

# Acid, Salt, Heat, Pressure: Testing the Limits of Biomolecule Preservation

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## Abstract

Experimental studies of the interactions between biomolecules and minerals under conditions simulating harsh planetary environments provide key insights into possible prebiotic processes and the search for life. Despite protection from cosmic rays, UV, and oxidative degradation, buried biosignatures may undergo diagenetic processes that decrease the concentration of organic matter. Additionally, other degradation mechanisms occur as a result of elevated temperatures, pressures, mineral-organic interactions, and fluid/brine processes. In this study, we aim to provide a fuller understanding of preservation potential by considering several variables, including pressure, temperature, the mineral matrix environment, and fluid chemistry (salinity, pH, composition). This research expands previous anhydrous work to investigate the influence of lower pressure regimes, especially in a combined fluid/brine environment with various mineral matrices. To test the preservation potential of various biomolecules, we subjected samples to temperature, pressure, fluid, and mineral matrix conditions representative of different environmental stressors. The starting materials included: 1) isolated organic compounds added to various mineral standards, 2) An endolithic and microbe-rich natural calcite deposited from a CO<sub>2</sub>-rich hot spring, 3) cyanobacteria necromass. Experiments were conducted in three different devices 1) a piston-cylinder press reaching up to 15 kbar and 550 °C, 2) high-volume batch reaction vessels generating up to 15 MPa pressure and 80 °C, and 3) ambient pressure, high temperature furnaces. Samples were analyzed by GC-MS and LC-MS, while ICP-MS, XRD, and Raman were used for additional characterization. The influence of pressure can be clearly identified. Similarly, fluid transport, complex thermal degradation, and oxidation mechanisms are identified.



# Acid, Salt, Heat, Pressure: Testing the Limits of Biomolecule Preservation

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## Introduction

"Who cares about germs or viruses or bare organic remnants halfway to life but never making it?  
Astronomers and biologists do!"

Isaac Asimov  
*Mars, We Love You*, Pyramid (1973)

Experimental studies of the interactions between biomolecules and minerals under conditions simulating harsh planetary environments provide key insights into possible prebiotic processes and the search for life (Fig. 1).

Despite protection from cosmic rays, UV, and oxidative degradation, buried biosignatures may undergo diagenetic processes that decrease the concentration of organic matter. Additionally, other degradation mechanisms occur as a result of elevated temperatures, pressures, mineral-organic interactions, and fluid/brine processes (Alleon et al. 2016, Montgomery et al. 2016, Bravenec et al. 2020, 2021).

Assessing the evolution of organic molecules in subsurface environments has significant implications for evaluating plausible scenarios for the origins of life and improved targeting of biosignatures and organic compounds during missions.

In this study, we aim to provide a fuller understanding of preservation potential by considering several variables, including pressure, temperature, the mineral matrix environment, and fluid chemistry (salinity, pH, composition).

