



<https://doi.org/10.70728/a.series.tab.v08.i09.199>

## FENILGLIOKSIL KISLOTA TIOSEMIKARBAZONINING KOBALT(II) BIS-XELATINI BAKTERIAL NISHONLARGA MOLEKULYAR DOCKING QILISH

**Akhatov Javokhirkhon**

*Termez state university, doctoral student*

*javohirxonahatov2@gmail.com*

*tel: +998937921898*

**Turaev Khayit**

*Termez state university, Doctor of Chemical science, professor*

**Ashurov Jamshid**

*Institute of Bioorganic Chemistry named after Academician Obid Sodikov*

*Academy of Sciences of the Republic of Uzbekistan*

*Doctor of Chemical science, professor*

**Annotatsiya.** Kobalt(II) bis-xelati  $\text{Co}(\text{PGTS})_2$  ning rentgen tuzilishidan o‘zgarmagan geometriyasi yettita bakterial nishonga molekulyar docking usulida tekshirildi. Oltitasi har qanday ball hisoblanishidan oldin muzlatilgan protokol asosida dock qilindi va oltitasi ham redocking validatsiyasidan o‘tdi. Kompleks oltitadan to‘rttasida erkin ligandning ikki shaklidan ham past ball oldi, eng kuchli ajralish *Bacillus subtilis* FtsZ (2RHL) da qayd etildi. Metall atomining turi natijani umuman o‘zgartirmadi. Vinardo funksiyasi bilan qayta ballash Vina tartibini faqat bitta nishonda takrorladi va bu xulosani sezilarli darajada cheklaydi.

**Kalit so‘zlar:** kobalt(II) kompleksi, tiosemikarbazon, molekulyar docking, AutoDock Vina, DNK giraza B, FtsZ, redocking validatsiyasi, konsensus ballash

**Аннотация.** Проведён молекулярный докинг бис-хелата кобальта(II)  $\text{Co}(\text{PGTS})_2$ , геометрия которого взята без изменений из рентгеновской структуры, к семи бактериальным мишеням. Шесть мишеней были докированы по протоколу, зафиксированному до вычисления каких-либо оценок, и все шесть прошли валидацию повторным докингом. Комплекс получил оценку ниже обеих форм свободного лиганда на четырёх мишенях из шести, наиболее выражено для FtsZ *Bacillus subtilis* (2RHL). Тип атома металла не изменил результат. Пересчёт Vinardo воспроизвёл порядок Vina лишь для одной мишени, что ограничивает вывод.

**Ключевые слова:** комплекс кобальта(II), тиосемикарбазон, молекулярный докинг, AutoDock Vina, ДНК-гираза B, FtsZ, валидация повторным докингом, консенсусная оценка

**Abstract:** The cobalt(II) bis-chelate  $\text{Co}(\text{PGTS})_2$ , taken unchanged from its X-ray structure, was docked against seven bacterial targets. Six were docked under a protocol frozen before any score was computed, and all six passed redocking validation. The complex scored below both free-ligand forms on four of the six targets, most strongly at *Bacillus subtilis* FtsZ (2RHL). Metal atom typing changed nothing. Vinardo rescoring reproduced the Vina ordering on only one target, which bounds the conclusion.

**Keywords:** cobalt(II) complex, thiosemicarbazone, molecular docking, AutoDock Vina, DNA gyrase B, FtsZ, redocking validation, consensus scoring

**INTRODUCTION.** Thiosemicarbazones supply, in one small molecule, a hard nitrogen donor, a soft sulfur donor and — when the parent carbonyl compound carries a further functional group — a third anchoring site. The ligand switches between thione and thiolate tautomers and between *E* and *Z* configurations about the  $\text{C}=\text{N}$  bond, so its coordination behaviour is set by the metal and the medium rather than by stoichiometry alone [4]. Complexes of 3d metal ions with this ligand class have long been examined for antimicrobial behaviour [15], and cobalt complexes in particular have attracted attention because the metal offers two accessible oxidation states and a well-defined octahedral preference [11].

In the  $\alpha$ -keto acid subclass the carboxylic acid provides an oxygen donor that completes an O,N,S tridentate pocket and fixes the protonation state at physiological pH. Phenylglyoxylic acid thiosemicarbazone ( $\text{H}_2\text{PGTS}$ ) is such a ligand and its crystal structure has been reported [17]; with cobalt it gives a 2:1 bis-chelate in which two tridentate arms enclose the metal, leaving no open coordination site.

