

# Retained Protein and Accessible Function at Biological Interfaces

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Author approved release edition. Literature analysis and proposed research; no new VARUNÉ experimental results are reported.

## Abstract

A protein can remain on a surface whilst losing the interaction for which it was deposited. Conversely, adsorption can enhance a useful response, and additional protein can sometimes improve function. This report examines how to distinguish these outcomes without treating surface loading as a universal performance measure. Three chapters of *Biomaterials Science* are connected with primary studies of adsorbed enzymes, fibrinogen, fibronectin, antibody capture, and Antarctic-derived antibiofilm proteins. The evidence supports separating retained molecular amount, accessible presentation, reaction and transport conditions, and the observation used to infer performance. The contribution proposed here is a decision-oriented comparison: identify which measurements would lead to a different choice between improving deposition, modifying presentation, reducing transport resistance, or replacing the candidate. Particular attention is given to spatial heterogeneity, conditioning by complex fluids, and the difference between soluble activity and coating performance. Four equations formalise quantities and assumptions; none establishes a mechanism for an untested VARUNÉ material. A staged research design is proposed around one accessible protein and one intended use. Its output would be an interpretable account of why a surface succeeds or fails, with independent preparations and unsuccessful results retained. No new experimental data, clinical efficacy, commercial rights, or confirmed partnership are reported.

## 1 Research question and contribution

The central question is: **when a protein-containing surface loses useful performance, what evidence can distinguish loss of material from loss of accessible function?** This is consequential because the same weak functional result can suggest incompatible engineering responses. Stronger attachment addresses release, but could worsen orientation. More protein addresses insufficient loading, but could increase crowding. A more protective matrix could preserve a molecule during storage whilst obstructing its substrate during use. Improving one property without identifying the limiting step can therefore produce an expensive version of the same failure.

The distinction is established in biomaterials research; it is not presented here as a new physical principle. The proposed contribution is its application to a VARUNÉ research decision: which explanation remains plausible, which additional observation would discriminate it, and what change would then be justified? This connects the wider research series on production, purification, and formulation to the final encounter between material and environment. A protein's identity before deposition does not completely specify the interface encountered by its target. [\[1-3\]](#)

Three applications are compared, with their mechanisms kept separate. An immobilised enzyme must admit substrate and release product. A capture surface must recognise an intended target whilst controlling nonspecific binding. An antibiofilm coating must change a defined microbial outcome under relevant exposure. Useful performance in one application cannot validate another. The shared analytical problem is the distance between what remains physically present and what the measurement can establish about function.

## 2 Scope and source method

This is a targeted critical narrative synthesis, not a systematic review or meta-analysis. The literature was selected to contrast mechanisms and expose alternative interpretations rather than estimate a pooled effect. The starting material was the supplied fourth edition of *Biomaterials Science*, using chapters 1.4.3A, 1.4.4, and 2.1.2. Selected passages were re-examined for this report: hydration and defects in nonfouling surfaces; attachment, release, ligand presentation, and mass transfer; and competitive adsorption, spreading, and accessible binding regions. These chapter texts had been examined in the preceding research instalment. This report does not add three new completed chapters to the cumulative reading count. [1-3]

Targeted online searches on 28 October 2026 included “PsyOmp38 silicone continuous flow antibiofilm 2025”, “immobilized enzyme orientation surface loading retained protein activity”, “fibrinogen adsorption conformation platelet adhesion surface density”, “antibody orientation immobilization antigen binding density”, and searches for exact titles identified from the chapters. Primary publisher pages, PubMed records, and accessible author-manuscript passages were prioritised. Studies were retained when they compared amount with function, manipulated presentation, or revealed a limitation relevant to an interface decision. General reviews were used for method interpretation, not as substitutes for candidate results.

Seven empirical studies are discussed directly. Access was uneven: publisher full text and targeted methods/results were available for the two antibiofilm papers; indexed manuscript passages supplemented the fibrinogen study; several older enzyme and antibody papers were available principally as abstracts or previews. Those restrictions are recorded in the references. No exhaustive supplement audit, complete raw-data reanalysis, or quantitative risk-of-bias scoring was performed. Reported correlations are reproduced only as source-specific observations. There is no pooled comparison across proteins, surfaces, or assay conditions.

