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Contingent Instructional Scaffolding for Teaching Tissue Scaffolds Using PrePostClass Assessments

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Abstract

The extracellular matrix (ECM) comprises hundreds of interacting proteins and connections that together form tissue scaffolds. Although understanding ECM structure and function is central to tissue engineering, instructional time is insufficient to teach all ECM components. Therefore, it is essential to build an instructional foundation that supports students in gradually developing the ability to reason about tissue scaffold protein components independently. Instructional scaffolding in education provides structured support calibrated to the learner and gradually fades as competence increases, grounded in the Zone of Proximal Development (ZPD) framework. Prior scaffolded learning in tissue engineering has improved capstone design skills, scientific writing, laboratory communication, and material characterization outcomes. However, a contingent instructional scaffolding approach that systematically calibrates support across the tissue engineering curriculum remains underdeveloped. This need becomes more pronounced in GenAI-aware settings, where out-of-class assignments may not reliably reflect students' independent progress. To address this limitation, we implemented an ECM-by-ECM protein instructional scaffold that organizes tissue engineering content around individual extracellular matrix proteins, with ZPD-aligned, low-stakes pre/post class formative assessments administered before and after each lecture, with instructional support contingently removed, in an upper-division course with data collected across three offerings from 2023 to 2025. The results support a scalable contingent scaffolding approach that aligns instructional support fading with learners' developmental trajectories. This approach is broadly applicable to interdisciplinary bioengineering concept-dense topics with limited instructional time, enabling students to develop independent reasoning and analysis skills.

1. Introduction

1.1 Teaching Tissue Scaffolds

In tissue engineering, scaffolds are the mechanical support to tissue in three-dimensional extracellular matrix (ECM) structures and facilitate biochemical signaling cues to regulate cellular attachment, proliferation, migration, and differentiation. Native extracellular matrices are composed of hundreds of interacting proteins, including structural components such as collagen and elastin, adhesive glycoproteins such as fibronectin and laminin, and proteoglycans that regulate hydration and growth factor presentation (Figure 1b). Current proteomic analyses estimate that more than 300 distinct extracellular matrix and ECM-associated proteins contribute to tissue-specific architecture and function [6]. Because extracellular matrix composition governs cell fate decisions across developmental, pathological, and regenerative contexts, mastery of scaffold biology is foundational to biotechnology innovation, biomedical engineering design, regenerative medicine, and translational bioengineering applications [5]. However, the complexity of ECM composition, structure–function relationships, and dynamic remodeling processes makes this topic conceptually dense, and students often struggle to integrate molecular detail with systems-level understanding.

Teaching conceptually dense, interdisciplinary, and hierarchical content in bioengineering presents persistent instructional challenges due to wide variation in students' prior knowledge, disciplinary backgrounds, and conceptual readiness [1], [2]. Instructors are expected to advance learners toward expert-level reasoning within constrained instructional time, despite heterogeneous preparation and uneven exposure to foundational concepts [3]. Courses such as tissue engineering are particularly challenging because they require students to integrate knowledge spanning cell biology, materials science, mechanics, and clinical translation, often within a single instructional sequence [4]. Unlike foundational cell biology introduced earlier in students' education, extracellular matrix structure and function are typically first encountered at the college level, where students face numerous interacting components simultaneously. To address these instructional challenges, the next section outlines the theoretical framework of contingent instructional scaffolding that guides this study.

1.2 Contingent Instructional Scaffolding

In education and pedagogy, scaffolding is the instructional support given to a student to develop autonomous learning strategies. Scaffolding instruction is grounded in sociocultural learning theory and the concept of the Zone of Proximal Development (ZPD) articulated by Lev Vygotsky, provides a principled framework for addressing instructional challenges in complex domains by linking instructional support to learners' evolving capabilities [7]. The ZPD framework describes the range between what a learner can do independently and what the learner can do with guidance and support. Three performance levels were defined: Zone 1, learner cannot do, representing tasks the student is unable to complete even after instructional support; Zone 2, learner can do with guidance (ZPD), representing tasks completed correctly following instructional support support; and Zone 3, learner can do unaided, representing independent performance without support and reflecting established competence (Figure 1a).

Early theoretical and empirical work by van de Pol et al., established that learning is most effective when support is provided contingently, targeting what learners can accomplish with guidance, and is gradually withdrawn as independent competence develops [8] [9] [10], [11] (Figure 1c). Rather than static instructional structure, contingent scaffolding emphasizes adaptive support calibrated to learners' current understanding. Seminal tutoring studies demonstrate that scaffolding is effective when supports are responsive to learners' moment-to-moment needs, progressively reduced as competence increases, and ultimately removed to promote autonomous performance [8]. Subsequent syntheses of classroom interaction and computer-supported learning environments confirm that maintaining contingency and appropriately timing the fading of support remain central challenges in practical implementation, particularly in concept-dense instructional settings [10], [11].