The bacterial targets examined here are established ones. DNA gyrase subunit B is a validated antibacterial target whose ATP site is chemically distinct from human topoisomerases [10]; the cell-division protein FtsZ is essential and structurally unlike its eukaryotic counterparts [2]; aminoacyl-tRNA synthetases are exploited clinically [13]; and the quorum-sensing regulator LasR of *Pseudomonas aeruginosa* offers a virulence-directed rather than growth-directed mechanism [3].

This article compares  $\text{Co}(\text{PGTS})_2$  with its own free ligand in two protonation states across seven bacterial entries. The purpose is narrow and worth stating at the outset. AutoDock Vina is not parameterised for transition metals, so a score for a metal complex is defensible only when no receptor atom can reach the metal; that condition was tested geometrically before any calculation and is satisfied here. What follows therefore describes a rigid organic scaffold whose shape the metal fixes, not cobalt chemistry, and the scores are predictions of an empirical function, not measured affinities.

## LITERATURE REVIEW AND METHODOLOGY

**Docking protocol.** All docking used AutoDock Vina 1.2.5 [7,18] with the vina scoring function, exhaustiveness 32, nine output modes, an energy range of  $3 \text{ kcal mol}^{-1}$  and three seeds (11, 22, 33). Each search box was centred on the centroid of the co-crystallised native ligand and padded 8 Å on each axis, with every edge clamped to 20–30 Å. Receptors were prepared from the deposited coordinates with PDBFixer [6]: missing side chains completed,

missing residues left unmodelled and recorded, hydrogens added at pH 7.4. H<sub>2</sub>PGTS was docked both as the neutral crystallographic Z isomer and as the carboxylate anion expected at pH 7.4; both were built with RDKit [9] and converted with Meeko [16], and both were treated as flexible. Every parameter above was fixed before any score was computed and none was altered afterwards.

**The metal centre.** Vina has no cobalt atom type. Substituting another metal type is defensible only if the metal is sterically unreachable, so this was measured rather than assumed: a probe sphere was rolled over the metal and the fraction of directions from which it could contact the metal without striking another atom was computed. For Co(PGTS)<sub>2</sub> that fraction is 0.0000 for an oxygen (1.52 Å), a carbon (1.70 Å) and a sulfur (1.80 Å) probe — the two tridentate arms enclose the metal completely. The primary runs therefore type the metal as Zn, and the choice was tested directly by repeating the identical search with the metal typed as Mg and as Fe.

**Structural input and the special position.** The geometry was taken unchanged from the X-ray structure and docked rigidly with zero rotatable bonds. The cobalt lies on a crystallographic two-fold axis at ( $\frac{1}{2}$ , y,  $\frac{1}{4}$ ) in C2/c, so the asymmetric unit holds only half the complex and a formula or burial computed from it would be wrong; the preparation step therefore expands the component by every space-group operation leaving the metal invariant, giving 45 atoms and the refined formula C<sub>18</sub>H<sub>14</sub>CoN<sub>6</sub>O<sub>4</sub>S<sub>2</sub>. The recovered sphere is octahedral CoN<sub>2</sub>O<sub>2</sub>S<sub>2</sub> with Co–N 1.881, Co–O 1.955 and Co–S 2.207 Å, each twice; the complex carries 31 heavy atoms and four polar hydrogens, the refined structure omitting the hydrazine N–H, which matters for the contact counts below.

**Redocking validation.** Each receptor's own co-crystallised ligand was redocked under the identical protocol and its top pose compared with the crystallographic pose, using a symmetry-aware (graph-isomorphism) RMSD with a 2.0 Å acceptance threshold set in advance. Targets that fail remain in every table, labelled, and their scores are qualified rather than removed. The control was not re-executed in the present run: redocking accuracy is a property of the receptor and of its own ligand and does not involve the compound under test, so these values come from an earlier execution of the identical protocol on these same receptors, which is stated rather than presented as output of this run. Three targets were re-executed before the control was stopped for computing time and reproduced the earlier values closely (1JIL 1.36 against 1.36 Å, 4URO 0.68 against 0.69 Å over three seeds; 4JRN 3.58 against 3.42 Å at one seed), and the prepared receptors are atom-for-atom identical to the earlier preparation in ten of twelve entries. One verdict is nonetheless *not* robust to a deviation of that size, and it is named in the results.