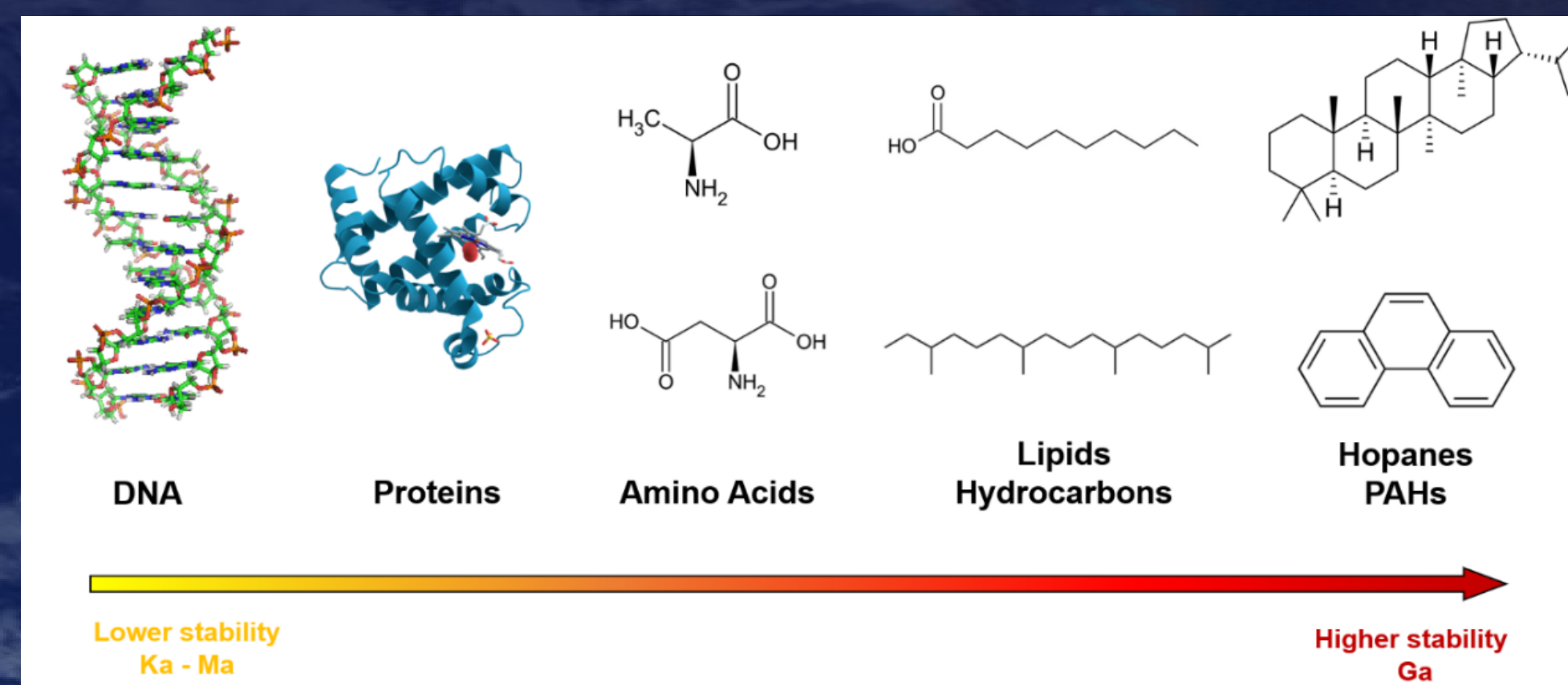


Fig. 1. Illustration of preservation potential of biomolecules in Ka (thousand) to Ga (billion) years.