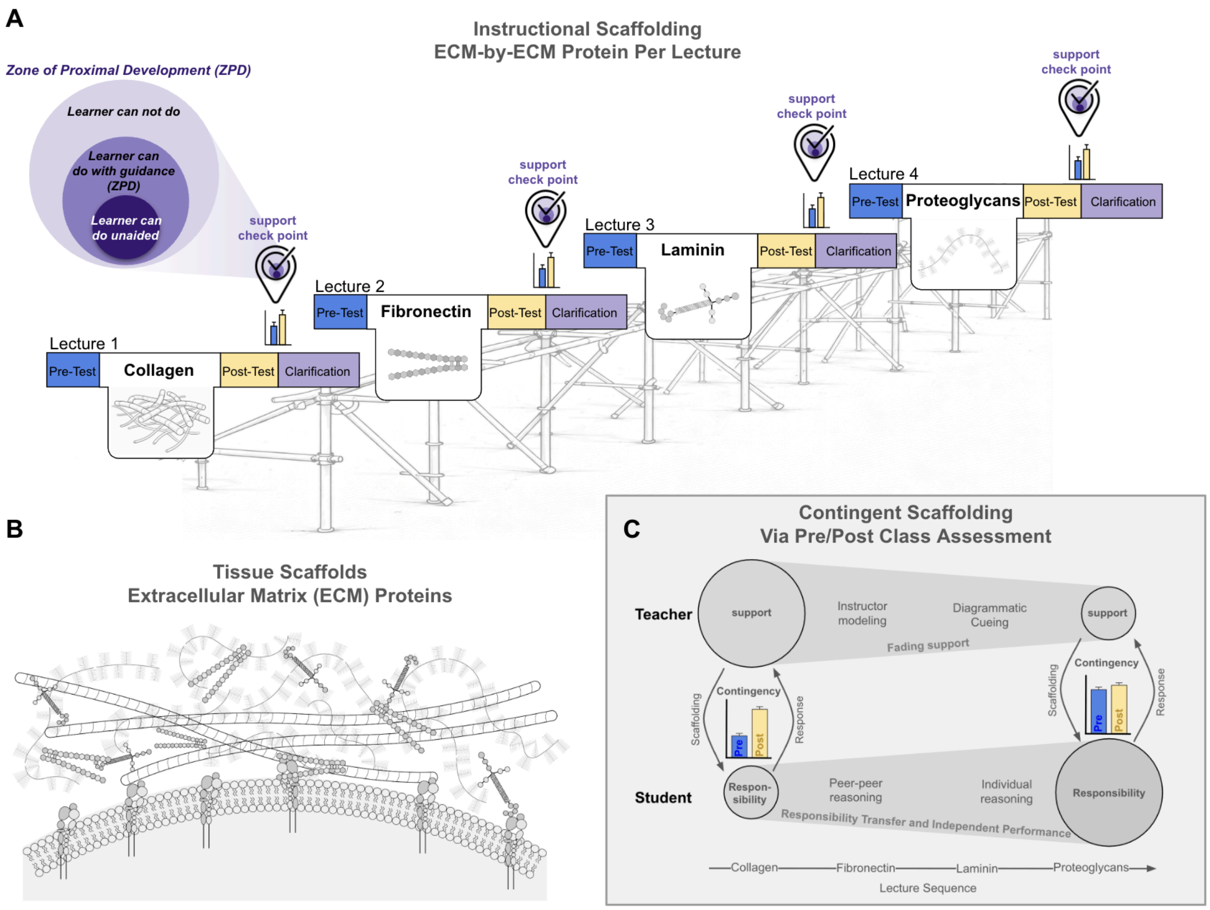

Figure 1. Contingent Instructional Scaffolding Across Extracellular Matrix (ECM) Tissue Scaffold Lectures. (A) ECM-by-ECM protein per lecture (Collagen, Fibronectin, Laminin, and Proteoglycans) using a repeated in-class assessment cycle: a brief Pre-Test (blue), targeted instruction with contingent prompts, a brief Post-Test (yellow), Clarification (purple), during which the instructor reviews class pre and post performance and addresses misconceptions or confusion before moving to the next lecture. Real-time support check points markers indicate where supports were calibrated based on Pre/Post Class assessments to infer the learner state within Zone of Proximal Development (ZPD) theory: Zone 1: learner cannot do, representing tasks the student is unable to complete even after instruction; Zone 2: learner can do with guidance (ZPD), representing tasks completed correctly following instructional support; and Zone 3 learner can do unaided: representing independent performance without support. Across lecture sequence, instructional support was progressively reduced as students demonstrated increased independent performance, reflecting a structured transfer of responsibility from teacher to student. The fading of support was contingent on observed learner performance via pre and post class assessment. (B) Representative ECM tissue scaffold proteins. (C) This study

implemented instructional scaffolding through instructor modeling and diagrammatic cueing, with support reduced contingently based on Pre/Post class assessments, illustrating the progressive transfer of responsibility and calibrated reduction of instructor support.

1.3 Instructional Scaffolding Application in Engineering Education

Engineering and bioengineering courses often require students to reason through concept-dense material that integrates multiple disciplinary frameworks. Within engineering education, scaffolded assessments have been used to structure students' engagement with complex tasks and to support the development of metacognitive regulation, teamwork, and analytical reasoning [12][13][14][15]. Sequenced exams, staged rubrics, structured deliverables, and guided collaboration illustrate how assessment tools can shape how students approach cognitively demanding work. In bioengineering education, scaffolding has been embedded within laboratory design, technical writing, evidence evaluation, and capstone experiences to support students' progression toward independent disciplinary reasoning [16][17][18][19][20][21][22]. These studies emphasize structured prompts, staged supports, and deliberate fading of guidance as competence develops. Across this literature, scaffolding is most effective when it is embedded in authentic disciplinary practice and gradually reduced so that responsibility shifts from instructor to student. Therefore, assessments are essential in informing students' level of understanding and helps guide adjustments in instructional support.

