Designing a Prognostics Framework for Pharmaceutical Development: Applying PHM Principles to Computational Drug Discovery for Novel HIV-1 C(SA) Protease Inhibitors

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ABSTRACT

Prognostics and Health Management (PHM) has transformed the engineering industry through accurate pre-initialisation failure prediction. Here, those principles are applied to pharmaceutical development by treating a physics-based computational chemistry model as a pre-synthesis failure predictor for candidate drug molecules. The target is the HIV-1 subtype C (South African, C(SA)) protease, the dominant strain across sub-Saharan Africa, against which the existing subtype-B-optimised protease inhibitors lose potency. Twenty pentacycloundecane (PCU) cage peptoid candidates in the C→N backbone orientation were evaluated against the HIV-1 C(SA) protease (PDB 3U71) by molecular dynamics and MM-GBSA binding free energy calculations (AMBER 20, ff19SB, n = 4 replicates), with nine FDA-approved protease inhibitors evaluated under an identical protocol as positive controls. Glu-PCU-Glu ranked first (ΔGbind = −89.24 ± 7.42 kcal mol^{-1}); the conformationally constrained Pro-PCU-Pro ranked last (−22.22 ± 2.20 kcal mol^{-1}). The MM-GBSA screen functions as a Stage 1 health check that rules out the primary failure mode, insufficient target affinity, before any synthesis cost is incurred. Three compounds, Glu-PCU-Glu, Cys-PCU-Cys and Tyr-PCU-Tyr, pass the check and are carried forward.

Keywords: prognostics and health management; pre-initialization failure prediction; computational drug discovery; MM-GBSA; HIV-1 C(SA) protease

1. INTRODUCTION

Prognostics and Health Management emerged in engineering to replace reactive maintenance with predictive decision-making. Jardine *et al,* in 2006 detailed a PHM framework as a method to monitor health-indicative metrics, predict the onset and mode of failure, and commit resources only to systems with acceptable, predicted reliability. Pharmaceutical development faces a structurally identical problem. The synthesis and biological evaluation of a candidate molecule is slow and expensive, and in the traditional workflow the failure modes of a candidate become apparent only after that investment has been made. This is a reactive paradigm, and it is especially costly in low-resource settings.

This letter applies the PHM philosophy to drug discovery by treating a physics-based binding free energy calculation as a pre-initialisation failure predictor. The molecular mechanics generalised Born surface area (MM-GBSA) method estimates how favourably a candidate engages its target before the candidate is ever synthesised; in PHM terms it is a condition-monitoring measurement, and the binding free energy it returns is the health metric. The target is the HIV-1 subtype C (South African) protease, which causes the majority of HIV infections in sub-Saharan Africa and against which the subtype-B-optimised clinical inhibitors are measurably less effective, UNAIDS (2023) & Ahmed *et al.,* (2013). The candidate chemotype is the penta-cyclo-undecane (PCU) cage peptoid, a rigid diamondoid cage carrying two amino acid-derived arms AS PER WORK BY Makatini, *et al*., (2011); the C→N backbone orientation is studied here.

Objective: The main objective of this work is the end-point failure prediction of twenty novel PCU cage peptoids, as potential HIV protease inhibitors against HIV_1C(SA) using MM-GBSA free energy method as a screen, and it is will also be used to identify synthesis candidates. This pre-synthesis molecular dynamics simulation is the drug discovery analogue of the pre-initialization failure prediction that prognostics and health management frameworks can bring to engineered systems and reduce costs and time of reactive failure-detection methods with a cheaper predictive approach.

2. COMPUTATIONAL METHODS

The HIV-1 C(SA) protease crystal structure (PDB 3U71) was prepared in AMBER 20 with the ff19SB force field; the catalytic Asp25/Asp25' dyad was treated as mono-protonated as detailed by Tian *et al.,* (2020). Twenty C→N PCU cage peptoids and nine FDA-approved protease inhibitors were parameterised with GAFF2 and AM1-BCC partial charges as done by Badaya & Sasidhar in a similar study in 2020. Each complex was solvated in explicit TIP3P water with 150 mM NaCl, minimised, heated to 298.15 K, equilibrated for 1 ns and simulated for 100 ns of production molecular dynamics; four independent replicates were generated per compound. Binding free energies were computed by single-trajectory MM-GBSA over 1,600 snapshots per replicate, with igb = 5, as: (Genheden & Ryde, 2015)

$$\Delta Gbind = \Delta Evdw + \Delta Eele + \Delta G\_GB + \Delta G\_SA \qquad (1)$$

*All energies are reported in kcal mol⁻¹.