**Interaction profiling and consensus.** Contacts of the best pose of each combination were assigned with PLIP [1]. The chelate's own coordination sphere is reported by PLIP as a metal-complex interaction of the ligand with itself; those records are excluded, so every contact counted is a protein contact. Two-dimensional diagrams reproduce the docked geometry directly, with coordination bonds assigned from the Cordero covalent radii [5]. As an independent check, every top pose was rescored without re-optimisation using the

Vinardo function [14] in smina [8]; Vinardo is reported because it disagrees with Vina on this compound set, not as corroboration. Vina and Vinardo values are never merged.

**Software.** AutoDock Vina 1.2.5; smina 2020.12.10; Open Babel 3.2.1 [12]; RDKit 2025.09.1; Meeko 0.6.1; PDBFixer 1.11 with OpenMM 8.3.1; gemmi 0.7.3; PLIP 2.4.0; spyrmsd 0.9.0. One reproducibility limitation should be stated plainly: the thread count was left unset, so Vina detects the cores and the multithreaded search is not deterministic. Re-running the free-ligand calculations reproduced seed-matched scores of an earlier execution to within 0.293 kcal mol<sup>-1</sup> (mean absolute deviation 0.017 over 108 comparisons, 48 exact). Nothing at 0.1 kcal mol<sup>-1</sup> resolution is affected, but a bit-exact rerun needs the thread count pinned to one — deliberately not changed after scores had been seen.

## RESULTS

**Target panel and validation.** Seven bacterial entries were assembled (1-jadval). One, 2VXY, carries no small-molecule native ligand from which a search box could be defined and could not be docked under the box rule fixed in advance; it is retained in the table with that status rather than omitted. The remaining six were docked and all six passed the redocking control, with mean symmetry-corrected RMSD values from 0.42 Å (2RHL) to 1.85 Å (6F86) (2-jadval). One qualification belongs with that statement: 6F86 passes by only 0.15 Å, a margin smaller than the run-to-run deviation reported above, so its verdict is borderline rather than comfortable. Every other target clears the threshold by at least 0.74 Å.

Table 1

**AutoDock Vina 1.2.5 scores (kcal mol<sup>-1</sup>, mean  $\pm$  SD over seeds 11/22/33) for H<sub>2</sub>PGTS, HPGTS<sup>-</sup> and Co(PGTS)<sub>2</sub>, with the score difference between the complex and the carboxylate anion and the number of PLIP-detected protein contacts**

| Target (protein)                       | Source organism                 | PDB ID | Re s. (Å) | Native ligand | H <sub>2</sub> PGTS | HPGTS <sup>-</sup> | Co(PGTS) <sub>2</sub> | $\Delta$ vs HPGTS <sup>-</sup> | Co contacts | HPGTS <sup>-</sup> contacts | Status     |
|----------------------------------------|---------------------------------|--------|-----------|---------------|---------------------|--------------------|-----------------------|--------------------------------|-------------|-----------------------------|------------|
| DNA gyrase subunit B (GyrB)            | <i>Escherichia coli O157:H7</i> | 6F86   | 1.9       | CWW           | -6.46 $\pm$ 0.01    | -6.08 $\pm$ 0.01   | -8.52 $\pm$ 0.01      | -2.44                          | 6           | 8                           | validated  |
| Tyrosyl-tRNA synthetase (TyrRS)        | <i>Staphylococcus aureus</i>    | 1JIL   | 2.2       | 485           | -7.20 $\pm$ 0.01    | -7.07 $\pm$ 0.00   | -8.82 $\pm$ 0.01      | -1.75                          | 10          | 6                           | validated  |
| Cell division protein FtsZ             | <i>Staphylococcus aureus</i>    | 4DXD   | 2.0       | 9PC           | -7.52 $\pm$ 0.00    | -7.72 $\pm$ 0.01   | -7.25 $\pm$ 0.02      | +0.47                          | 6           | 11                          | validated  |
| DNA gyrase subunit B (GyrB)            | <i>Staphylococcus aureus</i>    | 4URO   | 2.5       | NOV           | -6.26 $\pm$ 0.01    | -6.01 $\pm$ 0.00   | -8.60 $\pm$ 0.01      | -2.60                          | 6           | 5                           | validated  |
| Transcriptional activator protein LasR | <i>Pseudomonas aeruginosa</i>   | 2UV0   | 1.8       | OHN           | -7.88 $\pm$ 0.12    | -8.04 $\pm$ 0.01   | -6.47 $\pm$ 0.01      | +1.57                          | 5           | 7                           | validated  |
| Cell division protein FtsZ             | <i>Bacillus subtilis</i>        | 2RHL   | 2.4       | GDP           | -6.03 $\pm$ 0.01    | -6.88 $\pm$ 0.01   | -9.54 $\pm$ 0.01      | -2.67                          | 7           | 6                           | validated  |
| Cell division protein FtsZ             | <i>Bacillus subtilis</i>        | 2VXY   | 1.7       | —             | —                   | —                  | —                     | —                              | —           | —                           | not docked |

