Supplementary Information

## 1 Experimental part

Experiments were designed to prove the existence of stable intermediates inspired by the various mechanisms proposed by and Saladino<sup>1</sup>. There are also plenty of other mechanistic variants published by Springsteen, Krishnamurthy and others<sup>2,3,4</sup>. Those intermediates are predicted to be formed during from formamide or a formamide-clay mixture during thermal treatment, whereas the main purpose of this experimental approach was to detect them *via* an absolute detection method: Fourier transform infrared (FTIR) spectroscopy used in our laboratory for standard absorption.

#### 1.1 Experiment

The thermolysis was performed with pure formamide, formamide in the presence of three types of clays, and in the presence of clays and meteoritic iron. For each experiment, about 0.1 g of clay and, where applicable, about 0.01 g of meteoritic iron was inserted into a glass cell. The overview and characterization of the used materials. To this, 2.0 mL of formamide (>99.0%, Sigma Aldrich) was added. The pressure in the cell was reduced to 0.1 Torr and the cell filled with N<sub>2</sub> to approximately 500 Torr. The samples were then heated with a Kanthal wire up to 160 °C and kept at that temperature for 60 min.

### 1.2 Detection by FTIR

After thermolysis, the gas phase was expanded into an evacuated  $(1 \times 10^{-2} \text{ Torr})$  multipass cell to a pressure of about 0.5 Torr. The cell was inserted forms an optical path of 12 m and was connected to a high-resolution Fourier transform infrared spectrometer (Bruker IFS 125HR, Bruker Optics, Germany). The spectrometer was equipped with a KBr beamsplitter and nitrogen cooled LN-MCT (Hg-Cd-Te) detector. The spectra were measured in the range 680 – 4800 cm<sup>-1</sup> at a resolution of 0.02 cm<sup>-1</sup> with 300 scans co-added for each sample. Standard calibration measurements of pure gases (HCN, Messer Technogas, 186 ppm/He; CO, Linde Gas 5.3; CO<sub>2</sub>, Linde Gas 5.3 and NH<sub>3</sub>, Linde Gas 3.8) was performed for <u>quantification</u> purposes.

#### 1.3 Computational techniques

The biased condensed-phase dehydration reaction of formamide was reproduced by using the Born-Oppenheimer molecular dynamics approach and state-of-the-art metadynamics (MetD).<sup>5</sup> The results were obtained by exploiting the software suite for electronic structure and *ab initio* molecular dynamics (AIMD) simulations CP2K<sup>6</sup> in conjunction with PLUMED-2.3.3,<sup>7</sup> a patch which allows for the execution of MetD simulations. A cubic super-cell of side equal to 14.9 Å has been built in order to reproduce the experimental density of 1.13 g/cm<sup>3</sup>, since 45 formamide molecules (*i.e.*, 270 atoms) have been simulated. Periodic boundary conditions were applied. The temperature was kept fixed by means of the CSVR<sup>7</sup> thermostat at values around an average of 160 °C. As usual, the dynamics of ions was simulated classically using the Verlet algorithm. Wavefunctions of each atomic species were expanded in TZVP basis sets whereas the core electron interactions were treated by means of the Goedecker-Teter-Hutter pseudopotentials<sup>8</sup> using the GPW method.<sup>9,10</sup> A plane-wave cut-off of 400 Ry was imposed whereas a timestep for the nuclei dynamics of 0.5 fs has been adopted. The employed exchange-correlation DFT functional was the dispersion-corrected GGA functional PBE-D3.<sup>11 12,13</sup>

The MetD formalism here adopted employs the matrices of coordination numbers with the aim to define a given molecular state and exploits two Collective Variables (CV), S and Z, on which the free

energy relies upon. The former variable represents the progress along the chemical transformation whereas the latter the distance from an ideal pathway. During the calculations, the following parameters have been adopted for the coordination function (see Eq. 3 of ref. <sup>5</sup>: N = 6, M = 12, R<sup>0</sup> <sub>SS'</sub> = 1.8 Å for S,S'=O,C,N, 1.5 Å for S=O,C,N, S'=H, and 1.4 Å for S=S'=H. The parameter  $\lambda$  has been set such that  $\lambda D(R_k, R_{k,k'}) \approx 2.3$ . The MetD potential was composed of Gaussians with widths  $\sigma_s \sigma_s = 0.03$  and  $\sigma_z \sigma_z = 0.10$  and height 3 kcal/mol deposed every 50 fs.