## 3 What is actually being retained

A retained amount is meaningful only after specifying what it counts. A label can remain after a protein has fragmented. A surface-sensitive mass response may include coupled solvent or adsorbed environmental constituents. An antibody signal detects the regions recognised by that reagent, not necessarily all molecules of the intended protein. Contact angle detects wetting behaviour and is influenced by more than one possible surface change. Consequently, “the protein is still there” requires an operational definition and an analytical recovery check. [1-3]

For a defined parent protein, a simple retention ratio is useful:

$$R_{\Gamma}(t) = \frac{\Gamma_{\text{parent}}(t)}{\Gamma_{\text{parent}}(0)}$$

Equation 1. Retention of the specified parent protein relative to the initial measured surface state.

Here  $\Gamma_{\text{parent}}$  is the independently measured amount of the specified protein per area, for example  $\text{mol m}^{-2}$  or  $\text{mg m}^{-2}$ , and  $t$  is elapsed exposure time in a stated unit.  $R_{\Gamma}$  is dimensionless and requires a nonzero baseline. The numerator and denominator must use the same operational definition, recovery correction, and surface area convention. This is a definition, not an adsorption model. It does not imply intact structure, correct orientation, or useful function. If the assay cannot distinguish parent protein from retained fragments, the result must instead be labelled retention of that assay's signal.

The baseline also matters. Comparing material after fluid exposure with the amount nominally pipetted onto a surface confounds deposition yield with later retention. For mechanistic work, the baseline should normally follow preparation and the specified initial wash or equilibration. Manufacturing yield remains a separate quantity worth reporting. This prevents a poorly depositing process from appearing stable merely because the small fraction that survives initial handling is strongly bound.

### 3.1 Evidence from enzymes and capture surfaces

Fears and Latour examined lysozyme, xylanase, and glucose oxidase on chemically different model surfaces, combining structural measurements with activity assays. Their reported findings are especially useful because structural change and activity did not have a single relationship: small-enzyme activity varied despite relative structural stability, whilst adsorption-associated conformational change could enhance glucose oxidase activity. Thus, “native-like” and “useful” cannot be treated as synonyms for every interface. The direct evidence concerns these enzymes and model surfaces; it does not identify the preferred presentation of PsyOmp38. [4]

Shen and colleagues approached orientation more directly through site-specific attachment of nitroreductase variants. Surface-sensitive spectroscopy was combined with activity measurement, and attachment position was associated with orientation and activity. This supplies a stronger design logic than attributing every activity difference between two supports to orientation. However, a modified attachment site can also change other properties, so transfer to a new candidate still requires characterisation of the soluble variant and the immobilised state. The accessible abstract supports the directional relationship; detailed effect sizes and uncertainty are not reproduced here. [8]

Pei and colleagues provide a useful counterweight to the claim that loading is uninformative. In their antibody immobilisation study, antigen-binding capacity often followed the immobilisation response. Yet binding per immobilised antibody differed, and orientation-sensitive probing helped explain an exception. Loading can therefore be a useful process metric within a specified antibody, surface, and preparation window. It becomes unreliable when that empirical relationship is silently generalised beyond the conditions under which it was established. [7]

### 3.2 Evidence from proteins that mediate cell contact

Sivaraman and Latour separated the amount and conformation of adsorbed fibrinogen by varying both solution concentration and surface chemistry. Across their combined dataset, platelet adhesion

correlated strongly with adsorption-induced unfolding, reported as  $r^2 = 0.96$ , and poorly with amount,  $r^2 = 0.04$ . The valuable lesson is the experimental separation of otherwise correlated variables. These are in vitro, system-specific relationships; they neither establish universal causality from correlation nor predict clinical thrombosis for another material. [5]

Keselowsky and colleagues linked surface chemistry to fibronectin presentation, integrin binding, and cell adhesion. Different receptor interactions did not follow an identical ranking. This exposes a further problem with a single “active fraction”: a protein can be accessible to one interaction and less accessible to another. A functional state must be defined relative to the relevant binding partner and biological outcome. Antibody recognition is informative when its relationship to that outcome is demonstrated, not simply because it yields a convenient signal. [6]

## 4 A quantitative description with explicit limits

For a simple surface enzyme, a useful starting model is:

$$j_{\text{obs}} = \Gamma_{\text{cat}} k_{\text{cat}} \frac{C_s}{K_M + C_s} \eta$$

Equation 2. A conditional description of enzyme flux from a competent and appropriately presented catalytic population.

