nature portfolio

Peer Review File

Data-Driven De Novo Design of Super-Adhesive Hydrogels

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This file contains all reviewer reports in order by version, followed by all author rebuttals in order by version.

Version 0:

Reviewer comments:

Referee #1

(Remarks to the Author)
A. Summary of the key results
The proposed work is focused on the application of data-driven approaches to design and synthetize new materials with improved adhesive properties. The proposed approach combines data mining (ML), machine learning (ML) and experimental procedures to rationalize and predict adhesive hydrogels with superior properties for underwater environments minimizing the classical trial and error approach. The authors take inspiration from Nature, collecting and analyzing adhesive proteins from different sources and to have a large starting data-set. From these data, a descriptor strategy has been selected allowing the application of ML to predict hydrogel properties. As demonstrated by experimental validation, adhesive hydrogels with improved mechanical and adhesive properties were generated. The evaluation of the produced hydrogels for different application has been performed, including preliminary biomedical validation and environmental applications.

B. Originality and significance: if not novel, please include reference
The proposed work is highly innovative in the biomaterials, and in particular hydrogel field. The common trial-and-error method employed to produce hydrogel libraries for different applications still represent an uncovered gap to translate novel hydrogels.
However, the application of ML methods is limited by the lack of homogeneous data collection, the limited number of data available in literature or patents, the different polymers or hydrogel employed for the generation of hydrogels and finally but important the different characterization employed to evaluate the final properties of the hydrogels.
The presented concept is innovative and the idea to start with proteins already stored in data bank can overcome the problem of data availability. The application of ML in the generation of hydrogels starting from biomolecular data related to functional group is novel in the polymeric material field and could help the scientific community in the generation and application of AI based systems appliable also to polymers of different nature.

C. Data & methodology: validity of approach, quality of data, quality of presentation
• Validity of approach: the proposed data-mining and machine learning methods are well designed and structured. In general, the approaches employed to characterize mechanical and adhesive properties of the hydrogel are coherent with the aim of the work and with the reported aims.
The approaches employed for the experimental part lack of important information to fully evaluate the reported properties of the hydrogels and in general the efficacy of the proposed approach. In detail:
• the section on data mining of adhesive proteins reports on the selection of dataset including proteins with an average length between 300 to 500 amino acids. It is not clear if this range has been selected for specific “functional” reasons, or to allow the management of data.
• The impact of ML application and validation using random polymerization can be limited comparing other synthetic methods, considering that the interactions between chains with adhesive functional group favor the formation of hydrogels with major adhesive properties.
• Quality of data: the experimental part and the information reported, including the chemical characterization of the obtained hydrogels and the validation of the obtained hydrogels, are limited and lack of important information to evaluate the final properties of the hydrogels and consequently the accuracy of the proposed approach to obtain hydrogels with improved properties.
• Quality of presentation: the discussion on experimental results and their correlation with predicted data should be reorganized introducing structures of the hydrogels in the text, reporting qualitative data improving the discussion and

reorganizing the selection of figures and graphs cited and available in the extended data (in particular in the discussion of the experimental of the obtained hydrogels. Materials and methods should be implemented for each step to allow precise reproduction of the experimental and homogeneity / availability of experimental data.

D. Appropriate use of statistics and treatment of uncertainties
Use of statistics and treatment of uncertainties are appropriate for the DM and ML. statistics and treatment of uncertainties in the experimental part should be added and implemented.

E. Conclusions: robustness, validity, reliability
The conclusions are appropriate concerning the data collected and discussed for the dry part (DM and ML). The conclusions related to the experimental part, in particular on the evaluation of synthetic improvements and hydrogel validations are general and partially discussed.

F. Suggested improvements: experiments, data for possible revision

General comment: the manuscript is undoubtedly interesting and highly innovative. Furthermore, the proposed approach can open new perspectives that could be applied to other fields and to other polymers. However, several aspects should be revised or implemented. The manuscript should be revised to help the reader.
• The discussion on the experimental data should be implemented with structures and a clear assignation of the produced hydrogels in the text and not in the supplementary.
• The discussion of the experimental part should be implemented considering a) the chemical characterization of the produced hydrogels. NMR should be reported for the samples considering the entire spectra and an in-depth analysis using also the complete assignation of the spectra.
• The discussion on the obtained results should be implemented reporting the experimental results related to the chemical nature of obtained hydrogels (using for example NMR, and FTIR).
• In the paragraph related to the Validation the different methods and characterization should be implemented as following reported:
• adhesive testing should be reported considering and discussing quantitative data and qualitative data considering also the comparison between different samples at different time points.
• The hydrogels employed in the validation and characterization should be clearly represented, reported and identified; b) the discussion on biological validation is limited to few lines.
• To evaluate the impact of the proposed approach in the field of adhesive for medical application both the discussion and the experimental should be expanded (i.e biocompatibility, moisture vapor transmission rate (MVTR), cohesive / adhesive failure, permeability).

Materials and Methods:
• Hydrogel fabrication – the procedure should be implemented considering the different hydrogel formulations and well reported (quantities, mmol for each reagent, volumes…)
• Hydrogel underwater adhesive property characterization – volume of tested hydrogels should be reported. The experimental procedure should be reported considering also the number of experiments for each sample.
• Machine Learning. For non-linear models, materials and methods for each regression algorithm should be reported. The paragraph should contain just the methods and not the considerations (that should be included in the discussion)
• In the materials and methods experimental procedures on the characterization and validation should be introduced.

F) Reference: appropriate credit to previous work: YES

G) Clarity and context: lucidity of abstract/summary, appropriateness of abstract, introduction and conclusions.

The abstract well reports the aim of the work. However, the conclusions of the abstract should be revised considering the obtained results limiting the discussion on the correlation between predicted data and reported obtained data.
The introduction is well reported, with a clear introduction on the needs the state of art (still very limited and uncovered).
The conclusion should be revised considering the data reported and introducing the existing challenges in the field, in particular in the prediction of chemical features and in their correlation with the final properties of the hydrogels. Even if the conclusion in the field of adhesive is well understandable, the potential application in the biomedical field is not supported by data presented and discussed. This point should be modified in the light of the obtained experimental results.

(Remarks on code availability)
the code doesn't provide a README file with enough intructions for installing and running the application

Referee #2

(Remarks to the Author)
Paper Summary
This paper introduces a data-driven method to create high-performance adhesive hydrogels. By mining >20,000 adhesive protein sequences, they developed a descriptor strategy to statistically mimic protein sequence patterns using ideal random copolymerization. Machine learning, particularly Gaussian Processes and Random Forests, fine-tuned the designs,

achieving adhesive strengths over 1 MPa that outperform current products. These hydrogels adhere well to diverse range of surfaces in challenging environments.

Abstract
Good summary of paper but needs the inclusion of specific data points/key results. Authors should substantiate “Using machine learning, we optimized hydrogel formulations even with a small dataset, achieving unprecedented adhesive performance.” key results.

Introduction
The introduction is well-written and easy to follow, but it lacks critical context regarding previous research. It should provide a short discussion of similar DD/ML approaches used for designing hard materials, establishing the validity of this methodology. Readers shouldn’t be expected to simply accept that applying such techniques to hard materials is straightforward or standard practice. Authors should clarify how these methods are well-established for hard materials and why they are more challenging for soft materials like hydrogels.
Additionally, the introduction should address whether ML-based strategies have been previously applied to adhesives, for example some work in dry adhesives (https://doi.org/10.1016/j.mechmat.2023.104778 and https://doi.org/10.1016/j.eml.2022.101695). While these studies may not align precisely with the focus here, they highlight the broader context of DD approaches in adhesive design, which warrants discussion. Even broader is the discussion of previous DD/ML approaches for designing hydrogels. Including a broader discussion would clearly position the current work as an advancement in the field. A comparison to existing studies would highlight the novelty of this framework and better demonstrate its contribution. Improving the introduction in this way will make the work more compelling and accessible.
Figure 1 is excellent.

Data mining
I really like your approach. It’s robust, and your strategy as laid out is both logical and sound. The way you’ve mined the protein data and distilled it into functional classes strikes a great balance between simplifying the complexity and keeping the key chemical properties relevant to adhesion. Plus, the findings generated were then easily translated into synthetic hydrogels.
Exclusion of glycine, alanine, and proline – is there a danger that the exclusion of such groups might have other deleterious effects on other properties? i.e. how did you rule out unforeseen consequences to say flexibility or packing density or stability etc.?
If we think beyond adhesion, how generalisable is this approach to other types of soft materials/hydrogels? Could your approach be adapted to target additional properties, such as mechanical strength, elasticity, responsiveness to stimuli, etc?

Figure 2 is also excellent.

Synthesis
A lovely and systematic approach. The methodology is logical, with the robust use of Monte Carlo simulations adding strong validation to the process. The one-pot free radical polymerisation and straightforward testing method are both efficient and repeatable, making them ideal for high-throughput screening and practical application.
You chose six functional monomers for logical reasons, but how many in total could have fit this criterion? Were there potentially others?

Machine learning
This is the highlight of the paper. The use of ML, especially GP and RFR models with batched SMBO, is smart and effective. The workflow was both efficient and accurate. Your models were able to effectively identify new possibilities and impressively reveal key monomers as BA, PEA, and ATAC. Your approach will be much utilised.
Why do you think “Gaussian process (GP)30 194” and “random forest regression (RFR)31 195” were the most effective ML models? Why were the weakest ML models the weakest? Would this always be the case for ‘soft materials’?

Performance
Your performance testing was robust, and the results are impressive. You demonstrated the hydrogels’ versatility across different water types with multiple testing methods providing a thorough evaluation of adhesive performance. While the rubber duck demonstration is playful, it may indeed illustrate real-world conditions. However, the biocompatibility testing feels out of place here and could be omitted, allowing it to be expanded upon in a separate, more focused publication. Keeping the focus solely on adhesion performance would strengthen the manuscript, as the results are compelling enough to stand alone.
Section could benefit from some proper quantitative testing for the marine environment or burst pipe testing. The videos and demonstration are ideal to reach a wider audience but would have benefitted to have some hard data to support the rubber duck marine test and pipe burst test.

Summary
The last paragraph is weak Lines 324-330. Please revisit and think about the true potential of these superadhesive hydrogels, particularly for extreme environments.

Overall, this is an excellent paper that showcases an innovative data-driven approach to developing super-adhesive hydrogels. The authors are to be commended for their robust methodology, logical design, and comprehensive testing. The integration of ML is particularly impressive. Your high-performance hydrogels have real-world potential and your approach will benefit many researchers in the field. Minor revisions, including refining the introduction, potentially adding quantitative

data for marine and pipe bursting demo, and revisiting the summary.

Originality and significance: if not novel, please include reference
Yes. Original and significant. The authors have developed a methodology for the robust development of hydrogels that can be greatly utilised by many in the field.

Data & methodology: validity of approach, quality of data, quality of presentation
Excellent. High-quality presentation of the data in all figures. Valid approach. Logical and robust.

Appropriate use of statistics and treatment of uncertainties
yes.

Conclusions: robustness, validity, reliability
Room for improvement in the final summary of the paper.

Suggested improvements: experiments, data for possible revision
Stated above.

References: appropriate credit to previous work?
Could do with more thoughtful summation of previous work in the DD/ML in materials development—hard materials and other soft materials (hydrogels)

Clarity and context: lucidity of abstract/summary, appropriateness of abstract, introduction and conclusions
Abstract and conclusion need some work. It is a very well-written paper and easily understood.

Referee #3

(Remarks to the Author)
The manuscript by Takigawa, Li, Fan and Gong describes a concerted effort to develop high performance underwater adhesive by combining data mining, experiments and machine learning. The study started with analyzing protein sequence information upon reducing to 6 monomer types, followed with experimental inputs based on large library gel synthesis and characterization, and ended with machine learning to optimize systems with three monomer type. The resultant adhesives demonstrated quite impressive performance with potential technologically relevant applications. Present studies also accumulated quite large datasets and provided good experimental inputs, a key bottleneck for AI/ML in material development.
I have serious concerns regarding the connection made between what's made vs. protein sequence analysis. The gels were synthesized in the presence of crosslinker and at high monomer concentration (>2M total monomer) using UV initiator. The polymerization process is not living and is diffusion limited with high viscosity. It is reasonable to expect significant heterogeneity and to question the fidelity of sequence simulation based on Monte Carlo simulation to represent what the polymer strains in the final network may look like. Admittedly, synthesizing networks is difficult to characterize. However, this is the corner stone of the system design and selection, e.g. results shown in Figure 2. Control experiments need to be done to quantify to what level the reactions and sequence simulations share similarities and identify the differences. Given the gel formation, the crosslinking density and the polymer strain length are directly coupled, and adhesive performances depend on so many factors. The readers and community should be made aware of potential hidden caveat. Maybe it was just used as proxy or a convenient way to extract some information and perhaps there are some merits to predict short blocks simply by the system diversity. I do not see systematic characterization of gels or studies of adhesion mechanism. More in-depth understanding/characterization of the system are important part of work to bridge AI/ML with specific scientific topics.
Another question is regarding the logic behind 6 monomer type selected for protein sequence analysis vs. 3 monomer type in the final best performer. As reported earlier (Nature, 615, 251, 2023), 4 types of monomers were used to reduce protein sequences and the authors showed that increasing monomer type can significantly increase sequence space. With 6 monomers, the system flexibility associated with expanded sequence space may make many things possible. I am curious why the team did not loop back and see how the 3 monomer type-based sequence analysis looks like.
The team should also consider the possibility that the sequence coupling may not be the true origin and there are layered coupling among various factors. As is now, I got the impression that DM and ML can work like black magic. Is this true for basic material science, or it is just coincidental that the process worked, or we had wishful thinking to enforce that connection? At the age of AL, the scientific community needs to be guided/educated with all aspects possible, good and bad. IMO, it is more valuable to tell a layered, not so successful story than a perfect one.
Below are some specific comments:
1. The authors compile the protein dataset based on a key-word search from the NCBI database. Often key-word searches are not very reliable, and the results need to be cleaned systematically. Any data cleaning procedures should be listed in the SI, if they were conducted.
2. The authors categorize the protein sequences based on species, then use MSA to select one consensus sequence per species to perform analysis on. The rationale for this approach is not well explained. With explanation of the differences in protein properties and performance across different species, this categorization could be more insightful. Further, the sequence analysis methods chosen are simple enough to be done on the whole protein dataset, without introducing the uncertainty of selecting a single sequence to represent thousands of sequences. It seems that the sequence analysis would yield more statistically robust results without this MSA approach.
3. Similarly, the rationale behind this pairwise frequency approach in figures 2 and 3 should be explained. It is quite a

simplified sequence analysis method and thus should be justified.
4. Additional analysis should be done on figures 2c and 3c. The authors claim distinct patterns among different species, but they observe similar patterns between proteins and synthetic polymers. The proteins seem to have more smeared pairwise frequencies, as compared with the synthetic polymer, which has more discrete common pairs. Does this indicate less blocky sequences for the polymer counterparts? This difference should be addressed with further sequence analysis.
5. The authors explain that the “exact replication of functional class sequences would be ideal for mimicking protein functions.” This statement seems misleading in the context of this work. I believe networks synthesized here are ill defined. Even if this is not the case, it is important to convey that the statistical nature of this polymer class spans different levels, from the number of initial monomers to the sequence properties to the whole population.
6. A key result of the effort for ML-driven optimization was that some limit of performance had already been reached. Given this, the authors should explain the nature of this plateau. Is there a fundamental limit with this type of adhesive, or is this a function of the acrylate monomers selected? What is the nature of other hydrogel systems that surpass this limit? It seems like some additional insight would be useful in drawing meaningful conclusions from the ML effort.
7. The authors find that the three best performers R1-max, R2-max, and R3-max, from the ML optimization were all 3-monomer copolymers and excluded the other three monomers. This observation is quite consequential, as the sequence properties of a three-monomer as opposed to a six-monomer, random copolymer will be very different, especially from the perspective of their pairwise analysis. How can we decouple the conclusions about sequence properties from monomer feeding ratios? It is likely true that incorporating BA, PEA, and ATAC, as written, is key. However, among the best performers, how many exclude the other monomers, and thus change their sequence properties significantly?
Given this observation, the pairwise sequence analysis seems insufficient to understand the sequence level features that are necessary for optimal performance. Using figure 4c as a starting point, it may be useful to do some more detailed sequence analysis with further experimental inputs and more in-depth characterization to extract the average segment lengths for different compositions. This should be done for the Monte Carlo simulated sequences of the initial 180 compositions, as well as the ML-outputted sequences.
8. The adhesive performance testing in different conditions were done for R1-max, R2-max, and R3-max. How about the best performers from the initial 180 compositions, designed without ML optimization? Given the deviation from the six-monomer rational design, some experimental inputs/characterization in normal saline.
9. A major design rule derived from this work is: “The SHAP summary plot (Fig. 4c) reveals that high values of and significantly enhance . This is because both BA and PEA effectively expel water from the contact interface, and when neighboring with ATAC, they could enhance the electrostatic interactions with the negatively charged glass surface29,38-41.” It would be helpful to present the simulated sequences of the best performers like R1-max and analyze their sequence patterns to validate this claim. Also, comparing the sequence analysis of R1-max with the protein sequence analysis shown in Figure 2c can be beneficial. This comparison would provide further validation of the whole workflow and potentially highlight the differences in design rules between synthetic polymers and natural proteins.
10. The authors mentioned that “For copolymerization in DMS, the pairwise reactivity ratios deviate significantly from 1 (Supplementary Table 3), leading to blocky sequences (Extended Data Fig. 3).” However, based on the reactivity ratios provided in Extended Data Fig. 3 (e.g., rBA = 0.5, rPEA = 0.33 in DMS), an alternating sequence is expected rather than a blocky sequence.
11. The authors didn’t provide information regarding monomer conversion in the radical polymerization. Since monomer distribution depends on monomer conversion and the Monte Carlo simulation requires it as an experimental input, what values were used?
The hydrogel fabrication involves a two-weeks immersion in saltwater. Given that the polymers contain ester sidechain, it’s unclear whether these ester sidechains undergo hydrolysis in this process, especially when a nucleophilic monomer HEA is incorporated, which can alter the hydrogel's overall composition.

Others:
The labeling of Figure 4a is confusing, making it difficult to understand what are the differences between models RFR-GP, RFR-GP*, and GP-RFR.
In Fig. 5, all adhesive performance tests were conducted using R1-max, except for the high-pressure water leakage test, which was performed using R2-max. The rationale behind this choice is unclear.
As a side note, I found some wordings aren’t accurate although I fully understand they were used to enhance the impact of present studies. I personally prefer more objective assessment of the prior work even though they aren’t as trendy. For example, “Currently, the development of new soft materials heavily relies on experimental trial and error, often guided by researchers’ intuition and experience”. This is definitely an overstatement. There have been a lot of theory and modeling across various length scales. AI/ML are good tools but not going to save the day by itself.

Version 1:

Reviewer comments:

Referee #1

(Remarks to the Author)
A. Summary of the key results
This work focuses on applying data-driven approaches to design and synthesize new materials with enhanced adhesive properties. The proposed methodology integrates data mining, machine learning (ML), and experimental procedures to rationalize and predict the performance of adhesive hydrogels for underwater environments, reducing reliance on traditional trial-and-error methods. Inspired by nature, the authors analyze adhesive proteins from various sources to establish a large initial dataset. A descriptor-based strategy is then applied, enabling ML to predict hydrogel properties. Experimental

validation confirms the successful development of adhesive hydrogels with improved mechanical and adhesive characteristics. The study also evaluates these hydrogels for diverse applications, including preliminary biomedical validation and environmental use.

B. Originality and significance
This work is highly innovative in the field of biomaterials, particularly hydrogels. The conventional trial-and-error approach used to develop hydrogel libraries remains a significant challenge in translating novel hydrogels into practical applications. The manuscript aim to resolve an important gap in the field. The presented approach is innovative as it leverages protein data stored in existing databases, addressing the challenge of data availability. Using ML to generate hydrogels based on biomolecular functional group data represents an innovative contribution to the polymeric materials field. This approach could significantly aid the scientific community in developing AI-driven systems applicable to polymers of various types.

C. Data & methodology
Validity of Approach: <The proposed data-mining and ML methodologies are well-designed and structured. The approaches used to characterize the mechanical and adhesive properties of the hydrogels align with the study's objectives.
Quality of Data: the experimental work is robust, with comprehensive chemical characterization and validation of the synthesized hydrogels.
Quality of Presentation: The discussion of experimental results and their correlation with ML predictions has been refined and reorganized. Data and procedures are presented in the materials and methods and in SI sections.

D. Appropriate use of statistics and treatment of uncertainties
Use of statistics and treatment of uncertainties are appropriate.

E. Conclusions
The conclusions are well-founded, based on the collected and analyzed data. The revised manuscript includes improvements in hydrogel characterization, making the technical details accessible to the scientific community.

F. Suggested improvements
The manuscript has been improved.

G. Reference: appropriate credit to previous work: YES

H. Clarity and context: Abstract, introduction and conclusions are adeguate and clearly reported.

Referee #2

(Remarks to the Author)

None

Referee #3

(Remarks to the Author)
The authors have made a good effort to respond to previous review questions. But some key questions still linger.

1. The major concern still remains in terms of molecular connections between what's delivered via DM/ML vs. experimental results and explanations.

a. While I appreciate the characterizations of the resultant hydrogels, most if not all are bulk properties. There are quite gaps between monomer chemistry and distributions along a strand in a network to the bulk properties, let alone the adhesion. I also found the discussions on the rationale of these top performers are superficial in general. They mainly focused chemical identity, composition and occasionally are connected to the simulated sequences as shown in the SI Fig. 6b. This is a significant weakness and should be addressed with additional experiments. The authors specifically pointed out that for adhesive properties, the strands are the main motifs interacting with the substrate. Thus, how the polymers interact with the underlying substrate, e.g. glass surfaces, should be studied for a few key samples here. The authors should feel free to do whatever they see fit to bridge the gap. There are many studies from the adhesive community. But if this is my project, I will look into measuring adhesions of polymers via AFM so there is a base to compare with other known work on polymer adhesive properties. The short blocks of functional monomers appear to be a key design factor. The team should consider how this may affect the chemistry/functional groups at the interfaces. Ambient XPS under controlled humidity can help. ATR-FTIR can be useful as well and if the team can have access to polarized source, the results will be even more insightful.

b. I also appreciate the authors' efforts to monitor monomer conversions for ATAC/PEA pair in different solvents. The authors ascertained the reaction mixtures' miscibility by stating solution transparency/clarify. I feel the conclusions should have been drawn with more scientific vigor. Given the monomer concentration used for network formation is much higher than these control studies and the variety/chemical differences of the monomers, I think light scattering may be more convincing. (It is not ideal but an easier experiments than more advanced techniques to probe if the monomers may form aggregates or phase separate at length scales below 400 nm.) To clarify, with the high viscosity of monomer solutions and >98% conversion, the radicals have very low diffusivity. A slight aggregates will change the effective monomer ratio.

c. Based on what I read, the monomer conversion or near unity reactivity ratios were determined using a pair of two monomers. I want to bring one recent publication to the team attention that discuss potential complications of reactivity ratios in the presence of more monomers to account for other factors. The team should look into their system to account for the potential errors introduced by the reactivity ratios. (Mapping Composition Evolution through Synthesis, Purification, and Depolymerization of Random Heteropolymers, J. Am. Chem. Soc. 2024, 146, 617.)

d. During the DM, residues like glycine, alanine and proline were excluded. In fact, the selection appears to be arbitrary to me and can't be simply justified as stated in the main text and rebuttal. Although the team reasoned this by their lack of interactions with the substrates, these residues are important when it comes to the results shown Figure 2, for example the block length of different types of residues. The analysis including these residues should be done and compared.