Geometry optimization and frequency analysis of covalent and non-covalent formamide dimer were performed with the ORCA (V4.1.0) software<sup>14,15</sup> using the hybrid DFT method PW6B95-D3(BJ)<sup>16,12,13</sup> n gas phase. To speed up the calculations, resolution-of-identity (RI)<sup>17</sup>and chain-of-spheres (COSX)<sup>18</sup> approximations were used for Coulomb and exchange integrals, respectively. Quadrature grid was increased to grid4 whereas grid5 was used for final energy evaluation. The COSX grid was increased to GridX5. Calculations employed the def2-QZVP basis set<sup>19</sup> and auxiliary basis set def2/J<sup>20</sup>. Before performing frequency analysis, geometries were re-optimized using tighter convergence criteria (*TightOpt* keyword). Then, frequency analysis in the harmonic approximation was performed at 433 K (~160 °C). Vibrational frequencies were visually checked to confirm that the geometries correspond to local minima on potential energy surfaces. Gibbs free energies were calculated by adding the zeropoint energy (ZPE), thermal and entropic corrections to the total electronic energy. Non-covalent formamide dimer was prepared such that its most stable form —the one forming the two N-H...O hydrogen bonds— could be found during geometry optimization.

### 2 Characterization of the samples

Natural sodium clay (Na-clay) was acquired from Kunimine Industries (Japan) whilst ferruginous smectite (Fe-clay) was obtained from the Source Clay Repository of the Clay Mineral Society (Grand County, Washington, USA). Montmorillonite clay (H-clay) was purchased by Sigma Aldrich (Montmorillonite K 10, CAS 1318-93-0). Table S1 provides the elemental composition of the samples and mineralogy studied by standard X-ray diffractometry. The scanning electron microscope (SEM) photography of the samples is shown in Figure S1.

Species	Sample					
	Na-clay	H-clay	Fe-clay			
Na	2.51	0.24				
Mg	1.83	1.93	0.81			
Al	9.47	9.89	4.93			
Si	22.03	23.08	20.52			
Са	0.28		1.39			
Fe	0.82	0.81	8.58			
0	62.89	63.83	63.69			
Beidelite	x					
Quartz	x	x				
Montmorillonite		х				
Nontronite			x			

**Table S1:** Elemental analysis and mineralogy of the clay samples used in this study. "x" indicates positive detection of particular mineral by XRD.



Figure S1: Pictures taken by SEM of the clays employed. Panel A: Na-clay; Panel B: H-clay; Panel C: Fe-clay.

# 3 Supplementary to Results and Discussion

Thermolysis of the formamide was performed as described in the experimental part. The gas-phase products of thermolysis were transferred to the multipass cell and examined by high resolution infrared spectroscopy. The main products of liquid-phase formamide decomposition are ammonia and carbon monoxide, while HCN is detected as a trace. Concentration of these products can be significantly influenced by the peculiar interactions it establishes with the specific catalyst, as will be laid out in the following sections. Additionally, spectral signatures ascribed to the presence of non-covalent formamide dimer, aminoacetonitrile and formamidine have also been detected. The formation of these intermediates and their role in the following sections. Table S2 shows the detection of thermolysis products in all samples. All samples contain CO, CO<sub>2</sub>, HCN, NH<sub>3</sub>, methanol, formamide dimer and formamidine. Additionally, the thermolysis of pure formamide and formamide in presence of H-clay revealed the presence of aminoacetonitrile. The addition of meteoritic iron does not have a significant effect on the qualitative composition of the gas phase, although it alters the routes for further synthesis.