For a congeneric series of analogous biomolecules such as the cage peptoids, the configurational entropy term (−TΔS) is disregarded due to its computational expense and is in keeping with the single trajectory protocol as detailed by Genheden and Ryde in 2015. The term is approximately constant in such a series becoming negligible and having no impact on the relative ranking of free binding energy, the metric this study uses. Absolute ΔG_bind values are consequently systematically more negative than entropy-corrected experimental free energies; the screen is therefore used as a statistically reliable relative-ranking tool, not as a predictor of absolute affinity mirroring work by Lockhart *et al.,* (2016).

3. RESULTS

The full twenty-compound C→N screen is reported in the companion journal manuscript; this letter presents the result in the PHM framing. Table 1 lists nine representative cage peptoids, the three strongest, three mid-ranked and three weakest binders, spanning the full range of predicted affinity and the principal R-group chemical classes.

Glu-PCU-Glu is the strongest predicted binder at −89.24 ± 7.42 kcal mol⁻¹, with the carboxylate arms making a strong electrostatic contact with the catalytic dyad region. Pro-PCU-Pro is the weakest at −22.22 ± 2.20 kcal mol⁻¹; the conformationally locked proline backbone cannot adapt to the active site, and this variant is the failure-prone reference of the series. In PHM terms the screen has separated the candidate population into those that pass the Stage 1 affinity health check and those that fail it.

3.1 FDA-Approved Inhibitors as a Calibration Set

Nine FDA-approved HIV-1 protease inhibitors were evaluated under the identical protocol to calibrate the screen against compounds of known clinical performance (Table 2). Their predicted binding free energies cluster between −13.89 and −16.85 kcal mol⁻¹. In PHM terms these are reference systems of known condition, and the screen reproduces their expected ordering, which validates its use as a health monitor for the candidate cage peptoids. They act as a positive control and internal check for the model's validation as the calculated free binding energies must match what is observed in experiment as reported in literature in work by Ahmed et al., (2013) and Sayer et al., (2012), for the same target.

Table 1. Representative C→N PCU cage peptoids drawn from the complete twenty-compound screen

Compound	R-group class	ΔEvdw (kcal mol⁻¹)	ΔEele (kcal mol⁻¹)	ΔG_GB (kcal mol⁻¹)	ΔG_SA (kcal mol⁻¹)	ΔGbind (kcal mol⁻¹)	±SD (kcal mol⁻¹)
Glu-PCU-Glu	Acidic	-52.18	-621.34	+629.85	-45.57	**-89.24**	±7.42
Cys-PCU-Cys	Sulfur-containing	-38.74	-510.22	+510.47	-46.08	**-84.57**	±2.95
Tyr-PCU-Tyr	Aromatic	-44.86	-482.30	+494.28	-46.67	**-79.55**	±1.26
Gly-PCU-Gly	Aliphatic (non-polar)	-38.93	-448.74	+473.45	-43.27	**-57.49**	±3.55
Phe-PCU-Phe	Aromatic	-57.48	-395.22	+432.44	-37.22	**-57.48**	±6.58
Ser-PCU-Ser	Polar (uncharged)	-38.41	-447.19	+471.72	-42.22	**-57.10**	±3.44
Val-PCU-Val	Aliphatic (non-polar)	-33.17	-408.63	+438.22	-34.65	**-38.23**	±3.55
Lys-PCU-Lys	Basic	-31.84	-398.27	+428.52	-35.59	**-37.18**	±4.18
Pro-PCU-Pro	Aliphatic (non-polar)	-21.43	-312.85	+347.31	-35.25	**-22.22**	±2.20

The three strongest binders, three mid-ranked binders and three weakest binders are shown in table 1 above. With the free binding energy (ΔGbind), calculated according to equation 1- ΔGbind = ΔEvdw + ΔEele + ΔG_GB + ΔG_SA; means of four replicates; all energies in kcal mol⁻¹.

Table 2. The nine FDA-approved HIV-1 protease inhibitors evaluated under the identical MM-GBSA calibration set of known-condition reference systems.