The observed product flux  $j_{\text{obs}}$  has units  $\text{mol m}^{-2} \text{s}^{-1}$ .  $\Gamma_{\text{cat}}$  is the amount of chemically competent and appropriately presented enzyme per projected area,  $\text{mol m}^{-2}$ ; it excludes geometrically occluded sites but does not itself incorporate substrate concentration gradients.  $k_{\text{cat}}$  is the relevant turnover number,  $\text{s}^{-1}$ ;  $C_s$  is the substrate concentration at the outer surface of the enzyme-bearing layer,  $\text{mol m}^{-3}$ ; and  $K_M$  uses the same concentration unit. The dimensionless effectiveness factor  $\eta$  averages internal transport effects relative to the rate that the whole competent population would produce at that boundary concentration. For a nonporous monolayer whose sites all experience  $C_s$ ,  $\eta = 1$ . This avoids counting an internal concentration loss both in  $C_s$  and again in  $\eta$ . The model applies a standard Michaelis-Menten form to a surface population, assuming compatible kinetics and explicit treatment or absence of product inhibition, changing enzyme state, and cofactor limitations. [2,13]

The equation is useful mainly because it identifies quantities that an observed rate alone cannot determine.  $\Gamma_{\text{cat}}$  is not automatically equal to total retained protein.  $C_s$  is not automatically equal to the bulk concentration; internal gradients, if relevant, enter  $\eta$  rather than being counted a second time in  $C_s$ . A rate reduction could reflect fewer competent sites, slower turnover at those sites, or reduced substrate delivery. Fitting all these parameters to the same small rate dataset would produce an apparently detailed explanation with insufficient independent information.

If external delivery is important, a simple comparison of reaction demand and transport capacity is:

$$\text{Da}_{\text{ext}} = \frac{\Gamma_{\text{cat}} k_{\text{cat}} C_b / (K_M + C_b)}{k_m C_b}$$

Equation 3. A dimensionless external transport diagnostic evaluated using bulk substrate concentration.

$C_b$  is the bulk substrate concentration,  $\text{mol m}^{-3}$ , and  $k_m$  is a mass-transfer coefficient,  $\text{m s}^{-1}$ . Both numerator and denominator have flux units  $\text{mol m}^{-2} \text{s}^{-1}$ ; positive  $C_b$  and  $k_m$  are required. The numerator is an estimated reaction demand if the surface experienced bulk concentration, whilst the denominator is a simple delivery scale. A value much smaller than one is consistent with delivery

capacity exceeding that estimated demand. A large value raises a transport concern; it does not measure the actual surface concentration or prove diffusion limitation. Geometry, flow, substrate diffusivity, porous structure, and reaction assumptions still require attention. This diagnostic follows the mass-transfer caution in the immobilisation chapter, rather than supplying a fitted result for a VARUNÉ candidate. [2]

A deliberately hypothetical example illustrates the decision. If doubling enzyme loading produces little change in flux, adding still more enzyme is not automatically justified. First test whether greater delivery changes flux without changing retained amount. If it does, transport is a plausible limiting process. If it does not, crowding, molecular damage, substrate mismatch, or measurement saturation remain alternatives. The test should vary delivery within a regime that does not itself detach coating or change the target biology. For an antibiofilm protein with no established catalytic mechanism, these enzyme equations should not be used to invent a turnover constant.

## 5 Conditioning and spatial heterogeneity

A deposited surface subsequently exposed to a complex fluid acquires a new interface. Competitive adsorption can alter which molecular regions the target encounters, whilst the deposited protein may remain below that layer. The nonfouling chapter also shows why a chemical label such as hydrophilic is insufficient: architecture, density, defects, and exposure conditions matter. Resistance in a simplified fluid does not establish resistance in a more complex one. [1,3]

This creates two different design objectives. A sample-contact surface may need to resist almost all unintended binding. A capture surface must resist background whilst permitting intended recognition. Optimising the first objective without measuring the second could remove useful signal along with noise. Likewise, an adhesive or cell-supporting interface may require selected interactions that a broadly resistant layer prevents. “Nonfouling” is therefore a context-dependent performance claim, not a universal material virtue.

