

# wwPDB X-ray Structure Validation Summary Report (i)

### May 24, 2020 - 10:00 am BST

| PDB ID                 | : | 1SCN   |
|------------------------|---|--|
| $\operatorname{Title}$ | : | INACTIVATION OF SUBTILISIN CARLSBERG BY N-(TERT-BUTOXY |
|                        |   | CARBONYL-ALANYL-PROLYL-PHENYLALANYL)-O-BENZOL HY-      |
|                        |   | DROXYLAMINE: FORMATION OF COVALENT ENZYME-INHIBITOR    |
|                        |   | LINKAGE IN THE FORM OF A CARBAMATE DERIVATIVE          |
| Authors                | : | Steinmetz, A.C.U.; Demuth, HU.; Ringe, D.              |
| Deposited on           |   |  |
| Resolution             | : | 1.90 Å(reported)                                       |

This is a wwPDB X-ray Structure Validation Summary Report for a publicly released PDB entry.

We welcome your comments at validation@mail.wwpdb.org A user guide is available at https://www.wwpdb.org/validation/2017/XrayValidationReportHelp with specific help available everywhere you see the (i) symbol.

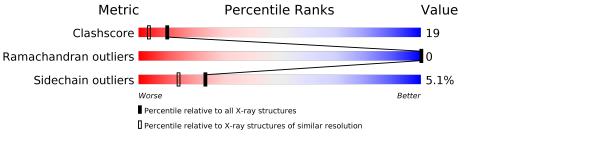
The following versions of software and data (see references (1)) were used in the production of this report:

# 1 Overall quality at a glance (i)

The following experimental techniques were used to determine the structure: X-RAY DIFFRACTION

The reported resolution of this entry is 1.90 Å.

Percentile scores (ranging between 0-100) for global validation metrics of the entry are shown in the following graphic. The table shows the number of entries on which the scores are based.



| Metric                | Whole archive $(\#Entries)$ | ${f Similar\ resolution}\ (\#{ m Entries,\ resolution\ range}({ m \AA}))$ |
|-----------------------|-----------------------------|---|
| Clashscore            | 141614                      | 6847 (1.90-1.90)  |
| Ramachandran outliers | 138981                      | 6760 (1.90-1.90)  |
| Sidechain outliers    | 138945                      | 6760 (1.90-1.90)  |

The table below summarises the geometric issues observed across the polymeric chains and their fit to the electron density. The red, orange, yellow and green segments on the lower bar indicate the fraction of residues that contain outliers for >=3, 2, 1 and 0 types of geometric quality criteria respectively. A grey segment represents the fraction of residues that are not modelled. The numeric value for each fraction is indicated below the corresponding segment, with a dot representing fractions <=5%

Note EDS was not executed.

| Mol | Chain | Length | Quality of chain |     |      |
|-----|-------|--------|------------------|-----|------|
| 1   | Б     | 070    |                  |     |      |
|     | E     | 276    | 68%              | 26% | 5% • |



# 2 Entry composition (i)

There are 5 unique types of molecules in this entry. The entry contains 2094 atoms, of which 0 are hydrogens and 0 are deuteriums.

In the tables below, the ZeroOcc column contains the number of atoms modelled with zero occupancy, the AltConf column contains the number of residues with at least one atom in alternate conformation and the Trace column contains the number of residues modelled with at most 2 atoms.