Contact counts exclude the chelate's own coordination sphere, which PLIP reports as an interaction of the ligand with itself. Protein names and source organisms are from the RCSB entry record with capitalisation normalised and the abbreviation used in the text added. Entries without scores could not be docked under the box rule fixed in advance and are retained with that status. Scores are predictions of an empirical function, not measured affinities. Native-ligand codes (RCSB chemical component dictionary): CWW, 4-(4-bromanylpyrazol-1-yl)-6-(ethylcarbamoylamino)-N-pyridin-3-yl-pyridine-3-carboxamide; 485, [2-AMINO-3-(4-HYDROXY-PHENYL)-PROPIONYLAMINO]- (3,4,5-TRIHYDROXY-6-METHYL-TETRAHYDRO-PYRAN-2-YL)- ACETIC ACID; 9PC, PC190723; NOV, novobiocin; OHN, N-3-oxo-dodecanoyl-L-homoserine lactone; GDP, guanosine 5'-diphosphate.

## Redocking validation of the receptors used in this study

| PDB ID | Native-ligand score | RMSD, plain (Å) | RMSD, symmetry-aware (Å) | Best of 9 poses (Å) | Rank of best | Verdict   |
|--------|---------------------|-----------------|--------------------------|---------------------|--------------|-----------|
| 6F86   | −8.02               | 1.85            | 1.85                     | 1.68                | 2            | validated |
| 1JIL   | −9.47               | 1.36            | 1.23                     | 0.35                | 2            | validated |
| 4DXD   | −10.05              | 1.05            | 1.05                     | 1.05                | 1            | validated |
| 4URO   | −9.71               | 0.69            | 0.58                     | 0.58                | 1            | validated |
| 2UV0   | −8.54               | 1.21            | 1.21                     | 1.21                | 1            | validated |
| 2RHL   | −9.12               | 0.89            | 0.42                     | 0.42                | 1            | validated |

The co-crystallised ligand of each entry was redocked under the identical protocol and its top pose compared with the crystallographic pose; values are means over the three seeds. Symmetry-aware RMSD is the graph-isomorphism value (spyrmsd) and governs the verdict at a 2.0 Å threshold fixed in advance. “Best of 9” is the lowest symmetry-aware RMSD anywhere in the nine output poses, with the rank at which it occurs — a low value at a high rank indicates a ranking rather than a sampling failure. Validation is a property of the receptor and its own native ligand, not of the docked compound; these values come from an earlier execution of the identical protocol, as stated in the methodology. Entries absent from this table had no small-molecule ligand to redock.

**Scores.** Co(PGTS)<sub>2</sub> scores between −9.54 and −6.47 kcal mol<sup>−1</sup> across the six docked targets (1-jadval, 1-rasm). Seed-to-seed reproducibility is high: the standard deviation over the three seeds does not exceed 0.13 kcal mol<sup>−1</sup> for any combination. The complex is scored below both free-ligand forms on four of the six targets; the exceptions are 4DXD, where it is 0.47 kcal mol<sup>−1</sup> above the carboxylate anion, and 2UV0, where it is 1.57 kcal mol<sup>−1</sup> above. The largest separation in favour of the complex is at *B. subtilis* FtsZ (2RHL), −9.54 ± 0.01 against −6.88 kcal mol<sup>−1</sup> for the anion, a difference of 2.67 kcal mol<sup>−1</sup>; the mean difference over the arm is −1.24 kcal mol<sup>−1</sup>. Deprotonation of the free ligand changes its score by at most 0.84 kcal mol<sup>−1</sup>, so the choice of free-ligand reference does not drive the comparison.