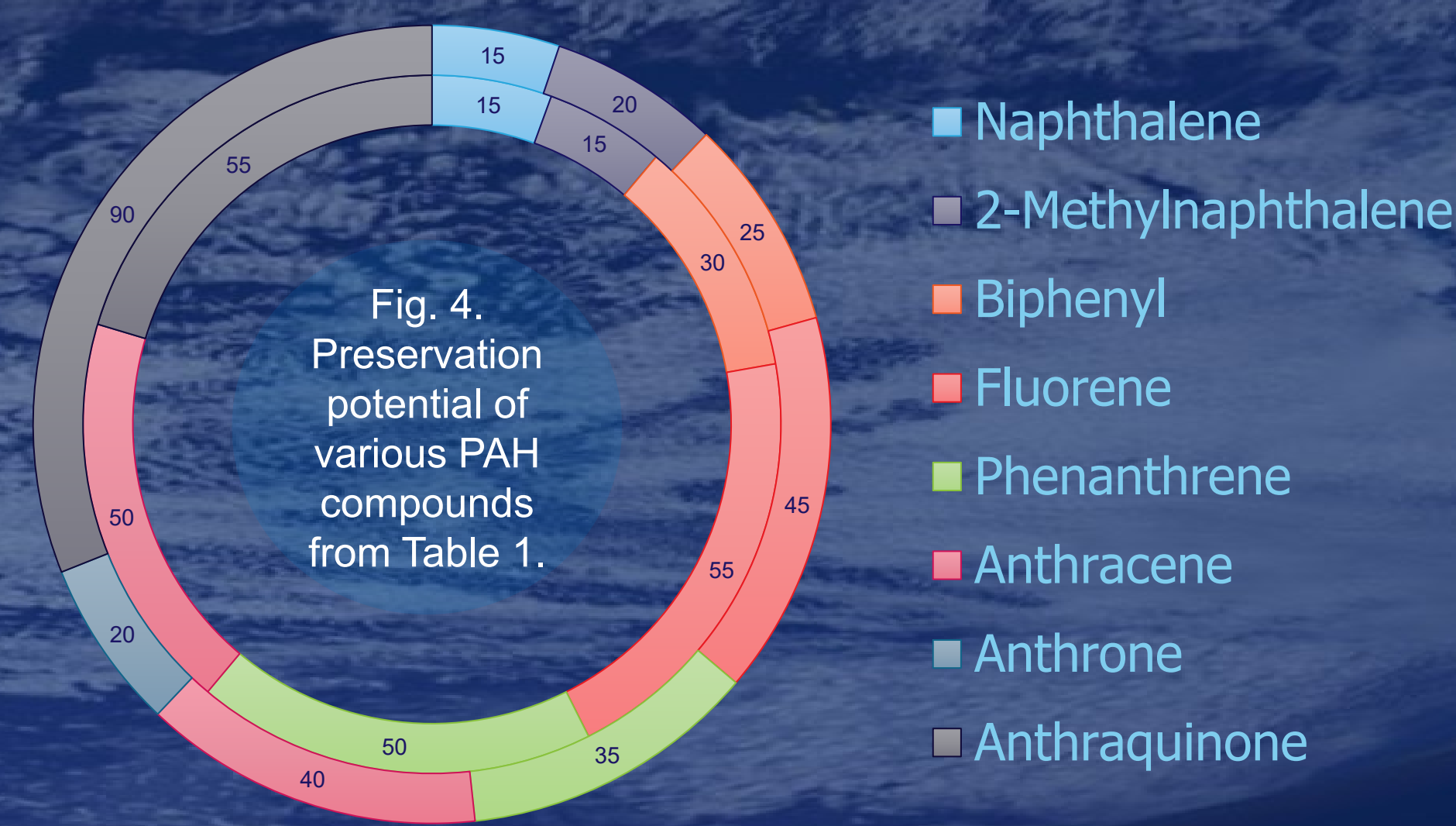
## PAHs

PAHs (Polycyclic Aromatic Hydrocarbons) are molecules with cores consisting of two or more fused benzene rings.

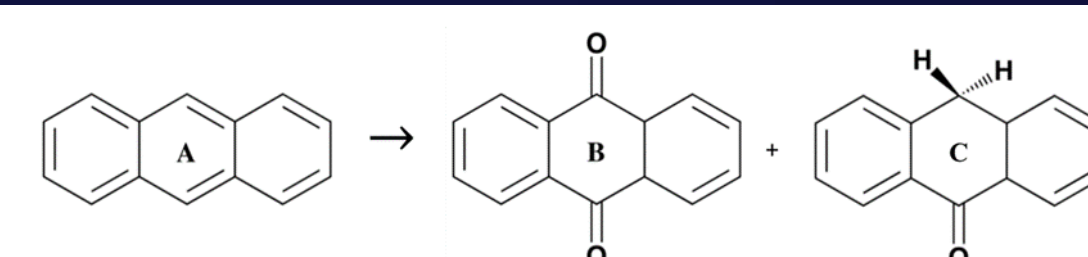
These aromatic molecules are estimated to be the most abundant compounds in the universe, comprising ~10 to 20% of all carbon (Plows et al. 2003). Although PAHs can occur in abiotic environments, identifying abundances of specific PAHs can be indicative of biological origin.

The enhanced stability of PAHs allows them to withstand the high pressures and temperatures inherent to burial environments, including metamorphic, diagenetic, and impact processes.