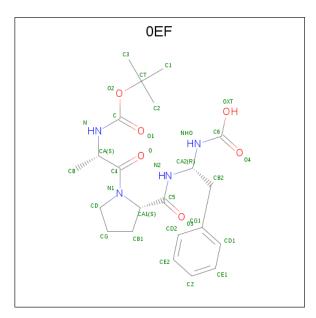
• Molecule 1 is a protein called SUBTILISIN CARLSBERG.

| Mol | Chain | Residues | Atoms         |           |          | ZeroOcc  | AltConf        | Trace |   |   |
|-----|-------|----------|---------------|-----------|----------|----------|----------------|-------|---|---|
| 1   | Е     | 274      | Total<br>1919 | C<br>1190 | N<br>332 | O<br>392 | ${ m S}{ m 5}$ | 0     | 0 | 0 |

There are 5 discrepancies between the modelled and reference sequences:

| Chain | Residue | Modelled | Actual | Comment  | Reference  |
|-------|---------|----------|--------|----------|------------|
| Е     | 103     | SER      | THR    | CONFLICT | UNP P00780 |
| Е     | 129     | ALA      | PRO    | CONFLICT | UNP P00780 |
| Е     | 158     | ASN      | SER    | CONFLICT | UNP P00780 |
| Е     | 161     | SER      | ASN    | CONFLICT | UNP P00780 |
| Е     | 212     | ASN      | SER    | CONFLICT | UNP P00780 |

• Molecule 2 is N-(tert-butoxycarbonyl)-L-alanyl-N-[(1R)-1-(carboxyamino)-2-phenylethyl]-L -prolinamide (three-letter code: 0EF) (formula:  $C_{22}H_{32}N_4O_6$ ).





| Mol | Chain | Residues | Atoms       |         |        |        | ZeroOcc | AltConf |
|-----|-------|----------|-------------|---------|--------|--------|---------|---------|
| 2   | Е     | 1        | Total<br>31 | C<br>22 | N<br>4 | O<br>5 | 0       | 0       |

• Molecule 3 is CALCIUM ION (three-letter code: CA) (formula: Ca).

| Mol | Chain | Residues | Atoms           | ZeroOcc | AltConf |
|-----|-------|----------|-----------------|---------|---------|
| 3   | Е     | 2        | Total Ca<br>2 2 | 0       | 0       |

• Molecule 4 is SODIUM ION (three-letter code: NA) (formula: Na).

| Mol | Chain | Residues | Atoms           | ZeroOcc | AltConf |
|-----|-------|----------|-----------------|---------|---------|
| 4   | E     | 1        | Total Na<br>1 1 | 0       | 0       |

• Molecule 5 is water.

| Mol | Chain | Residues | Atoms              | ZeroOcc | AltConf |
|-----|-------|----------|--------------------|---------|---------|
| 5   | Е     | 141      | Total O<br>141 141 | 0       | 0       |



# 3 Residue-property plots (i)

These plots are drawn for all protein, RNA and DNA chains in the entry. The first graphic for a chain summarises the proportions of the various outlier classes displayed in the second graphic. The second graphic shows the sequence view annotated by issues in geometry. Residues are colorcoded according to the number of geometric quality criteria for which they contain at least one outlier: green = 0, yellow = 1, orange = 2 and red = 3 or more. Stretches of 2 or more consecutive residues without any outlier are shown as a green connector. Residues present in the sample, but not in the model, are shown in grey.

Note EDS was not executed.

- Molecule 1: SUBTILISIN CARLSBERG



# 4 Data and refinement statistics (i)

Xtriage (Phenix) and EDS were not executed - this section is therefore incomplete.

| Property                               | Value   | Source    |  |
|--|---|-----------|--|
| Space group                            | P 21 21 21                                      | Depositor |  |
| Cell constants                         | 76.50Å $55.40$ Å $53.40$ Å                      | Depositor |  |
| a, b, c, $\alpha$ , $\beta$ , $\gamma$ | $90.00^{\circ}$ $90.00^{\circ}$ $90.00^{\circ}$ | Depositor |  |
| Resolution (Å)                         | 20.00 - 1.90                                    | Depositor |  |
| % Data completeness                    | (Not available) (20.00-1.90)                    | Depositor |  |
| (in resolution range)                  | (1000 available) (20.00 1.50)                   | Depositor |  |
| $R_{merge}$                            | (Not available)                                 | Depositor |  |
| R <sub>sym</sub>                       | (Not available)                                 | Depositor |  |
| Refinement program                     | TNT   | Depositor |  |
| $R, R_{free}$                          | 0.191 , (Not available)                         | Depositor |  |
| Estimated twinning fraction            | No twinning to report.                          | Xtriage   |  |
| Total number of atoms                  | 2094  | wwPDB-VP  |  |
| Average B, all atoms $(Å^2)$           | 20.0  | wwPDB-VP  |  |