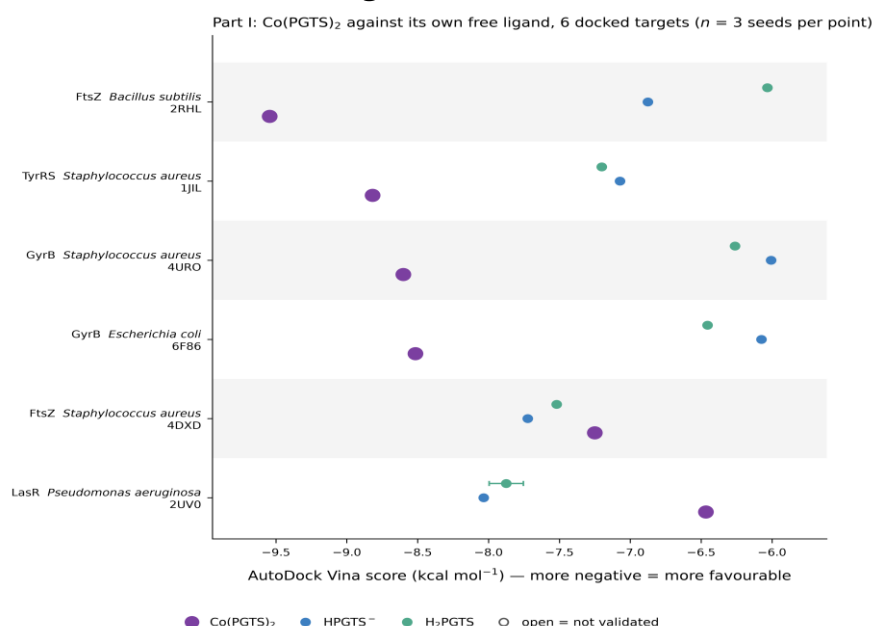


**Figure 1** .AutoDock Vina scores for the three compounds of this study — Co(PGTS)<sub>2</sub>, HPGTS<sup>−</sup> and H<sub>2</sub>PGTS — across the docked targets of this article, ordered by the complex score. Points are means over the three seeds and error bars the standard deviation, smaller than the symbol for most combinations; open symbols mark targets that did not pass redocking validation. Scores are predictions of an empirical function, not measured affinities.

**Binding modes.** At FtsZ of *B. subtilis* (2RHL) the complex occupies the interdomain nucleotide cleft and engages it from both chelate arms (2-rasm), while the free ligand reaches only part of the same pocket. At gyrase B of *S. aureus* (4URO) the complex bridges both subunits of the dimer, hydrogen-bonding to Arg84 of chain A and Tyr229 of chain D and  $\pi$ -stacking with Phe204 (D); the free ligand reaches Asn54, Gly125 and Glu58 near the entrance of the same pocket and contacts one subunit only.

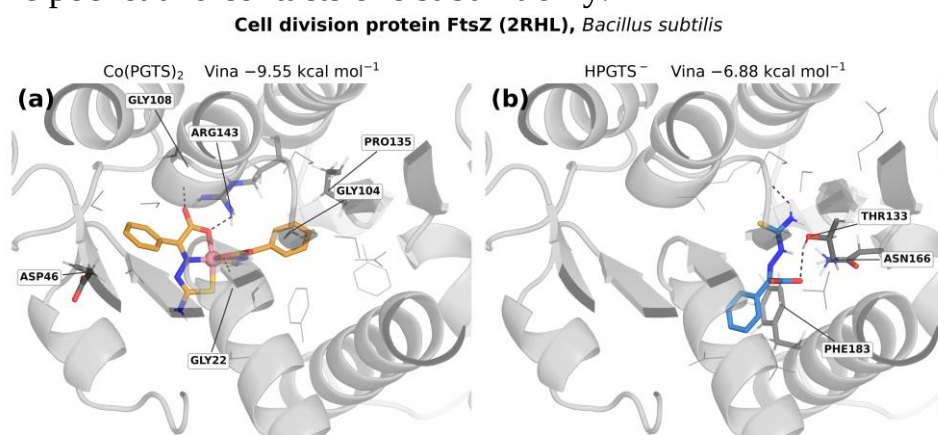


**Figure 2.**Best-scoring poses of Co(PGTS)<sub>2</sub> (a) and of its own free ligand HPGTS<sup>−</sup> (b) in *Bacillus subtilis* FtsZ (2RHL). Both panels share one camera, so the poses are directly comparable. The six Co–donor bonds are drawn explicitly: PyMOL perceives bonds by distance and does not bond metals, so they must be created from the coordinates; the recovered distances are Co–N 1.881, Co–O 1.955 and Co–S 2.207 Å, each twice. Contact residues are labelled with leader lines. Receptor cartoon grey and translucent; ligand carbons amber (complex) or blue (HPGTS<sup>−</sup>). Cobalt is the salmon sphere at the centre of the six donor bonds; further spheres are retained receptor cofactors.

**Metal-type sensitivity.** Re-docking at seed 11 with the metal typed as Mg and again as Fe changed no score at all: the largest absolute difference from the Zn runs is 0.0000 kcal mol<sup>−1</sup> over the twelve comparisons of this arm (3-jadval, 3-rasm). This is the expected consequence of an accessible fraction of exactly zero, and it is the clearest evidence in the study that the scoring function does not see the metal.