FDA-approved inhibitor	ΔGbind (kcal mol⁻¹)	±SD (kcal mol⁻¹)	Generation
Lopinavir (LPV)	**-16.85**	±0.31	2nd
Darunavir (DRV)	**-16.75**	±0.14	2nd
Atazanavir (ATV)	**-16.11**	±0.19	2nd
Tipranavir (TPV)	**-15.94**	±0.12	3rd
Amprenavir (APV)	**-15.13**	±0.22	2nd
Ritonavir (RTV)	**-15.06**	±0.26	1st
Saquinavir (SQV)	**-15.01**	±0.22	1st
Indinavir (IDV)	**-14.91**	±0.31	1st
Nelfinavir (NFV)	**-13.89**	±0.22	1st

As the single-trajectory approach does not include the configurational entropy penalty (Genheden & Ryde, (2015)), the comparisons are a relative ranking vs an absolute binding energy. The cage peptoid calculations rank better than the 9 FDA-drugs against the HIV_1 C(SA) protease target.

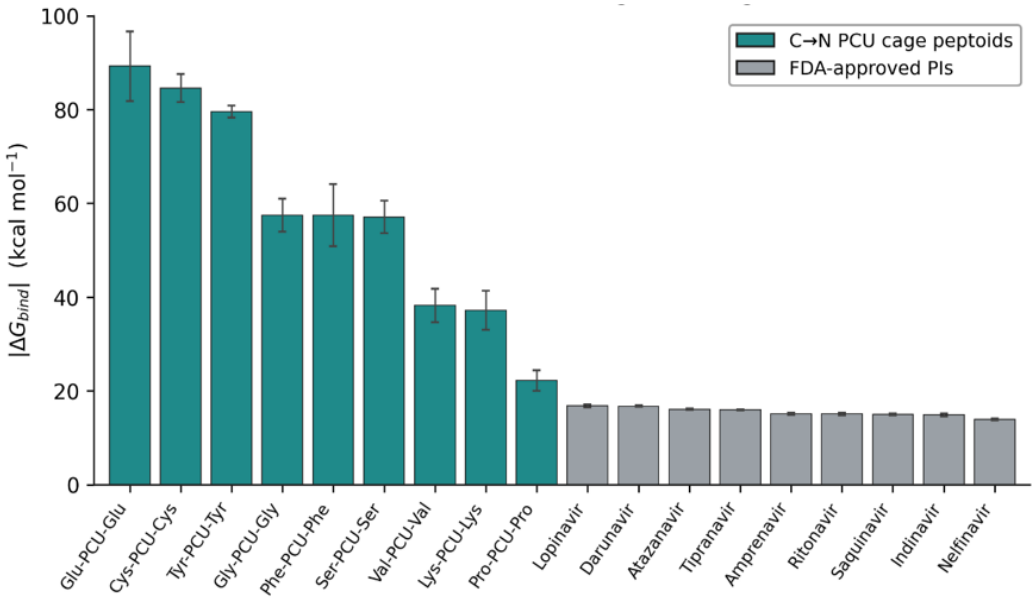

Figure 1. Predicted binding free energies of the nine representative C→N cage peptoids (teal) and the nine FDA-approved protease inhibitors (grey). The cage peptoids occupy a distinct and more favourable relative-ranking regime against the C(SA) enzyme.

3.2 Molecular Dynamics Stability Validation

A free binding energy calculation is reliable if the complex system remains structurally stable throughout the simulation duration. The condition-monitoring measurement acts as a second method validation check assessing the stability of the complex over the course of the 100ns production (≈0.35 nm) — while only the failure-prone Pro-PCU-Pro PCU shows the lowest guest mobility (0.16 nm), whereas the failure-prone Pro-PCU-Pro shows the largest guest excursion (0.50 nm), signalling a ligand that cannot maintain a productive pose as found by Badaya & Sasidhar in 2020.

