSmart, Lenexa, Kansas, USA); each GPIO port controls one relay (green lines in Fig. 4). The relay module requires an operating and controlling voltage of 5 V. The voltage of the GPIO output, however, is only 3.3 V when active (0 V when inactive). Therefore, transistor circuits were necessary, which were prepared according to the manufacturer's instructions. A power supply RS-35-5 35 W 5 VDC (Mean Well, New Taipei City, Taiwan) is used, which also provides power to the Raspberry Pi. Each relay closes or opens the 230 VAC current circuit of a magnetic valve, which accordingly is open or closed.

*Software*

The GPIO ports are enabled and configured to output by an additional bash script. This script runs at the end of the system boot process and is therefore an entry in the /etc/rc.local. After all ports that are necessary to control the relay module have been successfully enabled and configured, and only then, the script activates an additional GPIO port to which an LED is connected. Only after all needed ports have been successfully enabled and configured, the LED glows and indicates the readiness for use.

Each of the eight relays on the relay module is controlled by one GPIO port. Therefore, for each relay a bash script exists. Thus, eight wet–dry apparatuses can be used independently from each other. The script activates the corresponding GPIO port, which subsequently activates the connected relay, resulting in opening of the magnetic valve attached to this relay. Conversely, this means that the deactivation of the GPIO port leads to deactivation of the relay and closure of the magnetic valve. The activation and deactivation occur according to the instructions that have been entered by the user via a graphical user interface (see SOM-Fig. 2).
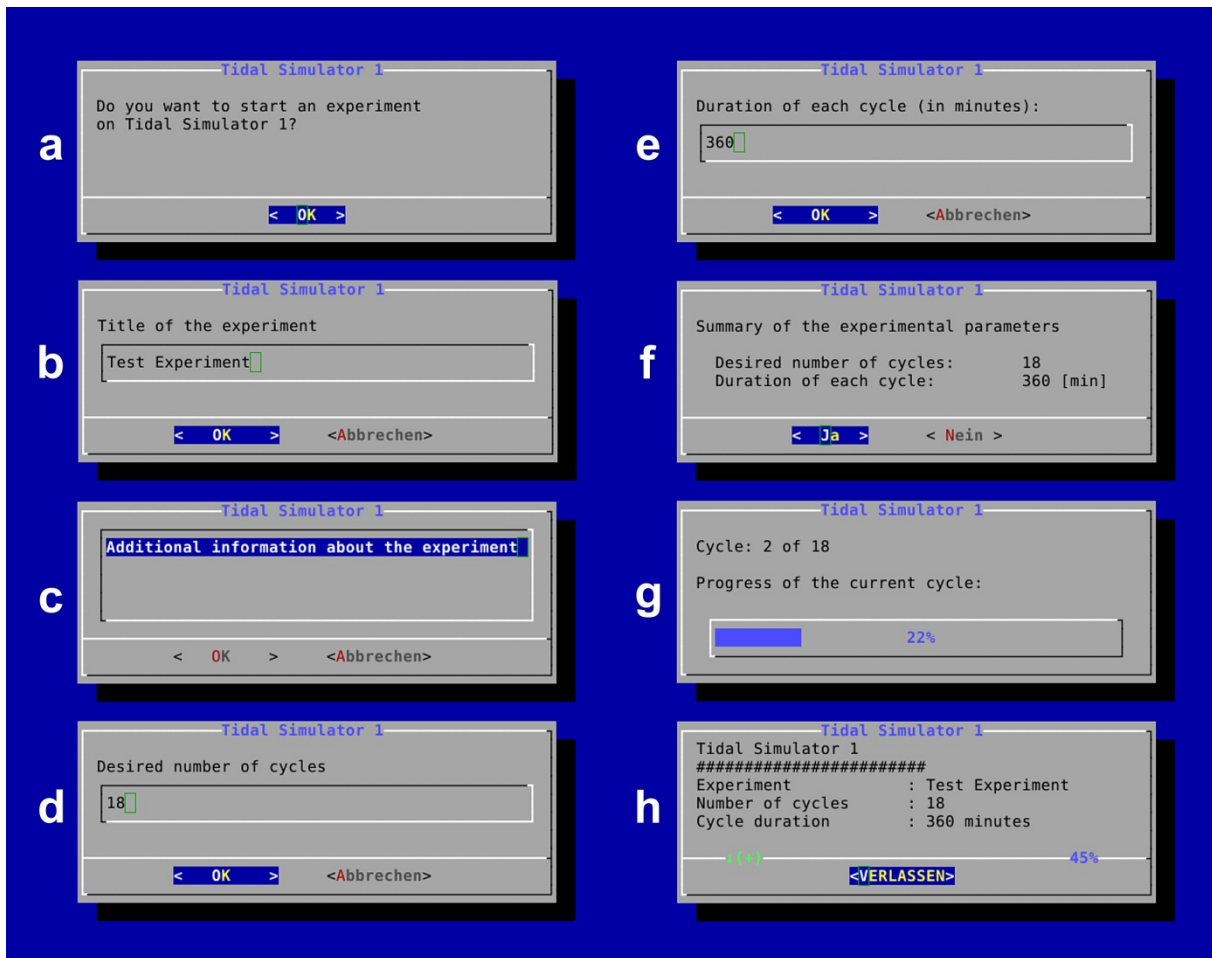