2. I strongly urge the team to revisit the introduction regarding the status of soft material design using DM, ML and bioinformation and more generally, work by the soft material community even before ML became the buzz word. For example, DM/ML and/or bioinformation have been used in soft material design by many, not only by the hard material community.

The team revised:
Traditionally, the development of new soft materials has relied on experimental trial and error, often guided by researchers' intuition and experience."

This is simply not true if one goes back to as simple as Flory-Huggins theory or as complex as scaling laws. Even for network synthesis, the formulations derived by the statistical mechanics have been around for a long time.

3. The authors should definitely check the references to make sure they are cited at the right place. T

Response Letter

We sincerely appreciate the reviewers' insightful comments and suggestions, which have been extremely helpful in improving our work. In the revised manuscript, we have carefully addressed all the concerns raised by the reviewers, incorporating additional analyses, clarifying key points, and refining our discussion to ensure a more comprehensive and rigorous presentation of our findings.

A point-by-point response to the reviewers is provided below. In this response, the reviewers' comments are shown in *italics*, our responses are in blue, and the changes in the Manuscript, Extended Data, and Supplementary Materials are highlighted in red.

Referee #1 (Remarks to the Author):

A. Summary of the key results

The proposed work is focused on the application of data-driven approaches to design and synthetize new materials with improved adhesive properties. The proposed approach combines data mining (DM), machine learning (ML) and experimental procedures to rationalize and predict adhesive hydrogels with superior properties for underwater environments minimizing the classical trial and error approach. The authors take inspiration from Nature, collecting and analyzing adhesive proteins from different sources and to have a large starting data-set. From these data, a descriptor strategy has been selected allowing the application of ML to predict hydrogel properties. As demonstrated by experimental validation, adhesive hydrogels with improved mechanical and adhesive properties were generated. The evaluation of the produced hydrogels for different application has been performed, including preliminary biomedical validation and environmental applications.

Response: We sincerely thank the reviewer for the insightful summarization of our work.

B. Originality and significance: if not novel, please include reference

The proposed work is highly innovative in the biomaterials, and in particular hydrogel field. The common trial-and-error method employed to produce hydrogel libraries for different applications still represent an uncovered gap to translate novel hydrogels.

However, the application of ML methods is limited by the lack of homogeneous data collection,

the limited number of data available in literature or patents, the different polymers or hydrogel employed for the generation of hydrogels and finally but important the different characterization employed to evaluate the final properties of the hydrogels.
The presented concept is innovative and the idea to start with proteins already stored in data bank can overcome the problem of data availability. The application of ML in the generation of hydrogels starting from biomolecular data related to functional group is novel in the polymeric material field and could help the scientific community in the generation and application of AI based systems appliable also to polymers of different nature.

Response: We are very grateful to the reviewer for the highly positive evaluation on the originality and significance of the work.

C. Data & methodology: validity of approach, quality of data, quality of presentation
• *Validity of approach: the proposed data-mining and machine learning methods are well designed and structured. In general, the approaches employed to characterize mechanical and adhesive properties of the hydrogel are coherent with the aim of the work and with the reported aims.*

Response: We thank the reviewer for the positive comment on the validity of our approach.

The approaches employed for the experimental part lack of important information to fully evaluate the reported properties of the hydrogels and in general the efficacy of the proposed approach. In detail:
• *the section on data mining of adhesive proteins reports on the selection of dataset including proteins with an average length between 300 to 500 amino acids. It is not clear if this range has been selected for specific "functional" reasons, or to allow the management of data.*

Response: We thank the reviewer for raising this point. We did not make any specific selection of the protein dataset. The average length of 300-500 is a result of statistical analysis conducted on the entire set of 24,707 adhesive proteins in the assembled database, as shown in Supplementary Fig. 2.

To clarify this point, we have revised the sentence in the manuscript (1st paragraph on page 5) as follows:

"This dataset includes proteins from 3,822 different organisms across archaea, bacteria, eukaryotes, viruses, and artificial proteins. Statistical analysis reveals that the average length of those adhesive proteins ranges from approximately 300 to 500 amino acids (Supplementary Fig. 2)."

• *The impact of ML application and validation using random polymerization can be limited comparing other synthetic methods, considering that the interactions between chains with adhesive functional group favor the formation of hydrogels with major adhesive properties.*

Response: We fully agree with this insightful comment by the reviewer. While random polymerization is highly efficient and cost-effective for preparing bulk materials, it does have limitations in terms of controlling monomer sequences. To expand the current method for broader applications targeting various functional materials, advanced synthetic methods that allow for more precise sequence control of functional monomers are required. Achieving this level of control remains one of the biggest challenges in polymer chemistry.

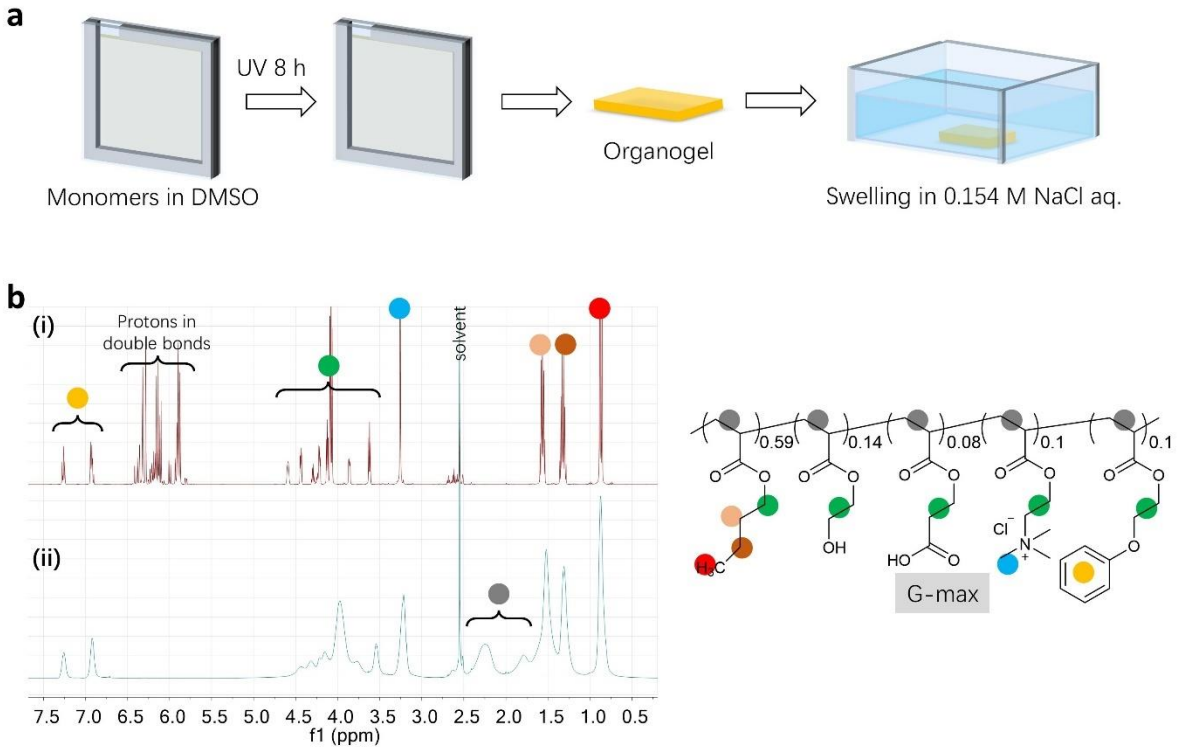
• *Quality of data: the experimental part and the information reported, including the chemical characterization of the obtained hydrogels and the validation of the obtained hydrogels, are limited and lack of important information to evaluate the final properties of the hydrogels and consequently the accuracy of the proposed approach to obtain hydrogels with improved properties.*

Response: We thank the reviewer for kindly pointing out the insufficiency of information regarding the chemical characterization and validation of obtained hydrogels. These procedures were carried out following strict protocols in our lab.

Monomer conversion was examined by structural characterization using NMR for representative samples. Due to the high reactivity of the selected monomers, the conversion exceeds 99% to form gels via free radical polymerization in DMSO. Also, we confirmed high

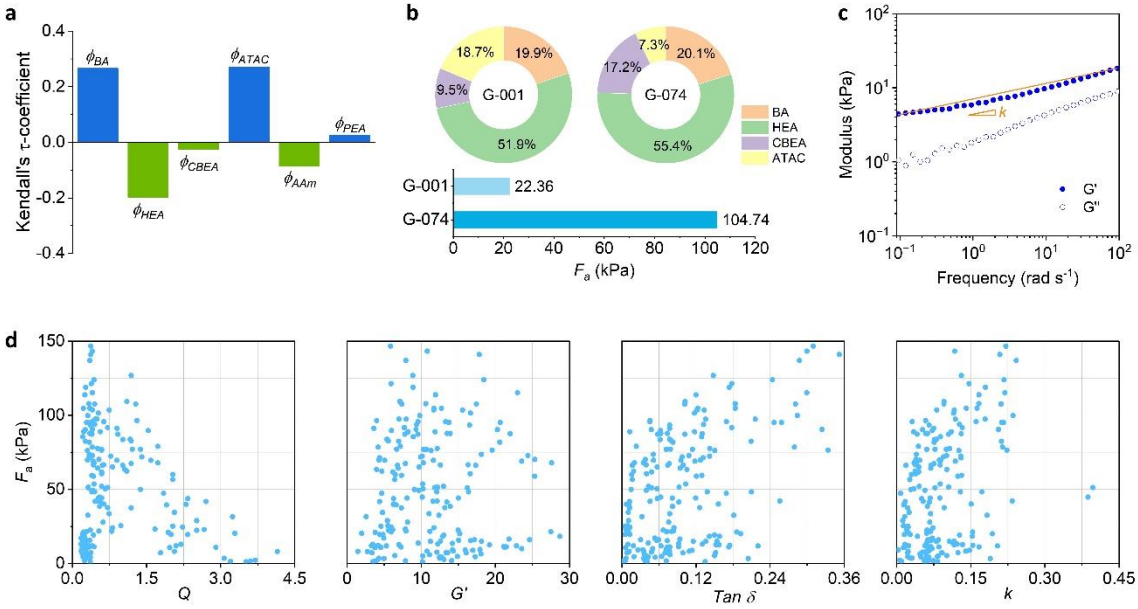
reproducibility in hydrogel structure formation, which leads to reproducible properties including swelling, rheological, and adhesive behaviors.

To address this point, we have now included chemical characterization data for the G-max gel, measured using NMR, in the newly revised Supplementary Fig. 9b (shown below). In addition, we added an illustration and description of the hydrogel fabrication process (revised Supplementary Fig. 9a).

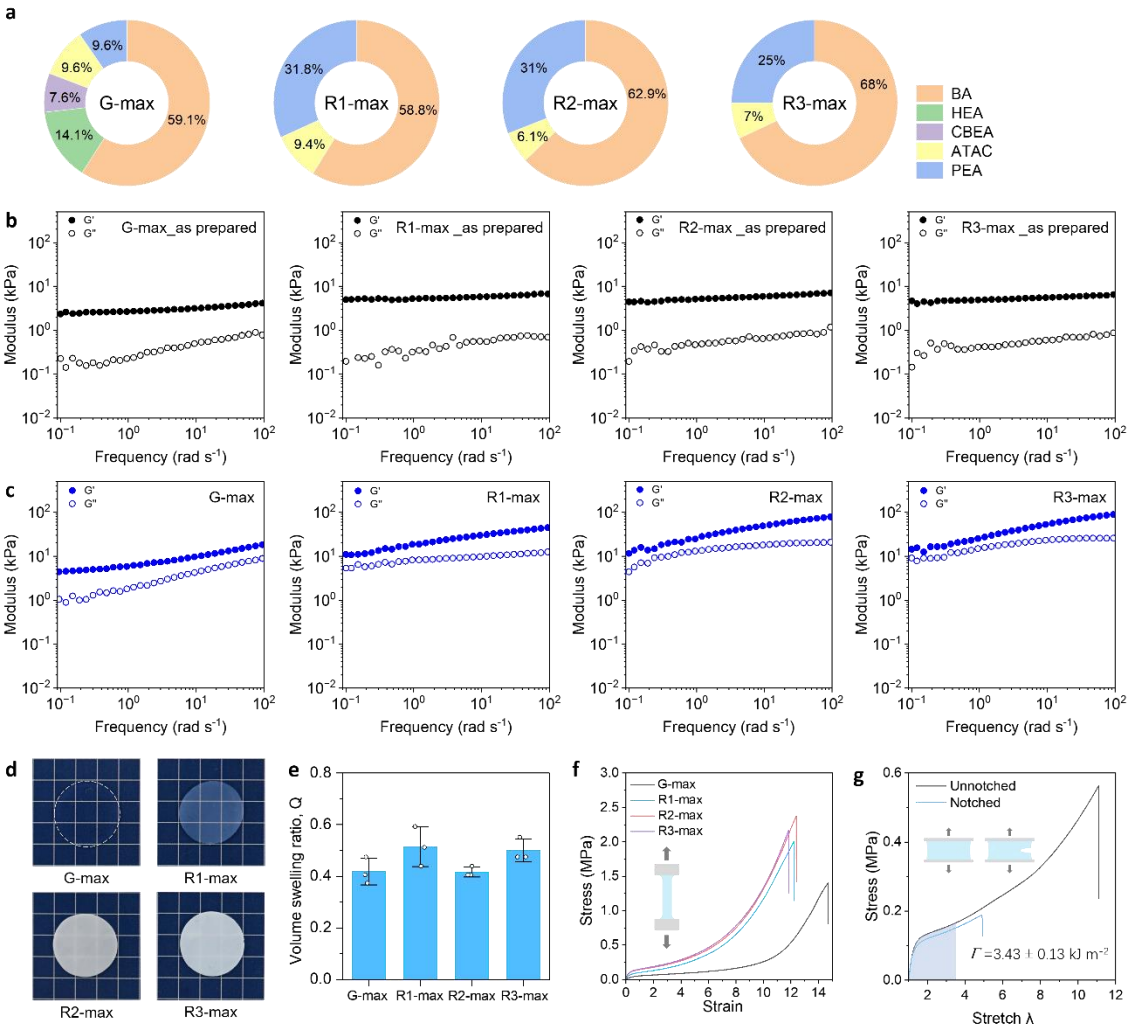


Supplementary Fig. 9. Fabrication of hydrogels. (a) Schematic diagram illustrating the hydrogel fabrication procedure. (b) ^{1}H NMR spectra of the monomer precursor solution for G-max (i) and the corresponding gel (ii) synthesized in DMSO. Based on the intensity of proton signals from double bonds, the monomer conversion ratio for gel formation was estimated to exceed 99%.

As to hydrogel property characterization, we have included swelling and rheological behaviors data for the 180 samples in new panels c and d of the revised Extended Data Fig. 3 (shown below). We also measured the rheological and tensile properties of G-max, R1-max, R2-max, and R3-max gels, which are now presented in the revised Extended Data Fig. 6 (shown below).



Extended Data Fig. 3. Analysis of the correlations between adhesive strength (F_a) and properties of 180 bioinspired hydrogels. (a) Correlation between monomer proportions (ϕ_i) and F_a, captured by Kendall's τ coefficients. These coefficients reveal that ϕ_{BA}, ϕ_{ATAC}, and ϕ_{PEA} have weak positive correlations with F_a, while ϕ_{HEA}, ϕ_{AAm}, and ϕ_{CBEA} show weak negative correlations. (b) Example illustrating the complex interplay between ϕ_i and F_a, due to the synergistic effects of different monomers. When comparing G-074 to G-001, ϕ_{BA} is roughly the same, ϕ_{HEA} and ϕ_{CBEA} increase, and ϕ_{ATAC} decreases. Despite the Kendall's τ coefficients suggesting that the F_a of G-074 should be lower than that of G-001, the actual F_a of G-074 is about five times higher. (c) Angular frequency dependence of the storage modulus (*G′*) and loss modulus (*G″*) of the G-042 hydrogel as a demonstration. The slope (*k*) of *G′* is calculated from the line connecting *G′* values at frequencies of 0.1 and 100 rad s^{-1}. A larger slope indicates higher viscoelasticity of the gel. (d) F_a as a function of network properties for the 180 bioinspired hydrogels. *Q*, *G′*, *Tan* δ = *G″*/*G′*, and *k* represent the volume swelling ratio, storage modulus at a frequency of 10^0 rad s^{-1}, loss factor at a frequency of 10^0 rad s^{-1}, and the slope of the *G′* curve, respectively. These results suggest that gels exhibiting shrinking behavior (*Q* < 1), moderate *G′*, large *Tan* δ, and moderate *k* tend to have higher F_a values. All characterizations were performed on gels equilibrated in normal saline (0.154 M NaCl). Adhesion tests were conducted using a tack test with a 10 N loading force and a 10-second contact time on a glass substrate to enable rapid screening and ensure consistent comparisons across the dataset.



Extended Data Fig. 6. Properties of the top-performing hydrogels from three rounds of machine learning (ML) optimization (R1-max, R2-max, R3-max) and comparison with the top-performing hydrogel identified via data mining (G-max). (a) Formulations of the four gels. (b) Angular frequency dependence of storage modulus (*G′*) and loss modulus (*G″*) for the four gels in their as-prepared state (using DMSO solvent). (c-f) Properties of the four hydrogels equilibrated in normal saline (0.154 M NaCl): (c) Angular frequency dependence of *G′* and *G″*, (d) Photographic images showing the gel appearance, (e) Volume swelling ratio (*Q*), and (f) Uniaxial tensile stress–strain curves at a stretch rate of 100 mm min^{-1}. (g) Pure shear stress–stretch ratio curves at a stretch rate of 100 mm min^{-1} for the R1-max hydrogel (equilibrated in normal saline) with and without a notch. The notched sample exhibited crack propagation at a critical stretch ratio (λ_c) of 3.4. The fracture energy (Γ) estimated from the pure-shear test is also shown. Experimental details are provided in the Supplementary Materials. Error bars represent the standard deviation of N = 3 measurements. In the as-prepared state, all gels were transparent (results not shown) and displayed similar rheological profiles with comparable storage moduli, indicating that synthesis in the cosolvent DMSO resulted in similar topological network structures, despite compositional differences. Upon equilibration in normal

saline, all gels underwent shrinkage due to inter- and intra-molecular interactions between polymer strands. The three ML gels (R1-max, R2-max, and R3-max) were translucent and exhibited a strong frequency dependence of G', with larger G'' compared to G-max, suggesting enhanced hydrophobic associations of polymer strands in normal saline. These properties contribute to the gels' higher strength and slightly reduced stretchability. Moreover, the ML gels also showed high toughness, as indicated by the large area under their stress–strain curves, which improves their adhesion by facilitating more efficient energy dissipation during the debonding process.

• *Quality of presentation: the discussion on experimental results and their correlation with predicted data should be reorganized introducing structures of the hydrogels in the text, reporting qualitative data improving the discussion and reorganizing the selection of figures and graphs cited and available in the extended data (in particular in the discussion of the experimental of the obtained hydrogels. Materials and methods should be implemented for each step to allow precise reproduction of the experimental and homogeneity / availability of experimental data.*

Response: We thank the reviewer for these valuable suggestions. For the initial 180 hydrogels, formulated based on data mining of adhesive proteins, we have performed swelling and rheological measurements in addition to adhesion tests. In the revised manuscript, we have included these results in revised Extended Data Fig. 3c and 3d (shown above).

Our findings suggest that, generally, a small swelling ratio (Q) appears to be a necessary but not sufficient condition for achieving high adhesive strength (F_a), while a large loss factor ($Tan\ \delta$) and viscoelasticity (k) are correlated with higher F_a. However, due to the inherent complexity of structure-property relationships in hydrogels, it remains challenging to derive optimal formulations from these results alone. As a result, we introduced machine learning techniques to facilitate the identification of optimal hydrogel formulations.

To address this point, we have included a schematic illustration of the hydrogel fabrication in Supplementary Fig. 9a (shown above). In addition, we have provided detailed protocols for hydrogel fabrication, swelling ratio characterization and rheological testing in the revised Supplementary Materials (on pages 3 and 4) as follows, to ensure better reproducibility of the experiments.

"**Hydrogel Fabrication**

All copolymer gels were synthesized via one-step free-radical copolymerization of monomers in the presence of a chemical crosslinker. The crosslinker concentration was set at 0.1 mol% relative to the total monomer content to balance the elasticity and deformability of the gels[32]. DMSO solutions containing functional monomers (total monomer concentration of 2.4 M) with compositions derived from data-mining and machine learning (Supplementary Table 2 and Table 7), chemical crosslinker (Glycerol 1,3-diglycerolate diacrylate, 2.4 mM), and UV initiator (2-oxoglutaric acid, 6 mM) were used. For example, to prepare the G-max gel, 1.819 g of BA, 0.413 g of HEA, 0.264 g of CBEA, 0.561 g of ATAC, 0.441 g of PEA, 8.4 mg of glycerol 1,3-diglycerolate diacrylate, and 8.8 mg of 2-oxoglutaric acid were added to a 10 mL volumetric flask. DMSO was then added to bring the volume to 10 mL. Then the resulting mixture (precursor solution) was transferred into a glove box to remove oxygen. After that, the precursor solution was poured into a reaction cell consisting of a pair of glass plates (10 cm × 10 cm, with a 0.5-mm spacing) and irradiated with UV light (365 nm wavelength, 4 mW cm^{-2} intensity) inside the glovebox for 8 hours to form gels (Supplementary Fig. 9a). After UV irradiation, over 99% of the monomers were converted into polymers, as confirmed by NMR (Supplementary Fig. 9b)."

"**Hydrogel Mechanical Property Characterization**

The volume swelling ratio Q is defined as the ratio of the sample volume at swelling equilibrium (V) in normal saline to that in the as-prepared state (V_0), expressed as $Q = V/V_0$. Since the size change of the gels is isotropic, the gel thickness was measured both in the as-prepared state (d_0) and at swelling equilibrium (d) using a thickness gauge (SDA-25, KOBUNSHI KEIKI CO., LTD). The swelling ratio was then calculated as $Q = (d/d_0)^3$.

Rheological tests were performed using an ARES-G2 rheometer (TA Instruments) in parallel-plate geometry at 24 °C. Samples equilibrated in normal saline were cut into discs with a diameter of 15 mm using a custom punch cutter. The samples were placed directly onto the plates without adhesive due to their inherent adhesion ability. A frequency sweep test was conducted at a shear strain of 0.1%, with frequencies ranging from 0.1 to 100 rad s^{-1}."

D. Appropriate use of statistics and treatment of uncertainties

Use of statistics and treatment of uncertainties are appropriate for the DM and ML. Statistics and treatment of uncertainties in the experimental part should be added and implemented.

Response: We thank the reviewer for raising this important point. All experimental

measurements were conducted at least three times, as indicated in the corresponding figure captions (e.g., error bars represent the standard deviation of $N = 3$ measurements). In the revised manuscript, we have emphasized this statement in the Methods section and all the relevant figure captions.

Furthermore, for greater transparency and to allow readers to better assess the variability of the data, we have added the original data points for related figures (Fig. 5a and 5c in the main text, Extended Data Figs. 2, 6, 7, and 8, and Supplementary Figs. 1, 12, 16, and 17).