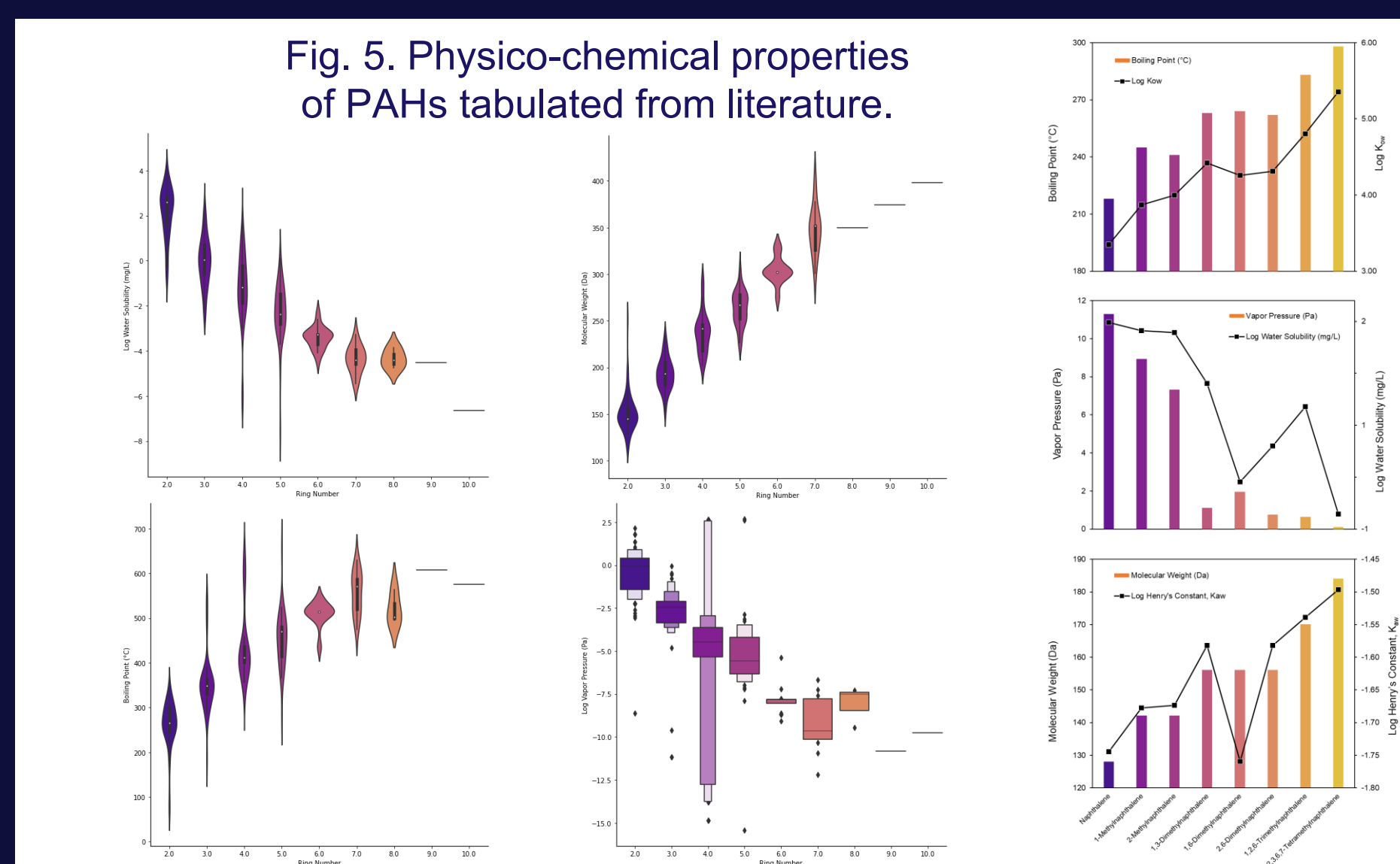
Table 1. Percent detection of PAH compound following thermal treatment at 150 °C for 200 hours in respective matrix environment.				
PAH	Experiment Set 1 (20 experiments) (%)	Experiment Set 2 (20 experiments) (%)	Average	±
Naphthalene	15	15	15	0
2-Methylnaphthalene	20	15	18	3
Biphenyl	25	30	28	3
Fluorene	45	55	50	5
Phenanthrene	35	50	43	8
Anthracene	40	50	45	5
Anthrone	20	0	10	10
Anthraquinone	90	55	73	18



Anthraquinone was the PAH most frequently detected, even when not added to the PAH mixture in experiment set 2 (Table 1). This could be due to both a higher resilience to degradation or mechanisms oxidizing other PAHs into anthraquinone:



Shale displayed the overall highest preservation potential in the presence of a destructive fluid environment (Fig. 2 and 3). Consideration of physico-chemical properties (molecular weight, boiling points, vapor pressures, water solubilities, Henry's Law constants,  $K_{OW}$ , pKa, and degree of alkylation) can provide a first order estimate of preservation potential (Fig. 5). For example, out of the PAHs tested in this study naphthalene has the lowest molecular weight and was most readily evaporated from the fluid solution (Table 1) leading to decreased preservation (Fig. 4).