Table 3

**Metal-type sensitivity of Co(PGTS)<sub>2</sub>**

| PDB ID | Zn (as docked)<br>mean $\pm$ SD, 3<br>seeds | Zn<br>seed 11 | Mg<br>seed 11 | Fe<br>seed 11 | $\Delta$ (Mg–Zn)<br>seed 11 | $\Delta$ (Fe–Zn)<br>seed 11 |
|--------|---------------------------------------------|---------------|---------------|---------------|-----------------------------|-----------------------------|
| 6F86   | $-8.52 \pm 0.01$                            | −8.51         | −8.51         | −8.51         | 0.00                        | 0.00                        |
| 1JIL   | $-8.82 \pm 0.01$                            | −8.82         | −8.82         | −8.82         | 0.00                        | 0.00                        |
| 4DXD   | $-7.25 \pm 0.02$                            | −7.23         | −7.23         | −7.23         | 0.00                        | 0.00                        |
| 4URO   | $-8.60 \pm 0.01$                            | −8.61         | −8.61         | −8.61         | 0.00                        | 0.00                        |

|      |                  |       |       |       |      |      |
|------|------------------|-------|-------|-------|------|------|
| 2UV0 | $-6.47 \pm 0.01$ | -6.47 | -6.47 | -6.47 | 0.00 | 0.00 |
| 2RHL | $-9.54 \pm 0.01$ | -9.55 | -9.55 | -9.55 | 0.00 | 0.00 |

AutoDock Vina has no cobalt atom type; the primary runs type the buried metal as Zn, and the sensitivity runs repeat the identical rigid-body search with the metal typed as Mg and as Fe. The Mg, Fe and  $\Delta$  columns are seed-11 values and the seed-11 Zn score is given alongside so the comparison is like for like; the second column is the three-seed mean of Table 1. Every difference is zero to four decimal places, the expected consequence of a metal no receptor atom can reach.

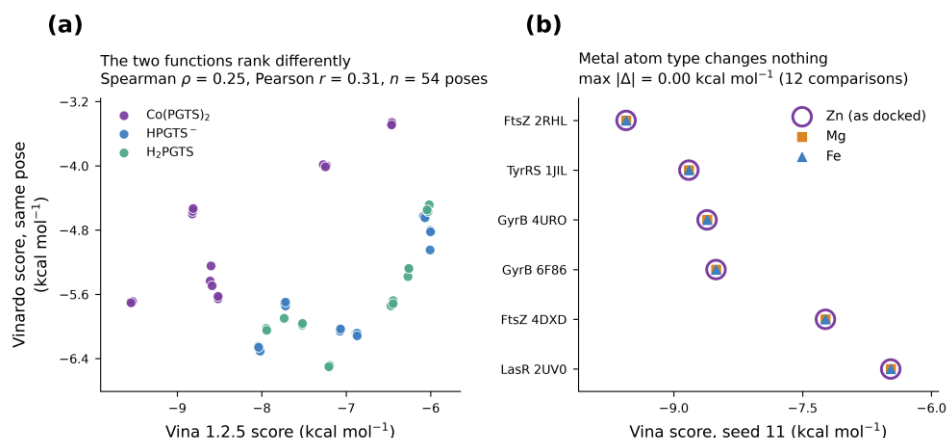


**Figure 3.** (a) Vinardo rescoring of the same top-ranked Vina poses — no re-search was performed, only the energy function differs. Each point is one pose; the two functions do not reproduce each other's ranking, which is the principal qualification of this study. (b) Metal-type sensitivity: the Zn (as docked), Mg and Fe points coincide exactly on every target, because the cobalt is fully enclosed by its two tridentate ligands and no receptor atom reaches it.

## Discussions

The metal-type result defines what the scores can and cannot mean. Because no receptor atom reaches the cobalt, the calculation describes a rigid organic scaffold held in a particular shape by a metal the scoring function is blind to. A favourable score is therefore evidence about shape and about the disposition of the carboxylate, thioamide and phenyl groups; it is not evidence about cobalt coordination, and it must not be read as such.

Rescoring with Vinardo changes the picture materially. Over the 54 rescored poses of this arm the rank correlation between the two functions is weak (Spearman  $\rho = 0.25$ , Pearson  $r = 0.31$ ). Vina ranks the complex first on four of the six targets; Vinardo ranks it first on only one, and the two functions select the same best compound on two of six (4-jadval, 3-rasm). This is the single most important qualification of the study: the preference for the complex is a property of one scoring function rather than a robust feature of the poses.