The results from figures 2 and the complementarity stability metrics from figure 4 below, allow for a similar conclusion to be independently drawn. The radius of gyration is essentially invariant across the strong and mid-ranked binders (1.73–1.84 nm), confirming that the protease homodimer remains compact and does not undergo domain separation, while Pro-PCU-Pro shows the largest solvent-accessible surface area and an early radius-of-gyration excursion, the structural signature of a poorly accommodated ligand. The intramolecular hydrogen-bond network of the protease is preserved throughout (approximately 92–98 bonds for the viable candidates) and collapses only for Pro-PCU-Pro. The native-contact analysis shows the same ordering (Figure S2), and the per-residue fluctuation profile confirms that only the mobile flap tips of the protease fluctuate appreciably while the catalytic core stays rigid (Figure S1). Critically, the equilibrated cage trajectories duration. Figure 2 below reports the root mean square deviation (RMSD) of the enzyme backbone, the PCU (guest) ligand and the full receptor-ligand complex for all the reported PCUs. The protease backbone equilibrates quickly and settles between 0.20 and 0.35 nm for every viable complex — Glu-, Gly- and Lys-PCU sit lowest (≈0.22–0.24 nm) and Tyr-PCU highest among the viable set

peptoid guest RMSD shows a weak positive correlation with the predicted binding free energy across the series (Pearson r = 0.35; Figure 3): the trend where more mobile ligands are weak binders is mainly driven two extremes –

1. **the rigid, low-mobility** Tyr-PCU at the strong-binding end and the failure-prone, highly mobile Pro-PCU-Pro at the weak-binding end. This places the structural stability analysis and the MM-GBSA health metric agreement and validates the model.

2. **directional,** specific pharmacophore contacts made by the strongest binders — the Glu carboxylate and Tyr phenol engaging the catalytic-site region, with 16 and 15 occupancy-weighted peptoid–protease hydrogen bonds respectively (Figure S3) — are consistent with the binding determinants previously reported by our research group, for PCU cage peptoids against the C(SA) protease (Makatini et al., (2011).

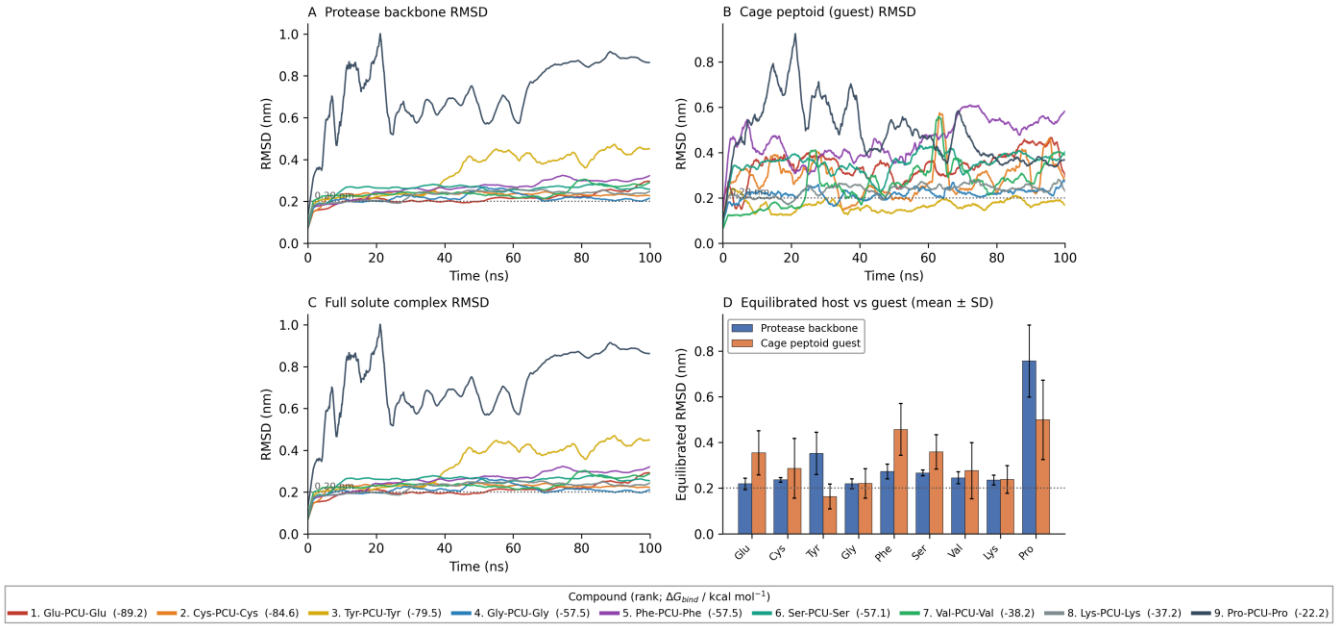

Figure 2. Root-mean-square deviation (RMSD) of the nine representative C→N cage peptoid complexes over 100 ns. (A) Protease backbone, (B) cage peptoid guest, (C) full solute complex, and (D) equilibrated host-versus-guest summary**.**