1.4 Contingent Scaffolding Challenges in Diagnosing ZPD Learning States through Homework Formative Assessment in GenAI-Aware Classrooms

The current challenge in contingent scaffolding application is the ongoing diagnosis of students' current level of competence to determine when and how instructional support should be adjusted or removed [11], [23]. Formative assessment is traditionally used to guide scaffolding decisions by showing what students can do on their own and where they still need support [24]. Viewed through a ZPD lens, formative assessment helps identify which learning zone students are in and guides calibration of instructional support. However, the growing use of generative AI (genAI) in formative assessments complicates this diagnostic process. When students rely on GenAI for out-of-class or unsupervised work, performance artifacts may no longer reflect students' independent understanding and may obscure their actual ZPD learning state. This distortion limits instructors' ability to accurately diagnose readiness for scaffold removal [26]. Consistent with this concern, prior work has shown that traditional homework performance is not reliably correlated with students' demonstrated learning gains, whereas frequent, low-stakes pre- and post-lecture self-assessments administered in class provide a more sensitive and valid measure of instructional impact and student understanding [25]. However, fewer studies describe how matched diagnostic pre/post class assessments can be systematically embedded within concept-dense engineering instruction in ways that remain scalable and instructionally coherent

[27]. Therefore, there is a need for reliable in-class assessment-driven diagnostics that allow instructional support to be adjusted based on student performance rather than homework.

1.5 Pre/Post In-Class Formative Assessments

Just-in-Time Teaching (JiTT) represents one of the earliest large-scale implementations of assessment-driven scaffolding in higher education [28]. Pre-class assessments are used in Just-in-Time Teaching to adjust instruction before class rather than simply to measure learning afterward. Across disciplines, this approach has been shown to increase student engagement and instructional alignment by allowing instructors to respond to documented gaps in understanding [29]. Studies that explicitly implemented paired pre- and post-lecture assessments consistently report positive learning effects, although the mechanisms and outcomes vary by field. In biomedical engineering education, pre/post lecture quizzes and formative assessments have been associated with measurable gains in conceptual understanding and learning outcomes compared to traditional lecture [30] or homework formats [25], while also supporting active learning environments. In computer science and programming education, pre/post testing has revealed heterogeneous learning gains across students, underscoring the need for instructional flexibility to address diverse learning needs [31]. In psychology education, mastery-oriented pre/post quizzes have been shown to focus student attention on key lecture concepts and to predict course examination performance [32]. In medical and clinical education, pre/post lecture assessments have led to statistically significant improvements in technical and procedural knowledge in specialized instructional settings [33]. Finally, in universal design-focused engineering education, pre/post assessments combined with metacognitive strategies have improved confidence and performance among first-generation and female students, suggesting potential benefits for retention and inclusivity [34]. Prior work of pre- and post-lecture assessments across disciplines, largely treats these assessments as evaluative tools used to document learning gains or justify instructional interventions.

1.6 Motivation: Pre/post Class Assessment-Driven Contingent Scaffolding in Tissue Engineering Education

Previous literature on pre- and post-class assessments provides limited clarity on how these assessments function within theory-driven contingent scaffolding frameworks, particularly in identifying students' current learning state through a Zone of Proximal Development (ZPD) lens. However, these assessments have the potential to serve as diagnostic indicators of learner readiness, informing when instructional support should be maintained, adjusted, or withdrawn in concept-dense curricula.

In this study, low-stakes in-class pre- and post-assessments are embedded within a staged instructional design that introduces one extracellular matrix (ECM) protein per lecture. This structure allows instructional support to be calibrated to a focused conceptual target and aligned with demonstrated student performance. By positioning pre- and post-assessments as mechanisms for diagnosing learning states and guiding contingent support, this study extends scaffolding theory into a conceptually dense bioengineering context.

2. Methods

2.1 Course Context

The scaffolding instructional intervention was implemented in a ten-week quarter graduate tissue engineering course of a multi-quarter sequence, which focused on extracellular matrix (ECM) scaffolds. Students had completed a prerequisite foundation course on tissue engineering and regenerative medicine with cell biology focus. This scaffolding intervention study took place from 2023 to 2025. The overall course structure was originally designed prior to 2023 and iteratively refined over more than ten years.

2.2 Instructional Scaffolding Design

Instructional scaffolding was implemented across the first four consecutive lectures spaced out evenly within the tissue engineering course. From the first lecture, students were provided with an instructional roadmap that outlined a staged progression through ECM concepts, keeping students oriented toward the learning goal and preventing off-track work. Instruction followed a deliberate scaffolded sequence in which one ECM protein was introduced per lecture, beginning with collagen and proceeding through fibronectin, laminin, and proteoglycans. Each lecture consistently addressed the same conceptual dimensions: biosynthesis and assembly, structural and mechanical properties, biological function, associated diseases, laboratory synthesis, and applications in tissue engineering and regenerative medicine.

The instructional scaffolding design followed a structured progression consistent with gradual release of responsibility, moving from instructor modeling to student independent reasoning. Scaffolding was executed through instructor modeling, prompt specificity, diagrammatic cueing (Examples in supplementary material figure), and opportunities for guided peer discourse. Instructor modeling involved explicit demonstration of expert reasoning processes, including think-aloud explanations of how specific ECM structural domains relate to biological function, stepwise interpretation of diagrams (e.g., identifying a binding domain and tracing its mechanical role). Verbal justification of how molecular features such as crosslinking regions contribute to tensile strength or adhesion. Prompt specificity provided students structured reasoning cues; high-specificity prompts included directed questions such as “Which structural domain enables adhesion?” or “How does this configuration differ from collagen in load-bearing capacity?”, whereas reduced-specificity prompts broader analysis such as “Explain how structure supports function.” Diagrammatic cueing referred to the presence or absence of visual supports in instructional materials, including fully labeled ECM components, highlighted structural regions, and annotated arrows linking structure to function in the first lecture. These visual guides were removed in later lectures. Guided peer discourse involved structured think–pair–share reasoning activities in which students articulated structure–function relationships aloud, compared interpretations of specific domains, and justified their analyses before whole-class discussion.