**SOM-Fig. 2.** In order to use the hardware efficiently, a convenient graphical user interface was implemented in the bash script which controls the wet–dry apparatuses. After the bash script is started, eight easy-to-use input and output boxes (a–h), which are based on the dialog command, open sequentially in the command line window. The first dialog box displays the identification number of the apparatus that will be controlled (a). After confirmation, the desired experiment name is entered (b). The next dialog box is an editor for optional data input (c). In the following two dialog boxes, the number of cycles (d) and the cycle duration (e) can be defined. Before starting the experiment, all set parameters can be checked (f). After confirmation, the experiment starts and its current status is shown (g). After completion of the experiment, a dialog box opens and displays the created log file, which is stored on the hard drive of the Raspberry Pi.
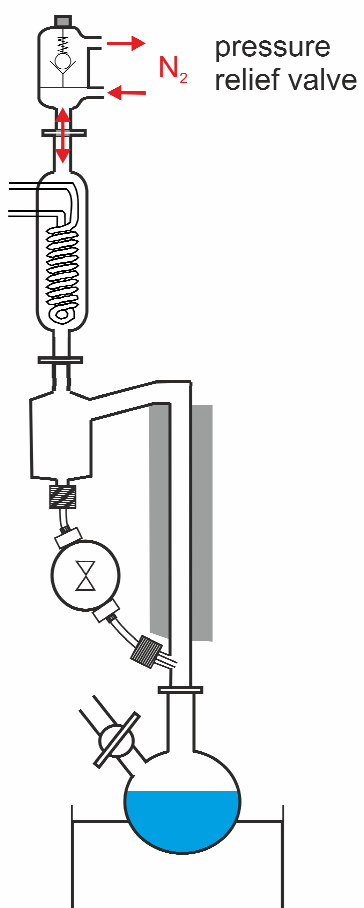
**SOM-Fig. 3.** Modified WDA used in the static atmosphere experiments with linoleic acid. In contrast to the standard setup (compare Fig. 2), a pressure relief valve was used. Furthermore, the nitrogen inlet was at the top of the apparatus and not at the Schlenk flask. This prevented spreading of the linoleic acid in the WDA when the valve opened.
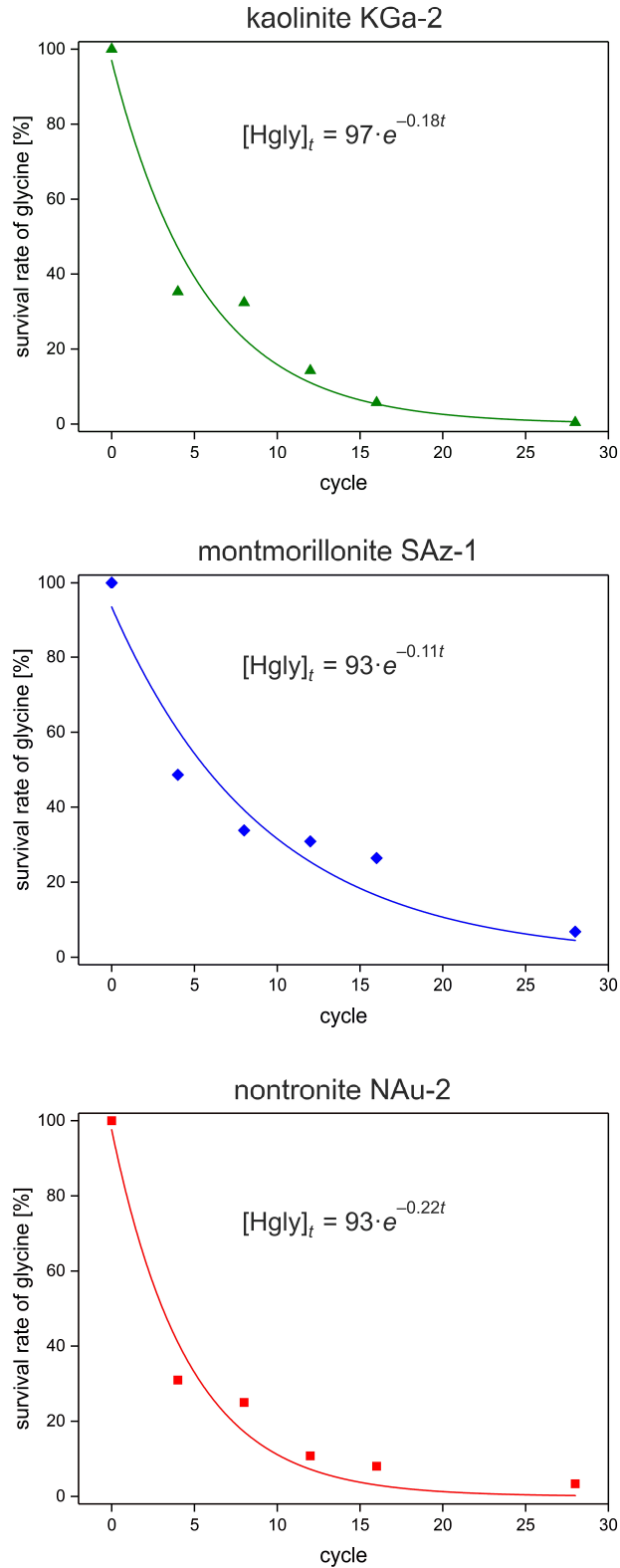
**SOM-Fig. 4.** Decrease of the glycine content as a function of the number of wet–dry cycles for three different clay minerals. A non-linear fit was performed on each data set, demonstrating that the glycine loss followed approximately first-order kinetics ($-\frac{d[\text{Hgly}]}{dt} = k \cdot [\text{Hgly}]$, from which follows $[\text{Hgly}]_t = [\text{Hgly}]_0 \cdot e^{-kt}$).