# 5 Model quality (i)

# 5.1 Standard geometry (i)

Bond lengths and bond angles in the following residue types are not validated in this section: NA, CA,  $0\mathrm{EF}$ 

The Z score for a bond length (or angle) is the number of standard deviations the observed value is removed from the expected value. A bond length (or angle) with |Z| > 5 is considered an outlier worth inspection. RMSZ is the root-mean-square of all Z scores of the bond lengths (or angles).

| Mal | Mol Chain |      | nd lengths    | Bond angles |                |  |
|-----|-----------|------|---------------|-------------|----------------|--|
|     | Chain     | RMSZ | # Z  > 5      | RMSZ        | # Z  > 5       |  |
| 1   | Е         | 0.80 | 4/1951~(0.2%) | 1.64        | 28/2662~(1.1%) |  |

All (4) bond length outliers are listed below:

| Mol | Chain | $\mathbf{Res}$ | Type | Atoms  | Z    | $\operatorname{Observed}(\operatorname{\AA})$ | Ideal(Å) |
|-----|-------|----------------|------|--------|------|---|----------|
| 1   | Ε     | 15             | LYS  | CE-NZ  | 8.77 | 1.71  | 1.49     |
| 1   | Е     | 54             | GLU  | CD-OE1 | 6.56 | 1.32  | 1.25     |
| 1   | Е     | 197            | GLU  | CD-OE1 | 6.48 | 1.32  | 1.25     |
| 1   | Е     | 112            | GLU  | CD-OE2 | 5.89 | 1.32  | 1.25     |

The worst 5 of 28 bond angle outliers are listed below:

| Mol | Chain | Res | Type | Atoms     | Z      | $\mathbf{Observed}(^{o})$ | $Ideal(^{o})$ |
|-----|-------|-----|------|-----------|--------|---------------------------|---------------|
| 1   | Е     | 181 | ASP  | CB-CG-OD2 | -12.98 | 106.62                    | 118.30        |
| 1   | Е     | 181 | ASP  | CB-CG-OD1 | 12.12  | 129.21                    | 118.30        |
| 1   | Е     | 167 | TYR  | CB-CG-CD1 | -9.94  | 115.03                    | 121.00        |
| 1   | Е     | 32  | ASP  | CB-CG-OD1 | -9.39  | 109.85                    | 118.30        |
| 1   | Е     | 140 | ASP  | CB-CG-OD2 | -7.93  | 111.16                    | 118.30        |

There are no chirality outliers.

There are no planarity outliers.

# 5.2 Too-close contacts (i)

In the following table, the Non-H and H(model) columns list the number of non-hydrogen atoms and hydrogen atoms in the chain respectively. The H(added) column lists the number of hydrogen atoms added and optimized by MolProbity. The Clashes column lists the number of clashes within the asymmetric unit, whereas Symm-Clashes lists symmetry related clashes.



| Mol | Chain | Non-H | H(model) | H(added) | Clashes | Symm-Clashes |
|-----|-------|-------|----------|----------|---------|--------------|
| 1   | Е     | 1919  | 0        | 1879     | 64      | 5            |
| 2   | Е     | 31    | 0        | 31       | 12      | 8            |
| 3   | Е     | 2     | 0        | 0        | 0       | 0            |
| 4   | Е     | 1     | 0        | 0        | 0       | 0            |
| 5   | Е     | 141   | 0        | 0        | 8       | 4            |
| All | All   | 2094  | 0        | 1910     | 73      | 9            |

The all-atom clashscore is defined as the number of clashes found per 1000 atoms (including hydrogen atoms). The all-atom clashscore for this structure is 19.