	aminoacetonitrile	NH₃	methanol	formamide dimer	formamidine	со	CO₂	HCN
Formamide	yes	yes	yes	yes	yes	yes	yes	yes
Formamide + H-clay	yes	yes	yes	yes	yes	yes	yes	yes
Formamide + H-clay + NiFe	no	yes	yes	yes	yes	yes	yes	yes
Formamide + Na-clay	no	yes	yes	yes	yes	yes	yes	yes
Formamide + Fe-clay	no	yes	yes	yes	yes	yes	yes	yes
Formamide + Fe-clay + NiFe	no	yes	yes	yes	yes	yes	yes	yes

Table S2: The presence of thermolysis products in the performed experiments.

All detections were confirmed by comparison with independently measured standards and a complete list of identified bands in each sample is given in Supplementary Information. Figure 2 in the main text shows the FTIR spectrum of the gas phase obtained after pure formamide thermolysis. Figures S2A, S2B and S2B show the same spectrum in detail.

We have already published a separate work which deals with the analysis of condensed phase thermolysis products and which details the formation of prebiotically-relevant molecules.<sup>21</sup> In that work, we describe in detail the formation of nucleic acid bases and glycine, and the effect of Fe-rich clays on the formation of these compounds in prebiotic conditions.



**Figure S2A**: The FTIR spectrum obtained from the gas phase after formamide thermolysis at 160  $^{\circ}$ C for 60 min, 600-1650 cm<sup>-1</sup> region.



**Figure S2B**: The FTIR spectrum obtained from the gas phase after formamide thermolysis at 160 °C for 60 min, 1650-2700 cm<sup>-1</sup> region.



**Figure S3**: The FTIR spectrum obtained from the gas phase after formamide thermolysis at 160 °C for 60 min, 2700-4800 cm<sup>-1</sup> region.



**Figure S4**: Global survey of formamide thermolysis FTIR spectra in presence of H-clay (A) are compared to spectra of formamidine hydrochloride evaporated to multipass cell at 80°C (B), aminoacetonitrile (C), gaseous formamide evaporated from a sample frozen by liquid nitrogen with characteristic dimer bands (D) and gas phase formamide vapours (E).



**Figure S5:** Detailed survey of formamidine band in formamide thermolysis FTIR spectra in presence of Fe-clay (A), where its bands were exceptionally visible. Spectrum of formamidine hydrochloride evaporated to multipass cell at 80°C (B) is provided for comparison.



**Figure S6:** Detailed survey of aminoacetonitrile band in formamide thermolysis FTIR spectra in presence of H-clay (A), where its bands were exceptionally visible. Spectrum of aminoacetonitrile band (B) and gaseous formamide evaporated from a sample frozen by liquid nitrogen with characteristic dimer spectral feature between 940 and 930 cm<sup>-1</sup> (C) is provided for comparison.



**Figure S7:** Detailed survey of aminoacetonitrile band in formamide thermolysis FTIR spectra in presence of H-clay (A), where its bands were exceptionally visible. Spectrum of aminoacetonitrile band (B) is provided for comparison.



**Figure S8:** Detailed survey of methanol band in formamide thermolysis FTIR spectra in presence of Hclay (A), where its bands were exceptionally visible. Spectrum of methanol vapours (B) is provided for comparison in this specific region.

#### 4. Detailed Reaction Mechanisms



**Figure S9**: Radical synthesis of nucleic acid bases. The figure was compiled from data published in Ferus et al.<sup>22</sup> Molecules shown in violet are also shown in Figure 1 in the main paper. The main intermediates in this mechanism are Aminohydroxyacetonitrile (AHAN), aminohydroxymalonitrile (AHMN), DAMN and AICN. This mechanism results in both purine nucleobases (adenine and guanine) and pyrimidine nucleobases (cytosine and uracil).