Average coverage can also conceal local failure. Two surfaces can contain the same total protein yet distribute it differently. One may present a relatively uniform functional layer; the other may combine overloaded islands with bare regions. A bulk assay cannot identify which pattern exists. Spatial co-registration of a coverage measurement and a relevant outcome could test whether failures cluster around poorly covered regions. It should include prespecified fields or a systematic sampling rule; choosing visually striking fields after the result is known would distort the comparison.

### 5.1 What the Antarctic coating evidence presently supports

Olimpo and colleagues deposited PsyOmp38 onto PDMS and investigated biofilm formation under continuous flow. They reported non-uniform antibiofilm performance and proposed heterogeneous deposition as an explanation. Wetting measurements supported altered surface behaviour, but did not independently quantify intact protein or accessible function. Figure 5 identifies two independent biological flow replicates, whereas the general statistics section states a minimum of three; the assay-specific count should be retained pending clarification. Cytotoxicity also appeared after the longer exposure examined in their cell experiments. These findings justify further characterisation; they do not establish durable medical-device protection or safety. [9]

Fionda and colleagues subsequently examined PsyOmp38 and Cold-Azurin against clinical *E. faecalis* and *K. pneumoniae* strains across different biofilm stages. Their effects varied by protein,

strain, and stage. This broadens biological context but does not establish a single performance specification for an immobilised coating. The full-text methods identify nine clinical strains and a reference strain for each species. These laboratory data should remain distinct from patient outcomes. The paper also describes *E. faecalis* as an ESKAPE member, although its own list names *E. faecium*; the species tested, rather than that grouping, is used here. [10]

## 6 Author analysis of the competing explanations

My analysis is that the valuable research unit is a **decision-changing discrepancy**. Retained amount and useful function disagree, and that disagreement can direct the next comparison. The aim is not to collect every possible characterisation result. It is to choose a small set whose possible outcomes distinguish engineering actions. This proposed approach needs testing against actual development decisions; it is not a validated diagnostic framework.

Table 1. Competing explanations and discriminating observations.

| Observed pattern                               | Plausible explanations                                                            | Decision-changing comparison                                                         |
|------------------------------------------------|-----------------------------------------------------------------------------------|--------------------------------------------------------------------------------------|
| Retained signal falls and function falls       | Release, degradation, or changing analytical recovery                             | Parent-protein and released-material accounting with recovery controls               |
| Parent protein remains but function falls      | Masking, altered presentation, molecular damage, or assay suppression             | Accessibility and independent function before and after defined conditioning         |
| More loading adds little function              | Transport limitation, crowding, inaccessible excess, or readout saturation        | Delivery variation and a loading series with independent retained-amount measurement |
| Failure is local despite adequate mean loading | Coverage defects, local flow differences, or target heterogeneity                 | Prespecified spatial sampling with registered coverage and outcome                   |
| Function improves with a second constituent    | Better presentation, surface blocking, direct companion activity, or interference | Companion-only, matched recovery, and orthogonal functional controls                 |

The first implication is that a coating has a history, not merely a composition. Identical final solution conditions do not guarantee identical surfaces if adsorption, drying, washing, or conditioning occurred in a different order. Consequently, preparation history should enter the material specification when it predicts the outcome. Treating history as nuisance variation can hide the cause of irreproducibility. [2,3]

The second implication is that accessibility must be target-relative. A small substrate can reach sites that a large target cannot. A probe antibody can bind an exposed region that is irrelevant to the desired biological interaction. A surface can therefore pass an accessibility assay whilst failing its intended use. The proposed research should declare what each probe is meant to represent and where the analogy is expected to fail. A disagreement between probes is potentially informative, rather than automatically a failed assay.

The third implication is that local uniformity and average molecular quality should be optimised separately. A uniformly inactive layer is not rescued by improving coverage. Conversely, a highly active molecule cannot compensate for unprotected regions if those regions govern failure. The useful experiment is a comparison that changes distribution whilst holding total loading and handling as nearly constant as feasible. If this cannot be achieved, the remaining confounding should be reported rather than concealed by an average concentration.