## Pressure Effect

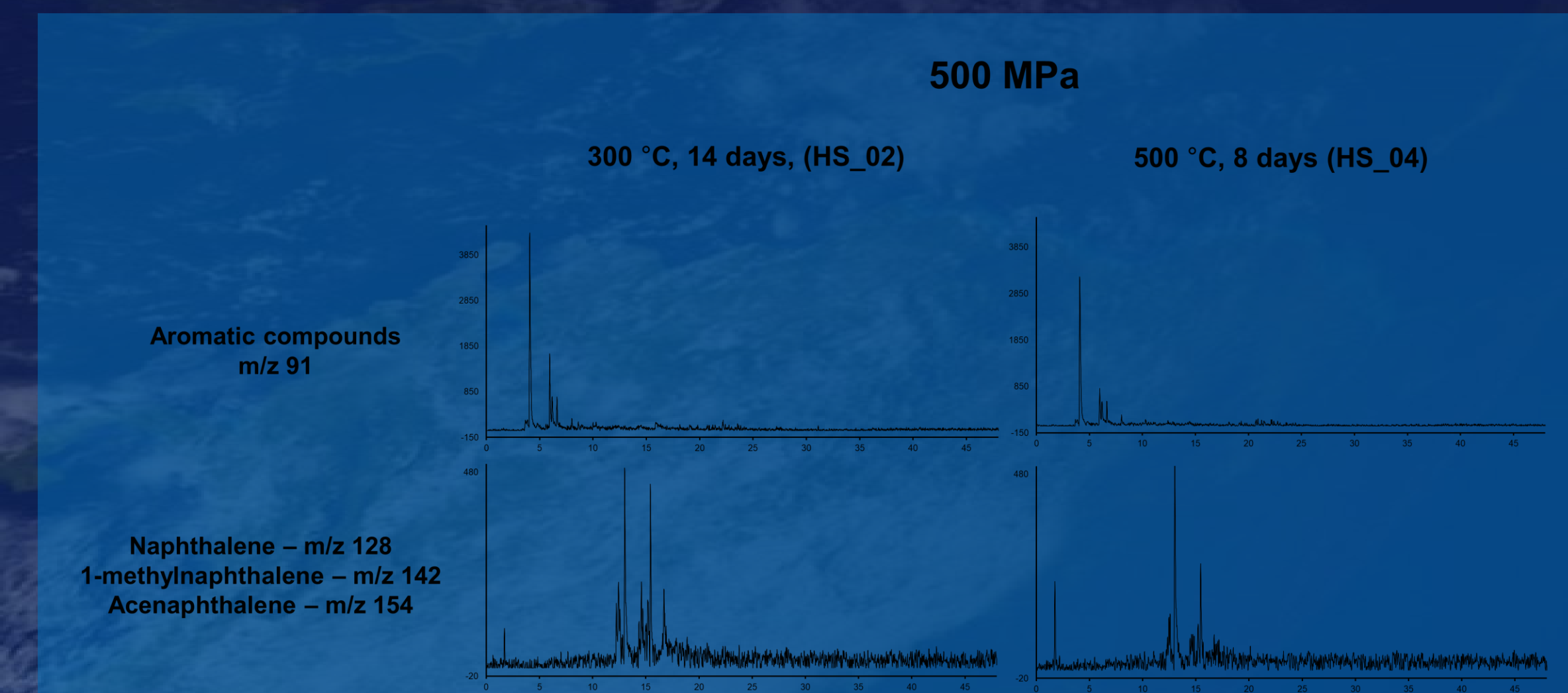


Fig. 6. Pressure and temperature treated endolith sample. Extracted ion chromatograms (XIC) from py-GC-MS data showing aromatic compounds (m/z 91) and mass fragments associated with selected PAHs (m/z 128, m/z 142, and m/z 154). These compounds show remarkable preservation potential even under subsurface conditions.