**Figure S10**: Formation of pyrimidine nucleic acid bases from formamide. This figure was compiled with mechanisms proposed by Saladino et al.<sup>1</sup> Molecules shown in violet are also shown in Figure 1 in the main paper. Here formamide reacts through a covalent formamide dimer. The initial step has been proposed by Yamada<sup>23,24</sup> and the mechanism is well summarized also by Yadav, Kumar and Krishnamurthy.<sup>25</sup> The synthesis is sequential and the first pyrimidine nucleobase to be formed is cytosine. Subsequent hydrolysis leads to uracil and then through hydroxymethyluracil to thymine.



**Figure S11**: Synthesis of adenine, guanine and cytosine from formamide. This figure was compiled with data from Ferus et al.<sup>26</sup> and Slavova&Enchev<sup>27</sup>. Molecules shown in violet are also shown in Figure 1 in the main paper. Formamide produces 2-iminoacetonitrile either through reaction with HCN or through a side-step with the synthesis of formamidine. Consequently, the synthesized 2-iminomalonitrile (AMN) is an intermediate, where the synthesis branches off towards DAMN, DAFN, AICA and guanine, or through AICN to adenine and hypoxanthine.



**Figure S12**: Synthesis of glycine from formamide. This figure was compiled with data from Ferus et al.<sup>26</sup> Molecules shown in violet are also shown in Figure 1 in the main paper. The synthesis of glycine proceeds from formamide through 2-iminoacetonitrile. This then reacts with H<sup>+</sup> to 2-aminoacetonitrile, which hydrolyses to glycine.



**Figure S13**: Synthesis of glycine from formamide. This figure was compiled with data from Pastorek et al.<sup>21</sup> Molecules shown in violet are also shown in Figure 1 in the main paper. The formation of urea was described in Pastorek et al.<sup>21</sup> and proceeds through HNCO.



**Figure S14**: Synthesis of purine from formamide via aminoacetonitrile. This figure was compiled with data from Hudson et al.<sup>2</sup> Molecules shown in violet are also shown in Figure 1 in the main paper. The synthesis of purine proceeds through 2-aminoacetonitrile, similar to the synthesis of glycine. If the 2-aminoacetonitrile reacts with formamide to 5-aminoimidazole, the synthesis then proceeds through formamide to purine.



**Figure S15**: Synthesis of formamide from formaldehyde and CO by reaction with ammonia. This figure was compiled with data from Ferus et al.<sup>28</sup> Molecules shown in violet are also shown in Figure 1 in the main paper. Formamide itself can be synthesized from ammonia and carbon monoxide or from formaldehyde with ammonia.

#### 5. Theoretical Calculations

In order to obtain an estimate of the free-energy supplied by the presence of clays, an *ab initio* molecular dynamics (AIMD) simulation coupled with a path-Collective-Variables metadynamics (MetD) has been employed. In particular, the condensed-phase reaction of the dehydration of formamide (*eq. 1*) has been computationally reproduced in absence of any catalyst with the aim of reconstructing the underlying free-energy surface. It turns out that a free-energy barrier of  $45 \pm 3$  kcal/mol separates the basins ascribed to reactants and products, as shown in Figure S8. Such a value is very close to that obtained in ref. <sup>5</sup> where the same chemical transformation has been simulated in liquid water. Mechanistically, in absence of catalysts a formamide molecule cooperate with the solvent in order to release a water molecule, as shown by the atomistic pictures of the reaction in Figure S8 (insets). Again, similarly to what was observed for the same reaction in aqueous environments,<sup>5</sup> an intermediate state is found during the interconversion of formamide into H<sub>2</sub>O and HCN. In particular, HNCHOH is formed, as shown in the inset 2 of Figure S8. Such a quantum-based analysis indicates that the free-energy contribution supplied by the Fe-containing clays is of at least 45 kcal/mol, quantitatively showing the crucial role they play as catalysts.

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