E. Conclusions: robustness, validity, reliability

The conclusions are appropriate concerning the data collected and discussed for the dry part (DM and ML). The conclusions related to the experimental part, in particular on the evaluation of synthetic improvements and hydrogel validations are general and partially discussed.

Response: We thank the reviewer for this insightful comment. In response to this feedback, as well as related comments from other reviewers, we have thoroughly revised the Conclusion section (on page 14 of the main text). The revised conclusion now provides a clearer summary of our key findings, outlines the remaining challenges, and offers a brief outlook for future developments. We hope this revision better captures the essence of our work and its potential impact moving forward.

"In summary, we introduced a data-driven approach that integrates the extraction of valuable sequence information from proteins, scalable polymer synthesis, and iterative machine learning to address longstanding challenges in the de novo design and development of soft materials. This approach enabled the discovery of hydrogels with underwater adhesive strengths exceeding 1 MPa, representing an order-of-magnitude improvement over previous benchmarks. Beyond adhesive hydrogels, this data-driven design framework offers a systematic, scalable, end-to-end approach for developing a wide range of functional soft materials. However, challenges remain, primarily due to limitations in monomer diversity, polymer synthesis technologies for controlling monomer sequences to a scale suitable for materials development, and dataset scalability. Overcoming these challenges will require expanding modular monomer libraries, advancing polymerization techniques, and developing physics-informed machine learning models capable of generalizing across sparse, multi-scale datasets."

F. Suggested improvements: experiments, data for possible revision

General comment: the manuscript is undoubtedly interesting and highly innovative. Furthermore, the proposed approach can open new perspectives that could be applied to other fields and to other polymers. However, several aspects should be revised or implemented. The manuscript should be revised to help the reader.

Response: We sincerely appreciate the reviewer's positive evaluation of our work. We have carefully followed the reviewer's suggestions, as detailed in the point-to-point replies below, to improve our manuscript.

- *The discussion on the experimental data should be implemented with structures and a clear assignation of the produced hydrogels in the text and not in the supplementary.*

Response: We thank the reviewer for this suggestion. In the revised manuscript, we have included the chemical characterization (via NMR) and network structure characterization (via swelling and rheology measurements) of the produced hydrogels, as mentioned in our response to previous comments. However, due to space limitations in the main text, these detailed characterizations are provided in the Extended Data Figures and Supplementary Materials.

Regarding the assignation of the produced hydrogels, we have included this information in Supplementary Tables 2 and 7. These tables contain 180 and 210 entries, respectively, making them quite lengthy. To adhere to the format requirements, we included the tables in separate Excel files.

To enhance readability, we have also included a new Supplementary Table 8 (shown below) in the Supplementary Materials (on page 35), which lists the names, formulations, volume swelling ratios, Young's modulus, storage modulus, and adhesive strengths of the four top-performing hydrogels from the training dataset and the three machine learning optimization rounds.

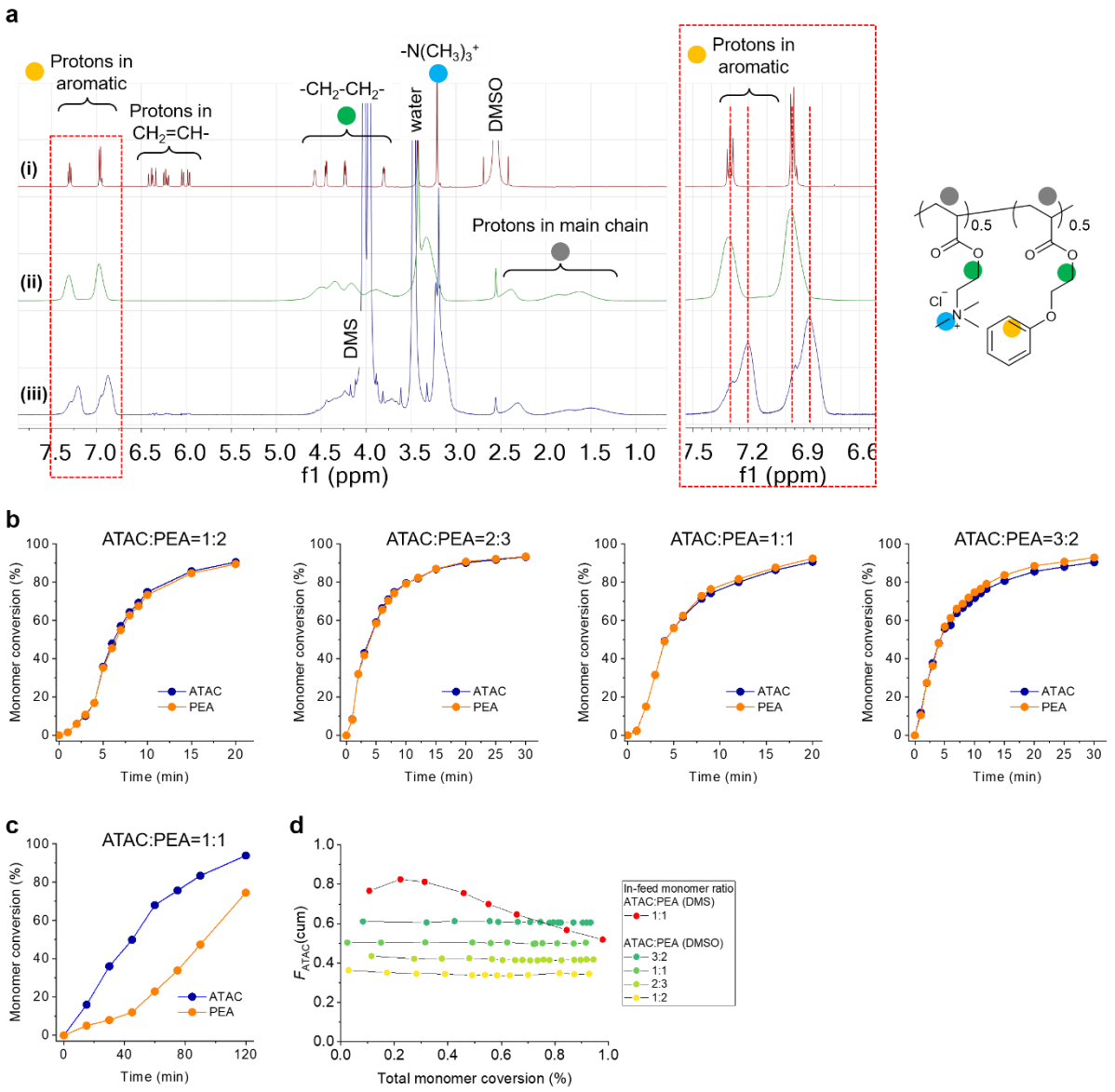
Supplementary Table 8. Formulations, volume swelling ratios (Q), Young's modulus (E), storage modulus (G') at a frequency of 10^0 rad s^{-1}, and adhesive strength (F_a) of the top-performing hydrogels from data mining and three machine learning optimization rounds. Tack tests were conducted under a 20 N loading force applied for 60 seconds on a glass substrate in normal saline (0.154 M NaCl). All hydrogels were equilibrated in normal saline.

No.	Φ_{HEA}	Φ_{BA}	Φ_{CBEA}	Φ_{ATAC}	Φ_{PEA}	Φ_{AAm}	Q	E (kPa)	G' (kPa)	F_a (MPa)
G-max	0.141	0.591	0.076	0.096	0.096	0.000	0.42±0.04	125.60±5.74	5.86	0.50±0.02
R1-max	0.000	0.588	0.000	0.094	0.318	0.000	0.51±0.06	320.76±7.93	18.72	1.02±0.01
R2-max	0.000	0.629	0.000	0.061	0.310	0.000	0.42±0.02	459.99±26.74	24.69	0.85±0.04
R3-max	0.000	0.680	0.000	0.070	0.250	0.000	0.50±0.04	538.95±4.72	25.35	0.86±0.01

• *The discussion of the experimental part should be implemented considering a) the chemical characterization of the produced hydrogels. NMR should be reported for the samples considering the entire spectra and an in-depth analysis using also the complete assignation of the spectra.*

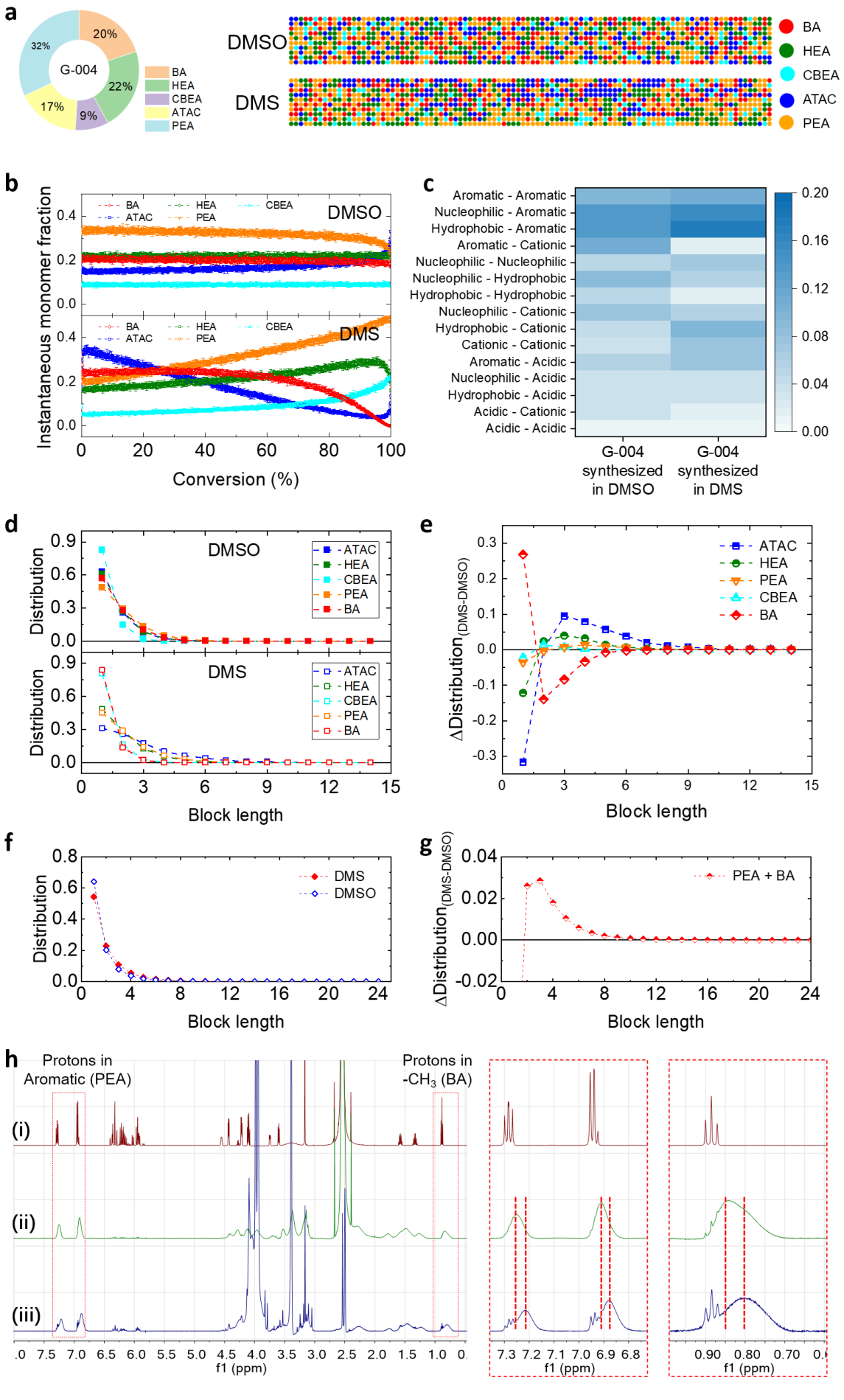
Response: We thank the reviewer for these suggestions. In response to previous comments, we have included the full NMR spectra in the revised Supplementary Figures 6a, 7h, and 9b. Specifically, Supplementary Fig. 6a now includes the spectra for the copolymerization of cationic ATAC and aromatic PEA in different solvents (shown below), Supplementary Fig. 7h shows the spectra for G-004 variants (shown below), and Supplementary Fig. 9b presents the spectra for G-042/G-max (see above).

These spectra confirm the quasi-ideal random copolymerization of the selected functional monomers, the variation of block lengths for copolymers polymerized in DMSO and DMS solvents, and a monomer conversion greater than 99% for gels synthesized in DMSO. The assignments of the peaks are clearly indicated in the figures, and further details are provided in the figure captions.



Supplementary Fig. 6. ^{1}H NMR studies on the time evolution of compositions during the copolymerization of cationic ATAC and aromatic PEA in different solvents. (a) ^{1}H NMR spectra of ATAC and PEA monomer precursor solutions at a 1:1 molar ratio (i) and the corresponding copolymers after full polymerization in DMSO (ii) and in DMS (iii). The portion highlighted by a red dashed frame in the full NMR spectra is enlarged on the right. For the copolymer synthesized in DMSO, the phenyl proton signals exhibit symmetric broadening around the peak of the phenyl protons of the aromatic monomers, indicating a statistically dispersed arrangement of cationic and aromatic monomers along the polymer chains. In contrast, for the copolymer synthesized in DMS, the phenyl proton signals show new, strong peaks at higher fields, suggesting the formation of long aromatic monomer sequences[5]. (b, c) Monomer conversion kinetics of ATAC and PEA at different feed molar ratios in DMSO (b) and at a 1:1 feed molar ratio in DMS (c). (d) Cumulative ATAC molar fraction in copolymers (F_{ATAC}) plotted against total monomer conversion for varying feed molar ratios in different solvents. In

DMSO, the copolymers maintained nearly the same monomer composition as the feed throughout polymerization, with no significant composition drift, indicating quasi-ideal random copolymerization. In contrast, in DMS at a 1:1 feed molar ratio, the ATAC fraction deviated significantly from 0.5 and varied with monomer conversion, demonstrating distinct composition drift during polymerization. In all NMR experiments, the total monomer concentration was 1.0 M.


a
G-004
20%
22%
9%
17%
32%
BA
HEA
CBEA
ATAC
PEA
DMSO
DMS
b
Instantaneous monomer fraction
Conversion (%)
c
Aromatic - Aromatic
Nucleophilic - Aromatic
Hydrophobic - Aromatic
Aromatic - Cationic
Nucleophilic - Nucleophilic
Nucleophilic - Hydrophobic
Hydrophobic - Hydrophobic
Nucleophilic - Cationic
Hydrophobic - Cationic
Cationic - Cationic
Aromatic - Acidic
Nucleophilic - Acidic
Hydrophobic - Acidic
Acidic - Cationic
Acidic - Acidic
G-004 synthesized in DMSO
G-004 synthesized in DMS
d
Distribution
Block length
e
ΔDistribution(DMS-DMSO)
Block length
f
Distribution
Block length
g
ΔDistribution(DMS-DMSO)
PEA + BA
Block length
h
Protons in Aromatic (PEA)
Protons in -CH3 (BA)
(i)
(ii)
(iii)
f1 (ppm)

Supplementary Fig. 7. Monte Carlo simulations (a-g) and ^{1}H NMR analysis (h) of monomer sequences for G-004 prepared in DMSO and DMS. (a) G-004 formulation and its simulated chain sequences in DMSO and DMS, based on reactivity ratios from Supplementary Table 3. Each chain consists of 1,000 monomers, presented in 10 rows, with each row showing the sequence of 100 monomers from left to right. (b) Instantaneous monomer fractions as a function of total conversion during polymerizations in DMSO and DMS. Each simulation involved 2 million monomers, with an average chain length of 1,000. The results show minor composition drift for polymerization in DMSO, but significant composition drift in DMS throughout the polymerization. (c) Pairwise frequency distribution of 15 distinct functional class pairs along encoded sequences. (d) Block length distributions for different monomers from the simulated sequences. (e) Differences in monomer block length distributions between copolymers prepared in DMS and DMSO, highlighting increased block lengths for ATAC, HEA, and PEA. (f) Block length distributions for combined aromatic PEA and hydrophobic BA in DMSO and DMS. (g) Difference between the two distribution curves in (f), suggesting longer segments of combined BA and PEA in DMS. Statistical results in (b-g) were obtained from five independent runs. (h) ^{1}H NMR spectra of G-004 monomer precursor solution (i) and copolymers synthesized in DMSO (ii) and in DMS (iii) after 8 hours of polymerization (conversion > 99% in DMSO and > 93% in DMS). The portions of the NMR spectra highlighted by red dashed frames are enlarged on the right. For PEA aromatic protons, a high-field shift in the peak indicates an increase in PEA block size. For BA methyl protons, a high-field shift suggests a higher frequency of adjacent BA-PEA units, corresponding to larger segments of combined BA and PEA monomers. These findings are consistent with the simulation results shown in (f, g). Overall, the simulations suggest that hydrogels prepared in DMSO maintain quasi-ideal random copolymerization, resulting in statistical monomer sequences that determine the hydrogel's adhesion properties. In contrast, hydrogels prepared in DMS exhibit blocky sequences of combined BA and PEA (both hydrophobic), which lead to aggregation of hydrophobic components and the opaque appearance (cf. Fig. 3f in the main text). Meanwhile, the increase in ATAC block length negatively impacts adhesion[1].

• *The discussion on the obtained results should be implemented reporting the experimental results related to the chemical nature of obtained hydrogels (using for example NMR, and FTIR).*

Response: We thank the reviewer for the suggestion. As mentioned in our response above, we have included the chemical characterization data, measured via NMR, for G-004 variants in Supplementary Fig. 7h and for G-max in the Supplementary Fig. 9b (see above), which serve

as typical examples.

• *In the paragraph related to the Validation the different methods and characterization should be implemented as following reported: adhesive testing should be reported considering and discussing quantitative data and qualitative data considering also the comparison between different samples at different time points.*

Response: We apologize for the lack of clarity on this point in the manuscript. For the experimental validation, we used the same adhesion test conditions for both the hydrogel dataset formulated from data mining and machine learning to ensure consistency in the data. The test conditions included: solvent medium (0.154 M NaCl aqueous solution), substrate type (glass), loading force (10 N), contact time (10 s), and retraction speed (10 mm/min).

After validating the ML-predicted formulations, we further tested the top-performing gels under a broader range of test conditions, including different solvent media, substrates, loading forces, and contact times. These results are shown in Fig. 5, with the legends indicating the test conditions.

To address this point, we have now specified tack test conditions in the revised figure captions of Figs. 3 and 4 in the main text, as well as in Extended Data Figs. 2, 3, and 7, and Supplementary Figs. 1, 12, 16, and 18. Furthermore, we have made the following revisions in the manuscript to enhance clarity.

1. In the 1st paragraph on page 10 of the main text:

"Experimental validation of the test sets followed the same protocol as for the training set to ensure consistency. Figure 4a plots the true F_a values for formulations proposed by different SMBO methods (Supplementary Table 4). For comparison, non-SMBO baselines, GP_enu and RFR_enu, which selected the top 5 PRED from an enumeration of ten million random formulations, failed to improve F_a beyond the training data. In contrast, all SMBO methods achieved higher F_a. GP_KB and RFR-GP emerged as the top performers, with RFR-GP yielding the highest F_a overall."

2. In the "Hydrogel underwater adhesive property characterization" subsection of the Methods section, starting on page 15 of the main text:

"The tack test was conducted using a SHIMADZU tester (Autograph AG-X) equipped with

Trapezium X software. In the experiment, a hydrogel (0.3 ~ 0.8 mm thickness) in its swelling equilibrium state was initially adhered to the probe using cyanoacrylate (super glue). For rapid screening, samples, including the data mining-driven hydrogels from the training round and the machine learning-driven hydrogels from the three optimization rounds, were prepared with a diameter of 15 mm for tack tests. For detailed investigations of adhesion capabilities, 10-mm diameter samples were used, as the adhesion of the 15-mm samples could exceed the measurable force range of the instrument. This change in diameter did not affect the adhesive strength results. The hydrogel on the probe was then immersed in a test solution (e.g., normal saline) for 5 minutes to achieve equilibrium before testing. The probe descended toward the substrate at a rate of 1 mm min^{-1} until a loading force of 10 N was reached, which was maintained for 10 seconds. The probe was then withdrawn at a rate of 10 m min^{-1} (Supplementary Fig. 10). These test conditions were used as a standard protocol unless otherwise specified. For repeated adhesion tests, the hydrogel was allowed to rest underwater for 5 minutes between consecutive tests, and a new glass substrate was introduced every 100 tests. During prolonged testing of attachment-detachment cycles (Supplementary Fig. 15), the loading force was set to 5 N with a 10-second contact time to minimize gel fatigue. All samples were measured at least three times. For hydrogel dataset construction, the highest adhesive strength recorded for each sample was reported as F_a, representing the maximum adhesion performance of the hydrogels under the specific measurement conditions."

- *The hydrogels employed in the validation and characterization should be clearly represented, reported and identified; b) the discussion on biological validation is limited to few lines.*

Response: We thank the reviewer for these suggestions. In response, we have now included detailed composition and characterization data for all the tested hydrogels in Supplementary Tables 2 and 7 as separate Excel files. To further enhance readability, we have also compiled the data for the four top-performing hydrogels into a new Supplementary Table 8.

Additionally, in accordance with the reviewer's recommendation, we have shortened the discussion on potential biological applications to improve focus and flow (please refer to the next response). The revised sentence (in the 2nd paragraph on page 13 of the main text) reads as follows:

"Furthermore, all these hydrogels exhibited good biocompatibility, as confirmed by subcutaneous implantation beneath the dorsal skin of mice (Extended Data Fig. 10),

suggesting potential applicability in the biomedical field. ~~This supports their applicability in biomedical field, such as wound dressing and bone tissue engineering.~~"

• *To evaluate the impact of the proposed approach in the field of adhesive for medical application both the discussion and the experimental should be expanded (i.e. biocompatibility, moisture vapor transmission rate (MVTR), cohesive / adhesive failure, permeability).*

Response: We sincerely thank the reviewer for this valuable feedback. We fully agree that biomedical applications are an important potential outcome for adhesive hydrogels. To address this, we included in vivo experiments to evaluate the biocompatibility of our samples, as we recognize that this is a critical factor for their applications in biomedical settings. However, as the primary focus of this work is on applying data-driven approaches to design and synthesize new materials with enhanced underwater adhesive properties, we believe that a more comprehensive exploration of biomedical evaluations would be better suited for a follow-up study. This approach aligns with Reviewer 2's suggestion to dedicate a separate paper to further investigations in this area. We deeply appreciate the reviewer's understanding and will certainly consider expanding these aspects in our future work.

Materials and Methods:

• *Hydrogel fabrication – the procedure should be implemented considering the different hydrogel formulations and well reported (quantities, mmol for each reagent, volumes…)*

Response: We are grateful for this suggestion. As mentioned in our response to a previous comment, we have provided a brief yet detailed description of the gel fabrication process, including specific quantities, in the revised "Hydrogel fabrication" subsection of the Methods section on page 15 of the main text. For ease of reference, this information has also been included in the Supplementary Materials. We hope that this revision enhances both the clarity and reproducibility of our work, and we appreciate the reviewer's insight in this regard.

• *Hydrogel underwater adhesive property characterization – volume of tested hydrogels*

should be reported. The experimental procedure should be reported considering also the number of experiments for each sample.

Response: We thank the reviewer for these comments. The size of the tested hydrogels is specified in the Methods section. Specifically, the tack test was performed using disc-shaped hydrogels with a diameter of 15 mm for the rapid screening tests (Supplementary Tables 2 and 7). The thickness of the hydrogels ranged from 0.3 to 0.8 mm, depending on the swelling ratio. All measurements were conducted at least three times, as indicated in the figure captions, where the error bars represent the standard deviation of $N = 3$ measurements.