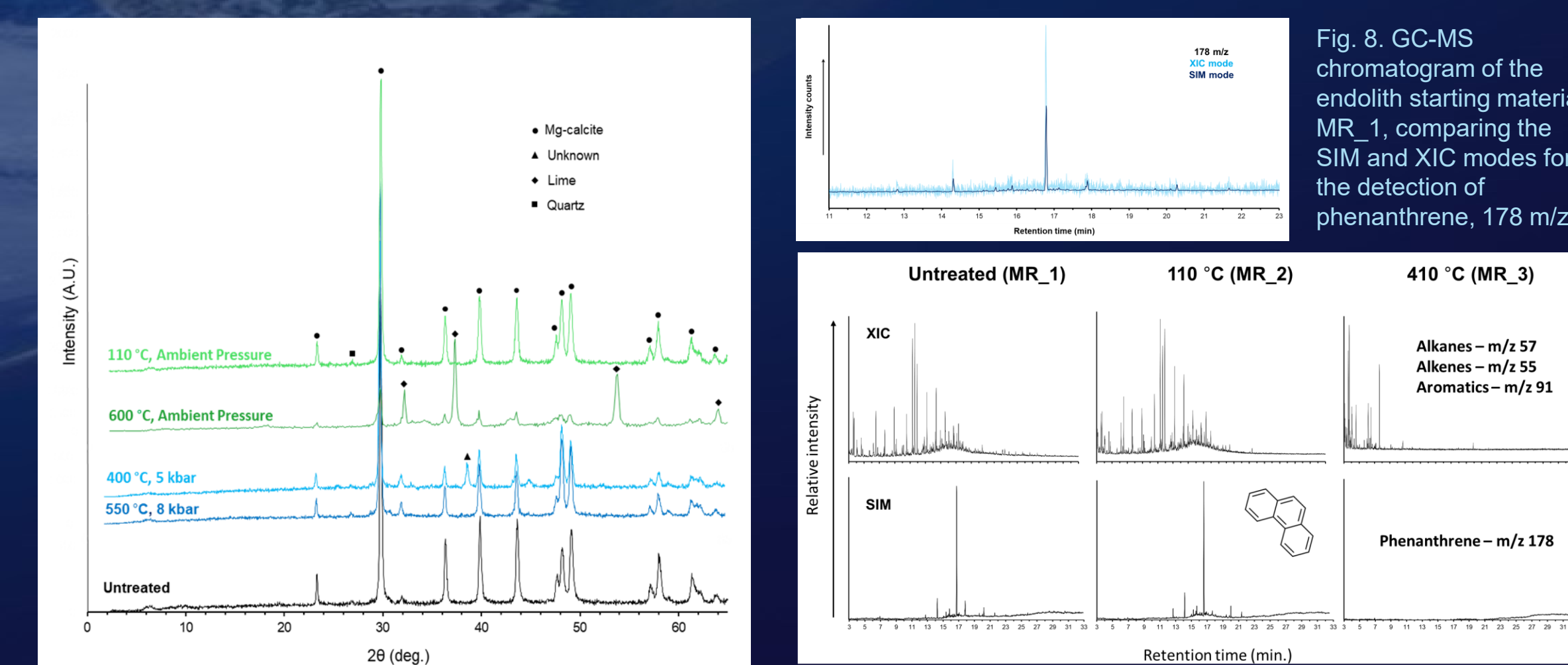


Fig. 7. XRD data for endolith samples run at various pressure and temperature conditions. The mineral matrix, consisting predominantly of calcite, is expected to be stable at the temperatures of this study and under the stabilizing effect of high-pressure conditions. The sample only decomposed at 600 °C after two hours in ambient pressure.

Fig. 8. GC-MS chromatogram of the endolith starting material, MR\_1, comparing the SIM and XIC modes for the detection of phenanthrene, 178 m/z.

Fig. 9. GC-MS XIC data from samples run at 110 °C and 410 °C compared to the untreated sample. The SIM for the PAH phenanthrene is also shown, which may be a compound indicative of biotic activity. Experiments were conducted in a piston cylinder press.

## Summary

- The early eluting peaks (Fig. 9) represent thermal degradation products rather than potential biosignatures.
- The influence of pressure can be clearly identified → it has a stabilizing and preservative influence on organics and biomolecule preservation.
- Aromatic stabilized compounds show remarkable preservation potential even at subsurface conditions.