Lecture notes were made available prior to each session, and primary literature readings were assigned to provide research exposure. Prior knowledge from earlier ECM proteins is explicitly leveraged in subsequent lectures, and instructional support is gradually reduced as students demonstrate increasing competence. Collagen was introduced first and serves as the entry point to the scaffolded sequence rather than a scaffolded outcome itself. Because collagen is the initial lecture in which the scaffold is established, there is no prior scaffolded baseline against which pre/post class change or Zone of Proximal Development (ZPD) shifts can be meaningfully assessed. For this reason, collagen is not included in the pre/post learning gain or ZPD analyses shown in Figures 2 and 3. Subsequent ECM proteins (fibronectin, laminin, and proteoglycans) are analyzed instead, as they explicitly build on the collagen scaffold and therefore allow measurement of scaffolded knowledge transfer and support fading. The implications of this design choice for pre/post and ZPD analysis are detailed in the Methods section.

2.3 Pre/post Class Assessment for Contingent Scaffolding

Central to contingent instructional scaffolding in the course design [10], [11], matched pre- and post-class assessments were administered during class time to monitor student understanding and inform the timing and degree of support reduction. Pre- and post-lecture assessments were used to monitor patterns of student performance across lectures. Observed trends informed instructional emphasis and clarification where needed. While the overall progression involved planned reduction of external supports, instructional attention remained responsive to areas of difficulty identified in the diagnostic data. For each lecture, students completed a brief pre-class check immediately before instruction and an identical post-class check immediately after instruction. Four repeated questions focused on the ECM protein structure and function for that lecture and spanned multiple levels of Bloom's taxonomy. The pre-class check functioned as a just-in-time diagnostic, allowing the instructor to identify prevalent misconceptions and tailor modeling, explanations, examples, and emphasis during the lecture. The post-class check verified consolidation of understanding and provided immediate evidence of whether instructional support could be faded or whether targeted clarification was required before advancing to the next topic. A clarification checkpoint was conducted prior to moving to the subsequent lecture to address persistent errors.

Initial implementations used Poll Everywhere for in-class polling; however, longitudinal analysis across offerings required substantial manual processing. Later offerings used www.PrePostClass.com, developed by the instructor to provide built-in pairing and analytics for pre and post responses. Across all years, the same questions were used for the pre and post checks within a lecture, with prompts, timing, and imagery held constant from 2023 and 2025 to enable aggregation.

To examine changes in dispersion across cohorts, we calculated pre- and post-instruction means, standard deviations, and sample variances for each ECM section. Variance was used to evaluate whether performance spread contracted or expanded following instruction.

2.4 Zone of Proximal Development (ZPD) Learning States Analysis

Student pre/post class responses were interpreted within a Zone of Proximal Development (ZPD) framework consisting of three performance levels: Zone 1: learner cannot do, representing tasks the student is unable to complete even after instruction; Zone 2: learner can do with guidance (ZPD), representing tasks completed correctly following instructional support; and Zone 3: learner can do unaided, representing independent performance without support.

To determine whether gains exceeded regressions within the same students, we applied McNemar's test for paired binary outcomes. Matched pre- and post-test responses were coded as correct (1) or incorrect (0) to enable this within-subject comparison. For each ECM protein and course offering, paired responses were summarized in a 2×2 contingency table using the following classification:

a: 1→1, correct at both pre and post, transition was interpreted as Zone 1 "learner can do unaided."
b: 1→0, regression, interpreted as Zone 3 "cannot do with support"
c: 0→1, gain during the instructional session, interpreted as Zone 2 "learner can do with guidance (ZPD)"
d: 0→0, incorrect at both pre and post, interpreted as Zone 3 "cannot do with support"

When the number of discordant pairs was small, we used the exact binomial version of the test. When discordant pairs were more numerous, we used the chi-square approximation with continuity correction. All tests were two-sided and evaluated at a conventional significance level. As a paired-change effect size, we reported the odds ratio of discordant pairs, defined as the ratio of regressions to gains. Values less than one indicate net improvement, while values greater than one indicate net regression.

2.5 Design and Outcomes

The primary endpoint was within-student change in correctness via the discordant pairs b and c from pre to post on the focal identification item for that lecture.

Secondary endpoints included the pre–post gap in percentage correct by ECM protein averaged across years, and the distribution of ZPD learning states. We report the ZPD index as

$$\text{ZPD Index}=c/(c+d)$$

which represents the proportion of students who were initially incorrect and demonstrated learning during the instructional session. Responses of “I do not know” were coded as incorrect.

2.7 Zero Cell Handling

When one of the discordant McNemar cells ($b = 1 \rightarrow 0$ or $c = 0 \rightarrow 1$) contains zero observations, the odds ratio becomes undefined due to division by zero. In such cases, we applied the Haldane–Anscombe correction by adding a small constant to both discordant cells before calculating the odds ratio. Odds ratios and log-scale confidence intervals were computed using the corrected counts and then reported on the original odds ratio scale.

2.8 Reporting

For each ECM protein, we report the McNemar p value, the corrected odds ratio with confidence interval, and the gain dominance metric $c/(b+c)$. Together, these statistics provide an inference on within-student change, its direction, and its magnitude, while remaining robust in cases with small or zero discordant counts.