Table.4

### Which compound each scoring function ranks first, per target

| PDB ID | Best compound, Vina   | Best compound, Vinardo | Co(PGT S) <sub>2</sub> , Vina | Co(PGTS ) <sub>2</sub> , Vinardo | HPGTS <sup>-</sup> , Vinardo | H <sub>2</sub> PGTS, Vinardo | Same winner ? |
|--------|-----------------------|------------------------|-------------------------------|----------------------------------|------------------------------|------------------------------|---------------|
| 6F86   | Co(PGTS) <sub>2</sub> | H <sub>2</sub> PGTS    | -8.52                         | -5.64                            | -4.63                        | -5.71                        | no            |
| 1JIL   | Co(PGTS) <sub>2</sub> | H <sub>2</sub> PGTS    | -8.82                         | -4.57                            | -6.04                        | -6.50                        | no            |

|      |                       |                       |       |       |       |       |     |
|------|-----------------------|-----------------------|-------|-------|-------|-------|-----|
| 4DXD | HPGTS <sup>-</sup>    | H <sub>2</sub> PGTS   | -7.25 | -4.00 | -5.72 | -5.97 | no  |
| 4URO | Co(PGTS) <sub>2</sub> | Co(PGTS) <sub>2</sub> | -8.60 | -5.39 | -4.89 | -5.31 | yes |
| 2UV0 | HPGTS <sup>-</sup>    | HPGTS <sup>-</sup>    | -6.47 | -3.48 | -6.27 | -5.99 | yes |
| 2RHL | Co(PGTS) <sub>2</sub> | HPGTS <sup>-</sup>    | -9.54 | -5.70 | -6.09 | -4.53 | no  |

Vinardo values are rescores of the same Vina poses (smina --score\_only); no re-search was performed. Vina and Vinardo numbers are never merged, and the two functions are not averaged into a consensus score.

A second observation points the same way. The complex makes *fewer* protein contacts than the free ligand across this arm — 40 against 43 in total, and 12 hydrogen bonds against 18 — while scoring more favourably on most targets. Part of that gap is a property of the deposited model rather than of the chemistry: the refined structure lacks the hydrazine N–H hydrogens, so the prepared complex offers two fewer hydrogen-bond donors than the ligand would if those hydrogens were located. A redetermination that places them could narrow the difference.

Three further limitations bound the work. The receptors were rigid, and an unmodelled internal loop lies within 8 Å of the search box in two of the six docked targets (6F86, an 18-residue gap flanked by Leu98–Gly117; 4URO, a 19-residue gap flanked by Val105–Gly125). Solvation on binding, change of coordination number and ligand-exchange equilibria in aqueous medium are outside the model entirely. And no biochemical or cell-based assay was performed: the values reported here are docking scores, not binding constants, and they are not evidence of inhibition or of biological activity.

## CONCLUSIONS

A fixed, fully logged docking protocol was applied to Co(PGTS)<sub>2</sub> in its unmodified X-ray geometry and to its free ligand in two protonation states across seven bacterial targets, six of which could be docked and all six of which passed redocking validation, one of them only marginally. With the Vina scoring function the preformed cobalt bis-chelate is scored below both free-ligand forms on four of the six targets, most strongly at *B. subtilis* FtsZ and at staphylococcal gyrase B. Three findings bound that observation: the metal is sterically inaccessible and therefore invisible to the scoring function, so the result describes a rigid organic scaffold rather than cobalt chemistry; the second scoring function reproduces the ordering on only one of six targets; and the complex makes fewer protein contacts than its own free ligand, in a way traceable to missing hydrogens in the deposited model. The calculations are consequently useful for prioritising *B. subtilis* FtsZ and staphylococcal gyrase B for experimental testing, and not as evidence of activity. No biochemical assay was performed.