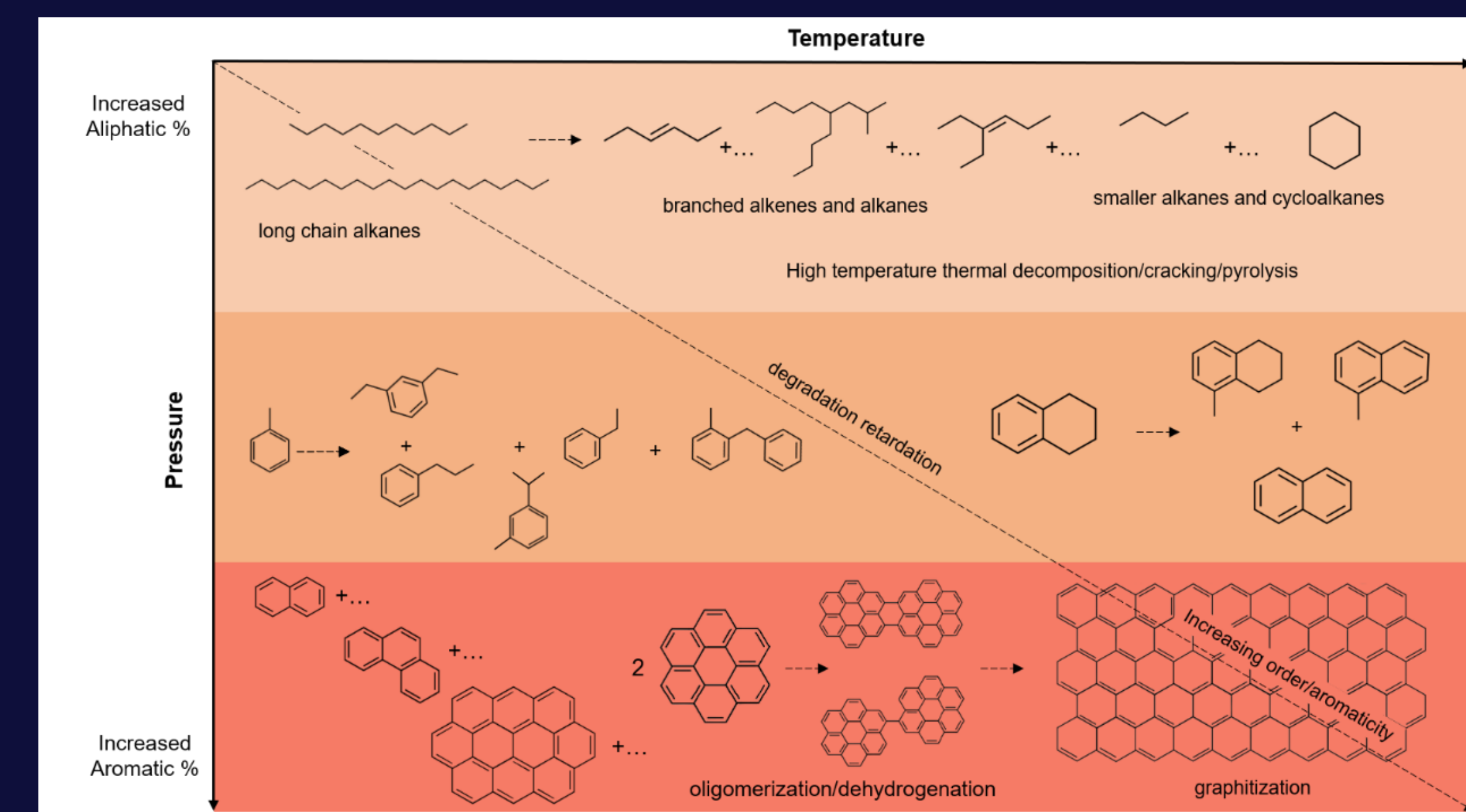
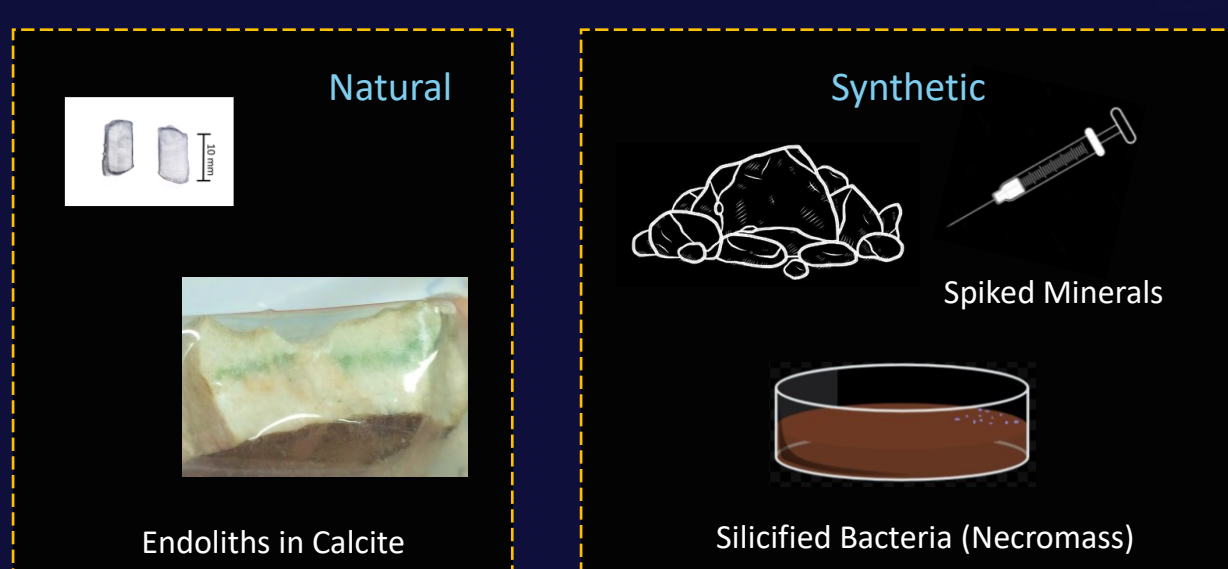