3. Ethical Approval

This work includes results from course assessments and participant feedback collected for educational improvement. All participants received an informed consent statement approved by the University of California San Diego Institutional Review Board (IRB-807339). The project was reviewed by the IRB and determined to be exempt.

4. Results and Discussion

4.1 Class Learning Gains via Pre/Post Class Assessments

To characterize learning gains, we used real-time in-class assessment across ECM topics by comparing matched pre and post checks administered under identical scaffolding instruction over three course offerings from 2023 to 2025 with an average of 50 participating students (Fig. 1). An example measurement item uses an unannotated ECM tissue scaffold image and asks students to identify the specific ECM protein that will be covered in the lecture that day. We measured the percent of students answering correctly on a timed pre check and again on a matched post check for each lecture. Pooled across years, post accuracy exceeded pre accuracy for all topics (Fig. 2a). Across the ordered sequence of topics taught one per lecture, the difference between pre and post progressively decreased, consistent with growing unaided recognition and a shrinking pool of students positioned for large immediate gains. The largest and only statistically significant increase occurred for Fibronectin (pre 47–50% to post

~78–80%, p = 0.024). Laminin showed a moderate, non-significant rise (p = 0.154), and Proteoglycan showed a small, non-significant change (p = 0.500) (Fig. 2b).

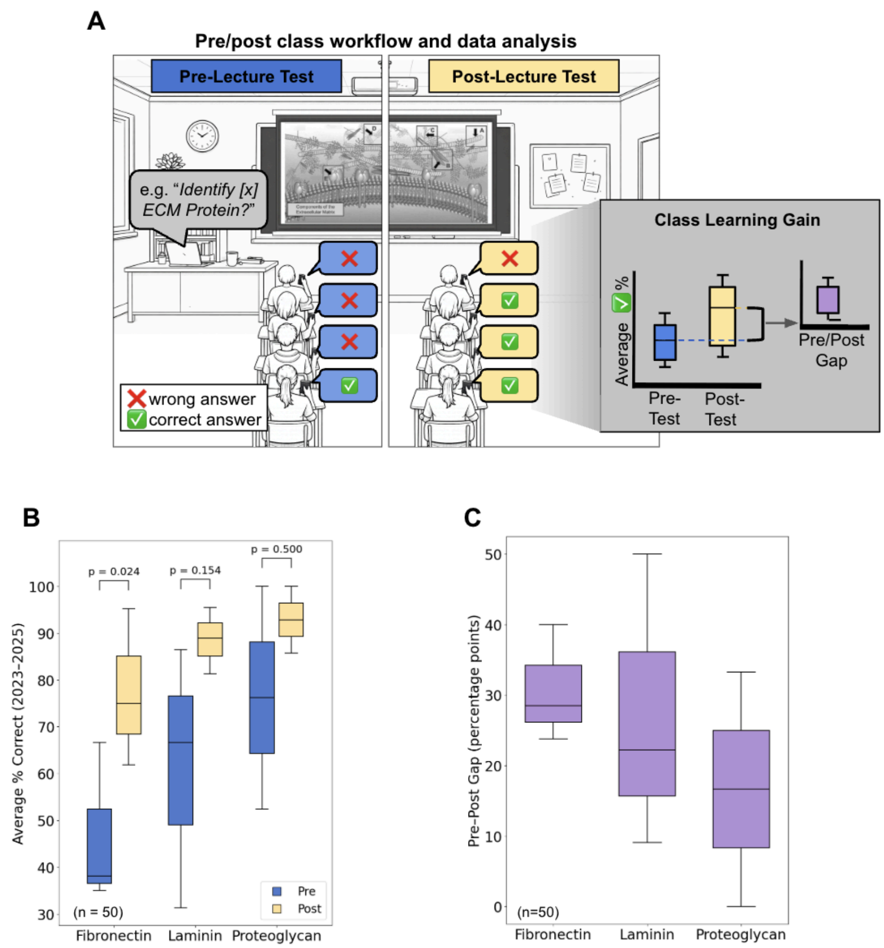

Figure 2. Pre/Post Class learning gains across ECM lectures, 2023–2025. (a) Schematic of Pre/Post class test workflow and data analysis. Each lecture begins with a pre-lecture test, followed by instruction, and then a post-lecture test. An example teaching activity for the scaffolding support check asks students to identify the target ECM protein in a figure. The inset illustrates cohort-level change in average percent correct answers on the pre- and post-tests and the pre–post gap. (b) Average percent correct for ECM protein identification and characterization, computed as the mean of course-year (2023-2025) averages Fibronectin, Laminin, Proteoglycan with an average sample number of participants (n = 50). Error bars show the standard deviation (SD) across years. Above each pair is the paired t test across years for that protein comparing year-level pre vs. post means (Fibronectin p = 0.024, Laminin p = 0.154, Proteoglycan p = 0.5). (c) Average pre–post gap (average post minus pre, percentage points) for each ECM protein, reported as the mean of year-level gaps with SD across years.