A fourth implication concerns reversibility. If a conditioning-associated loss of function can be reversed without replacing the retained protein, irreversible molecular destruction becomes less likely. Yet recovery after washing does not by itself prove unmasking: the wash may also alter ionic conditions, remove an inhibitor, or change the assay environment. Reversal experiments require matched handling controls and measurement after the same final equilibration. This is a proposed reasoning test, not evidence that a particular coating can be regenerated.

## 7 Why an attractive curve can mislead

A common loading model is:

$$\Gamma(C) = \Gamma_{\max} \frac{KC}{1 + KC}$$

Equation 4. The Langmuir concentration form and its restricted interpretation.

$\Gamma$  and  $\Gamma_{\max}$  use the same surface-amount unit.  $C$  is solution concentration, and  $K$  has reciprocal concentration units so  $KC$  is dimensionless. An equilibrium interpretation requires reversible adsorption onto equivalent sites, an appropriate monolayer description, and the absence of interactions that invalidate the independent-site assumption. Latour explains why protein adsorption can yield a similar curve through spreading, reorientation, aggregation, or other processes that violate those assumptions. A good fit is therefore insufficient evidence for an equilibrium constant or adsorption free energy. [11]

For VARUNÉ, an empirical curve may still be operationally useful. If it predicts loading reproducibly within a defined preparation window, it can support process control. Its fitted parameter should then be described as an empirical parameter for that window. A claim about molecular affinity requires additional evidence of reversibility and model adequacy. This separation preserves useful prediction without attaching a stronger mechanism than the data support.

The strongest objection to the report's approach is that extensive mechanistic characterisation can delay a useful empirical product. That objection is valid when the intended performance is reproducible, the comparator is beaten under relevant conditions, and unresolved mechanism does not undermine the claim. The answer is proportionality: collect extra mechanistic evidence when its possible outcomes change formulation, qualification, manufacturing, or interpretation. Do not require an atomistic explanation merely to demonstrate a bounded performance improvement.

## 8 Proposed VARUNÉ research package

The initial package should concern one accessible protein, one support, and one intended environment. Selection depends on a partner-confirmed problem, acceptable rights, a funded scope, and a qualified scientific lead. A coating study should not automatically become a device-development programme. The immediate objective is to determine whether a performance gap is primarily associated with deposition, presentation, environmental exposure, or measurement.

The design should first establish that the measurements can answer that question. Retained amount needs recovery controls and specificity for the nominated material. Functional measurement needs a support-only control, a preparation-reagent control where relevant, and an independent readout that does not share the most likely interference. Labelling or probing should itself be evaluated for effects on the molecule and surface. An operationally accessible epitope is a proxy unless its link to useful function is demonstrated. Combined structural, accessibility, and activity methods can reduce ambiguity, but their interpretations remain conditional. [12]

Table 2. A staged comparison and its decision output.

| Stage                     | Main comparison                                                         | Required interpretation                                           |
|---------------------------|-------------------------------------------------------------------------|-------------------------------------------------------------------|
| Measurement qualification | Known additions, blank supports, and recovery/interference controls     | Identify what each signal measures and its usable range           |
| Preparation comparison    | A bounded loading or deposition contrast using independent preparations | Separate nominal input, deposited amount, and useful function     |
| Environmental challenge   | Defined conditioning compared with matched handling                     | Attribute changes to exposure rather than uncontrolled processing |
| Mechanism discrimination  | A targeted accessibility, transport, or spatial comparison              | Resolve the leading alternative explanations                      |
| Confirmation              | Repeat the chosen candidate against the relevant comparator             | Test reproducibility and the pre-agreed practical requirement     |

Independent coating preparations are the experimental units for a preparation claim. Multiple fields, wells, or time points from one preparation are subsamples or repeated measurements. Their additional information must not be converted into an inflated independent sample count. Run order and surface position should be randomised where practical, analysis rules fixed before examining outcomes, and measurement performed blind to preparation when feasible. Sample size should follow the minimum useful difference, expected preparation variance, and planned analysis; no universal replicate count is asserted here.

For a capture surface, the acceptance question is useful target recovery and specificity in the intended matrix. For a catalytic surface, it is process output under the required flow, substrate, and reuse conditions. For an antibiofilm coating, it is the prespecified microbial endpoint and duration under an agreed challenge. A better contact angle or a larger retained-mass signal is not a substitute. Acceptance margins, uncertainty intervals, and the permitted failure frequency should be agreed before results are available.