Solid traces show the full 100 ns trajectory for all nine complexes, each coloured by affinity rank (running mean over 150 frames). The 0.20 nm reference marks the threshold below which the protease is considered equilibrated. Every viable complex equilibrates within ≈0.20–0.35 nm, whereas only the failure-prone Pro-PCU-Pro rises well above this range, confirming that each ΔG_bind value is drawn from a structurally stable, equilibrated ensemble. Compounds are coloured by affinity rank (legend, bottom).

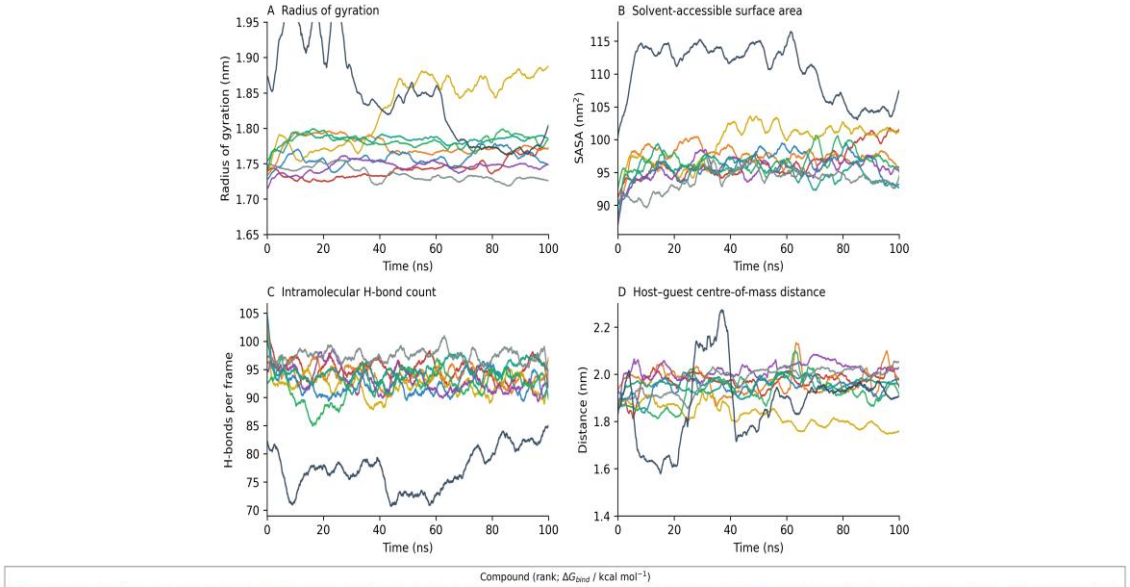

Figure 3. ***Complementary structural-stability metrics for the nine representative C→N cage peptoid complexes over 100 ns.*** (A) Radius of gyration, (B) solvent-accessible surface area, (C) intramolecular hydrogen-bond count, and (D) host–guest centre-of-mass distance. Solid traces show the full 100 ns trajectory for all nine complexes, each coloured by affinity rank (running mean over 150 frames). *Across every metric the viable candidates form a tight, stable cluster while the failure-*

prone Pro-PCU-Pro is the consistent outlier (largest SASA, fewest hydrogen bonds and an early radius-of-gyration excursion), corroborating the affinity ranking through independent structural evidence. Compounds are coloured by affinity rank (legend, bottom).

The complementary metrics reinforce this picture. Solvent-accessible surface area is tightly clustered for the viable candidates (≈94–100 nm²) but markedly larger for Pro-PCU-Pro (≈111 nm²), the structural signature of a ligand that cannot bury itself in the pocket; correspondingly, the native-contact retention is highest for the strong, low-scatter binder Tyr (≈105) and lowest for Pro (≈41), with the remaining candidates retaining ≈69–81 contacts. Taken together, the hydrogen-bond, SASA and native-contact analyses converge on the same conclusion as the binding free energy: the viable candidates form compact, well-buried, contact-rich complexes, while Pro-PCU-Pro fails on every structural axis simultaneously and serves as the failure reference of the series.

