DiffBond: A Method for Predicting Intermolecular Bond Formation

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Abstract—Many tools that explore models of protein complexes are also able to analyze interactions between specific residues and atoms. A comprehensive exploration of these interactions can often uncover aspects of protein-protein recognition that are not obvious using other protein analysis techniques. This paper describes DiffBond, a novel method for searching for intermolecular interactions between protein complexes while differentiating between three different types of interaction: hydrogen bonds, ionic bonds, and salt bridges. DiffBond incorporates textbook definitions of these three interactions while contending with uncertainties that are inherent in computational models of interacting proteins. We used it to examine the barnasebarstar, Rap1a-raf, and Smad2-Smad4 complexes, as well as a subset of protein complexes formed between three-finger toxins and nAChRs. Based on electrostatic interactions established by previous experimental studies, DiffBond was able to identify ionic and hydrogen bonds with high precision and recall, and identify salt bridges with high precision. In combination with other electrostatic analysis methods, DiffBond can be a useful tool in helping predict influential amino acids in protein-protein interactions and characterizing the type of interaction.

I. INTRODUCTION

Deducing the role of chemical bonds is a crucial part of understanding how protein-protein complexes achieve selective binding. In structural biology, this effort occurs frequently after the structure of a protein-protein complex is determined. First, bonds are identified with the application of the appropriate chemical and geometric constraints. Next, hypotheses are developed about what role certain bonds play in stabilizing particular parts of the complex. Finally, mutational experiments that remove or alter specific bonds can begin to test these hypotheses by establishing the resulting change in binding affinity. Once the effect of those mutations are evaluated, new mutational experiments can be devised, until the role of bonding in the apparatus of recognition is explained.

Unfortunately, a high resolution structure of most protein complexes is unavailable, so the closest alternative is to develop hypotheses from computational models of the interacting proteins. This approach must contend with additional uncertainties: First, the presence of intermolecular bonds will be constrained by the limits of bond geometry, which have been carefully measured in the chemical literature [1]–[3]. Second, the parts of a bond that form in vivo might be separated distantly enough in the model that their potential for assembly might not be discovered. Finally, intermolecular bonds may exhibit trends in length and angle that are atypical of the general bonds surveyed in the literature. There are few tools that perform an in-depth search of intermolecular bonds while attributing a biochemical reason to the interaction. To perform such a search while contending with existing constraints, this paper aims to assess the predictability of intermolecular bonds based on standard descriptions in the chemical literature.

Our approach is to treat the textbook chemical measurements of salt bridges, ionic interactions, and hydrogen bonds as a predictor for the presence of intermolecular bonds, and verify these predictors against experimentally established findings. In the case of hydrogen bonds, standard bond angles and bond lengths are extremely well defined [1], but in the case of ionic bonds, it is far less so. Coulomb's law defines attraction or repulsion between charged atoms at any distance, but the degree of attraction or repulsion is modulated by the presence and geometry of the dielectric between them. High dielectric aqueous environments attenuate the electric field, whereas low dielectric environments within a protein enhance it [4]. Thus, the concept of ionic bond lengths must always be an approximation based on assumptions of a biological environment, as several groups have done [2], [5]. Salt bridges, being the co-occurrence of both a hydrogen bond and an ionic bond, must also exist in the presence of these assumptions [5], [6]. Following these conventional definitions of bond geometry, we created DiffBond, a basic classifier for predicting the presence of salt bridges, ionic bonds, and hydrogen bonds.

Many methods deduce influential amino acids from computational models and predict the effects of mutation. Some provide information about the stability of a mutation by analyzing heuristic energy changes, rigidity-based mutation analysis, or molecular dynamic simulations [7]–[9]. Other methods compute and predict interactions from the computational model and infer mutation stability based on these interactions, such as hydrogen bond location prediction [1]. Like hydrogen bond location prediction, this method predicts existence and location of interactions between complexes. However, DiffBond is the first to predict the formation and location of intermolecular salt bridges and ionic bonds, and analyze these results in conjunction with hydrogen bond predictions. This bottom-up approach not only provides enough specificity to classify each interaction, but also is flexible enough to merge with other methods to improve prediction of influential amino acids.