The worst 5 of 73 close contacts within the same asymmetric unit are listed below, sorted by their clash magnitude.

| Atom-1           | Atom-2          | Interatomic<br>distance (Å) | Clash<br>overlap (Å) |
|------------------|-----------------|-----------------------------|----------------------|
| 1:E:15:LYS:NZ    | 1:E:15:LYS:CE   | 1.71                        | 1.51                 |
| 2:E:447:0EF:N2   | 2:E:447:0EF:CA2 | 1.84                        | 1.36                 |
| 1:E:158:ASN:HD22 | 1:E:158:ASN:N   | 1.53                        | 1.03                 |
| 1:E:242:SER:H    | 1:E:245:GLN:NE2 | 1.56                        | 1.03                 |
| 1:E:158:ASN:H    | 1:E:158:ASN:ND2 | 1.63                        | 0.96                 |

The worst 5 of 9 symmetry-related close contacts are listed below. The label for Atom-2 includes the symmetry operator and encoded unit-cell translations to be applied.

| Atom-1         | Atom-2                  | $\begin{array}{c} {\rm Interatomic}\\ {\rm distance}~({\rm \AA}) \end{array}$ | Clash<br>overlap (Å) |
|----------------|-------------------------|---|----------------------|
| 2:E:447:0EF:C3 | 5:E:852:HOH:O[4_477]    | 0.79  | 1.41                 |
| 2:E:447:0EF:C1 | 5:E:650:HOH:O[4_477]    | 1.07  | 1.13                 |
| 1:E:249:ARG:CG | $2:E:447:0EF:C2[4_577]$ | 1.38  | 0.82                 |
| 1:E:249:ARG:CD | 2:E:447:0EF:C2[4_577]   | 1.44  | 0.76                 |
| 2:E:447:0EF:CT | 5:E:852:HOH:O[4_477]    | 1.62  | 0.58                 |

### 5.3 Torsion angles (i)

#### 5.3.1 Protein backbone (i)

In the following table, the Percentiles column shows the percent Ramachandran outliers of the chain as a percentile score with respect to all X-ray entries followed by that with respect to entries of similar resolution.

The Analysed column shows the number of residues for which the backbone conformation was analysed, and the total number of residues.



| Mol | Chain | Analysed      | Favoured  | Allowed | Outliers | Percentiles |  |
|-----|-------|---------------|-----------|---------|----------|-------------|--|
| 1   | Ε     | 272/276~(99%) | 263~(97%) | 9~(3%)  | 0        | 100 100     |  |

There are no Ramachandran outliers to report.

#### 5.3.2 Protein sidechains (i)

In the following table, the Percentiles column shows the percent sidechain outliers of the chain as a percentile score with respect to all X-ray entries followed by that with respect to entries of similar resolution.

The Analysed column shows the number of residues for which the sidechain conformation was analysed, and the total number of residues.

| Mol | Chain | Analysed       | Rotameric | Outliers | Percentiles |  |
|-----|-------|----------------|-----------|----------|-------------|--|
| 1   | Ε     | 198/199~(100%) | 188~(95%) | 10~(5%)  | 24 14       |  |

5 of 10 residues with a non-rotameric side chain are listed below:

| Mol | Chain | $\mathbf{Res}$ | Type |
|-----|-------|----------------|------|
| 1   | Е     | 158            | ASN  |
| 1   | Е     | 163            | ASN  |
| 1   | Е     | 188            | SER  |
| 1   | Е     | 101            | SER  |
| 1   | Е     | 172            | ASP  |

Some sidechains can be flipped to improve hydrogen bonding and reduce clashes. 5 of 11 such sidechains are listed below:

| Mol | Chain | Res | Type |
|-----|-------|-----|------|
| 1   | Е     | 141 | ASN  |
| 1   | Е     | 158 | ASN  |
| 1   | Е     | 238 | HIS  |
| 1   | Е     | 97  | ASN  |
| 1   | Е     | 212 | ASN  |

#### 5.3.3 RNA (i)

There are no RNA molecules in this entry.