4. Discussion

The result demonstrates a working prognostics framework for pre-synthesis pharmaceutical screening. The MM-GBSA calculation is the condition-monitoring measurement, the binding free energy is the health metric, and the screen delivers an end-point failure prediction: it identifies, before any synthesis, which candidates will fail at the affinity endpoint. The internal calibration against nine FDA-approved inhibitors confirms that the screen behaves as a validated health monitor.

The molecular dynamics stability analysis (Section 3.2) adds a second, corroborating validation metric to the framework. In PHM terms, a single sensor reading is rarely trusted in isolation; a robust health monitor cross-validates the primary measurement against an independent indicator of the same underlying condition. Here, the structural stability of each complex, backbone RMSD, radius of gyration, hydrogen-bond integrity and native-contact retention, serves as that independent indicator, and it agrees with the binding free energy ranking most clearly at the extremes of the series: the candidates that pass the affinity check are also the ones that remain structurally stable, while the failure-prone Pro-PCU-Pro fails on every structural axis simultaneously. This concordance raises confidence that the Stage 1 prediction reflects genuine target engagement rather than a numerical artefact of an unstable pose, and it allows for a better-informed synthesis candidate prediction, before any synthesis cost is incurred.

Binding affinity is, however, only one axis of candidate viability. Pharmacokinetics, toxicity and metabolic stability are independent failure-risk axes that a complete prognostics framework must also address. The present study is therefore explicitly the first stage of a multi-stage pipeline, the analogue of first-level fault detection in an engineered PHM system. The Stage 1 affinity screen rules out the primary failure mode; subsequent stages will apply in silico ADMET profiling to the surviving candidates before any experimental commitment, exactly as a staged engineering qualification proceeds through structural, then thermal, then fatigue screening.

Computational calculations precede experimental validation at this: the computational model exists to determine, ahead of synthesis, which compounds justify the cost of laboratory work. Synthesis and IC_{50} evaluation of the three candidates that pass the Stage 1 screen are the planned next phase.

The wider value to the PHM community is the transfer of method. Drug discovery and engineering health management share a single structure: in both, a quantitative physics-based model predicts end-point failure so that resources flow only to systems that will perform. The framework demonstrated here shows that the prognostics paradigm, developed for engineered assets, applies directly and usefully to the pre-synthesis triage of drug candidates.

5. Conclusions and Future Directions

This letter has presented a prognostics framework for pre-synthesis pharmaceutical screening, applied to twenty C→N PCU cage peptoid candidates against the HIV-1 C(SA) protease. The MM-GBSA screen, calibrated against nine FDA-approved inhibitors, functions as a Stage 1 health check that delivers an end-point failure prediction and separates viable candidates from the failure-prone reference, Pro-PCU-Pro, before any synthesis cost is incurred.

As future directions, three candidates pass the Stage 1 affinity claclulatins and are carried forward into the multi-stage pipeline: Glu-PCU-Glu, the top-ranked candidate; Cys-PCU-Cys, whose thiol additionally offers covalent-engagement potential; and Tyr-PCU-Tyr, whose low inter-replicate scatter makes it the highest-confidence prediction. The next pipeline stages are in silico ADMET profiling of these survivors to address the pharmacokinetic and toxicity failure-risk axes, followed by chemical synthesis and IC_{50} evaluation against the recombinant HIV-1 C(SA) protease. The framework is target-agnostic and is expected to transfer to other therapeutic targets.

Acknowledgements

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Biographies

Charmaine A. .Kahiya holds a BSc degree in biochemistry and microbiology from Rhodes University,South Africa (2012), a BSc Honours degree in biological sciences from the University of Zimbabwe, Zimbabwe,(2014), and an MSc degree (cum laude) in medical sciences with a focus on pharmaceutical chemistry from the University of KwaZulu-Natal, under the Catalysis and Peptide Research Unit, South Africa, (2020). She is currently working toward a PhD degree in pharmaceutical chemistry at the Catalysis and Peptide Research Unit, University of KwaZulu-Natal, South Africa. Her current research interests include computational pharmaceutical chemistry, molecular dynamics simulation, and metal chelation strategies for antiviral drug discovery, with a focus on NOTA-mediated zinc chelation at the HIV-1 integrase HHCC motif as a resistance-distinct mechanism against HIV-1 subtype C, and PCU cage peptoid inhibitors of the HIV-1 C(SA) protease. Alongside her research, she is an active advocate for women and girls in science, contributing through the OWSD Zimbabwe Executive Committee which implemented the UNESCO STEM Pilot Project and now is on the second phase, conducted across all ten provinces of Zimbabwe. She contributes to national STEM policy consultations, and she mentors girls and young women pursuing careers in science. She is a member of UNESCO's, Organization for Women in Science for the Developing World and serves on its Zimbabwe Executive Committee (2022-2028) and serves on the World Bank Zimbabwe Women in STEMM working Group. She is a recipient of the Schlumberger Faculty for the Future Fellowship (2023–2027) and the Mandela Washington Fellowship (2023), is a 2024 Stanford Spark Africa alumna. She was featured among the 25 Women in STEM in Zimbabwe You Must Know by Girls in STEM Trust in 2024 and was nominated as the Women's University in Africa Most Inspiring Woman in STEM in 2024.