Across the three ECM components, post-instruction variance decreased relative to pre-instruction levels, indicating greater cross-cohort alignment following the scaffolded sequence (Table 1). For Fibronectin, variance declined modestly from 300.79 to 281.78, suggesting that while cohort performance remained somewhat dispersed, variability showed slight convergence after instruction. In contrast, Laminin exhibited a substantial reduction in variance, from 774.03 to 50.91, reflecting a marked shift from widely dispersed cohort performance pre-instruction to tightly clustered outcomes post-instruction. This pronounced contraction in spread indicates that outcomes not only improved in mean level but also became more concentrated around a shared central performance range. The consistent decrease in variance across proteins indicates that instructional gains were accompanied by reduced dispersion, with performance becoming more tightly clustered. Thus, beyond increases in mean scores, the post-instruction pattern reflects greater alignment around a common level of conceptual understanding. While these variance estimates are descriptive due to the limited number of cohorts, the consistent contraction in spread supports the interpretation that learning outcomes became more uniform following the scaffolded sequence.

Protein	Phase	Mean (%)	SD (%)	Variance
Fibronectin	Pre	46.60	17.34	300.79
Fibronectin	Post	77.37	16.79	281.78
Laminin	Pre	61.47	27.82	774.03
Laminin	Post	88.57	7.13	50.91
Proteoglycan	Pre	76.20	33.65	1132.88
Proteoglycan	Post	92.85	10.11	102.25

Table 1. Pre- and post-instruction mean, standard deviation (SD), and variance for each ECM protein across cohorts.

To probe the magnitude of learning gains across years, we summarized the pre–post gap for each topic (post minus pre, averaged over available years with SD) shown in Fig. 2c. The average gap decreased across the sequence, from 31 percentage points for the first topic to 27 points for the middle topic and 16 points for the final topic, with wide year-to-year variability. This pattern is consistent with consolidation of recognition as scaffolded instruction progresses, where a larger fraction of students are already correct at the start of later lectures and fewer are positioned for large immediate gains.

Learning gains increased and pre–post gaps decreased across the ordered ECM-by ECM protein in the lecture sequence, consistent with scaffolded instruction that builds cumulative recognition while shrinking the pool of students positioned for large immediate gains. Teaching one ECM protein per lecture acts as deliberate scaffolding that chunks complex content into manageable units, lowers extraneous load, and makes misconceptions visible in real time through paired pre and post checks. Used this way, formative assessments provide contingent support and structured opportunities for practice, then enable a planned transfer of responsibility as competence stabilizes and hints are withdrawn. Across sessions, this cycle strengthens metacognitive monitoring, sustains engagement, and promotes independent recognition and application without overwhelming learners with simultaneous constructs.

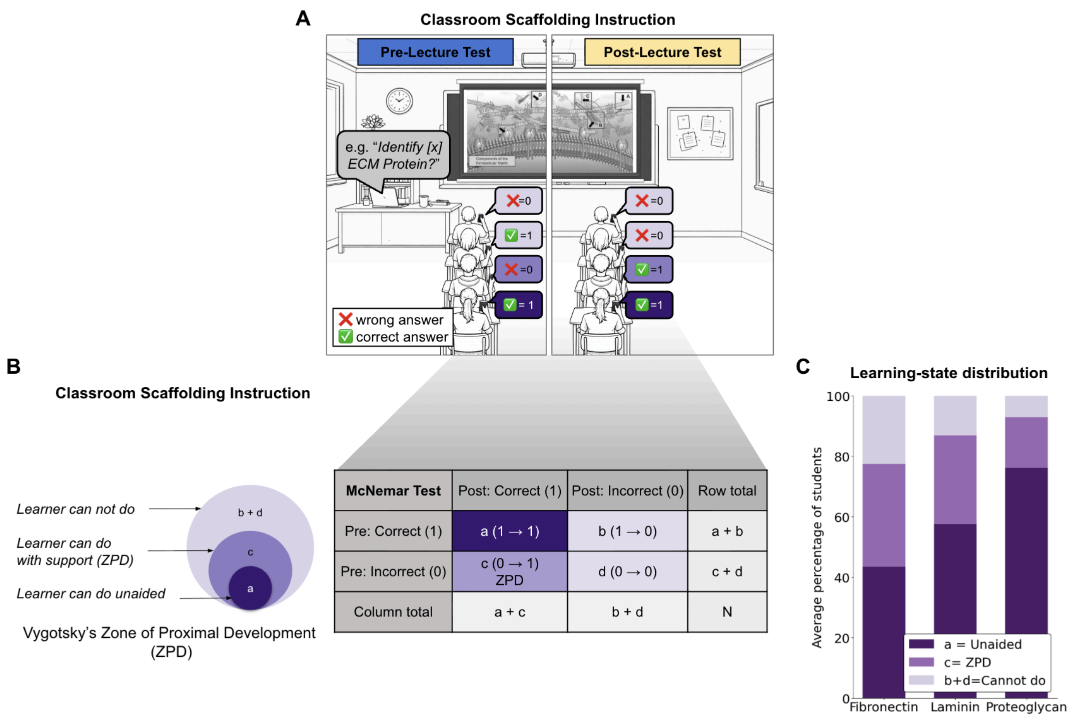

Figure 3. ZPD framing of the pre/post classroom assessment and learning-state distributions. (a) Classroom scaffolding workflow used in each lecture. Students completed a matched pre and post in-class check on the same item. Each response was coded as correct (1) or incorrect (0), and pre–post responses were later paired at the student level to classify within-lecture learning states. (b) Vygotsky's Zone of Proximal Development (ZPD) overview with the paired pre/post 2×2 table defining learning states: unaided success a = 1→1, regression b = 1→0, ZPD gain c = 0→1, and cannot do d = 0→0. Row and column totals correspond to post and pre outcomes, respectively. (c) Learning-state distribution by ECM proteins (Fibronectin, Laminin, Proteoglycan), averaged across years (2023–2025). Bars show the mean percentage of students classified as unaided = a , ZPD = c, and cannot do= b+d.