To further clarify this, we have added the following sentences to the end of the 1st paragraph of the "Hydrogel underwater adhesive property characterization" subsection in the Methods section on page 16 of the main text:

"All samples were measured at least three times. For hydrogel dataset construction, the highest adhesive strength recorded for each sample was reported as F_a, representing the maximum adhesion performance of the hydrogels under the specific measurement conditions."

• *Machine Learning. For non-linear models, materials and methods for each regression algorithm should be reported. The paragraph should contain just the methods and not the considerations (that should be included in the discussion).*

Response: We are very grateful to the reviewer for these thoughtful suggestions. To enhance clarity, we have added a new table (revised Supplementary Table 5 on page 32 of the Supplementary Materials) that defines the regression models used in this study.

Furthermore, in the "Machine learning methods" subsection (on page 19 of the main text), we have refined the content by removing extraneous considerations (shown below). The revised section now focuses solely on describing the models, algorithms, and implementation details, ensuring a more concise and logical presentation.

"First, we employed an exploitation-only enumeration approach (GP_enu and RFR_enu). This approach involved feeding the trained model with a random sampling of 10 million ϕ_i values, generated as random floats within the interval [0.0, 1.0) from a continuous uniform distribution for each monomer proportion. The proportion vector was then normalized to ensure

the sum of the compositions equal to 1.0. We ranked the proportion vectors based on their predicted F_a and selected the top 5 from GP_enu and RFR_enu individually for the experimental validation. ~~This facile approach was deemed suboptimal due to its reliance on limited, biased training data, making it less effective for extrapolating new formulations inside a multi-dimensional global space (Fig. 4a). To tackle this problem, one has to strike a balance between exploitation and exploration, which involves intelligently selecting new data points with a high likelihood of surpassing the current maximum F_a.~~"

• *In the materials and methods experimental procedures on the characterization and validation should be introduced.*

Response: We thank the reviewer for this comment. In response to previous feedback and suggestions, we have strengthened the Methods section and Supplementary Materials by providing more detailed descriptions of the experimental procedures for characterization and validation. We hope these additions improve the clarity and reproducibility of our work.

F) Reference: appropriate credit to previous work: YES

Response: We thank the reviewer for this assessment.

G) Clarity and context: lucidity of abstract/summary, appropriateness of abstract, introduction and conclusions.
The abstract well reports the aim of the work. However, the conclusions of the abstract should be revised considering the obtained results limiting the discussion on the correlation between predicted data and reported obtained data.
The introduction is well reported, with a clear introduction on the needs the state of art (still very limited and uncovered).
The conclusion should be revised considering the data reported and introducing the existing challenges in the field, in particular in the prediction of chemical features and in their correlation with the final properties of the hydrogels. Even if the conclusion in the field of adhesive is well understandable, the potential application in the biomedical field is not

supported by data presented and discussed. This point should be modified in the light of the obtained experimental results.

Response: We are truly grateful to the reviewer for these insightful suggestions. In response, we have revised the Abstract to better highlight the inherent challenges faced in the field, as well as the key findings of our work. Moreover, we have thoroughly revised the Conclusion section following the reviewer's suggestion. The updated Abstract and Conclusion are now as follows:

"**Abstract**

Data-driven methodologies have revolutionized the discovery and prediction of hard materials with well-defined atomic structures, such as crystal structures and high-entropy alloys, by utilizing standardized datasets, accurately predicting properties, and efficiently exploring design space[1-3]. However, their application to soft materials remains challenging due to the inherently complex and multiscale structure–property relationships[4-6]. Here, we present a data-driven approach that integrates data mining, experimentation, and machine learning to develop high-performance adhesive hydrogels from scratch, tailored for demanding underwater environments. By leveraging protein databases, we devised a descriptor strategy to statistically replicate protein sequence patterns in polymer strands via ideal random copolymerization, enabling targeted hydrogel design and dataset construction. Using machine learning, we optimized hydrogel formulations from an initial dataset of 180 bioinspired hydrogels, achieving a remarkable improvement in adhesive strength, with a maximum value exceeding 1 MPa. These super-adhesive hydrogels hold immense potential across diverse applications, from biomedical engineering to deep-sea exploration, marking a significant advancement in the data-driven innovation for soft materials."

"**Conclusion**

In summary, we introduced a data-driven approach that integrates the extraction of valuable sequence information from proteins, scalable polymer synthesis, and iterative machine learning to address longstanding challenges in the de novo design and development of soft materials. This approach enabled the discovery of hydrogels with underwater adhesive strengths exceeding 1 MPa, representing an order-of-magnitude improvement over previous benchmarks. Beyond adhesive hydrogels, this data-driven design framework offers a systematic, scalable, end-to-end approach for developing a wide range of functional soft materials. However, challenges remain, primarily due to limitations in monomer diversity, polymer synthesis technologies for controlling monomer sequences to a scale suitable for

materials development, and dataset scalability. Overcoming these challenges will require expanding modular monomer libraries, advancing polymerization techniques, and developing physics-informed machine learning models capable of generalizing across sparse, multi-scale datasets."

Remarks on code availability: the code doesn't provide a README file with enough instructions for installing and running the application.

Response: We sincerely appreciate the reviewer's feedback on this aspect. We have carefully revised the README file and added the suggested instructions for installing and running the application. We hope this improves the clarity and ease of use for future users.

Referee #2 (Remarks to the Author):

Paper Summary

This paper introduces a data-driven method to create high-performance adhesive hydrogels. By mining >20,000 adhesive protein sequences, they developed a descriptor strategy to statistically mimic protein sequence patterns using ideal random copolymerization. Machine learning, particularly Gaussian Processes and Random Forests, fine-tuned the designs, achieving adhesive strengths over 1 MPa that outperform current products. These hydrogels adhere well to diverse range of surfaces in challenging environments.

Response: We sincerely appreciate the reviewer for the insightful evaluation of our work.

Abstract

Good summary of paper but needs the inclusion of specific data points/key results. Authors should substantiate "Using machine learning, we optimized hydrogel formulations even with a small dataset, achieving unprecedented adhesive performance." key results.

Response: We are grateful to the reviewer for this thoughtful suggestion. In response, we have revised the Abstract by incorporating data points and highlight key results, as shown below:

"**Abstract**

Data-driven methodologies have revolutionized the discovery and prediction of hard materials with well-defined atomic structures, such as crystal structures and high-entropy alloys, by utilizing standardized datasets, accurately predicting properties, and efficiently exploring design space[1-3]. However, their application to soft materials remains challenging due to the inherently complex and multiscale structure–property relationships[4-6]. Here, we present a data-driven approach that integrates data mining, experimentation, and machine learning to develop high-performance adhesive hydrogels from scratch, tailored for demanding underwater environments. By leveraging protein databases, we devised a descriptor strategy to statistically replicate protein sequence patterns in polymer strands via ideal random copolymerization, enabling targeted hydrogel design and dataset construction. Using machine learning, we optimized hydrogel formulations from an initial dataset of 180 bioinspired hydrogels, achieving a remarkable improvement in adhesive strength, with a maximum value exceeding 1 MPa. These super-adhesive hydrogels hold immense potential across diverse applications, from

biomedical engineering to deep-sea exploration, marking a significant advancement in the data-driven innovation for soft materials."

Introduction

The introduction is well-written and easy to follow, but it lacks critical context regarding previous research. It should provide a short discussion of similar DD/ML approaches used for designing hard materials, establishing the validity of this methodology. Readers shouldn't be expected to simply accept that applying such techniques to hard materials is straightforward or standard practice. Authors should clarify how these methods are well-established for hard materials and why they are more challenging for soft materials like hydrogels.

Additionally, the introduction should address whether ML-based strategies have been previously applied to adhesives, for example some work in dry adhesives (doi.org/10.1016/j.mechmat.2023.104778 and doi.org/10.1016/j.eml.2022.101695). While these studies may not align precisely with the focus here, they highlight the broader context of DD approaches in adhesive design, which warrants discussion. Even broader is the discussion of previous DD/ML approaches for designing hydrogels. Including a broader discussion would clearly position the current work as an advancement in the field. A comparison to existing studies would highlight the novelty of this framework and better demonstrate its contribution. Improving the introduction in this way will make the work more compelling and accessible.

Figure 1 is excellent.

Response: We sincerely thank the reviewer for these valuable comments and suggestions. We fully agree with the suggested flow and recognize the importance of acknowledging similar DD/ML approaches used for designing hard materials. In response, we have extended the discussion on this aspect in the Abstract (see above) and cited important works in the field, including "MatterGen" in *Nature* (2025) [doi.org:10.1038/s41586-025-08628-5], *Nature* **624**, 80-85 (2023), and *Science* **376**, eabn3103 (2022), as Refs. 1-3.

We are also grateful to the reviewer for providing references on dry adhesives, which indeed present very interesting work. Given the reference limits and relevance, we have cited the study on the design of directional adhesive pillars using deep learning-based optimization (doi.org/10.1016/j.mechmat.2023.104778) as Ref. 23, as a representative example of gecko-inspired dry adhesive studies.

Furthermore, we have highlighted the challenges in designing soft materials, extended the

background on adhesive hydrogel development, and emphasized the end-to-end nature of our data-driven approach, which remains rare in the field. The updated Introduction is provided below. We hope these revisions help make the work more compelling and accessible, as the reviewer envisioned.

"**Introduction**

Designing soft materials, such as gels and elastomers, is a complex task. It requires not only selecting suitable types and amounts of building blocks (e.g., monomers) but also determining their arrangement within the constituent polymer chains. This complexity leads to a gigantic design space with countless possible combinations. Moreover, soft materials exhibit intricate behaviors due to the interplay of weak molecular interactions and thermal motion, resulting in complex structure–property relationships across multiple time and length scales, with mesoscale structures playing an important role[7]. These challenges make it difficult to develop accurate predicative theories or computational models for materials properties, thus hindering the rational design of soft materials[8-10].

Traditionally, the development of new soft materials has relied on experimental trial and error, often guided by researchers' intuition and experience. To reduce experimental demands, data-driven strategies are becoming increasingly essential[11,12]. Emerging tools, such as data mining and machine learning, are transforming the field by advancing the analysis of complex behaviors, enhancing material property prediction, and driving theory and modeling development[5,13-16].

Effectively integrating these tools into an end-to-end data-driven design framework is crucial for accelerating soft material discovery. A critical first step is the creation of high-quality datasets, which is complicated by the vast number of potential material designs and limited experimental resources[17,18]. Adhesive hydrogels, for example, are a type of soft material widely sought for high-end applications, yet achieving instant, strong, and repeatable underwater adhesion remains a longstanding challenge[19,20]. Previous studies on this material have identified a plethora of monomer types, making it a daunting task to form a consistent dataset or forge a simple design principle for optimizing performance[19].

Biological soft tissues, as naturally evolved soft materials, exemplify complex structures tailored for specific functions[21]. Studying these structures can help reduce the design space for synthetic soft materials[22], such as gecko-inspired dry adhesives[23,24]. Particularly, adhesive proteins, found in diverse organisms (e.g., archaea, bacteria, eukaryotes, and viruses), enable adhesion in wet environments. Despite their diversity, these proteins share common sequence patterns that offer valuable insights into designing underwater adhesives[25]. However, the precise control of monomer sequences—crucial for replicating protein functions—remains a

significant challenge in synthesis."

Data mining

I really like your approach. It's robust, and your strategy as laid out is both logical and sound. The way you've mined the protein data and distilled it into functional classes strikes a great balance between simplifying the complexity and keeping the key chemical properties relevant to adhesion. Plus, the findings generated were then easily translated into synthetic hydrogels. Exclusion of glycine, alanine, and proline – is there a danger that the exclusion of such groups might have other deleterious effects on other properties? i.e. how did you rule out unforeseen consequences to say flexibility or packing density or stability etc.?

Response: We are truly grateful to the reviewer for this insightful feedback. We agree that every amino acid in proteins plays a critical role in their function. Small amino acids, such as glycine, alanine, and proline, are indeed known to contribute to flexibility, folding, and stability in proteins. Incorporating monomers with similar functional groups into synthetic hydrogels could potentially influence adhesion and other material properties.

In this study, we chose to exclude these small amino acids in order to simplify the system by reducing the number of monomer types (and thus the dimensions) for the hydrogel dataset creation. This decision was based on the assumption that small functional groups would likely have a lesser impact on adhesion compared to larger, more interaction-prone functional groups. However, we acknowledge that excluding these amino acids may influence the adhesion properties, and we appreciate the reviewer's suggestion. We will consider investigating the role of small functional group monomers in hydrogel adhesion and performance in future work, as this could provide valuable insights for optimizing hydrogel properties across a broader range of functionalities.

To address this point and enhance clarity, we have revised a paragraph in the "Data Mining of Adhesive Proteins for Hydrogel Design" section (in the 3rd paragraph on page 5 of the main text) as follows:

"To reduce the dimensionality of the variables, the 20 amino acids were classified into six classes based on their chemical properties: hydrophobic, nucleophilic, acidic, cationic, amide, and aromatic (Supplementary Fig. 4). The consensus sequences were then encoded into functional class sequences. For simplicity, we excluded glycine, alanine, and proline from the

hydrophobic class in the encoding, as their smaller sizes likely play a less significant role in interfacial contact and interactions compared to other amino acids[31].”

If we think beyond adhesion, how generalizable is this approach to other types of soft materials/hydrogels? Could your approach be adapted to target additional properties, such as mechanical strength, elasticity, responsiveness to stimuli, etc?
Figure 2 is also excellent.

Response: We thank the reviewer for these excellent questions. In our view, the introduced end-to-end approach could indeed be adapted to the development of other bioinspired hydrogels and functional soft materials. The machine learning tools presented in this work can also be readily applied to optimize material properties, as long as high-quality dataset are available.

In particular, random heteropolymers provide a versatile platform for achieving target functions related to intrinsically disordered proteins (IDPs), as demonstrated by the Xu group (e.g., *Science* **2018**, 359, 1239-1243 and *Nature* **2023**, 615, 251-258, Refs. 33 and 22 in the manuscript). Properties such as adhesion, coacervation, and protein stabilization are closely tied to IDPs, and our method could be extended to explore these functionalities further.

To address this point, we have briefly summarized our vision for this direction in the conclusion (please refer to our response below regarding the summary section).

Synthesis
A lovely and systematic approach. The methodology is logical, with the robust use of Monte Carlo simulations adding strong validation to the process. The one-pot free radical polymerisation and straightforward testing method are both efficient and repeatable, making them ideal for high-throughput screening and practical application.
You chose six functional monomers for logical reasons, but how many in total could have fit this criterion? Were there potentially others?

Response: We sincerely thank the reviewer for the insightful comment and thoughtful questions. While achieving ideal random copolymerization is challenging, we believe there are still many candidates within the monomer library that could satisfy this criterion. For instance, we have identified a range of cationic and hydrophobic monomers that can undergo ideal

copolymerization under specific conditions (*Nat. Commun.* **2019**, 10, 5127; *Sci. China Chem.* **2021**, 64, 1560; *Polym. Chem.* **2024**, 15, 2104). These findings indicate that the current set of six monomers is not exhaustive, and expanding the modular monomer library could further enhance the versatility of the approach. Future work could explore incorporating additional monomers to target a broader range of properties, while still maintaining synthetic feasibility.

Machine learning

This is the highlight of the paper. The use of ML, especially GP and RFR models with batched SMBO, is smart and effective. The workflow was both efficient and accurate. Your models were able to effectively identify new possibilities and impressively reveal key monomers as BA, PEA, and ATAC. Your approach will be much utilised.

Why do you think "Gaussian process (GP)30 194" and "random forest regression (RFR)31 195" were the most effective ML models? Why were the weakest ML models the weakest? Would this always be the case for 'soft materials'?

Response: We are truly grateful to the reviewer for the feedback and the thoughtful questions.

To evaluate model performance, we assessed both training and test errors using Root Mean Squared Error (RMSE). An ideal model should exhibit low test error while avoiding excessive overfitting, which would be indicated by a significant gap between training and test errors.

As shown in Extended Data Figure 4, Gaussian Process (GP) and Random Forest Regression (RFR) demonstrated the lowest test errors, highlighting their strong generalization to unseen data. Although XGBoost (XGB) showed the lowest training error, the substantial gap between training and test errors suggests significant overfitting, limiting its predictive power. Based on these observations, GP and RFR emerged as the most effective machine learning models for this study.

This effectiveness can be attributed to the fact that GP and RFR are non-parametric models, which makes them well-suited to handling complex, non-linear interactions between features. This is particularly important in the discovery of soft materials, where structure-property relationships are often intricate and highly non-linear. GP is particularly effective when the dimensionality of the explored space is moderate, as was the case in this study with a six-dimensional dataset. On the other hand, XGB tends to perform better with high-dimensional datasets, which made it less suitable for the lower-dimensional data used in this work.

Lasso and Ridge regression performed poorly due to their assumption of linear relationships between input variables and target properties. However, soft material properties typically exhibit non-linear and multi-scale effects, rendering these models insufficient for capturing the underlying complexity. The high training and test errors for both Lasso and Ridge regression indicate underfitting, where the models failed to capture meaningful patterns from the data.

While GP and RFR were the most effective models in this study, we acknowledge that they may not always be the best choices for soft materials discovery. The optimal ML model depends on factors such as dataset dimensionality, noise levels, and computational constraints. For significantly higher-dimensional datasets, GP may face challenges due to its computational complexity, and tree-based methods like XGB or deep learning models might be more appropriate. In cases with substantial noise or sparse measurements, ensemble methods like RFR or boosting models such as XGB may outperform GP due to their robustness to noisy data. Additionally, given that GP scales poorly with increasing data points, models like Gradient Boosting Machines (GBM) could offer a more computationally efficient alternative for larger datasets.

To enhance clarity, we have revised the 1st paragraph of the "Machine learning for hydrogel adhesion optimization" section (on page 9) in the main text, as well as the figure caption of Extended Data Fig. 4, to briefly incorporate these considerations. The revised texts are as follows:

"Next, we employed machine learning to explore hydrogel formulations with higher adhesive strength, starting with the 180-hydrogel dataset. By benchmarking nine ML models (Supplementary Tables 5 and 6), Gaussian process (GP)[37] and random forest regression (RFR)[38] emerged as the most effective base models for predicting F_a from ϕ_i, as they achieved low test error while minimizing overfitting (Extended Data Fig. 4)."

"**Extended Data Fig. 4. Machine learning (ML) trained models.** (a) Error plots showing the 90%/10% training-test split for nine ML models. Training data points are represented by blue dots, while test data points are shown in red. The dashed line indicates where the predicted values match the experimental data (truth). (b) Root mean squared errors (RMSEs) depicting the prediction accuracy across the nine ML models trained on the dataset of 180 bioinspired hydrogels, assessed via 10-fold cross-validation. A lower test error, combined with minimized overfitting (i.e., a smaller gap between training and test errors), indicates a more effective regression model."

Performance

Your performance testing was robust, and the results are impressive. You demonstrated the hydrogels' versatility across different water types with multiple testing methods providing a thorough evaluation of adhesive performance. While the rubber duck demonstration is playful, it may indeed illustrate real-world conditions. However, the biocompatibility testing feels out of place here and could be omitted, allowing it to be expanded upon in a separate, more focused publication. Keeping the focus solely on adhesion performance would strengthen the manuscript, as the results are compelling enough to stand alone.

Response: We sincerely appreciate the reviewer for the insightful comments and suggestions.

A related suggestive comment regarding potential biomedical applications was also raised by Reviewer 1, and we agree that these applications represent an important potential outcome for adhesive hydrogels. In this manuscript, we included a preliminary in vivo experiment to assess the biocompatibility of our hydrogel samples, as this is a critical factor for their potential biomedical applications. We acknowledge that focusing primarily on adhesion performance would strengthen the manuscript, particularly given the compelling findings. As such, we plan to expand the biomedical aspects in future work to provide a more comprehensive exploration. We are grateful for your input on this matter.

To further address this point, we have simplified the related discussion in the 2nd paragraph on page 13 of the main text, as follows:

"Furthermore, all these hydrogels exhibited good biocompatibility, as confirmed by subcutaneous implantation beneath the dorsal skin of mice (Extended Data Fig. 10), suggesting potential applicability in the biomedical field. ~~This supports their applicability in biomedical field, such as wound dressing and bone tissue engineering.~~"

Section could benefit from some proper quantitative testing for the marine environment or burst pipe testing. The videos and demonstration are ideal to reach a wider audience but would have benefitted to have some hard data to support the rubber duck marine test and pipe burst test.

Response: We thank the reviewer for these thoughtful suggestions. In response, we have calculated the burst flow rate of the water leakage at the outlet of the hole, which is approximately 5.4 m/s, and included this data in the revised caption of Fig. 5 as follows.

"(e) Photographic images of R2-max (6 cm × 6 cm in size, ~0.37 mm thickness) sealing a 20 mm hole at the base of a 3-meter-tall PC pipe to halt high-pressure water leakage (burst flow rate at the outlet of hole was ~5.4 m s^{-1})."

Moreover, given the challenges of mimicking complex marine environmental conditions, we performed systematic adhesion tests in simplified artificial seawater (0.7 M NaCl) instead. The results are provided in Fig. 5c and Supplementary Fig. 17. We hope these data provide useful insights for designing real-world applications.

Summary

The last paragraph is weak Lines 324-330. Please revisit and think about the true potential of these super adhesive hydrogels, particularly for extreme environments.

Response: We are truly grateful to the reviewer for this valuable suggestion. We are excited about the super-adhesive properties of our newly developed hydrogels, and as a novel type of adhesive material, we anticipate they could have broad applications. However, given the complexity of developing real-world products and the challenges of conducting experiments under extreme environments, we were only able to conduct prototypical examinations in our lab. We hope that these preliminary findings will provide useful insights for potential applications.

To address this point, we have thoroughly revised the Conclusion section (shown below) to provide a clearer summary of our key findings, outline the remaining challenges, and offer a brief outlook on future developments. We hope this revision more effectively captures the essence of our work and its potential impact moving forward.

"In summary, we introduced a data-driven approach that integrates the extraction of valuable sequence information from proteins, scalable polymer synthesis, and iterative machine learning to address longstanding challenges in the de novo design and development of soft materials. This approach enabled the discovery of hydrogels with underwater adhesive strengths exceeding 1 MPa, representing an order-of-magnitude improvement over previous benchmarks. Beyond adhesive hydrogels, this data-driven design framework offers a systematic, scalable, end-to-end approach for developing a wide range of functional soft materials. However, challenges remain, primarily due to limitations in monomer diversity,

polymer synthesis technologies for controlling monomer sequences to a scale suitable for materials development, and dataset scalability. Overcoming these challenges will require expanding modular monomer libraries, advancing polymerization techniques, and developing physics-informed machine learning models capable of generalizing across sparse, multi-scale datasets."

Overall, this is an excellent paper that showcases an innovative data-driven approach to developing super-adhesive hydrogels. The authors are to be commended for their robust methodology, logical design, and comprehensive testing. The integration of ML is particularly impressive. Your high-performance hydrogels have real-world potential and your approach will benefit many researchers in the field. Minor revisions, including refining the introduction, potentially adding quantitative data for marine and pipe bursting demo, and revisiting the summary.

Originality and significance: if not novel, please include reference
Yes. Original and significant. The authors have developed a methodology for the robust development of hydrogels that can be greatly utilised by many in the field.