Fig. 10. Schematic model showing the process of progressive molecular degradation and increased aromaticity under high-pressure conditions. Thermal degradation (pyrolysis, thermolysis, cracking) reactions are shown for high-temperature/low-pressure.

## Graphical Abstract

### 1. Starting Material



### 5. Data Analysis

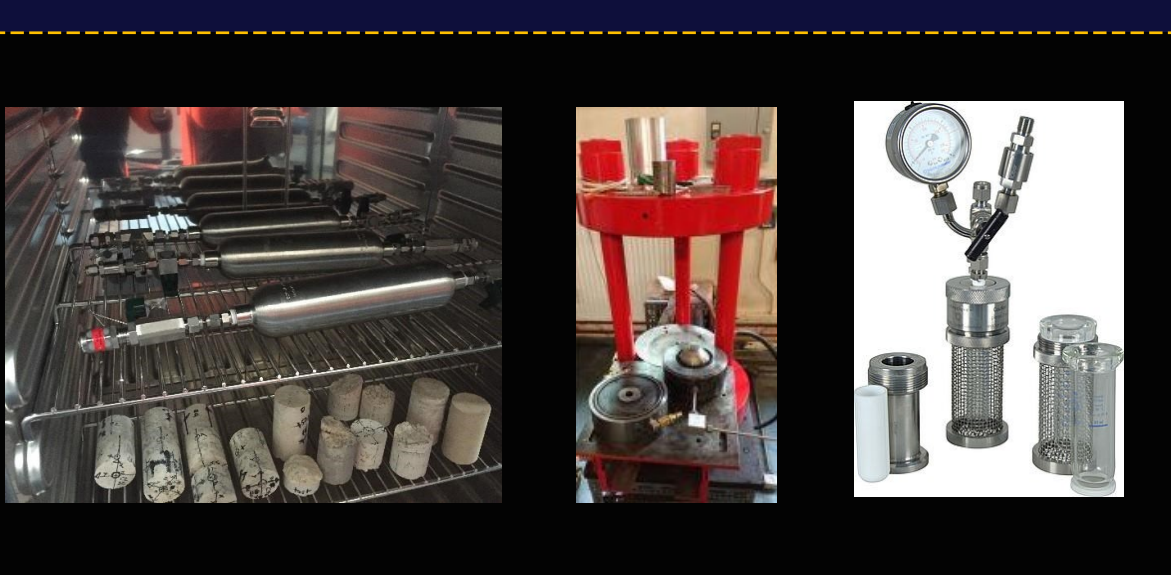
Principal component analysis  
Biosignature degradation  
Preservation potential  
Mineral matrix effects  
Etc.

### 4. Instrumental Analysis

Py-GC-MS, Liq-GC-MS, LC-MS,  
ICP-MS, Raman, EPMA, XRD



### 2. Subsurface Experiments



### 3. Preparation

Extraction,  
Purification,  
Separation,  
Derivatizations