4.2 Learning-state analysis in a ZPD frame

To further explain why the pre to post gap shrinks across the sequence, we classified each matched response into four learning states that mirror instructional support needs (see methods section 2.4) (Figure 3a). Changes in student performance were evaluated between pre and post for each protein concept via McNemar test, a nonparametric test for paired binary outcomes (Fig. 3b).

The McNemar test evaluates whether the number of students transitioning from incorrect to correct responses (c=0→1) differs from those transitioning from correct to incorrect responses (b=1→0), thereby isolating instructional effects while accounting for within-student pairing.
We then averaged c/(b+c) by protein, computed a pooled exact McNemar test on summed discordants, and reported the continuity-corrected odds ratio; Haldane–Anscombe corrected odds ratio OR= (b+0.5)/(c+0.5), where values below 1 favor improvement (Table 2).

Across years, improvement among discordants was strongest for Fibronectin and Proteoglycan. Fibronectin showed a mean ZPD share c/(b+c) of 1.00 with zero 1→0 losses in any year, a highly significant pooled McNemar result (b=0, c=22, $p=4.8\times10^{-7}$), and OR=0.022. Proteoglycan likewise had a mean ZPD share of 1.00 in the one evaluable year with discordants (b=0, c=7, p=0.0156), OR=0.067.

Laminin exhibited consistently high ZPD movement among discordants, mean c/(b+c) = 0.905±0.164 across three years, with a significant pooled test (b=2, c=19, $p=2.2\times10^{-4}$) and OR=0.128, indicating far more 0→1 gains than 1→0 regressions. Collagen improved overall but was mixed across years, mean c/(b+c)=0.708±0.505, with a nonsignificant pooled test (b=7, c=13, p=0.263) and OR=0.556, reflecting both gains and some early regressions. The normalized across-year view shows that most students who changed their answer did so in the ZPD direction, especially for Fibronectin and Proteoglycan, with Laminin close behind.

ECM protein	**a** (2023-2025)	**b** (2023-2025)	**c (ZPD)** (2023-2025)	**d** (2023-2025)	**Total discordant** (b+c)	**Mean c/(b+c) across years ± SD**	**Exact McNemar p (pooled)**	**OR**
Fibronectin	27	0	22	13	22	1.000 ± 0.000	4.8e-07	0.022
Laminin	33	2	19	5	21	0.905 ± 0.164	2.2e-04	0.128
Proteoglycan	23	0	7	2	7	1.000 ± 0.000	0.0156	0.067

Table 2. Learning-state counts and accuracies by ECM protein and year. Pooled learning-state and McNemar summary by ECM protein across years. For each protein, the table reports pooled counts for the four pre–post states a = 1→1 (unaided correct), b = 1→0 (regressed), c = 0→1 (improved with support, ZPD), and d = 0→0 (remained incorrect), plus the number of years that contributed discordant pairs. Mean c/(b+c) ± SD summarizes, across years, the share of improvements among discordants. Total discordant is b+c pooled across years. Exact McNemar p is computed on pooled discordants. OR is the Haldane–Anscombe corrected odds ratio calculated as (c+0.5)/(b+0.5).

To complete a more granular analysis of student's learning states, we recast matched responses into three pooled learning states (Fig. 3c). Aligned with instructional support needs: a = unaided (correct at both pre and post), c = ZPD (moved from incorrect at pre to correct at post with instructional support), and b+d = cannot answered correctly with instructional support (regressed or remained incorrect). We then averaged state proportions across years for each topic, excluding the first-lecture baseline. Across the ordered topics, the unaided (a) share rose steadily from roughly 43.4% to 57.6% to 76.2%, while ZPD (c) declined from 34.0% to 29.3% to 16.7%, and cannot do even with support (b+d) fell from 22.6% to 13.1% to 7.2%. This pattern indicates consolidation of independent performance as the scaffold progresses: early lectures place more students within instructional reach, mid-course shows partial consolidation, and by the final topic most students answered correctly without support. In short, ECM-by-ECM protein per lecture scaffolding shifts the cohort from c toward a, while b+d contracts, consistent with the decreasing pre–post gaps observed earlier.

The third lecture on Laminin showed fewer ZPD transitions than the other proteins, which may be related to its variance pattern discussed in the previous section. It had the highest pre-instruction variance (774.03) and the strongest reduction post-instruction (50.91), reflecting a 93.4% contraction in dispersion. This suggests that instruction reduced unevenness across cohorts and brought performance to a more consistent level. When baseline performance is widely distributed, some portions of the cohort may already be performing at higher levels, limiting observable upward movement within the ZPD framework. Thus, the comparatively lower ZPD count for Laminin may reflect convergence of student performance rather than reduced instructional effect, indicating a shift toward more independent reasoning rather than reliance on instructional support.

The learning-state view shows that most students who changed their answers did so in the productive direction, with improvements ($0 \rightarrow 1$) consistently outnumbering regressions ($1 \rightarrow 0$). This pattern, together with odds ratios below one and significant pooled McNemar results for several topics, suggests that the scaffolded, one-protein-per-lecture design delivers real instructional lift. Variability for the first lecture likely reflects a true baseline condition with no prior scaffold to build on, after which state c contracts and state a expands as concepts consolidate and ceiling effects emerge. For engineering education, this means that tightly scoped, in-class formative checks can both trigger just-in-time support early in a sequence and document the planned shift toward independent performance later. The approach is practical at scale and yields interpretable learning state metrics that can guide when to reteach, when to fade supports, and how to pace subsequent topics.