## 9 Contributions to existing programmes

This research could help an originating laboratory or material owner decide whether to improve its coating process or reconsider the underlying candidate. The proposed deliverable is an interface-characterisation package linking deposited amount, local coverage, accessible function, release, and relevant performance. Its value lies in resolving a development choice, rather than in supplying another favourable image.

Table 3. Possible contributions and conditions for value.

| Programme context                            | Proposed useful contribution                                          | Condition before activation                                              |
|----------------------------------------------|-----------------------------------------------------------------------|--------------------------------------------------------------------------|
| PsyOmp38 or related protein work             | Separate coverage limitations from presentation or retention failure  | Originating team confirms the unresolved need and grants suitable access |
| Enzyme process development                   | Identify whether immobilisation or substrate delivery limits output   | A customer supplies the actual workflow and comparator                   |
| Environmental capture or analytical surfaces | Test target recovery alongside nonspecific binding after conditioning | A programme defines the sample matrix and downstream decision            |

Potential funding would come from an authorised research sponsor, a material or device developer, or a buyer-funded comparison, if that organisation considers the question valuable. No sponsor interest, budget, available licence, or contract is established by this report. Background protein rights, rights to modified surfaces, access to results, and reuse of analytical methods would need separate agreement. A general characterisation service is not automatically a defensible product; value may instead reside in a reproducible process improvement or a validated application-specific method.

Stop or redirect if the team already has a satisfactory answer, if access or rights prevent useful work, if measurement remains non-discriminating, or if the relevant comparator already meets the requirement without a meaningful remaining gap. Six reports do not imply six laboratory programmes. This package should compete for the single active research slot against the other proposals in the series.

## 10 Limitations and conclusion

This report connects model surfaces, biochemical assays, and candidate antibiofilm studies that differ substantially in chemistry and intended use. Their agreement supports a general reason to distinguish amount from function; it does not establish common molecular mechanisms. Abstract-only evidence limits assessment of some primary controls, and targeted reading may favour studies with clear mechanistic contrasts. Surface-specific methods can disturb the state they aim to measure. Proposed equations are definitions or conditional models, not fitted descriptions of VARUNÉ materials.

The resulting research position is precise: retained protein should be treated as one part of the explanation for interface performance. A useful programme will establish when retention predicts function, when it does not, and which additional observation changes the next action. The near-term VARUNÉ contribution would be an experimentally defensible decision about deposition, presentation, or use conditions. Demonstrating that contribution requires new work, qualified review, and an explicit comparator. The present report provides the literature argument and the proposed decision structure.