## REFERENCES

1. Adasme, M., Linnemann, K. L., Bolz, S. N., Kaiser, F., Salentin, S., Haupt, V. J., & Schroeder, M. (2021). PLIP 2021: Expanding the scope of the protein–ligand interaction profiler to DNA and RNA. *Nucleic Acids Research*, 49, W530–W534. <https://doi.org/10.1093/nar/gkab294>

2. Anderson, D. E., Gueiros-Filho, F. J., & Erickson, H. P. (2004). Assembly dynamics of FtsZ rings in *Bacillus subtilis* and *Escherichia coli* and effects of FtsZ-regulating proteins. *Journal of Bacteriology*, 186, 5775–5781. <https://doi.org/10.1128/JB.186.17.5775-5781.2004>
3. Bottomley, M. J., Muraglia, E., Bazzo, R., & Carfi, A. (2007). Molecular insights into quorum sensing in the human pathogen *Pseudomonas aeruginosa* from the structure of the virulence regulator LasR bound to its autoinducer. *Journal of Biological Chemistry*, 282, 13592–13600. <https://doi.org/10.1074/jbc.M700556200>
4. Casas, J. S., García-Tasende, M. S., & Sordo, J. (2000). Main group metal complexes of semicarbazones and thiosemicarbazones: A structural review. *Coordination Chemistry Reviews*, 209, 197–261. [https://doi.org/10.1016/S0010-8545\(00\)00363-5](https://doi.org/10.1016/S0010-8545(00)00363-5)
5. Cordero, B., Gómez, V., Platero-Prats, A. E., Revés, M., Echeverría, J., Cremades, E., Barragán, F., & Alvarez, S. (2008). Covalent radii revisited. *Dalton Transactions*, 2832–2838. <https://doi.org/10.1039/B801115J>
6. Eastman, P., Swails, J., Chodera, J. D., McGibbon, R. T., Zhao, Y., Beauchamp, K. A., et al. (2017). OpenMM 7: Rapid development of high performance algorithms for molecular dynamics. *PLOS Computational Biology*, 13, e1005659. <https://doi.org/10.1371/journal.pcbi.1005659>
7. Eberhardt, J., Santos-Martins, D., Tillack, A. F., & Forli, S. (2021). AutoDock Vina 1.2.0: New docking methods, expanded force field, and Python bindings. *Journal of Chemical Information and Modeling*, 61, 3891–3898. <https://doi.org/10.1021/acs.jcim.1c00203>
8. Koes, D. R., Baumgartner, M. P., & Camacho, C. J. (2013). Lessons learned in empirical scoring with smina from the CSAR 2011 benchmarking exercise. *Journal of Chemical Information and Modeling*, 53, 1893–1904. <https://doi.org/10.1021/ci300604z>
9. Landrum, G., et al. (n.d.). *RDKit: Open-source cheminformatics* [Computer software]. <https://www.rdkit.org>
10. Maxwell, A. (1997). DNA gyrase as a drug target. *Trends in Microbiology*, 5, 102–109. [https://doi.org/10.1016/S0966-842X\(96\)10085-8](https://doi.org/10.1016/S0966-842X(96)10085-8)
11. Munteanu, C. R., & Suntharalingam, K. (2015). Advances in cobalt complexes as anticancer agents. *Dalton Transactions*, 44, 13796–13808. <https://doi.org/10.1039/C5DT02101D>
12. O'Boyle, N. M., Banck, M., James, C. A., Morley, C., Vandermeersch, T., & Hutchison, G. R. (2011). Open Babel: An open chemical toolbox. *Journal of Cheminformatics*, 3, 33. <https://doi.org/10.1186/1758-2946-3-33>
13. Qiu, J., Gao, C., & Wang, X. (2014). Aminoacyl-tRNA synthetases as antibacterial drug targets. *Current Topics in Medicinal Chemistry*, 14, 1273–1282. <https://doi.org/10.2174/1568026614666140523115936>
14. Quiroga, R., & Villarreal, M. A. (2016). Vinardo: A scoring function based on AutoDock Vina improves scoring, docking, and virtual screening. *PLOS ONE*, 11, e0155183. <https://doi.org/10.1371/journal.pone.0155183>

15. Rosu, T., Gulea, A., Nicolae, A., & Georgescu, R. (2007). Complexes of 3d<sup>n</sup> metal ions with thiosemicarbazones: Synthesis and antimicrobial activity. *Molecules*, 12, 782–796. <https://doi.org/10.3390/12040782>

16. Santos-Martins, D., et al. (n.d.). *Meeko: Preparation of small molecules for AutoDock* [Computer software]. <https://github.com/forlilab/Meeko>

17. Tillaeva, N. S., et al. (2022). Synthesis and crystal structure of phenylglyoxylic acid thiosemicarbazone. *Zeitschrift für Naturforschung B*, 77, 117–123. <https://doi.org/10.1515/znb-2021-0170>

18. Trott, O., & Olson, A. J. (2010). AutoDock Vina: Improving the speed and accuracy of docking with a new scoring function, efficient optimization, and multithreading. *Journal of Computational Chemistry*, 31, 455–461. <https://doi.org/10.1002/jcc.21334>