### 5.4 Non-standard residues in protein, DNA, RNA chains (i)

There are no non-standard protein/DNA/RNA residues in this entry.

# 5.5 Carbohydrates (i)

There are no carbohydrates in this entry.

# 5.6 Ligand geometry (i)

Of 4 ligands modelled in this entry, 3 are monoatomic - leaving 1 for Mogul analysis.

In the following table, the Counts columns list the number of bonds (or angles) for which Mogul statistics could be retrieved, the number of bonds (or angles) that are observed in the model and the number of bonds (or angles) that are defined in the Chemical Component Dictionary. The Link column lists molecule types, if any, to which the group is linked. The Z score for a bond length (or angle) is the number of standard deviations the observed value is removed from the expected value. A bond length (or angle) with |Z| > 2 is considered an outlier worth inspection. RMSZ is the root-mean-square of all Z scores of the bond lengths (or angles).

| ſ | Mol  | Type           | Chain | Res Link |   | Bo       | ond leng | ths      | B        | ond ang | gles     |
|---|------|----------------|-------|----------|---|----------|----------|----------|----------|---------|----------|
|   | WIOI | туре           | Chain | nes      |   | Counts   | RMSZ     | # Z  > 2 | Counts   | RMSZ    | # Z >2   |
|   | 2    | $0\mathrm{EF}$ | Е     | 447      | 1 | 32,32,33 | 6.32     | 5 (15%)  | 41,44,46 | 2.79    | 16 (39%) |

In the following table, the Chirals column lists the number of chiral outliers, the number of chiral centers analysed, the number of these observed in the model and the number defined in the Chemical Component Dictionary. Similar counts are reported in the Torsion and Rings columns. '-' means no outliers of that kind were identified.

| Mol | Type           | Chain | $\mathbf{Res}$ | Link | Chirals | Torsions    | Rings   |
|-----|----------------|-------|----------------|------|---------|-------------|---------|
| 2   | $0\mathrm{EF}$ | Е     | 447            | 1    | -       | 12/30/42/43 | 0/2/2/2 |

All (5) bond length outliers are listed below:

| Mol | Chain | Res | Type            | Atoms   | Z     | $\operatorname{Observed}(\operatorname{\AA})$ | $\operatorname{Ideal}(\operatorname{\AA})$ |
|-----|-------|-----|-----------------|---------|-------|---|--|
| 2   | Ε     | 447 | $0\mathrm{EF}$  | CA2-N2  | 34.73 | 1.84  | 1.45                                       |
| 2   | Е     | 447 | $0 \mathrm{EF}$ | CD-N1   | 4.20  | 1.55  | 1.47                                       |
| 2   | Е     | 447 | $0 \mathrm{EF}$ | CB2-CA2 | 3.42  | 1.56  | 1.53                                       |
| 2   | Е     | 447 | 0EF             | C-N     | 2.97  | 1.42  | 1.34                                       |
| 2   | Ε     | 447 | $0 \mathrm{EF}$ | CB2-CG1 | -2.96 | 1.44  | 1.51                                       |

The worst 5 of 16 bond angle outliers are listed below:



| 1 | SCN           |  |
|---|---------------|--|
| _ | $\sim \sim 1$ |  |

| Mol | Chain | Res | Type | Atoms      | Z     | $\mathbf{Observed}(^{o})$ | $Ideal(^{o})$ |
|-----|-------|-----|------|------------|-------|---------------------------|---------------|
| 2   | Е     | 447 | 0EF  | CT-O2-C    | -7.95 | 108.76                    | 120.99        |
| 2   | Е     | 447 | 0EF  | CB2-CA2-N2 | -7.57 | 85.43                     | 111.28        |
| 2   | Е     | 447 | 0EF  | CA1-C5-N2  | -4.95 | 105.56                    | 116.58        |
| 2   | Е     | 447 | 0EF  | O2-C-O1    | -4.74 | 116.98                    | 125.62        |
| 2   | Е     | 447 | 0EF  | CD-N1-CA1  | -4.34 | 105.12                    | 112.00        |

There are no chirality outliers.