Tricia Naicker received the MSc degree (cum laude) in chemistry in 2008 and the PhD degree in pharmaceutical chemistry in 2011, both from the University of KwaZulu-Natal, Durban, KwaZulu-Natal, South Africa, where her doctoral work on tetrahydroisoquinoline-based organocatalysts was the first report on asymmetric organocatalysis from Africa. She subsequently held an Oppenheimer-funded postdoctoral fellowship in asymmetric catalysis with Prof. K. A. Jørgensen at Aarhus University, Aarhus, Denmark. She joined the University of KwaZulu-Natal as an academic in 2013 and is currently a Professor in the Discipline of Pharmaceutical Sciences and Director of the Catalysis and Peptide Research Unit, having been the institution's youngest associate professor. Her research interests include method development in the synthesis of biologically important intermediates and drugs, with a focus on antibacterial agents. She is a member of the Organization for Women in Science for the Developing World and has served on the Executive Committee of its South African National Chapter. She received the University of KwaZulu-Natal Vice-Chancellor's Award in 2019 and the Raikes Medal of the South African Chemical Institute in 2020, being the first woman from the university to receive the medal, and she holds national awards from the South African Department of Science and Technology and the National Research Foundation.

Glenn E. M. Maguire was born in Northern Ireland. He received the PhD degree in physical organic chemistry from

Queen's University Belfast, Belfast, Northern Ireland, in 1993, under the supervision of Prof. A. P. de Silva. He carried out postdoctoral research at the School of Chemistry and Biochemistry, University of California, Los Angeles, USA, with Prof. Donald J. Cram, and at the School of Molecular Biology and Pharmacology, Washington University Medical School, St. Louis, Missouri, USA, with Prof. George W. Gokel. He joined the School of Chemistry and Physics, University of KwaZulu-Natal, Durban, KwaZulu-Natal, South Africa, in 1999, and is a co-founder and principal investigator of the Catalysis and Peptide Research Unit. His research interests span catalysis, organic, analytical, medicinal, computational and structural chemistry.

Hendrik G. Kruger received the PhD degree from Potchefstroom University (now North-West University), Potchefstroom, South Africa, in 1996, under the supervision of Prof. F. J. C. Martins and Prof. A. M. Viljoen, working in polycyclic cage chemistry. He is currently a Research Professor in the Catalysis and Peptide Research Unit, University of KwaZulu-Natal, Durban, KwaZulu-Natal, South Africa. His research interests include organic synthesis, polycyclic cage chemistry, NMR structure elucidation, computational chemistry, chiral and peptide synthesis, and the development of tuberculosis and HIV protease inhibitor drugs.

José Rogério A. Silva received the Licentiate degree in chemistry in 2010, the MSc degree in chemistry in 2012, and the PhD degree in chemistry in 2015, all from the Federal University of Pará, Belém, Pará, Brazil, completing part of his doctoral research as a visiting scholar at the University of Florida, Gainesville, Florida, USA, under Prof. Adrian Roitberg. He subsequently held a CAPES-funded postdoctoral research position in the Graduate Program in Chemistry at the Federal University of Pará. Since 2016, he has been a Professor in the Faculty of Chemistry, Institute of Exact and Natural Sciences, Federal University of Pará, where he is a member of the Laboratory of Computer Modeling of Molecular Biosystems (CompMBio), and he holds a research affiliation with the Catalysis and Peptide Research Unit, University of KwaZulu-Natal, Durban, South Africa. His research interests include theoretical and computational chemistry, computational biophysics and biochemistry, molecular docking, molecular dynamics simulation, and combined quantum mechanics/molecular mechanics (QM/MM) methods applied to enzyme catalysis and drug discovery.