SOM-Table 1. *Identification of glycine and glycine peptides by high-resolution/high-accuracy mass spectrometry in samples exposed to 28 wet–dry cycles without clay mineral*

| Molecule | Elemental composition | $[M + H]^+$ experimental | $[M + H]^+$ calculated | $\Delta$ (ppm) |
|---|---|---|---|---|
| Hgly | $C_2H_5N_1O_2$ | 76.03922 | 76.03930 | −1.1 |
| DKP | $C_4H_6N_2O_2$ | 115.05022 | 115.05020 | +0.2 |
| Gly$_2$ | $C_4H_8N_2O_3$ | 133.06081 | 133.06077 | +0.3 |
| Gly$_3$ | $C_6H_{11}N_3O_4$ | 190.08244 | 190.08223 | +1.1 |
| Gly$_4$ | $C_8H_{14}N_4O_5$ | 247.10382 | 247.10369 | +0.5 |
| Gly$_5$ | $C_{10}H_{17}N_5O_6$ | 304.12511 | 304.12515 | −0.1 |
| Gly$_6$ | $C_{12}H_{20}N_6O_7$ | 361.14653 | 361.14662 | −0.2 |
| Gly$_7$ | $C_{14}H_{23}N_7O_8$ | 418.16787 | 418.16808 | −0.5 |
| Gly$_8$ | $C_{16}H_{26}N_8O_9$ | 475.18934 | 475.18954 | −0.4 |
| Gly$_9$ | $C_{18}H_{29}N_9O_{10}$ | 532.21068 | 532.21101 | −0.6 |
| Gly$_{10}$ | $C_{20}H_{32}N_{10}O_{11}$ | 589.23247 | 589.23247 | ±0.0 |

$M$ is the molecular mass, and $\Delta$ is the difference between the experimental and the calculated $[M + H]^+$ values.

SOM-Table 2. *Identification of glycine and glycine peptides by high-resolution/high-accuracy mass spectrometry in samples exposed to 28 wet–dry cycles with montmorillonite*

| Molecule | Elemental composition | $[M + H]^+$ experimental | $[M + H]^+$ calculated | $\Delta$ (ppm) |
|---|---|---|---|---|
| Hgly | $C_2H_5N_1O_2$ | 76.03918 | 76.03930 | −1.6 |
| DKP | $C_4H_6N_2O_2$ | 115.05020 | 115.05020 | ±0.0 |
| Gly$_2$ | $C_4H_8N_2O_3$ | 133.06080 | 133.06077 | +0.2 |
| Gly$_3$ | $C_6H_{11}N_3O_4$ | 190.08235 | 190.08223 | +0.6 |
| Gly$_4$ | $C_8H_{14}N_4O_5$ | 247.10386 | 247.10369 | +0.7 |
| Gly$_5$ | $C_{10}H_{17}N_5O_6$ | 304.12521 | 304.12515 | +0.2 |
| Gly$_6$ | $C_{12}H_{20}N_6O_7$ | 361.14659 | 361.14662 | −0.1 |
| Gly$_7$ | $C_{14}H_{23}N_7O_8$ | 418.16792 | 418.16808 | −0.4 |
| Gly$_8$ | $C_{16}H_{26}N_8O_9$ | 475.18932 | 475.18954 | −0.5 |
| Gly$_9$ | $C_{18}H_{29}N_9O_{10}$ | 532.21108 | 532.21101 | +0.1 |
| Gly$_{10}$ | $C_{20}H_{32}N_{10}O_{11}$ | 589.23215 | 589.23247 | −0.5 |

$M$ is the molecular mass, and $\Delta$ is the difference between the experimental and the calculated $[M + H]^+$ values.