5. Limitations

This study does not include an external control condition in which ECM proteins were taught without scaffolded sequencing. However, the ordered design provides an internal reference point.

The first lecture establishes the scaffold without prior scaffolded exposure, whereas subsequent lectures build on it while holding instructional format and assessment structure constant. Although comparison within this scaffold sequence strengthens interpretation, it does not replace a randomized control. In addition, some yearly analyses included small numbers of discordant pairs, indicating the need for multi-item assessments and replication across institutions to improve generalizability.

6. Conclusion

An instructional scaffold that teaches one ECM protein per lecture, pairs pre and post checks with contingent prompts, and then fades support produced consistent gains in accuracy and movement into the ZPD state across offerings, while keeping assessments in class minimized risks of GenAI contamination and preserved a clean readout of independent performance. The pattern replicated across years, indicating reliability in GenAI-aware settings and practicality for conceptually dense material. By calibrating support to students' ZPD and withdrawing it as understanding consolidates, the cohort shifted from needing guidance to unaided performance, and the shrinking pre/post gap across the sequence tracked this consolidation. This workflow yields actionable checkpoints for instructors: diagnose with a pre check, teach to observed gaps, verify with a matched post check, and include a brief clarification step before advancing. The approach is scalable across institutions, and offers a concrete template for improving equity and rigor by calibrating the fading support only when learners can succeed unaided. Future work should adopt multi-item question banks per concept, direct comparison of contingent scaffolding study of in-class versus homework assessment, examine retention and transfer beyond immediate post checks, and test generalizability across courses and institutions.

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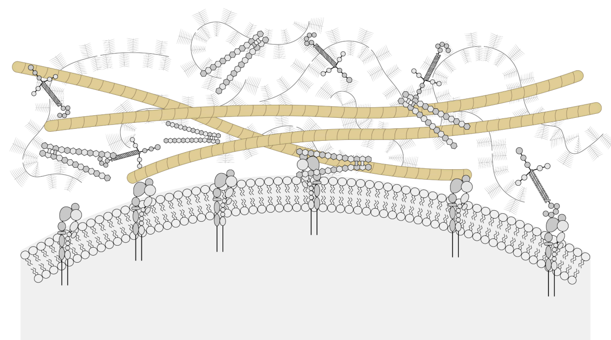
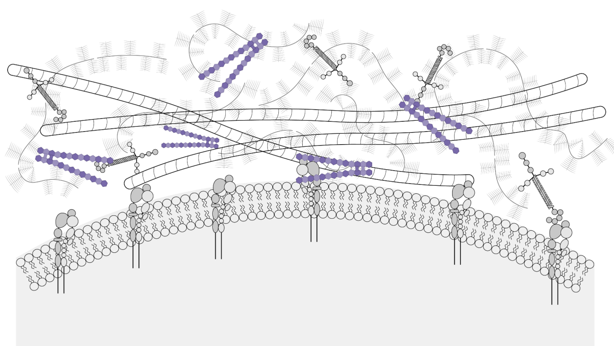
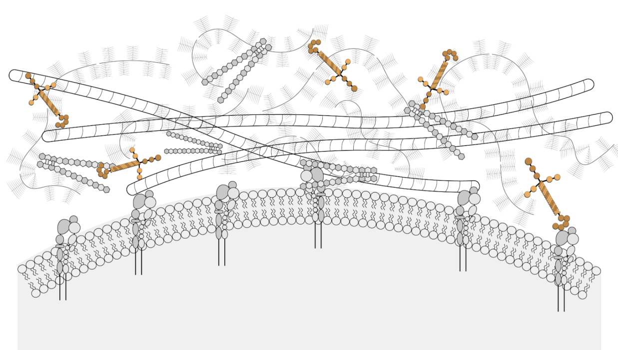
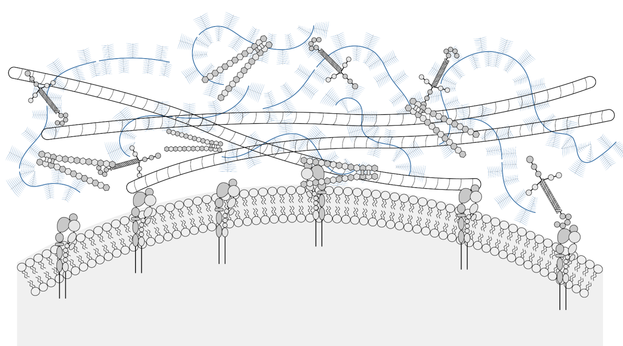
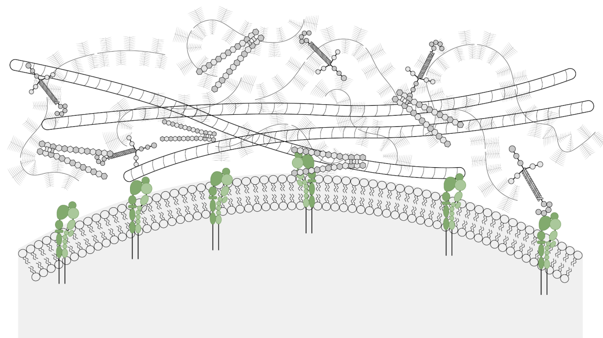
Supplementary Material











Supplementary Figure. Diagrammatic cueing in ECM instruction. Selective color highlighting is used to direct student attention to one extracellular matrix component at a time while the rest of the diagram remains visually muted. The cues are gradually removed across panels, supporting a transition from guided attention to independent structural reasoning.