We evaluated the classifier on a small dataset of protein complexes with well documented chemical bonds, measuring how frequently the classifier agreed with the authors' findings

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on each complex. While such a dataset cannot be representative of all intermolecular bonds within the space of protein complexes, it can determine whether intermolecular bonds infringe conventional norms.

II. METHODS

DiffBond identifies a list of all bonds that have the geometric and electrostatic capacity to make intermolecular bonds and affect binding affinity between two proteins that form a protein complex. First, we identify pairs of residues that are able to form three types of intermolecular interactions: salt bridge, ionic bond, and hydrogen bond. Identified residue pairs must satisfy geometric and electrostatic criteria for forming any of these three interactions based on textbook and literature measurements. We then form lists of bond predictions for each type of interaction as output. Finally, the resulting lists can be interpreted to help identify significant residues or aid in experimental design of mutations. This paper explores our software which encompasses two methods for identifying protein mutations that affect binding affinity, and discusses prospects for applying them in conjunction to mutation testing.

A. Scanning for Bond Formation

We outline a method to scan for intermolecular bonds between two proteins that form a protein complex, especially at the interface. We first generate a fully connected graph between all atoms from one protein to all atoms from the other protein; to identify specific and significant connections within the graph, we filter the connected graph using biochemical criteria like distance, residue charge, and contacting atoms.

A list of possible hydrogen bonds were compiled using HBPlus [1], which takes two hydrogen coordinates and looks at several criteria to decide if a hydrogen bond is possible and likely; criteria include minimum bond angles between atoms at 90°, maximum distance depending on the type of bond with 3.9Å for donor-acceptor pairs and 2.5Å for hydrogen-acceptor pairs, and minimum covalent separation of 3 covalent bonds. In addition, we also searched for amino-aromatic hydrogen bonds. These criteria values have been used in previous hydrogen bond interaction studies [1], [10], [11].

A list of ionic bonds were compiled by searching within a distance constraint for oppositely charged amino acids, namely interactions between arginine, histidine, and lysine with aspartate or glutamate [12]. In this study, we define an ionic bond as residues whose charged atoms, namely a positive N (nitrogen) in basic residues or negative O (oxygen) in acidic residues, are less than a cutoff distance [3]. Our software allows variable distance as an input parameter, but for the purposes of this paper, we use 5 angstroms. The 5Å cutoff is strict enough to yield only amino acids that are biochemically likely to form a bond, while the N-O atom pairs make sure the residue side chains are oriented towards each other. Ionic bonds, rarely, can form over long distances between 5-10Å in length [3] and so we also provide ionic bond predictions at 7.5Å and 10Å in Supplemental Text S1.

We also compile a list of salt bridges in the same way that we compute ionic bonds. Barlow and Thornton define salt bridges as a pair of oppositely charged residues whose side chain N-O atoms are within a cutoff distance of 4Å [2]. Fig 1 shows an example of oppositely charged side chains, glutamate and arginine of a Barnase-Barstar protein complex, within 4Å distance of each other. A cutoff at 4Å is a well defined distance that only considers "good" salt bridge geometries [13]. This 4Å measurement of salt bridge length aligns well with the textbook definition where salt bridges are a co-occurrence of both a hydrogen bond and an ionic bond [5], [6]. At less than 4Å, oppositely charged atoms are likely to interact; this also creates an environmental condition where water molecules cannot fit between the interacting residues. This implies the formation of a hydrogen bond which agrees with textbook measurements [5], [14].

Our bond scanning method consists of searching for neighboring amino acids that satisfy specific electrostatic and distance criteria; this design allows us to not only search within dimers, but also scan among higher oligomers consisting of many subunits.



Fig. 1. Sidechain visualization of Arg59 on Barnase (green) and Glu76 on Barstar (teal). Arg59 and Glu76 are within 4Å and are oppositely charged amino acids, so they are predicted to form a salt bridge by DiffBond.

B. Computing Electrostatic Isopotential Surfaces

An electrostatic isopotential surrounding a protein, at some potential p, is a subset of the electrostatic isopotential field that has potential equal to p. The electrostatic isopotential is a surface that outlines an area within an electrostatic potential at some threshold electrostatic potential k (kT/e). This isopotential threshold creates a surface where one side of the surface has isopotentials less than k and the other side has isopotentials greater than k. When the surface does not have infinite dimensions, it can be said to describe a geometric solid with measurable volume.