## References and access record

- [1] Zhang P, Ratner BD, Hoffman AS, Jiang S. Nonfouling Surfaces. In: Wagner WR, Sakiyama-Elbert SE, Zhang G, Yaszemski MJ, editors. *Biomaterials Science An Introduction to Materials in Medicine*. 4th ed. Elsevier; 2020. Chapter 1.4.3A, main text pp. 507-512; supplied physical PDF pp. 506-511. Selected chapter passages re-examined for this report; references were not individually rechecked.
- [2] Frey SJ, Hoffman AS, Hubbell JA, Kane RS. Surface-Immobilized Biomolecules. In: *Biomaterials Science*. 4th ed.; 2020. Chapter 1.4.4, main text pp. 539-549; supplied physical PDF pp. 536-546. Selected attachment, presentation, and transport passages re-examined.
- [3] Horbett TA, Latour RA. Adsorbed Proteins on Biomaterials. In: *Biomaterials Science*. 4th ed.; 2020. Chapter 2.1.2, main text pp. 645-659; supplied physical PDF pp. 639-653. Selected competition, spreading, conformation, and activity passages re-examined. Printed and physical page offsets are chapter-specific.
- [4] Fears KP, Latour RA. Assessing the influence of adsorbed-state conformation on the bioactivity of adsorbed enzyme layers. *Langmuir*. 2009;25:13926-13933. <https://doi.org/10.1021/la900799m>. Publisher/PubMed abstract and indexed manuscript material checked; direct PMC opening encountered a browser check. Full methods and supplements were not comprehensively audited.
- [5] Sivaraman B, Latour RA. The relationship between platelet adhesion on surfaces and the structure versus the amount of adsorbed fibrinogen. *Biomaterials*. 2010;31:832-839. <https://doi.org/10.1016/j.biomaterials.2009.10.008>. PubMed metadata, abstract, and indexed author-manuscript results/conclusions checked; direct PMC opening encountered a browser check. Correlations are source-reported, not recalculated.
- [6] Keselowsky BG, Collard DM, García AJ. Surface chemistry modulates fibronectin conformation and directs integrin binding and specificity to control cell adhesion. *Journal of Biomedical Materials Research A*. 2003;66:247-259. <https://doi.org/10.1002/jbm.a.10537>. Publisher/PubMed abstract checked, with supporting discussion in [3]; complete methods were not reanalysed.
- [7] Pei Z, Anderson H, Myrskog A, Dunér G, Ingemarsson B, Aastrup T. Optimizing immobilization on two-dimensional carboxyl surface: pH dependence of antibody orientation and antigen binding capacity. *Analytical Biochemistry*. 2010;398:161-168. <https://doi.org/10.1016/j.ab.2009.11.038>. Publisher abstract, preview methods, and conclusion checked; full paper not accessed.
- [8] Shen L, Schroeder M, Ogorzalek TL, et al. Surface orientation control of site-specifically immobilized nitro-reductase NfsB. *Langmuir*. 2014;30:5930-5938. <https://doi.org/10.1021/la5016862>. PubMed abstract and bibliographic record checked; no numerical effect-size extraction from the full paper.
- [9] Olimpo D, D'Angelo C, Imbimbo P, et al. Novel insights into the struggle against biofilm: the PsyOmp38 protein from the Antarctic marine bacterium *Psychrobacter* sp. TAE2020. *Microbial Biotechnology*. 2025;18:e70249. <https://doi.org/10.1111/1751-7915.70249>. Publisher full text available; sections 2.15-2.17, 3.3, 3.5, Figure 5 caption, and discussion selectively checked. Replication inconsistency unresolved; no complete supplement audit.
- [10] Fionda M, Trecca M, Olimpo D, et al. New proteins from cold: antibiofilm activity of Cold-Azurin and PsyOmp38 against *Enterococcus faecalis* and *Klebsiella pneumoniae*. *Probiotics and Antimicrobial Proteins*. 2026;18:8024-8043. <https://doi.org/10.1007/s12602-026-11009-7>. Publisher full text available; abstract, species/strain methods, and relevant results/discussion checked selectively. No clinical efficacy inferred.

[11] Latour RA. The Langmuir isotherm: a commonly applied but misleading approach for the analysis of protein adsorption behavior. *Journal of Biomedical Materials Research A*. 2015;103:949-958. <https://doi.org/10.1002/jbm.a.35235>. Publisher abstract checked; detailed model caution also verified in supplied chapter [3], pp. 651-652.

[12] Thyparambil AA, Wei Y, Latour RA. Experimental characterization of adsorbed protein orientation, conformation, and bioactivity. *Biointerphases*. 2015;10:019002. <https://doi.org/10.1116/1.4906485>. Indexed author-manuscript introduction, method description, example table, and conclusion checked; methodological source rather than an independent candidate-validation study.

[13] IUPAC. Michaelis-Menten equation. *Compendium of Chemical Terminology*, 5th ed.; 2025, online version 5.0.0. <https://doi.org/10.1351/goldbook.11546>. Official indexed definition checked; direct page opening returned an access error. Equation 2 adapts the standard kinetic form to a stated surface and transport model.

Online access date for all web sources: 28 October 2026. References [4-10] are empirical primary studies. References [1-3] provide textbook foundations; [11-12] support model and measurement interpretation. The proposed comparisons and decision structure are the report's analytical synthesis, offered for author and scientific review.

## Authorship and research provenance

Varun Sharma is the sole author and approved this edition for release on 28 October 2026. No new experimental data are reported; published findings remain attributable to the cited investigators.

Competing interests. Varun Sharma founded VARUNÉ Bio, which may benefit commercially from applications discussed here. Funding and contributor declarations remain subject to author confirmation.

This report shares concepts and sources with the five companion reports dated 28 October 2026; it presents a distinct literature analysis.