Data & methodology: validity of approach, quality of data, quality of presentation
Excellent. High-quality presentation of the data in all figures. Valid approach. Logical and robust.

Appropriate use of statistics and treatment of uncertainties
yes.

Conclusions: robustness, validity, reliability
Room for improvement in the final summary of the paper.

Suggested improvements: experiments, data for possible revision
Stated above.

References: appropriate credit to previous work?
Could do with more thoughtful summation of previous work in the DD/ML in materials

development—hard materials and other soft materials (hydrogels)

Clarity and context: lucidity of abstract/summary, appropriateness of abstract, introduction and conclusions
Abstract and conclusion need some work. It is a very well-written paper and easily understood.

Response: We sincerely thank the reviewer for the overall positive evaluation of our work and for the insightful suggestions. In response, we have made revisions as indicated in our responses above, which we believe significantly improve the quality of the manuscript.

Referee #3 (Remarks to the Author):

The manuscript by Takigawa, Li, Fan and Gong describes a concerted effort to develop high performance underwater adhesive by combining data mining, experiments and machine learning. The study started with analyzing protein sequence information upon reducing to 6 monomer types, followed with experimental inputs based on large library gel synthesis and characterization, and ended with machine learning to optimize systems with three monomer type. The resultant adhesives demonstrated quite impressive performance with potential technologically relevant applications. Present studies also accumulated quite large datasets and provided good experimental inputs, a key bottleneck for AI/ML in material development.

Response: We are very grateful to the reviewer for the thoughtful comments and positive evaluation of our work.

I have serious concerns regarding the connection made between what's made vs. protein sequence analysis. The gels were synthesized in the presence of crosslinker and at high monomer concentration (>2M total monomer) using UV initiator. The polymerization process is not living and is diffusion limited with high viscosity. It is reasonable to expect significant heterogeneity and to question the fidelity of sequence simulation based on Monte Carlo simulation to represent what the polymer strains in the final network may look like. Admittedly, synthesizing networks is difficult to characterize. However, this is the corner stone of the system design and selection, e.g. results shown in Figure 2. Control experiments need to be done to quantify to what level the reactions and sequence simulations share similarities and identify the differences. Given the gel formation, the crosslinking density and the polymer strain length are directly coupled, and adhesive performances depend on so many factors. The readers and community should be made aware of potential hidden caveat. Maybe it was just used as proxy or a convenient way to extract some information and perhaps there are some merits to predict short blocks simply by the system diversity. I do not see systematic characterization of gels or studies of adhesion mechanism. More in-depth understanding/characterization of the system are important part of work to bridge AI/ML with specific scientific topics.

Response: We sincerely thank the reviewer for the insightful comments and valuable

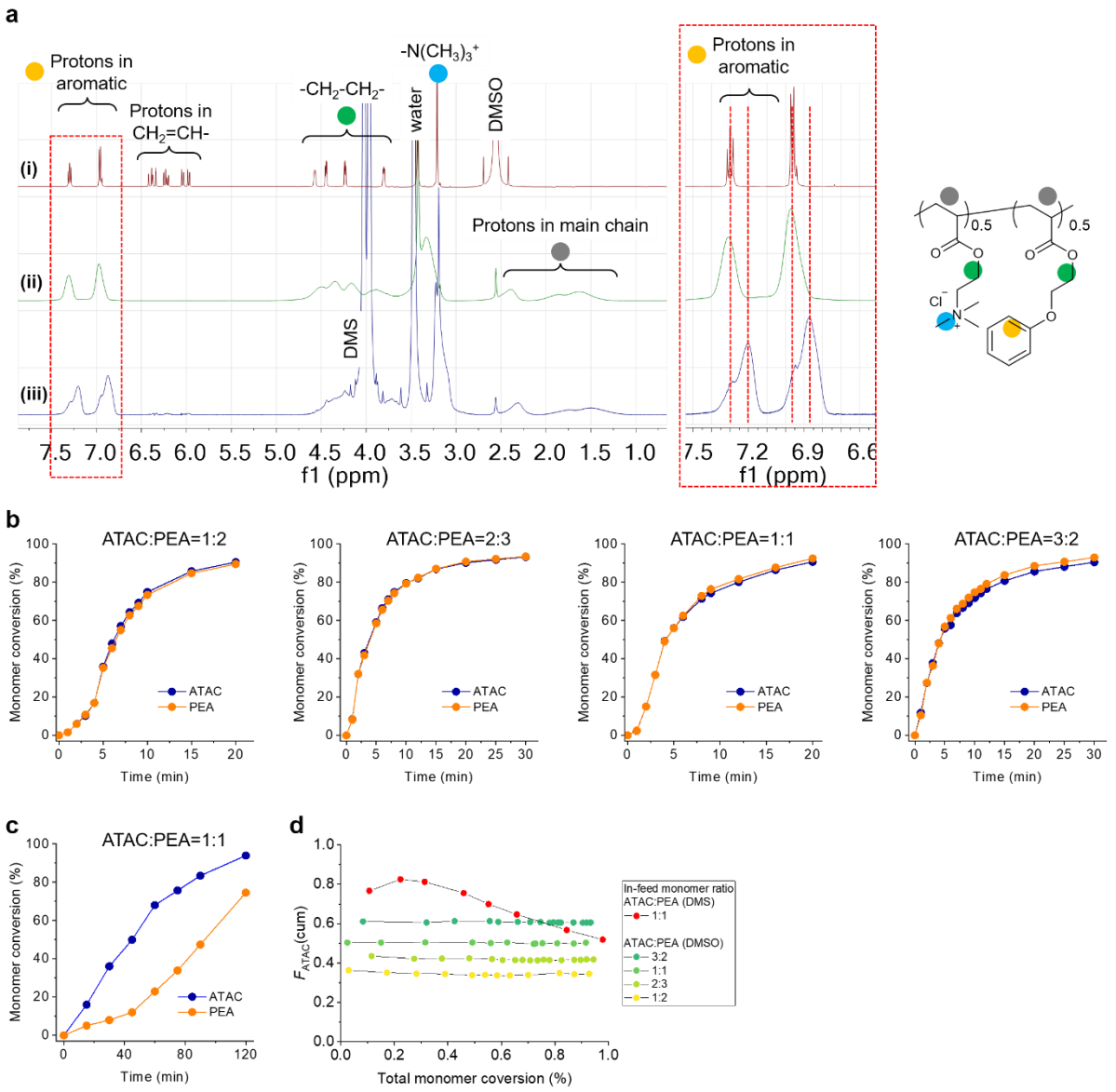
suggestions, which have prompted us to carefully reflect on aspects of our study that require further clarification. This has helped us ensure a balanced and rigorous interpretation of the results.

First, as the reviewer pointed out, composition drift is a common occurrence in free radical polymerization due to differences in monomer reactivity ratios. This process, along with crosslinking and gel formation, can lead to heterogeneity in copolymer sequences and network structures, raising concerns about the fidelity of Monte Carlo simulations in depicting sequence features of synthesized gels. We appreciate the reviewer's input and in fact, had already considered this aspect when designing our experiments.

To mitigate composition drift, we carefully selected monomers capable of undergoing ideal azeotropic copolymerization in their common good solvent (DMSO). For example, we examined the monomer conversions of ATAC and PEA, which exhibit significant hydrophobicity differences, at different molar ratios but a fixed total monomer concentration of 1 M. We confirmed that both monomers maintained the same conversion ratios throughout polymerization, demonstrating minimal composition drift (revised Supplementary Fig. 6, shown below).

For all formulations, both the initial precursor solutions and the final gels exhibited high transparency, suggesting that DMSO is a good solvent for the selected monomer systems. Additionally, we confirmed the high reproducibility in hydrogel structure formation, which leads to reproducible properties including swelling, rheological, and adhesive behaviors.

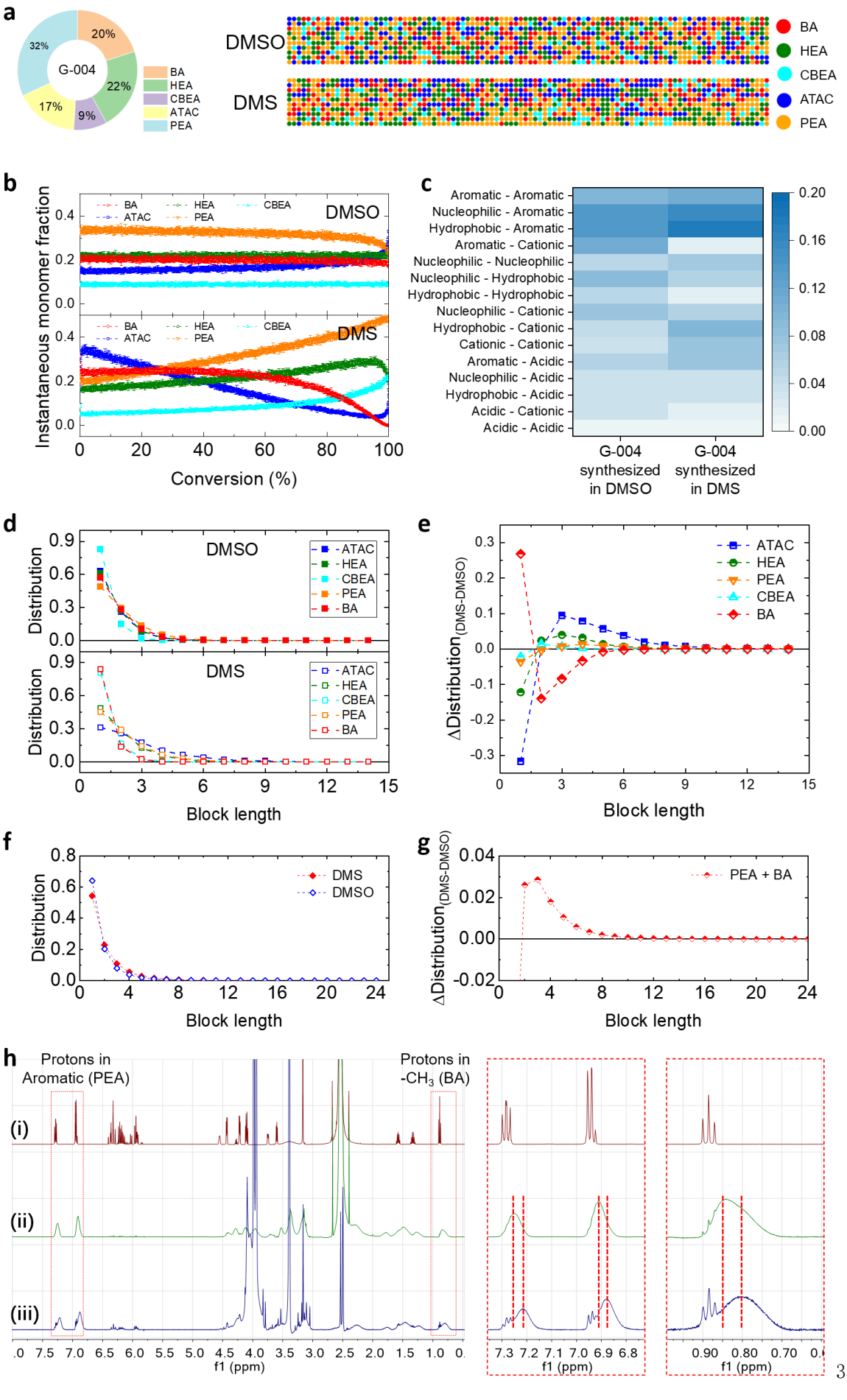
Based on these findings, we believe the monomer distribution remains stable during polymerization, which can be reliably described using the well-established Mayo−Lewis equation. As a result, Monte Carlo simulations based on the Mayo−Lewis model provide meaningful insights into sequence characteristics, which would otherwise be challenging to obtain due to the lack of suitable sequence identification techniques for hydrogels (as the reviewer also noted).



Supplementary Fig. 6. ^{1}H NMR studies on the time evolution of compositions during the copolymerization of cationic ATAC and aromatic PEA in different solvents. (a) ^{1}H NMR spectra of ATAC and PEA monomer precursor solutions at a 1:1 molar ratio (i) and the corresponding copolymers after full polymerization in DMSO (ii) and in DMS (iii). The portion highlighted by a red dashed frame in the full NMR spectra is enlarged on the right. For the copolymer synthesized in DMSO, the phenyl proton signals exhibit symmetric broadening around the peak of the phenyl protons of the aromatic monomers, indicating a statistically dispersed arrangement of cationic and aromatic monomers along the polymer chains. In contrast, for the copolymer synthesized in DMS, the phenyl proton signals show new, strong peaks at higher fields, suggesting the formation of long aromatic monomer sequences[5]. (b, c) Monomer conversion kinetics of ATAC and PEA at different feed molar ratios in DMSO (b) and at a 1:1 feed molar ratio in DMS (c). (d) Cumulative ATAC molar fraction in copolymers (F_{ATAC}) plotted against total monomer conversion for varying feed molar ratios in different solvents. In

DMSO, the copolymers maintained nearly the same monomer composition as the feed throughout polymerization, with no significant composition drift, indicating quasi-ideal random copolymerization. In contrast, in DMS at a 1:1 feed molar ratio, the ATAC fraction deviated significantly from 0.5 and varied with monomer conversion, demonstrating distinct composition drift during polymerization. In all NMR experiments, the total monomer concentration was 1.0 M.

Second, our sequence simulations show good agreement with experimental findings. For example, when comparing two variants of G-004 synthesized in DMSO and DMS, Monte Carlo simulations predicted that from DMSO to DMS, the block length of PEA slightly increases while the block length of BA decreases. However, the segments composed of both PEA and BA exhibited increasing lengths, leading to stronger associations of hydrophobic contents. This was confirmed by NMR analysis and further supported by the fact that the corresponding hydrogel displayed an opaque appearance. These findings are now presented in the newly added Supplementary Fig. 7 (see below).


a
G-004
20%
22%
9%
17%
32%
BA
HEA
CBEA
ATAC
PEA
DMSO
DMS
b
Instantaneous monomer fraction
Conversion (%)
c
Aromatic - Aromatic
Nucleophilic - Aromatic
Hydrophobic - Aromatic
Aromatic - Cationic
Nucleophilic - Nucleophilic
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Hydrophobic - Hydrophobic
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Acidic - Acidic
G-004 synthesized in DMSO
G-004 synthesized in DMS
d
Distribution
Block length
e
ΔDistribution(DMS-DMSO)
Block length
f
Distribution
Block length
g
ΔDistribution(DMS-DMSO)
PEA + BA
Block length
h
Protons in Aromatic (PEA)
Protons in -CH3 (BA)
(i)
(ii)
(iii)
f1 (ppm)

Supplementary Fig. 7. Monte Carlo simulations (a-g) and ^{1}H NMR analysis (h) of monomer sequences for G-004 prepared in DMSO and DMS. (a) G-004 formulation and its simulated chain sequences in DMSO and DMS, based on reactivity ratios from Supplementary Table 3. Each chain consists of 1,000 monomers, presented in 10 rows, with each row showing the sequence of 100 monomers from left to right. (b) Instantaneous monomer fractions as a function of total conversion during polymerizations in DMSO and DMS. Each simulation involved 2 million monomers, with an average chain length of 1,000. The results show minor composition drift for polymerization in DMSO, but significant composition drift in DMS throughout the polymerization. (c) Pairwise frequency distribution of 15 distinct functional class pairs along encoded sequences. (d) Block length distributions for different monomers from the simulated sequences. (e) Differences in monomer block length distributions between copolymers prepared in DMS and DMSO, highlighting increased block lengths for ATAC, HEA, and PEA. (f) Block length distributions for combined aromatic PEA and hydrophobic BA in DMSO and DMS. (g) Difference between the two distribution curves in (f), suggesting longer segments of combined BA and PEA in DMS. Statistical results in (b-g) were obtained from five independent runs. (h) ^{1}H NMR spectra of G-004 monomer precursor solution (i) and copolymers synthesized in DMSO (ii) and in DMS (iii) after 8 hours of polymerization (conversion > 99% in DMSO and > 93% in DMS). The portions of the NMR spectra highlighted by red dashed frames are enlarged on the right. For PEA aromatic protons, a high-field shift in the peak indicates an increase in PEA block size. For BA methyl protons, a high-field shift suggests a higher frequency of adjacent BA-PEA units, corresponding to larger segments of combined BA and PEA monomers. These findings are consistent with the simulation results shown in (f, g). Overall, the simulations suggest that hydrogels prepared in DMSO maintain quasi-ideal random copolymerization, resulting in statistical monomer sequences that determine the hydrogel's adhesion properties. In contrast, hydrogels prepared in DMS exhibit blocky sequences of combined BA and PEA (both hydrophobic), which lead to aggregation of hydrophobic components and the opaque appearance (cf. Fig. 3f in the main text). Meanwhile, the increase in ATAC block length negatively impacts adhesion[1].

Third, as the reviewer highlighted, both chemical structure and topological features (e.g., polymer chain length and network connectivity) influence adhesive performance. To reduce complexity, we focused on polymer chemistry while maintaining consistent polymer chain length and network connectivity by synthesizing gels with a fixed crosslinker density (0.1 mol%) at a total monomer concentration of 2.4 M for dataset generation and ML validation.

To further examine network structure effects, we fabricated the top-performing formulations identified through data mining and machine learning—G-max and R1-max—at

varying crosslinker densities (0.05–0.5 mol%). Figure R1 compares the adhesion strength (F_a) and debonding work, showing a decreasing trend in adhesion as crosslinker density increases. This can be attributed to reduced gel stretchability at higher crosslinker densities, which limits energy dissipation during debonding.

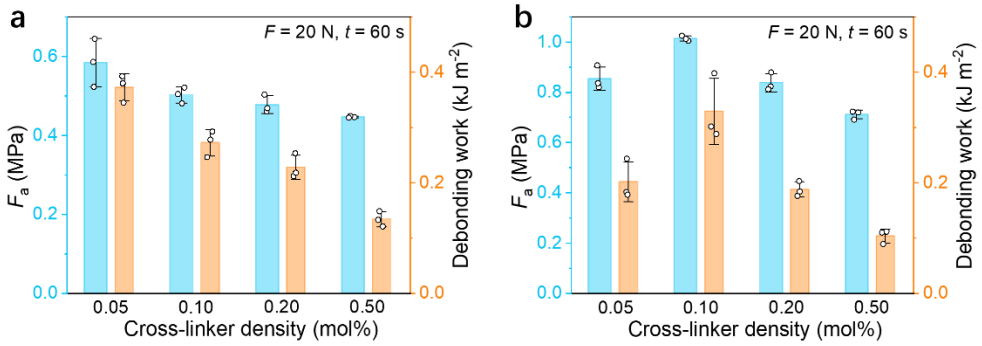
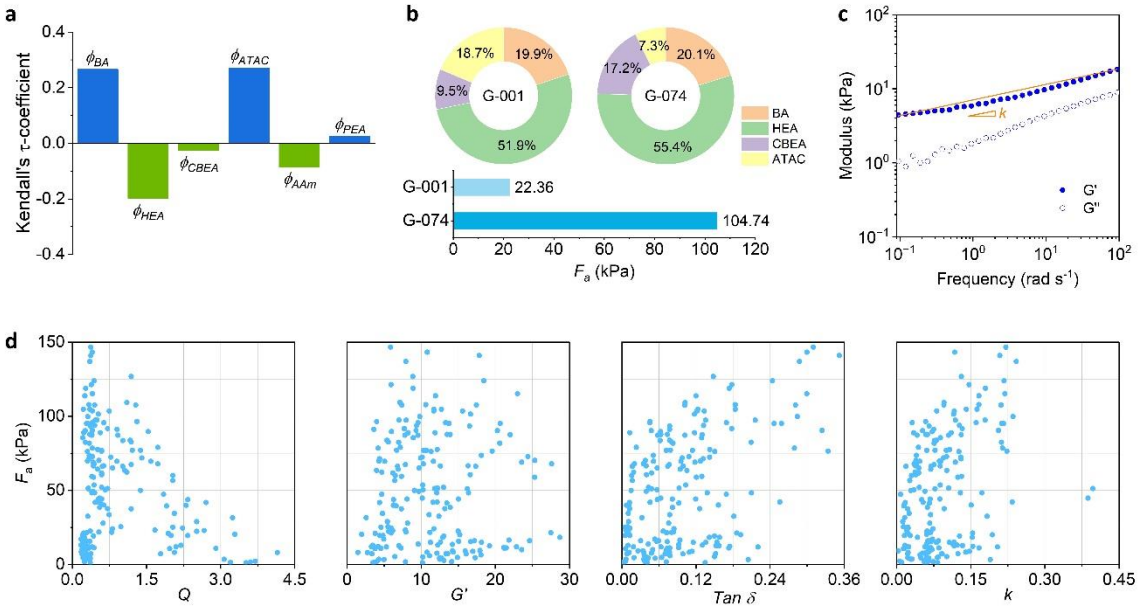


Figure R1. Comparison of adhesion strength (F_a) and debonding work for (a) G-max and (b) R1-max hydrogels with different crosslinking densities.

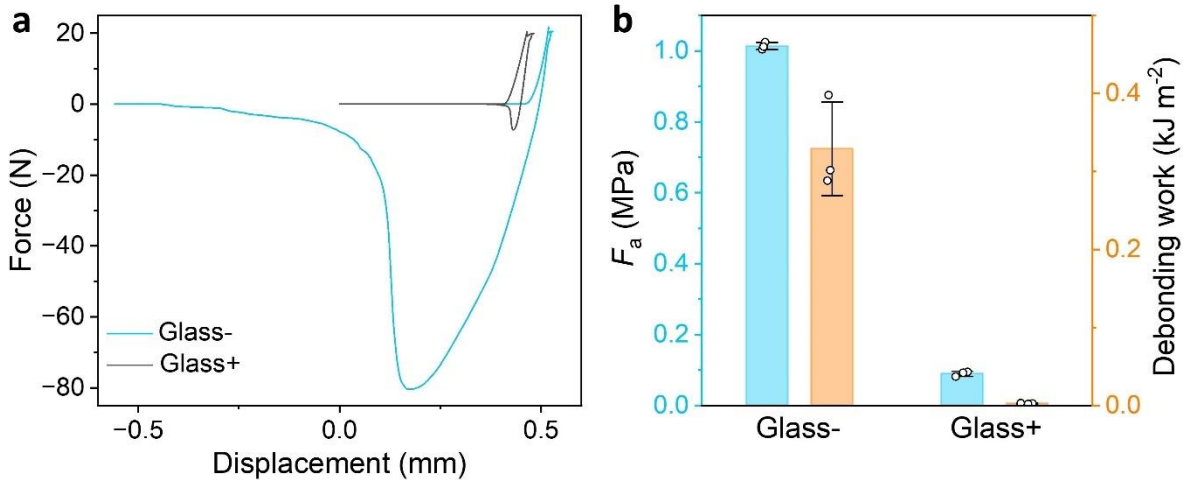
Fourth, we conducted a systematic characterization of the gels. Correlations between F_a and gel swelling/rheological behaviors are now presented in Extended Data Fig. 3 (shown below), with additional results summarized in Supplementary Tables 2, 7, and 8. Specifically, Extended Data Fig. 3 highlights the complex structure-property relationships regarding hydrogel adhesion.