The following figures provide the per-residue and contact-level molecular dynamics analyses that underpin the stability validation in Section 3.2.

Appendix

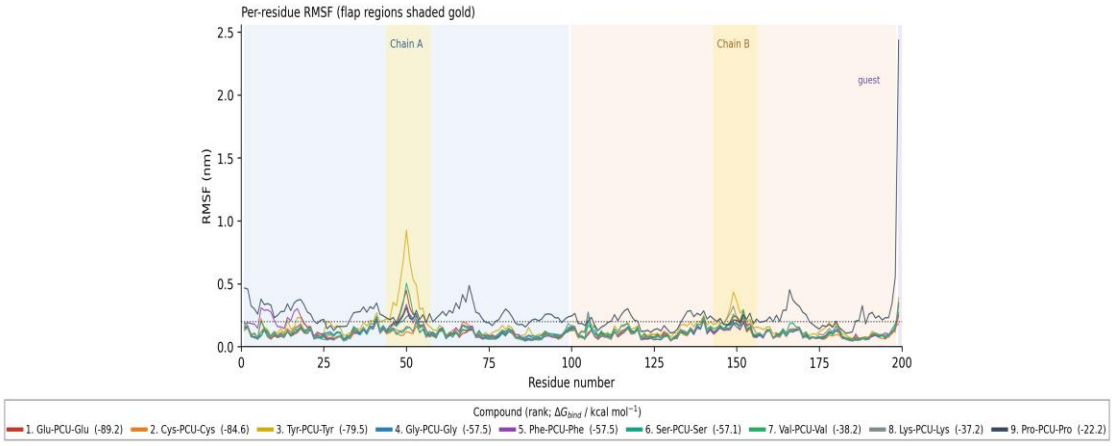

Figure S1. Per-residue root-mean-square fluctuation (RMSF) of all nine representative C→N cage peptoid complexes, overlaid and coloured by affinity rank, with the structural domains shaded: protease chain A (residues 1–99), chain B (residues 100–198), the flap regions (gold), and the cage peptoid guest (residue 199). *Only the mobile flap tips exceed the 0.20 nm flexibility threshold; the catalytic core remains rigid, confirming that ligand binding does not destabilise the active site.*

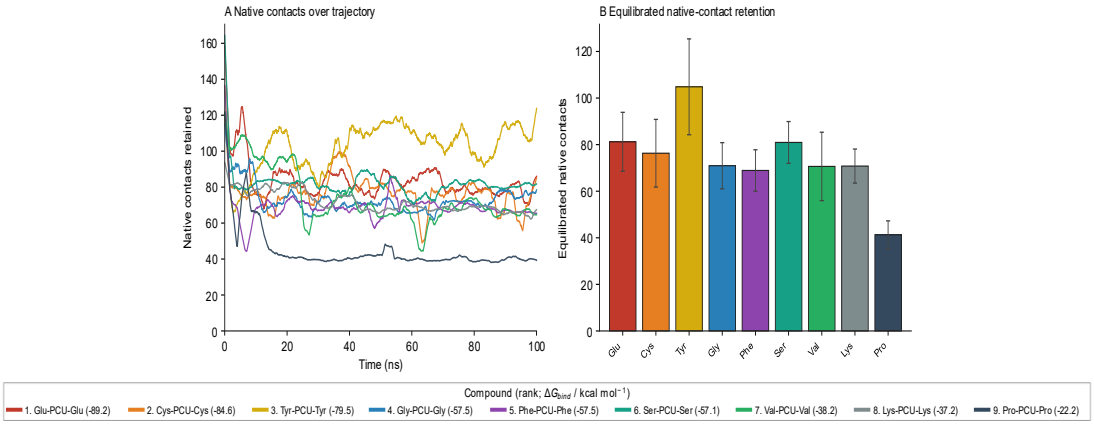

Figure S2. Native-contact analysis for the nine PCU complexes. (A) Native contacts retained over the 100 ns trajectory (running mean), and (B) the equilibrated native-contact retention per compound (mean ± SD). The ranking of the native contacts mirrors the binding free energy ranking, confirming that the strongest binders preserve their docked interface most faithfully.