5 of 12 torsion outliers are listed below:

| Mol | Chain | Res | Type | Atoms           |
|-----|-------|-----|------|-----------------|
| 2   | Ε     | 447 | 0EF  | CA-C4-N1-CD     |
| 2   | Е     | 447 | 0EF  | O-C4-N1-CD      |
| 2   | Е     | 447 | 0EF  | N2-CA2-CB2-CG1  |
| 2   | Е     | 447 | 0EF  | CA2-CB2-CG1-CD1 |
| 2   | Е     | 447 | 0EF  | CA2-CB2-CG1-CD2 |

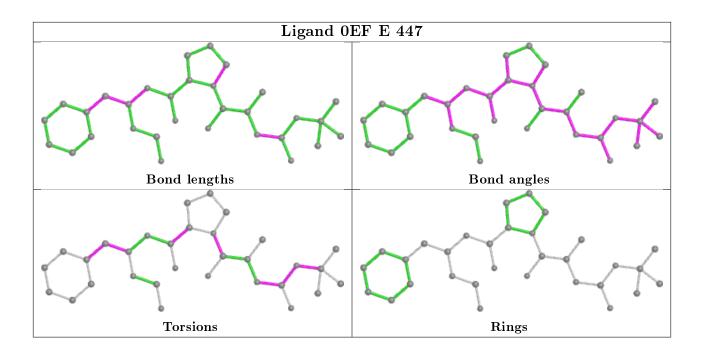
There are no ring outliers.

1 monomer is involved in 20 short contacts:

| Mol | Chain | Res | Type            | Clashes | Symm-Clashes |
|-----|-------|-----|-----------------|---------|--------------|
| 2   | Ε     | 447 | $0 \mathrm{EF}$ | 12      | 8            |

The following is a two-dimensional graphical depiction of Mogul quality analysis of bond lengths, bond angles, torsion angles, and ring geometry for all instances of the Ligand of Interest. In addition, ligands with molecular weight > 250 and outliers as shown on the validation Tables will also be included. For torsion angles, if less then 5% of the Mogul distribution of torsion angles is within 10 degrees of the torsion angle in question, then that torsion angle is considered an outlier. Any bond that is central to one or more torsion angles identified as an outlier by Mogul will be highlighted in the graph. For rings, the root-mean-square deviation (RMSD) between the ring in question and similar rings identified by Mogul is calculated over all ring torsion angles. If the average RMSD is greater than 60 degrees and the minimal RMSD between the ring in question and sufficient the ring is also greater than 60 degrees, then that ring is considered an outlier. The outliers are highlighted in purple. The color gray indicates Mogul did not find sufficient equivalents in the CSD to analyse the geometry.





## 5.7 Other polymers (i)

There are no such residues in this entry.

# 5.8 Polymer linkage issues (i)

There are no chain breaks in this entry.



# 6 Fit of model and data (i)

# 6.1 Protein, DNA and RNA chains (i)

EDS was not executed - this section is therefore empty.

# 6.2 Non-standard residues in protein, DNA, RNA chains (i)

EDS was not executed - this section is therefore empty.

## 6.3 Carbohydrates (i)

EDS was not executed - this section is therefore empty.

## 6.4 Ligands (i)

EDS was not executed - this section is therefore empty.

### 6.5 Other polymers (i)

EDS was not executed - this section is therefore empty.