Extended Data Fig. 3. Analysis of the correlations between adhesive strength (F_a) and

properties of 180 bioinspired hydrogels. (a) Correlation between monomer proportions (ϕ_i) and F_a, captured by Kendall's τ coefficients. These coefficients reveal that ϕ_{BA}, ϕ_{ATAC}, and ϕ_{PEA} have weak positive correlations with F_a, while ϕ_{HEA}, ϕ_{AAm}, and ϕ_{CBEA} show weak negative correlations. (b) Example illustrating the complex interplay between ϕ_i and F_a, due to the synergistic effects of different monomers. When comparing G-074 to G-001, ϕ_{BA} is roughly the same, ϕ_{HEA} and ϕ_{CBEA} increase, and ϕ_{ATAC} decreases. Despite the Kendall's τ coefficients suggesting that the F_a of G-074 should be lower than that of G-001, the actual F_a of G-074 is about five times higher. (c) Angular frequency dependence of the storage modulus (*G′*) and loss modulus (*G″*) of the G-042 hydrogel as a demonstration. The slope (*k*) of *G′* is calculated from the line connecting *G′* values at frequencies of 0.1 and 100 rad s^{-1}. A larger slope indicates higher viscoelasticity of the gel. (d) F_a as a function of network properties for the 180 bioinspired hydrogels. *Q*, *G′*, *Tan* δ = *G″*/*G′*, and *k* represent the volume swelling ratio, storage modulus at a frequency of 10^0 rad s^{-1}, loss factor at a frequency of 10^0 rad s^{-1}, and the slope of the *G′* curve, respectively. These results suggest that gels exhibiting shrinking behavior (*Q* < 1), moderate *G′*, large *Tan* δ, and moderate *k* tend to have higher F_a values. All characterizations were performed on gels equilibrated in normal saline (0.154 M NaCl). Adhesion tests were conducted using a tack test with a 10 N loading force and a 10-second contact time on a glass substrate to enable rapid screening and ensure consistent comparisons across the dataset.

Fifth, to further elucidate the adhesion mechanism, we conducted adhesion tests on glass with a positively charged surface. The newly added Supplementary Fig. 12 (shown below) demonstrates that R1-max exhibits poor adhesion on positively charged glass, indicating that its strong and repeatable adhesion is driven by enhanced electrostatic interactions between ATAC and the negatively charged surface.



Supplementary Fig. 12. Effect of substrate surface charge properties on the adhesion of the R1-max hydrogel. (a) Force – displacement curves from the tack tests for R1-max (10 mm diameter, ~0.4 mm thickness) on glass substrates with negatively and positively charged surfaces in normal saline (0.154 M NaCl), conducted under a 20 N loading force with 60-second contact time. (b) Extracted adhesive strength (F_a) and debonding work.

Finally, given the complexity of structure-property relationships discussed above, we emphasize that maintaining data consistency and capturing latent correlations are critical to ensuring the effectiveness of our end-to-end data-driven approach.

To further address the reviewer's comments, we have incorporated the following revisions throughout the manuscript, in addition to presenting the newly added Extended Data and Supplementary Figures.

1. In the first two paragraphs on page 7 of the main text:

"Six functional monomers (Fig. 3a) were selected based on their pairwise reactivity ratios, which are close to 1 when copolymerized in the cosolvent dimethyl sulfoxide (DMSO) via free radical polymerization, as confirmed by ^{1}H NMR analysis (Supplementary Fig. 5 and Supplementary Table 3). Due to these near-unity reactivity ratios, no significant composition drift occurred during copolymerization in DMSO (Supplementary Figs. 6 and 7)."

"Monte Carlo simulations based on the Mayo–Lewis model were performed to analyze the sequence properties of the six functional monomers in the corresponding 180 heteropolymers, using the measured reactivity ratios (Supplementary Table 3) and the derived formulations of monomer proportions (ϕ_i) (Supplementary Table 2)[35]."

2. In the 4th paragraph on page 8 of the main text:

"To improve F_a, the correlations between F_a and ϕ_i were assessed using Kendall's τ coefficients[36], and the dependence of F_a on hydrogels' swelling and rheological behaviors was also characterized (Extended data Fig. 3). We found that ϕ_{ATAC}, ϕ_{BA}, and ϕ_{PEA} have weak positive correlations with F_a, while ϕ_{HEA}, ϕ_{AAm}, and ϕ_{CBEA} show weak negative correlations. Nevertheless, these weak correlations, along with the intricate structure-property relationships (Extended data Fig. 3), cannot be directly used to predict hydrogel formulations for optimal adhesion, highlighting the complex synergistic effects of monomer species, sequences, and network structures."

Another question is regarding the logic behind 6 monomer type selected for protein sequence analysis vs. 3 monomer type in the final best performer. As reported earlier (Nature, 615, 251, 2023), 4 types of monomers were used to reduce protein sequences and the authors showed that increasing monomer type can significantly increase sequence space. With 6 monomers, the system flexibility associated with expanded sequence space may make many things possible. I am curious why the team did not loop back and see how the 3 monomer type-based sequence analysis looks like.

Response: We are very grateful to the reviewer for this insightful suggestion and for providing the reference on the four-component heteropolymer strategy for mimicking proteins (now cited as Ref. 22).

In this study, we selected six functional monomers to represent corresponding functional classes of amino acids, capturing key functional group diversity relevant to adhesion. Notably, the best-performing gels identified through machine learning utilized only three of these monomers, even though the bioinspired training set did not contain such three-component hydrogels. We emphasize that this outcome reflects an optimization under specific conditions, such as adhesion to glass in normal saline. If the target substrate or environment changes, the optimal functional monomer types and formulations are also likely to change.

In response to the reviewer's suggestion, we have conducted sequence analysis of the three-component gels and incorporated the results and related discussions in Supplementary Figs. 11 and 14. For further details, please refer to our responses to points 7 and 9 in the specific comments below.

The team should also consider the possibility that the sequence coupling may not be the true origin and there are layered coupling among various factors. As is now, I got the impression that DM and ML can work like black magic. Is this true for basic material science, or it is just coincidental that the process worked, or we had wishful thinking to enforce that connection? At the age of AL, the scientific community needs to be guided/educated with all aspects possible, good and bad. IMO, it is more valuable to tell a layered, not so successful story than a perfect one.

Response: We deeply appreciate the reviewer for sharing this perspective. We acknowledge that sequence coupling is an important factor to consider, alongside other key contributors to hydrogel adhesion performance. Moreover, we fully agree on the importance of transparency in presenting both the successes and limitations of data-driven approaches in materials science. While AI and ML are powerful tools, they are not panaceas. Their effectiveness in developing hard materials has been demonstrated, but their success in soft materials design should not be taken for granted due to the inherent complexity arising from multiscale structures and interactions.

As discussed above, hydrogels, as representative soft materials, exhibit intricate structure-property relationships driven by complex interplay among various factors. This complexity poses significant challenges to rational design. Data-driven approaches offer a means to efficiently explore the vast design space and identify promising formulations by capturing high-dimensional correlations—patterns that would be difficult to discern through traditional trial-and-error methods based on experience and intuition.

The success of data-driven approaches, however, relies on high-quality data. In our case, given the time-intensive nature of experimental procedures, we established a hydrogel dataset of a manageable size from scratch. Biomimicry provided valuable design insights, allowing us to construct a dataset of 180 bioinspired hydrogels by extracting characteristic sequences from adhesive proteins. This small yet highly consistent dataset proved to be well-suited for machine learning, enabling further optimization beyond the initial bioinspired formulations. We believe this outcome provides valuable guidance for future studies in soft materials design.

Below are some specific comments:

1. The authors compile the protein dataset based on a key-word search from the NCBI database.

Often key-word searches are not very reliable, and the results need to be cleaned systematically. Any data cleaning procedures should be listed in the SI, if they were conducted.

Response: We thank the reviewer for this comment. In our study, to ensure inclusiveness of data, no data cleaning procedures were applied. However, we implemented methods such as ranking species based on the number of adhesive proteins they contain and applying multiple sequence alignment (MSA) to extract representative characteristic sequences. These procedures ensure statistical significance.

To clarify this point, we have revised the relevant sentence in the "Data Mining of Adhesive Proteins" subsection on page 17 of the main text as follows:

"A total of 24,707 protein sequences were gathered from 3,822 different organisms, including bacteria, viruses, eukaryotes, and animals, without further data cleaning."

2. The authors categorize the protein sequences based on species, then use MSA to select one consensus sequence per species to perform analysis on. The rationale for this approach is not well explained. With explanation of the differences in protein properties and performance across different species, this categorization could be more insightful. Further, the sequence analysis methods chosen are simple enough to be done on the whole protein dataset, without introducing the uncertainty of selecting a single sequence to represent thousands of sequences. It seems that the sequence analysis would yield more statistically robust results without this MSA approach.

Response: We thank the reviewer for these thoughtful comments. The rationale for categorizing protein sequences by species and applying multiple sequence alignment (MSA) to extract consensus sequences was to identify common structural motifs while reducing noise from individual sequence variations. Since adhesive proteins exhibit evolutionary adaptations specific to each species, analyzing them collectively without categorization could obscure functionally relevant patterns. By grouping sequences by species, we aimed to highlight conserved sequence features that may contribute to adhesion across different biological contexts.

We agree that applying sequence analysis to the entire dataset without MSA could provide a broader statistical perspective. However, adhesive proteins often exhibit high sequence

variability within a species, and treating each sequence individually may introduce excessive variability, making it difficult to extract meaningful trends. MSA helps filter out mutations and individual sequence deviations, ensuring that the most conserved and functionally relevant segments are retained. This approach mitigates uncertainties associated with sequence variations and provides a more biologically relevant representation of each species' adhesive mechanism.

To clarify this point, we have revised the relevant sentence in the 2nd paragraph on page 5 of the main text as follows:

"To identify the most representative protein sequences and minimize the impact of individual variations, we ranked all species by the number of adhesive proteins they contain and selected the top 200 species for further analysis (Fig. 2a and Supplementary Fig. 3). We then performed multiple sequence alignment using Clustal Omega[28] to determine consensus sequences for each species (e.g., Extended Data Fig. 1), which are believed to play a crucial role in maintaining protein stability and adhesion throughout evolution[29,30]."

3. Similarly, the rationale behind this pairwise frequency approach in figures 2 and 3 should be explained. It is quite a simplified sequence analysis method and thus should be justified.

Response: We thank the reviewer for this comment. After extracting characteristic sequences, we needed to develop a strategy to translate them into hydrogel design. Given the inherent heterogeneity of the sequences, even at the coarse functional class level, ideal random copolymerization was chosen as a suitable approach to enforce monomer heterogeneity. However, this method provides limited control over precise monomer sequencing. While directly using monomer compositions extracted from the sequences as a 0th-order descriptor for hydrogel formulations was an initial consideration, we found that simply disregarding all sequence features would be insufficient.

Pairwise frequency analysis is indeed a fundamental sequence analysis method. From our previous study on two-component hydrogels composed of PEA and ATAC (*Nat. Commun.* **2019**, *10*, 5127), we observed that sequences with a higher occurrence of heterogeneous PEA–ATAC pairs exhibited stronger adhesion. Based on this insight, we hypothesized that incorporating relative compositions derived from pairwise frequency analysis would be a more effective strategy. Admittedly, this choice was guided by our prior experience and intuition, but the

results in Figs. 2 and 3 strongly support its effectiveness.

To clarify this point, we have revised the 5th and 6th paragraphs on page 5 of the main text as follows:

"Based on these insights, we devised a strategy for hydrogel design using six functional monomers to represent six functional classes of amino acids. While directly replicating functional class sequences offers a straightforward way to mimic protein primary structures and functions, achieving precise control over monomer sequences in synthetic polymers remains a significant challenge. Therefore, we aimed to statistically replicate the sequence features of functional classes through ideal random copolymerization of the six functional monomers, which has minimal composition drift during polymerization and enables statistically controlled sequences[22,32-34].

For this purpose, we employed a relative composition approach to capture the neighboring preferences of amino acid functional classes within the synthetic polymer chains. Specifically, we counted the occurrences of 21 distinct pair types for the six functional classes of amino acids, denoted as n_{ij} (where *i*, *j* = 1, …, 6), along the functional class sequences for each species and ranked them in descending order. The top five pairs, collectively accounting for approximately 50% of all occurrences, were used to compute the monomer proportions of each functional class as $\phi_i = N_i / \sum_i N_i$, where $N_i = \sum_j (n_{ij} + n_{ji})$ for each species (Extended Data Fig. 1, Supplementary data 1 and 2). These relative compositions served as descriptors for the corresponding species. From the top 200 species, we derived 180 unique compositions after removing 20 duplicates (Supplementary Table 2), which were then used for hydrogel synthesis."

4. Additional analysis should be done on figures 2c and 3c. The authors claim distinct patterns among different species, but they observe similar patterns between proteins and synthetic polymers. The proteins seem to have more smeared pairwise frequencies, as compared with the synthetic polymer, which has more discrete common pairs. Does this indicate less blocky sequences for the polymer counterparts? This difference should be addressed with further sequence analysis.

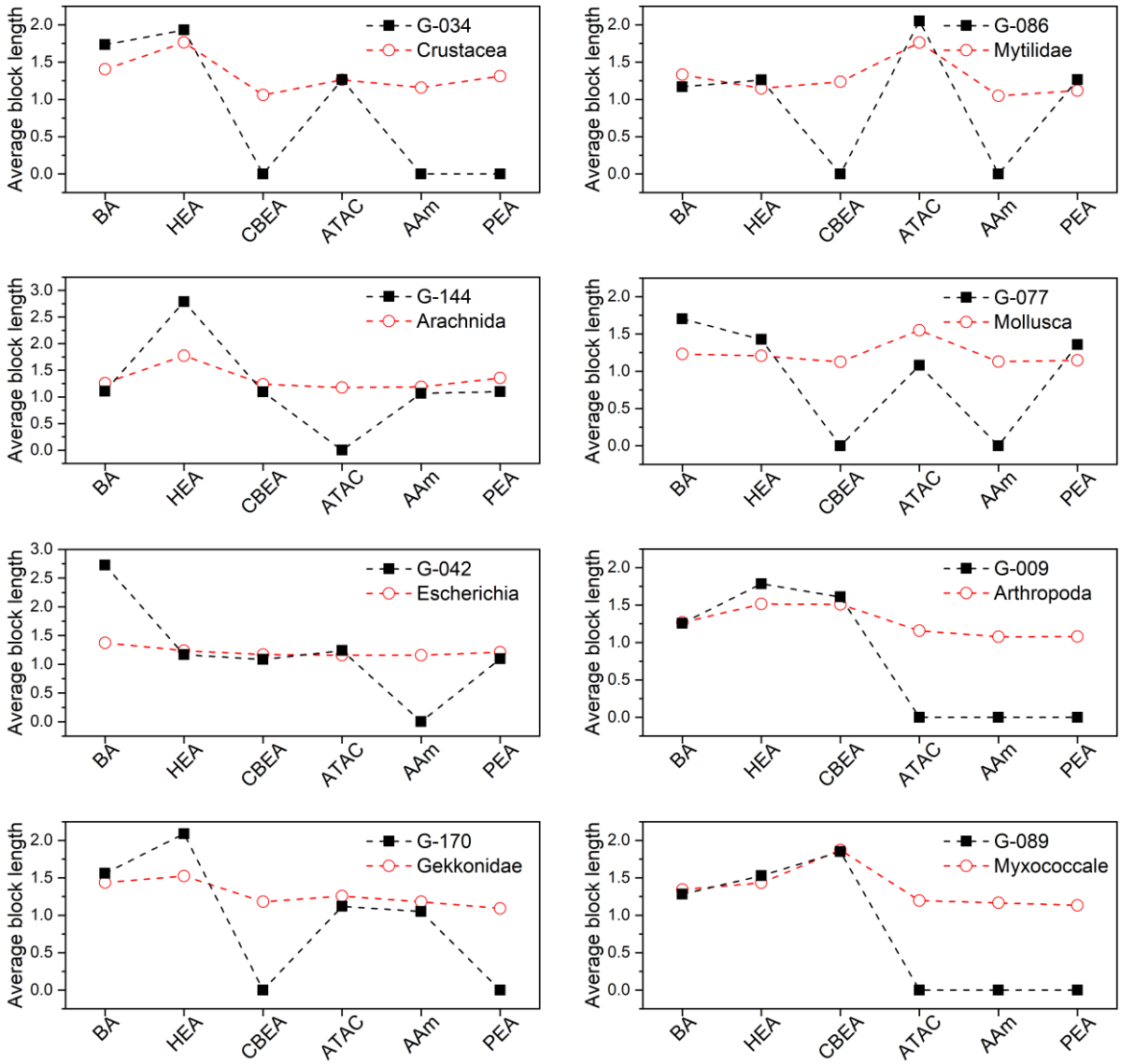
Response: We thank the reviewer for this thoughtful suggestion.

First, we clarify that the gel formulations presented in Fig. 3c are derived from the corresponding species in Fig. 2c, ensuring that they share similar pairwise frequency

distribution patterns in the heatmaps. This highlights distinct sequence features that vary across species.

Second, we note that in the gel formulation process, we implemented a relative composition strategy. Specifically, we ranked functional group pairs by their occurrence in the encoded functional class sequences and selected only the top five for relative composition computation. This modification altered the functional class distribution in the final formulations and, consequently, influenced the pairwise frequencies. Notably, while all consensus sequences contain amide groups, most hydrogel formulations do not include acrylamide. As a result, the heatmap of synthetic polymers appears more discrete, whereas the heatmap of proteins appears more smeared, as the reviewer correctly pointed out.

Regarding blockiness comparisons, we have plotted the average block lengths of functional monomers for synthetic polymers in Fig. 3c (obtained from Monte Carlo simulations) and the corresponding biological species in Fig. 2c. These results are now presented in a new Supplementary Fig. 8 (shown below). Due to the enhanced sampling strategy (i.e., selecting the top five pairs from pairwise frequency sorting), some functional monomers are absent in the extracted formulations. Additionally, the average block length differences vary depending on monomer types and species. Nevertheless, the observed agreement provides another degree of statistical similarity beyond pairwise frequency analysis.



Supplementary Fig. 8. Comparison of average block lengths between functional monomers, including BA (hydrophobic), HEA (nucleophilic), CBEA (acidic), ATAC (cationic), AAm (amide), and PEA (aromatic), in heteropolymer sequences (generated via Monte Carlo simulations) associated with the gel formulations in Fig. 3c and the functional classes in the consensus sequences of corresponding biological species in Fig. 2c of the main text. Due to the implementation of enhanced sampling (i.e., selecting the top five pairs from the pairwise frequency sorting), some functional monomers are absent from the extracted formulations. Nevertheless, the comparison shows reasonable agreement in average block lengths, providing another degree of statistical similarity in addition to pairwise frequency.

To further address this point, we have revised the caption of Fig. 2c (on page 6) and Fig. 3c (on page 7) as follows.

"(c) Pairwise frequency distribution of the 21 functional class pairs along encoded sequences, shown for the total dataset and for representative species (including those with commonly studied adhesive proteins in the literature), categorized according to their biological classifications within the database."

"(c) Pairwise frequency distribution of monomer pairs in heteropolymer sequences obtained from Monte Carlo simulations, shown for all 180 derived formulations and for specific gel formulations (denoted by gel index) corresponding to representative species in Fig. 2c."

5. The authors explain that the "exact replication of functional class sequences would be ideal for mimicking protein functions." This statement seems misleading in the context of this work. I believe networks synthesized here are ill defined. Even if this is not the case, it is important to convey that the statistical nature of this polymer class spans different levels, from the number of initial monomers to the sequence properties to the whole population.

Response: We thank the reviewer for this insightful comment and for sharing the perspective on the network structures.

In our view, hydrogels consist of crosslinked copolymer strands, forming network structures. Interfacial adhesion is governed by the contact between these copolymer strands and the substrate surface. While proteins have intricate three-dimensional structures that dictate their functions, we expect that the wet adhesion of adhesive proteins primarily arises from the interfacial interactions of their amino acid groups. Therefore, replicating the amino acid sequence—i.e., the primary structure—provides a reasonable starting point for mimicking protein adhesion functions.

To address this point, we have revised the 5th paragraph on page 5 of the main text as follows, to emphasize that our approach specifically targets "protein primary structures."

"Based on these insights, we devised a strategy for hydrogel design using six functional monomers to represent six functional classes of amino acids. While directly replicating functional class sequences offers a straightforward way to mimic protein primary structures and functions, achieving precise control over monomer sequences in synthetic polymers remains a significant challenge. Therefore, we aimed to statistically replicate the sequence features of functional classes through ideal random copolymerization of the six functional monomers, which has minimal composition drift during polymerization and enables statistically

controlled sequences[22,32-34].”

Furthermore, we fully agree with the reviewer’s insight regarding the statistical nature of heteropolymer classes, which span from the number of initial monomers to sequence properties. In addition, hydrogels introduce another layer of complexity due to their network structures. To elaborate on this aspect, we have included a new paragraph on page 8 of the main text, as shown below.

“To improve F_a, the correlations between F_a and ϕ_i were assessed using Kendall’s τ coefficients36, and the dependence of F_a on hydrogels’ swelling and rheological behaviors was also characterized (Extended data Fig. 3). We found that ϕ_{ATAC}, ϕ_{BA}, and ϕ_{PEA} have weak positive correlations with F_a, while ϕ_{HEA}, ϕ_{AAm}, and ϕ_{CBEA} show weak negative correlations. Nevertheless, these weak correlations, along with the intricate structure-property relationships (Extended data Fig. 3), cannot be directly used to predict hydrogel formulations for optimal adhesion, highlighting the complex synergistic effects of monomer species, sequences, and network structures.”

6. A key result of the effort for ML-driven optimization was that some limit of performance had already been reached. Given this, the authors should explain the nature of this plateau. Is there a fundamental limit with this type of adhesive, or is this a function of the acrylate monomers selected? What is the nature of other hydrogel systems that surpass this limit? It seems like some additional insight would be useful in drawing meaningful conclusions from the ML effort.

Response: We thank the reviewer for these insightful questions. The observed performance plateau in ML-driven optimization suggests a potential limit within the constraints of our material design space. This limit could arise from several factors. One possibility is the intrinsic limitation of acrylate-based hydrogels in underwater adhesion, even though they performed best in our previous study on two-component hydrogels (*Nat. Commun.* **2019**, *10*, 5127).

While the selected acrylate monomers strike a balance between hydrophobicity, electrostatic interactions, and polymer flexibility, they may not achieve adhesion beyond a certain threshold due to the restricted nature of available physical and chemical interactions. Expanding the monomer library to incorporate other functional groups (e.g., zwitterionic, fluorinated, or bioinspired catechol-containing monomers) and exploring alternative adhesive

strategies (e.g., dynamic covalent bonds, substrate-specific adaptations, supramolecular interactions, or interfacial roughening) could provide new avenues for enhancing adhesion performance.

Beyond acrylate monomers, we have also tested other functional monomers with identical side groups to those presented in this study but featuring a hydrophobic methyl group on the vinyl head. However, the increased hydrophobicity led to severe deswelling of the corresponding gels when the solvent was exchanged with normal saline. This further highlights the complexity of structure-property relationships in hydrogels, as illustrated in the revised Extended Data Fig. 3 (see above).

Regarding insights gained from the ML effort, we found that this end-to-end, black-box-like approach serves as an effective starting point for soft materials design. As optimization progresses, additional meaningful data points are collected, collectively revealing suggestive correlations through interpretability studies. These insights help uncover underlying mechanisms, accelerating materials development beyond conventional trial-and-error approaches.

Moreover, the observed performance limit does not necessarily represent the absolute maximum for hydrogel adhesion but rather the best outcome achievable within the explored formulation space. Future studies could focus on broader monomer selections, refined polymer architectures, and physics-informed ML strategies.

We have incorporated this discussion into the Conclusion section (on page 14) as follows:

"However, challenges remain, primarily due to limitations in monomer diversity, polymer synthesis technologies for controlling monomer sequences to a scale suitable for materials development, and dataset scalability. Overcoming these challenges will require expanding modular monomer libraries, advancing polymerization techniques, and developing physics-informed machine learning models capable of generalizing across sparse, multi-scale datasets."

To further address this point, we have revised the related sentence in the 3rd paragraph on page 10 of the main text as follows:

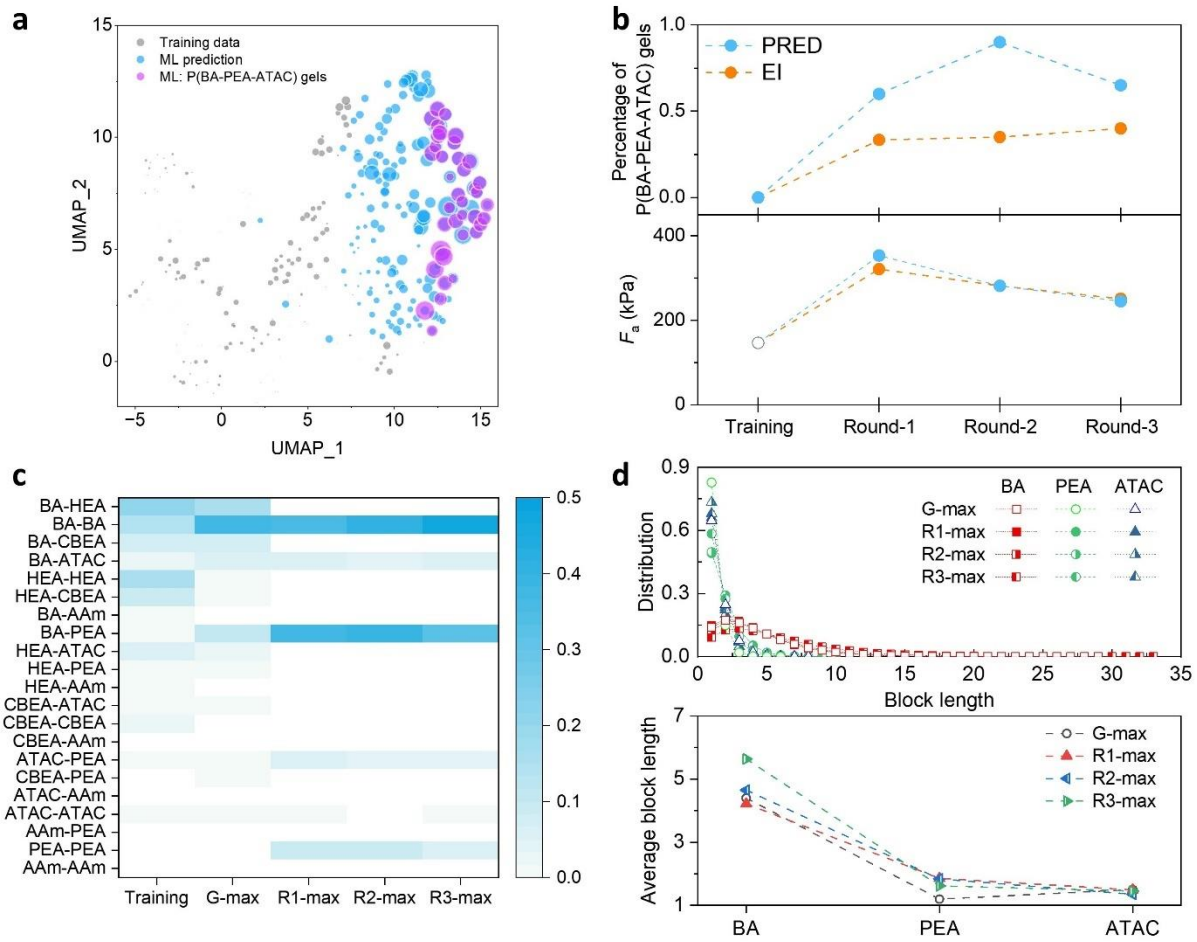
"The validation outcomes expanded our hydrogel dataset. To assess the exploration capabilities of RFR-GP and GP_KB within the SMBO framework, we conducted two additional rounds of ML optimization and experimental validation. Although new high-F_a formulations

were identified, none surpassed the maximum F_a achieved in the first round (Extended Data Fig. 5). We suspect that the functionalities of the adopted monomer species may account for the observed performance plateau, and further optimization rounds were not pursued."

7. The authors find that the three best performers R1-max, R2-max, and R3-max, from the ML optimization were all 3-monomer copolymers and excluded the other three monomers. This observation is quite consequential, as the sequence properties of a three-monomer as opposed to a six-monomer, random copolymer will be very different, especially from the perspective of their pairwise analysis. How can we decouple the conclusions about sequence properties from monomer feeding ratios? It is likely true that incorporating BA, PEA, and ATAC, as written, is key. However, among the best performers, how many exclude the other monomers, and thus change their sequence properties significantly? Given this observation, the pairwise sequence analysis seems insufficient to understand the sequence level features that are necessary for optimal performance. Using figure 4c as a starting point, it may be useful to do some more detailed sequence analysis with further experimental inputs and more in-depth characterization to extract the average segment lengths for different compositions. This should be done for the Monte Carlo simulated sequences of the initial 180 compositions, as well as the ML-outputted sequences.

Response: We are very grateful to the reviewer for these insightful questions and thoughtful suggestions. In response, we have conducted a comprehensive study on the three-component hydrogels consisting of BA, PEA, and ATAC, referred to as P(BA-PEA-ATAC) gels.

Specifically, the initial 180-hydrogel dataset derived from adhesive proteins does not contain this type of gel. It appeared only in the machine learning optimization rounds, where it accounted for 46.7%, 62.5%, and 52.5% of all tested hydrogels in the three rounds. Notably, the gels with the highest adhesive strength (F_a) in each optimization round were all P(BA-PEA-ATAC) gels, demonstrating their dominant performance. We have summarized these results in a new Supplementary Fig. 14 (shown below) on page 21 of the Supplementary Materials.



Supplementary Fig. 14. (a) UMAP representation of the relationship between adhesive strength (F_a) and reduced monomer proportions, highlighting the adhesive strength of ML-predicted three-component gels comprising BA, PEA, and ATAC monomers, namely, P(BA-PEA-ATAC) gels. The training dataset does not include this type of gels. (b) Percentage of three-component P(BA-PEA-ATAC) gels among the tested gels, along with the highest F_a values in the training and three ML optimization rounds, distinguishing results from the PRED and EI sorting schemes. In each ML round, the gels with the highest F_a are all P(BA-PEA-ATAC) gels. (c) Pairwise frequency distribution of 21 functional class pairs in the training dataset of 180 hydrogels, the five-component G-max, and the top-performing three-component ML-driven hydrogels, obtained from Monte Carlo simulations based on formulations and pairwise reactivity ratios. (d) Comparison of block length distributions and corresponding average block lengths for BA, PEA, and ATAC in the top-performing hydrogels, demonstrating similar statistical sequence features.

Furthermore, we analyzed the sequence features of the top-performing hydrogels using Monte Carlo simulations, as experimental characterization at this level of detail remains challenging (as the reviewer also noted). Supplementary Fig. 14c presents a comparison of

pairwise frequency distributions across the entire training dataset of 180 hydrogels, the five-component G-max, and the three-component ML-driven hydrogels, highlighting distinct pair features shared by the four top-performing gels.

In addition to pairwise sequence analysis, we compared the block length distributions and average block lengths of BA, PEA, and ATAC in the four top-performing gels. Interestingly, these gels exhibit striking similarities in these statistical sequence features, particularly the small block length of ATAC. This raises an intriguing question about how adhesion performance depends on block length. However, experimentally studying this aspect is difficult, and we plan to dedicate a molecular dynamics simulation investigation to this topic in future work.

We believe that the additional analysis stimulated by the reviewer's suggestions has significantly improved our manuscript, and we truly appreciate the reviewer's input on this matter.

To further address this point, we have revised the 2nd paragraph on page 11 of the main text as follows:

"Indeed, the hydrogels with the highest F_a from each ML round, denoted as R1-max, R2-max, and R3-max, are all exclusively composed of these three monomers (Fig. 4d) and share similar statistical sequence features as indicated by Monte Carlo simulations (Supplementary Figs. 11 and 14)."

8. The adhesive performance testing in different conditions were done for R1-max, R2-max, and R3-max. How about the best performers from the initial 180 compositions, designed without ML optimization? Given the deviation from the six-monomer rational design, some experimental inputs/characterization in normal saline.

Response: We thank the reviewer for this question. The best performer from the initial 180 compositions is denoted as G-max. Its material properties, including volume swelling ratio, tensile and rheological behaviors, as well as adhesion performance, have been systematically characterized, along with the top-performing hydrogels from the ML-driven rounds. These results are summarized in the new Supplementary Table 8 (shown below), and the corresponding data can be found in Fig. 5c, Extended Data Figs. 6 and 7, and Supplementary Fig. 17.

Moreover, the top 10 performers from the training dataset were also examined for their

maximum adhesion performance, with the results presented in Extended Data Fig. 7.

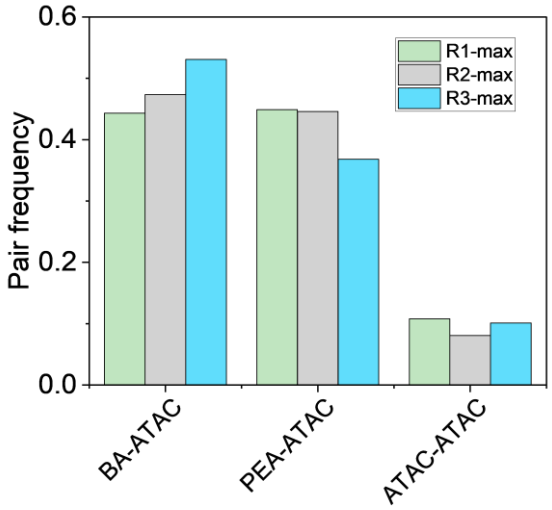
Supplementary Table 8. Formulations, volume swelling ratios (*Q*), Young's modulus (*E*), storage modulus (*G'*) at a frequency of 10^0 rad s^{-1}, and adhesive strength (F_a) of the top-performing hydrogels from data mining and three machine learning optimization rounds. Tack tests were conducted under a 20 N loading force applied for 60 seconds on a glass substrate in normal saline (0.154 M NaCl). All hydrogels were equilibrated in normal saline.

No.	Φ_{HEA}	Φ_{BA}	Φ_{CBEA}	Φ_{ATAC}	Φ_{PEA}	Φ_{AAm}	*Q*	*E (kPa)*	*G' (kPa)*	F_a *(MPa)*
G-max	0.141	0.591	0.076	0.096	0.096	0.000	0.42±0.04	125.60±5.74	5.86	0.50±0.02
R1-max	0.000	0.588	0.000	0.094	0.318	0.000	0.51±0.06	320.76±7.93	18.72	1.02±0.01
R2-max	0.000	0.629	0.000	0.061	0.310	0.000	0.42±0.02	459.99±26.74	24.69	0.85±0.04
R3-max	0.000	0.680	0.000	0.070	0.250	0.000	0.50±0.04	538.95±4.72	25.35	0.86±0.01

As shown in Fig. 5c, the adhesive performance of G-max (~0.5 MPa) is inferior to that of the hydrogels optimized through machine learning (~1.0 MPa), when measured under the same conditions. This highlights the effectiveness of the ML optimization in identifying superior formulations that go beyond the initial rational design.

9. A major design rule derived from this work is: "The SHAP summary plot (Fig. 4c) reveals that high values of and significantly enhance. This is because both BA and PEA effectively expel water from the contact interface, and when neighboring with ATAC, they could enhance the electrostatic interactions with the negatively charged glass surface29,38-41." It would be helpful to present the simulated sequences of the best performers like R1-max and analyze their sequence patterns to validate this claim. Also, comparing the sequence analysis of R1-max with the protein sequence analysis shown in Figure 2c can be beneficial. This comparison would provide further validation of the whole workflow and potentially highlight the differences in design rules between synthetic polymers and natural proteins.

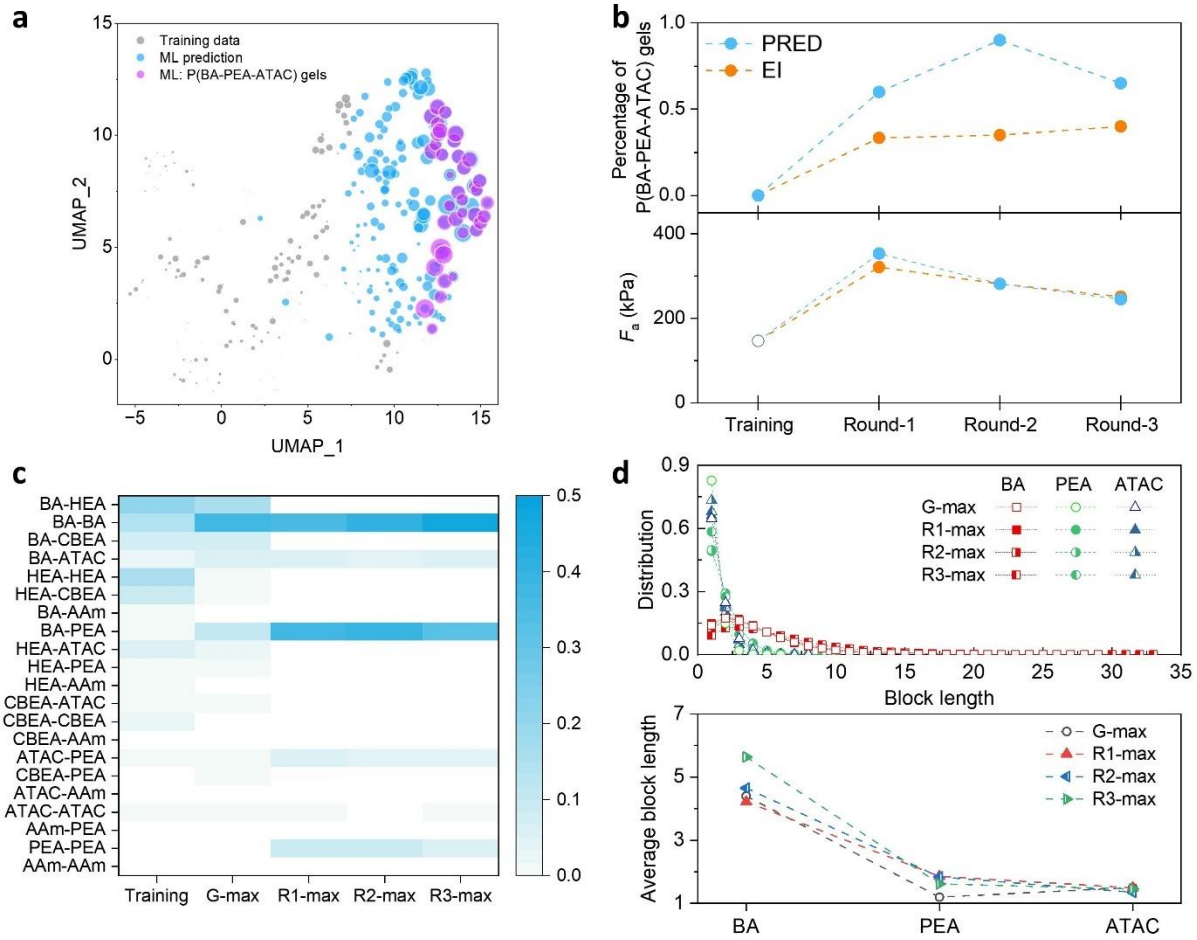
Response: We sincerely thank the reviewer for these thoughtful suggestions. In response, we have simulated and analyzed the sequences of the best-performing hydrogels predicted by ML, namely, R1-max, R2-max, and R3-max. The comparison of pairwise frequency distributions is now shown in a new Supplementary Fig. 11 on page 18 of the Supplementary Materials (shown below). These distributions exhibit higher fractions of heterogeneous pairs (BA-ATAC and PEA-ATAC) in all three gels.



Supplementary Fig. 11. Pairwise frequency distribution of pairs containing ATAC from copolymer sequences generated via Monte Carlo simulations using R1-max, R2-max, and R3-max formulations. The results reveal higher fractions of heterogeneous pairs compared to the homogeneous pair of ATAC.

Furthermore, we examined the adhesion of R1-max on glass with a positively charged surface, which exhibited a poor adhesive strength (Supplementary Fig. 12, shown below). These findings support our claim that the proximity of BA and PEA to ATAC enhances electrostatic interactions with negatively charged glass surfaces, leading to strong adhesion.

Regarding the comparison of sequence features between the best-performing hydrogels and adhesive proteins, we have included the results in a new Supplementary Fig. 14 (on page 21). The training set consists of 180 bioinspired hydrogels derived from adhesive proteins. This figure highlights distinct pairwise features identified by machine learning during the optimization process.



Supplementary Fig. 14. (a) UMAP representation of the relationship between adhesive strength (F_a) and reduced monomer proportions, highlighting the adhesive strength of ML-predicted three-component gels comprising BA, PEA, and ATAC monomers, namely, P(BA-PEA-ATAC) gels. The training dataset does not include this type of gels. (b) Percentage of three-component P(BA-PEA-ATAC) gels among the tested gels, along with the highest F_a values in the training and three ML rounds, distinguishing results from the PRED and EI sorting schemes. In each ML round, the gels with the highest F_a are all P(BA-PEA-ATAC) gels. (c) Pairwise frequency distribution of 21 functional class pairs in the training dataset of 180 hydrogels, the five-component G-max, and the top-performing three-component ML-driven hydrogels, obtained from Monte Carlo simulations based on formulations and pairwise reactivity ratios. (d) Comparison of block length distributions and corresponding average block lengths for BA, PEA, and ATAC in the top-performing hydrogels, demonstrating similar statistical sequence features.

We note that extracted protein sequences exhibit a greater diversity of functional class pairings (Supplementary Fig. 14), reflecting a fundamental biological principle—adaptability over universal optimization, as discussed in the manuscript. In contrast, ML-driven prediction

works as an optimization problem with specified conditions, such as a target substrate (glass) and working environment (underwater). Therefore, we emphasize that the derived design principle is specifically "for achieving strong underwater hydrogel adhesion to glass surfaces using the selected functional monomers." Moreover, while our approach can be readily adapted for targeting other substrates (e.g., metals, plastics, or soft tissues) and working environments (e.g., dry, submerged, or oil-covered surfaces), it may yield different top-performing hydrogels and design principles.

To address these points, we have made the following revisions in the manuscript, starting in the last paragraph on page 10 of the main text:

"To assess the influence of ϕ_i on F_a, we used SHAP (SHaply Additive exPlanations)[44] with the RFR model trained on the final 341-hydrogel dataset. The SHAP summary plot (Fig. 4c) reveals that high values of ϕ_{BA} and ϕ_{PEA} significantly enhance F_a. This is because both BA and PEA effectively expel water from the contact interface, and when neighboring with ATAC (cf. Supplementary Fig. 11), they could enhance the electrostatic interactions with the negatively charged glass surface (Supplementary Fig. 12)[32,45-48]. In contrast, high values of ϕ_{HEA}, ϕ_{CBEA}, and ϕ_{AAm} tend to reduce F_a. Interestingly, ϕ_{ATAC} has a dual effect (Supplementary Fig. 13): lower levels diminish electrostatic interactions, while excessive ϕ_{ATAC} increases hydrogel swelling, limiting polymer-surface contact and thus lowering F_a. Therefore, a moderate amount of ATAC is crucial."

"These insights, consistent across all three ML rounds, establish a clear design principle for achieving strong hydrogel adhesion to glass surfaces using the selected functional monomers: incorporating BA, PEA, and ATAC is key. This combination leverages both hydrophobic effects and electrostatic interactions to enhance underwater adhesion to negatively charged surfaces. Indeed, the hydrogels with the highest F_a from each ML round, denoted as R1-max, R2-max, and R3-max, are all exclusively composed of these three monomers (Fig. 4d) and share similar statistical sequence features as indicated by Monte Carlo simulations (Supplementary Figs. 11 and 14)."

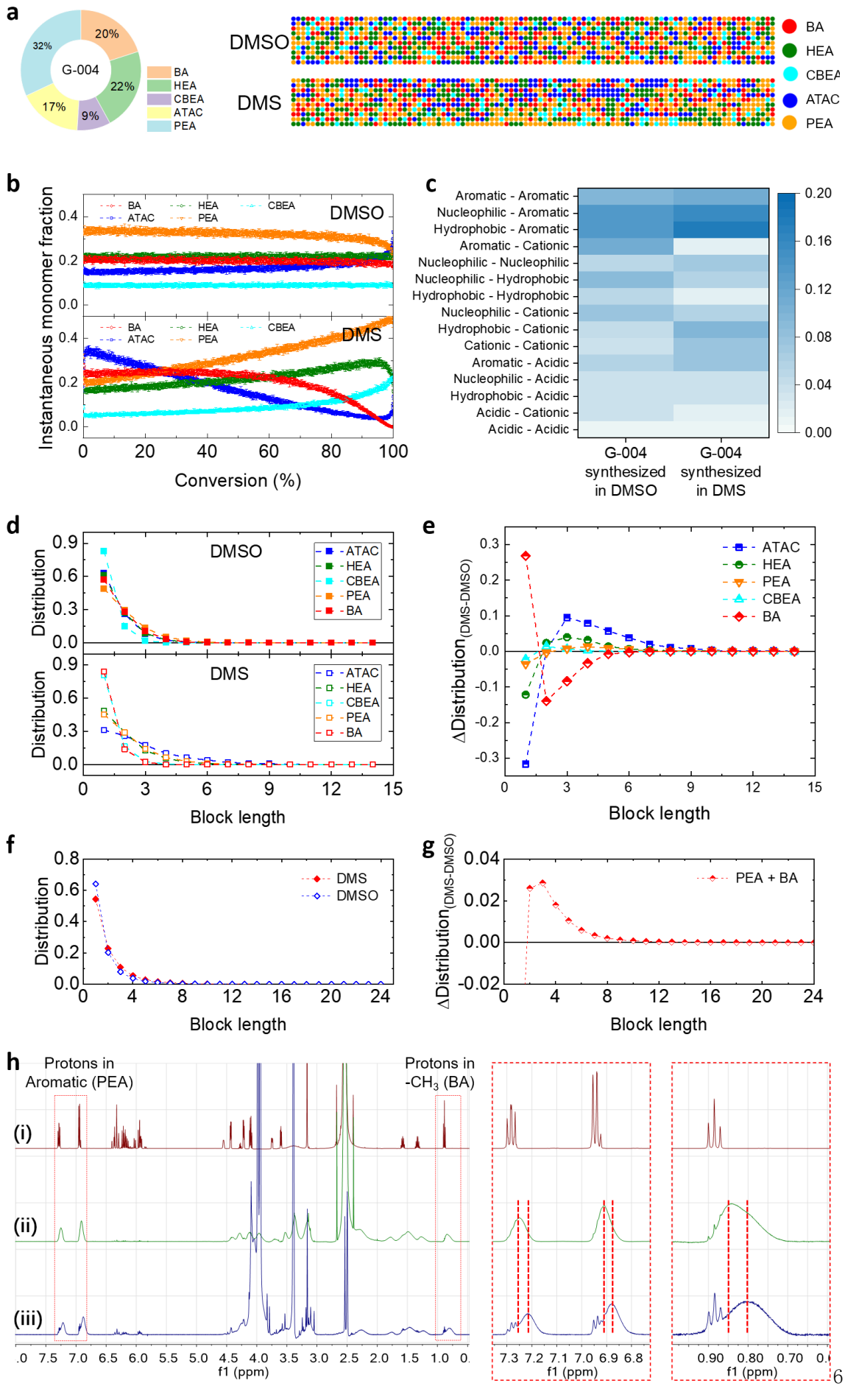
10. The authors mentioned that "For copolymerization in DMS, the pairwise reactivity ratios deviate significantly from 1 (Supplementary Table 3), leading to blocky sequences (Extended Data Fig. 3)." However, based on the reactivity ratios provided in Extended Data Fig. 3 (e.g., rBA = 0.5, rPEA = 0.33 in DMS), an alternating sequence is expected rather than a blocky sequence.

Response: We thank the reviewer for this insight.

For the copolymerization of two types of monomers, when their reactivity ratios, r_1 and r_2, are both small and near 0, the monomers are unable to homopolymerize, and thus can only alternate with each other, leading to an alternating sequence.

In the case of copolymerization in DMS, the mentioned reactivity ratio values are not very close to 0. Moreover, G-004 contains three additional functional monomers (i.e., 20% BA, 22% HEA, 9% CBEA, 17% ATAC, and 32% PEA), and the synergistic effects of these monomers influence the copolymer sequence.

To illustrate the sequence differences between hydrogels synthesized in DMSO and DMS, we have added the results from Monte Carlo simulations and ^{1}H NMR analysis of G-004 variants in a new Supplementary Fig. 7 (on pages 13 and 14). Specifically, the Monte Carlo simulations show an increase in block lengths in DMS, except for BA. Notably, the block length of combined PEA and BA (both hydrophobic) increases, which is corroborated by NMR measurements. This increase in block length promotes the association of hydrophobic components, aligning with the translucent appearance of the G-004 hydrogel synthesized in DMS.


a
G-004
20%
22%
9%
17%
32%
BA
HEA
CBEA
ATAC
PEA
DMSO
DMS
b
Instantaneous monomer fraction
Conversion (%)
c
Aromatic - Aromatic
Nucleophilic - Aromatic
Hydrophobic - Aromatic
Aromatic - Cationic
Nucleophilic - Nucleophilic
Nucleophilic - Hydrophobic
Hydrophobic - Hydrophobic
Nucleophilic - Cationic
Hydrophobic - Cationic
Cationic - Cationic
Aromatic - Acidic
Nucleophilic - Acidic
Hydrophobic - Acidic
Acidic - Cationic
Acidic - Acidic
G-004 synthesized in DMSO
G-004 synthesized in DMS
d
Distribution
Block length
e
ΔDistribution(DMS-DMSO)
Block length
f
Distribution
Block length
g
ΔDistribution(DMS-DMSO)
PEA + BA
Block length
h
Protons in Aromatic (PEA)
Protons in -CH3 (BA)
(i)
(ii)
(iii)
f1 (ppm)

Supplementary Fig. 7. Monte Carlo simulations (a-g) and ^{1}H NMR analysis (h) of monomer sequences for G-004 prepared in DMSO and DMS. (a) G-004 formulation and its simulated chain sequences in DMSO and DMS, based on reactivity ratios from Supplementary Table 3. Each chain consists of 1,000 monomers, presented in 10 rows, with each row showing the sequence of 100 monomers from left to right. (b) Instantaneous monomer fractions as a function of total conversion during polymerizations in DMSO and DMS. Each simulation involved 2 million monomers, with an average chain length of 1,000. The results show minor composition drift for polymerization in DMSO, but significant composition drift in DMS throughout the polymerization. (c) Pairwise frequency distribution of 15 distinct functional class pairs along encoded sequences. (d) Block length distributions for different monomers from the simulated sequences. (e) Differences in monomer block length distributions between copolymers prepared in DMS and DMSO, highlighting increased block lengths for ATAC, HEA, and PEA. (f) Block length distributions for combined aromatic PEA and hydrophobic BA in DMSO and DMS. (g) Difference between the two distribution curves in (f), suggesting longer segments of combined BA and PEA in DMS. Statistical results in (b-g) were obtained from five independent runs. (h) ^{1}H NMR spectra of G-004 monomer precursor solution (i) and copolymers synthesized in DMSO (ii) and in DMS (iii) after 8 hours of polymerization (conversion > 99% in DMSO and > 93% in DMS). The portions of the NMR spectra highlighted by red dashed frames are enlarged on the right. For PEA aromatic protons, a high-field shift in the peak indicates an increase in PEA block size. For BA methyl protons, a high-field shift suggests a higher frequency of adjacent BA-PEA units, corresponding to larger segments of combined BA and PEA monomers. These findings are consistent with the simulation results shown in (f, g). Overall, the simulations suggest that hydrogels prepared in DMSO maintain quasi-ideal random copolymerization, resulting in statistical monomer sequences that determine the hydrogel's adhesion properties. In contrast, hydrogels prepared in DMS exhibit blocky sequences of combined BA and PEA (both hydrophobic), which lead to aggregation of hydrophobic components and the opaque appearance (cf. Fig. 3f in the main text). Meanwhile, the increase in ATAC block length negatively impacts adhesion[1].

11. The authors didn't provide information regarding monomer conversion in the radical polymerization. Since monomer distribution depends on monomer conversion and the Monte Carlo simulation requires it as an experimental input, what values were used?
The hydrogel fabrication involves a two-weeks immersion in saltwater. Given that the polymers contain ester sidechain, it's unclear whether these ester sidechains undergo hydrolysis in this process, especially when a nucleophilic monomer HEA is incorporated, which can alter the hydrogel's overall composition.

Response: We thank the reviewer for this excellent question regarding monomer conversion and the comment on the hydrolysis effect.

The monomer conversion was measured using NMR, and the results indicated a conversion rate of over 99% for copolymerization in DMSO, which can be attributed to the high reactivity of the acrylate monomers (revised Supplementary Figs. 6a, 7h, and 9b). Additionally, we confirmed that the monomer conversion of pairs such as ATAC and PEA, which exhibit significant hydrophobicity differences, remained consistent throughout the polymerization process (revised Supplementary Fig. 6b). Based on these findings, we believe that the monomer distribution remains stable throughout polymerization, and the monomer conversion was set to 100% in our Monte Carlo simulations for sequence analysis.

To clarify this point, we have added a subsection in the Supplementary Materials (on page 3) regarding the setting of the Monte Carlo simulations as follows:

"**Monte Carlo Simulations**

Monte Carlo simulations were performed using the Compositional Drift program[3]. Unless otherwise specified, the statistical results presented are based on sequences generated by simulations involving a total of 2 million monomers, with an average chain length of 100 and 100% conversion. These simulations were conducted using the gel formulations (Supplementary Tables 2 and 7) and the measured pairwise reactivity ratios (Supplementary Table 3) of functional monomers. Simulations with different average chain lengths (up to 1,000) were also tested, with results showing only marginal differences."

For the concern about hydrolysis during the two-week saltwater immersion, we agree that hydrolysis of ester side chains may occur over prolonged soaking periods. However, our long-term underwater adhesion experiments demonstrated that the hydrogel maintained its adhesion strength for over one year (Fig. 5b and Supplementary Fig. 16). This suggests that hydrolysis

effects are negligible within the time frame relevant to our study. The long-term stability of swelling and modulus further supports this conclusion (Figure R2). We are grateful to the reviewer for the input on this matter.

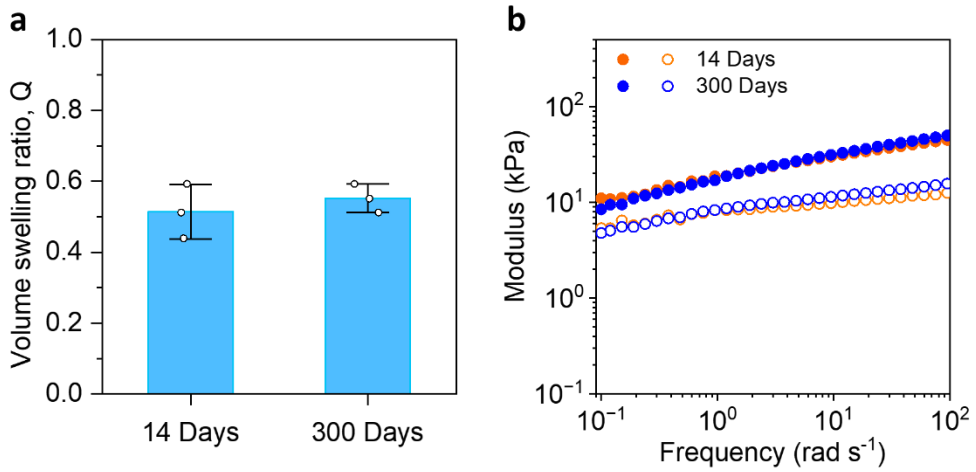


Figure R2. Comparison of (a) volume swelling ratio and (b) rheological properties of the R1-max hydrogel after immersion in normal saline for 14 days and 300 days. The results demonstrate that hydrolysis effects are negligible within the time frame relevant to our study.

Others:

The labeling of Figure 4a is confusing, making it difficult to understand what are the differences between models RFR-GP, RFR-GP, and GP-RFR.*

Response: We thank the reviewer for this comment. We acknowledge that the labeling in Figure 4a may be unclear, as the detailed descriptions of the models were provided in the Methods section due to space limitations in the main text.

To enhance readability and improve clarity, we have revised the relevant sentences in the "Machine Learning for Hydrogel Adhesion Optimization" section (on page 9) as follows:

"Next, we employed machine learning to explore hydrogel formulations with higher adhesive strength, starting with the 180-hydrogel dataset. By benchmarking nine ML models (Supplementary Tables 5 and 6), Gaussian process (GP)[37] and random forest regression (RFR)[38] emerged as the most effective base models for predicting F_a from ϕ_i, as they achieved low test error while minimizing overfitting (Extended Data Fig. 4)."

"To enhance efficiency, we explored several batched SMBO methods, using trained base models as hypothetical value providers (*P*) and GP, RFR, extra-trees (ETR)[39], and gradient boosting machine (GBM)[40] as EI maximizers (*M*), collectively denoted as "*P-M*." In parallel, we implemented traditional Bayesian optimization methods, using kriging believer (GP_KB)[41], maximum/minimum constant liar (GP_CLmax, GP_CLmin)[41], and local penalization (GP_LP)[41,42] as heuristics for determining batch points."

The caption of Fig. 4 (on page 11) has also been updated to include a reference for further clarification as follows:

"Fig. 4. Machine learning optimization of underwater adhesive hydrogels. (a) Adhesive strength (Fa) of hydrogels fabricated based on predictions from various models trained on the 180-hydrogel dataset. The model nomenclature and detailed descriptions are provided in the Methods section. All adhesion measurements were performed under the test conditions of 10 N loading force, 10 seconds contact time, on a glass substrate, and in normal saline."

Furthermore, we have added a new Supplementary Table 5 (on page 32 of the Supplementary Materials) to provide a concise summary of the models.

Supplementary Table 5. Definitions of the regression models used in this work.

Type	Subcategory	Method	Description
Linear		Lasso	Performs linear regression with L1 regularization, leading to feature selection by shrinking some coefficients to zero.
		Ridge	A linear regression method with L2 regularization, which helps prevent overfitting by penalizing large coefficients.
Nonlinear	Non-parametric methods	KNN	A non-parametric method that predicts a target value based on the average (or weighted average) of its k-nearest neighbors.
	Kernel-based methods	GP	A Bayesian regression method that models the distribution over functions, providing uncertainty estimates in predictions. Matern Kernel is used.
		KRR	Combines Ridge Regression with kernel methods (radial basis functions) to capture non-linear relationships in data.
		SVR	A regression technique using Support Vector Machines, aiming to find a hyperplane that best fits the data within a margin.
	Tree-ensemble methods	ETR	Similar to Random Forest but introduces more randomness in tree splits to reduce variance further.
		XGB	A powerful gradient boosting algorithm that optimizes decision trees to maximize accuracy and efficiency.
		RFR	Uses an ensemble of decision trees to improve prediction accuracy and reduce variance through bagging.

In Fig. 5, all adhesive performance tests were conducted using R1-max, except for the high-pressure water leakage test, which was performed using R2-max. The rationale behind this choice is unclear.

Response: We thank the reviewer for pointing out the omission of the rationale behind our selection. We chose R1-max to demonstrate the adhesive performance, as it exhibited the highest adhesive strength (F_a) overall in normal saline. However, for the high-pressure water leakage test, which was performed using tap water, we selected R2-max because it exhibited a higher F_a in deionized water (Fig. 5c).

To clarify this point, we have revised the related sentences in the "Performance of super-adhesive hydrogels in diverse aqueous conditions" section (on pages 12 and 13) as follows:

"In artificial seawater (0.7 M NaCl solution), all three ML-driven hydrogels displayed similar levels of strong adhesion (Fig. 5c and Supplementary Fig. 17). However, in deionized water, R2-max outperformed the other two, exhibiting cavitation during debonding (Supplementary Fig. 18)."

"Figure 5e highlights the effectiveness of R2-max, the hydrogel with the highest adhesion in deionized water (Fig. 5c), in sealing a 20-mm hole at the base of a 3-meter-tall polycarbonate pipe filled with tap water."

As a side note, I found some wordings aren't accurate although I fully understand they were used to enhance the impact of present studies. I personally prefer more objective assessment of the prior work even though they aren't as trendy. For example, "Currently, the development of new soft materials heavily relies on experimental trial and error, often guided by researchers' intuition and experience". This is definitely an overstatement. There have been a lot of theory and modeling across various length scales. AI/ML are good tools but not going to save the day by itself.

Response: We truly appreciate the reviewer for this valuable feedback and for sharing perspectives on the objective assessment of the prior work, as well as the importance of theory and modeling in advancing the field of soft materials. We completely agree with these insights and apologize for any unintentional overstatements in our original manuscript.

In the revision, we have carefully ensured that statements throughout the manuscript are objective and pertinent. Specifically, the sentence pointed out by the reviewer has been revised to soften the tone, as follows (in the 2nd paragraph of the Introduction section on page 2):

"Traditionally, the development of new soft materials has relied on experimental trial and error, often guided by researchers' intuition and experience."

Furthermore, we are committed to applying theory and modeling to deepen our understanding of the intricate structure-property relationships and guide material design. To highlight this point, we have added the following sentence at the end of the Conclusion section (on page 14 of the main text):

"Overcoming these challenges will require expanding modular monomer libraries, advancing polymerization techniques, and developing physics-informed machine learning models capable of generalizing across sparse, multi-scale datasets."

Response Letter

We are very grateful to referee #1 for their insightful summary and positive evaluation of our manuscript. We also sincerely thank referee #3 for their thoughtful and constructive feedback, which has greatly enhanced the quality of our work. We have carefully addressed all concerns raised and revised the manuscript accordingly.

A point-by-point response to the reviewer #3 is provided below. In this response, the reviewers' comments are shown in *italics*, our responses are in blue, and the changes in the Manuscript, Extended Data, and Supplementary Materials are highlighted in red.

Referee #3 (Remarks to the Author):

The authors have made a good effort to respond to previous review questions. But some key questions still linger.

1. The major concern still remains in terms of molecular connections between what's delivered via DM/ML vs. experimental results and explanations.

a. While I appreciate the characterizations of the resultant hydrogels, most if not all are bulk properties. There are quite gaps between monomer chemistry and distributions along a strand in a network to the bulk properties, let alone the adhesion. I also found the discussions on the rationale of these top performers are superficial in general. They mainly focused chemical identity, composition and occasionally are connected to the simulated sequences as shown in the SI Fig. 6b. This is a significant weakness and should be addressed with additional experiments. The authors specifically pointed out that for adhesive properties, the strands are the main motifs interacting with the substrate. Thus, how the polymers interact with the underlying substrate, e.g. glass surfaces, should be studied for a few key samples here. The authors should feel free to do whatever they see fit to bridge the gap. There are many studies from the adhesive community. But if this is my project, I will look into measuring adhesions of polymers via AFM so there is a base to compare with other known work on polymer adhesive properties. The short blocks of functional monomers appear to be a key design factor. The team should consider how this may affect the chemistry/functional groups at the interfaces. Ambient XPS under controlled humidity can help. ATR-FTIR can be useful as well and if the team can have access to polarized source, the results will be even more insightful.

Response: We thank the referee for this insightful and constructive feedback. We

agree that elucidating the interplay of monomer chemistry, interfacial interactions, network architecture, and viscoelastic dissipation is critical for optimizing adhesive performance. Bridging molecular structure with macroscopic properties, given the complex structure-property relationships, remains a central challenge in soft materials research.

Our study establishes a data-driven framework that combines data mining, experimentation, and machine learning to guide de novo design of adhesive hydrogels. While interfacial mechanisms are highly relevant, detailed mechanistic studies fall outside the current scope. Our primary aim is to develop an efficient discovery pipeline, with mechanistic investigations planned for future work.

We appreciate the referee's suggestions to use techniques such as AFM, ambient XPS, and ATR-FTIR to probe strand – substrate interactions. These approaches will be invaluable for future studies exploring molecular-level adhesion mechanisms.

We underscore that uncovering structure – property relationships in complex soft materials is a multifaceted challenge. While a comprehensive mechanistic analysis is beyond this manuscript's scope, our work offers a strong foundation for predictive design and future in-depth investigation.

b. I also appreciate the authors' efforts to monitor monomer conversions for ATAC/PEA pair in different solvents. The authors ascertained the reaction mixtures' miscibility by stating solution transparency/clarify. I feel the conclusions should have been drawn with more scientific vigor. Given the monomer concentration used for network formation is much higher than these control studies and the variety/chemical differences of the monomers, I think light scattering may be more convincing. (It is not ideal but an easier experiments than more advanced techniques to probe if the monomers may form aggregates or phase separate at length scales below 400 nm.) To clarify, with the high viscosity of monomer solutions and >98% conversion, the radicals have very low diffusivity. A slight aggregates will change the effective monomer ratio.

Response: We thank the referee for these thoughtful observations and valuable suggestions. We agree that evaluating monomer miscibility via light scattering offers a rigorous approach and that increased viscosity at high monomer conversion (>98%), along with potential aggregation, could influence effective reactivity ratios.

Our previous work employed dynamic light scattering (DLS) to examine monomer

solution structures (Polym. Chem. 2024, 15, 2104). In that study, DMSO solutions containing hydrophilic and hydrophobic monomers at concentrations comparable to those in the present study showed no detectable aggregation, whereas aggregation was observed by adding non-cosolvent. Accordingly, we selected DMSO as an effective cosolvent for diverse monomer mixtures.

Although DLS was not conducted for the specific monomers used here, the high transparency and frequency-independent rheological behavior of the as-prepared gels suggest minimal aggregation in both monomer mixtures and resulting copolymers, supporting a random copolymerization mechanism. Furthermore, Figure S6b shows that cationic ATAC and aromatic PEA monomers were consumed at comparable rates across various feed ratios, even at high conversions. This indicates reactivity ratios near unity and minimal viscosity effects under our reaction conditions.

To clarify these points, we have revised the relevant discussion in the second paragraph on page 8 in the main text as follows.

"In their as-prepared state, all gels were transparent and exhibited frequency-independent storage moduli (*G′*) (Extended Data Fig. 6a), indicating negligible inter- or intramolecular aggregation in DMSO. Despite compositional differences, comparable *G′* values suggest similar network topologies."

c. Based on what I read, the monomer conversion or near unity reactivity ratios were determined using a pair of two monomers. I want to bring one recent publication to the team attention that discuss potential complications of reactivity ratios in the presence of more monomers to account for other factors. The team should look into their system to account for the potential errors introduced by the reactivity ratios. (Mapping Composition Evolution through Synthesis, Purification, and Depolymerization of Random Heteropolymers, J. Am. Chem. Soc. 2024, 146, 6178.)

Response: We thank the referee for highlighting this important point and for referencing the recent publication (J. Am. Chem. Soc. 2024, 146, 6178). We agree that determining reactivity ratios in multicomponent monomer systems introduces additional complexity and potential sources of error.

In this study, we evaluated reactivity ratios using binary monomer pairs to simplify interpretation and ensure control over copolymer composition. We recognize that more complex copolymerization systems, as discussed in the reference work, may require deeper analysis.

We will examine the insights from this reference and consider their implications for our system in future work. Incorporating these considerations will enhance our design principles and improve predictive control in multicomponent formulations.

d. During the DM, residues like glycine, alanine and proline were excluded. In fact, the selection appears to be arbitrary to me and can't be simply justified as stated in the main text and rebuttal. Although the team reasoned this by their lack of interactions with the substrates, these residues are important when it comes to the results shown Figure 2, for example the block length of different types of residues. The analysis including these residues should be done and compared.

Response: We thank the referee for their insightful observation regarding the exclusion of glycine, alanine, and proline during data mining (DM). We acknowledge the concern that this selection may appear arbitrary and insufficiently justified in the main text and previous rebuttal. Our decision to exclude these residues was based on their limited contribution to direct adhesive interactions with substrates, as informed by prior studies on protein–substrate binding mechanisms.

We prioritized residues with stronger adhesive relevance to streamline the identification of sequence patterns critical for hydrogel design. Despite this simplification, the final results of our approach—including the successful identification and design of high-performance adhesive hydrogels — demonstrate the practical effectiveness of our methodology.

We agree that these residues may contribute to sequence architecture and secondary structure modulation, warrant further exploration. We plan to investigate their roles in future studies to deepen our understanding of sequence–property relationships.

To clarify our rationale, we have revised the corresponding sentence on page 4 as follows.

"For consistency in the encoding, glycine, alanine, and proline were excluded from the hydrophobic class due to their smaller side chains, which are hypothesized to have a less significant role in interfacial contacts and interactions compared to other amino acids."

2. I strongly urge the team to revisit the introduction regarding the status of soft material design using DM, ML and bioinformation and more generally, work by the soft material

community even before ML became the buzz word. For example, DM/ML and/or bioinformation have been used in soft material design by many, not only by the hard material community.

The team revised:

Traditionally, the development of new soft materials has relied on experimental trial and error, often guided by researchers' intuition and experience."

This is simply not true if one goes back to as simple as Flory-Huggins theory or as complex as scaling laws. Even for network synthesis, the formulations derived by the statistical mechanics have been around for a long time.

Response: We thank the referee for their thoughtful and valuable feedback. We fully agree that the soft materials community has a long and distinguished history of employing theoretical and computational tools to guide material design, well before the emergence of modern data mining, machine learning, and bioinformatics approaches.

Our original phrasing was not intended to overlook these foundational contributions but rather to emphasize the continued challenge posed by the complex structure-property relationships in soft materials. These complexities often hinder the development of quantitative theoretic models and accurate simulations, thereby necessitating extensive experimental trial and error. We envision that integrating emerging data-driven approaches into a cohesive, end-to-end framework offers a promising path to reduce experimental burden and accelerate material discovery.

To clarify this point, we have revised the sentence noted by the referee on page 2 as follows.

"These complexities hinder the development of accurate predictive theories or computational models, often rendering soft material discovery reliant on experimental trial and error."

3. The authors should definitely check the references to make sure they are cited at the right place.

Response: We thank the referee for this helpful comment. In response, we have carefully reviewed all references in the revised manuscript to ensure they are cited accurately and appropriately throughout the text.