To generate an electrostatic isopotential surface, we first solve the overall potential field of a protein using DelPhi,



Fig. 2. Intersection using CSG a) Two proteins with oppositely charged electrostatic fields. b) When the proteins are in complex, the oppositely charged fields overlap forming an intersection region shown in orange. c) The intersection region represents the degree to which the field of one protein complements the field of the other.

an application that takes a 3-D coordinate molecule as input, computes solutions to the Poisson-Boltzmann equation for the input molecule, and outputs the potential field of the whole system [15], [16]. An algorithm called Marching Cubes then takes the potential field and some isopotential threshold k and generates an approximation of the isopotential surface representing the protein at k [17].

VASP-E is a tool that implements Marching Cubes and is able to manipulate, and calculate the volume of electrostatic isopotentials [18]. As implemented in VASP-E, marching cubes takes the potential field from DelPhi's output and first aligns it to a lattice grid. We then approximate the isopotential by determining which grids the isopotential intersects within the lattice. We use the intersections to further improve the isopotential approximation [17]. This technique produces a high-resolution approximation of the electrostatic potential surface of a protein [18].

1) Interface Field Comparison: VASP-E extends the method for manipulating electrostatic surfaces to perform a comparison of interface fields; VASP-E allows us to manipulate isopotential surfaces using constructive solid geometry (CSG) [19]. Based on VASP-E implementation, CSG can calculate the union, intersection, and difference of volumetric objects [18]. In this method, we use a series of CSG operations to perform a comparison of the region of electrostatic interaction between two proteins, called the interface field. We first generate isopotential surfaces for two proteins that form a protein complex. For one protein, the isopotential surface is generated at +k kT/e. On the other protein, a surface is generated at -k. The intersection between the +k and -k surfaces is the interface region where the positively charged region of one protein overlaps with the negatively charged region of the other protein as seen in Fig. 2. In other words, the intersection of +k and -k represent the degree to which the field of one protein complements the field of the other; the greater the volume of this intersection region, the more electrostatically complementary the proteins are.

2) *Nullification:* DelPhi is able to solve the potential field of a molecule while ignoring the charge contribution by an amino acid in a process called nullification [20]. Nullification of amino acids will affect the overall electrostatic surface of a protein. For example, if a large positive charge is normally observed in a potential surface, a lack of this positive charge by nullification will result in volume decrease of the potential. Important to note, the nullification of an amino acid outside of the protein-protein interface region will usually result in no surface volume change since volume is only measured around the interface region.

In interface field comparison, nullification allows us to generate volume differences for each amino acid. We use the interface field comparison method to generate differences in volume between an un-nullified interface field and a nullified interface field. An example of the effect of nullification on volume of an electrostatic surface can be seen in Fig. 3. Nullification at residue 59 removed a large electrostatic region at the interface between barnase and barstar.

C. Interpreting Data

We introduced DiffBond, a method that outputs lists of bonds for three interactions: salt bridges, ionic bonds, and hydrogen bonds. We also discuss a method for identifying residue mutations that are likely to change the electrostatic complementarity between proteins. Nullifying a residue and comparing the interface field outputs an intersection region representing the complementarity of two proteins. We discuss how we can interpret these outputs to identify residues that are significant to electrostatic interactions and can be strong candidates for mutation testing.

1) Nullification Graphs: Using a similar method design to VASP-E, we apply nullification to each amino acid in a complex and perform wildtype-mutant comparison on each to generate a volume difference for each amino acid nullification [18]. We define two conservative prediction thresholds to identify electrostatically influential amino acid interactions. We first find the two amino acids, i and j, that maximize or minimize volume difference. For maximum volume difference Ω at i, we assign an upper prediction threshold of $P = \Omega/2$; likewise, for minimum volume difference ω at j, we assign a lower prediction threshold of $p = \omega/2$. If the nullification of an amino acid x increases the volume difference above P, then x is predicted to reduce complimentarity of the complex and reduce affinity; a decrease in volume difference by amino acid y below p aligns with a prediction that y improves



Fig. 3. Effect of Nullification on Barnase-barstar a) Wildtype barnase electrostatic surface at isopotential of +1 kT/e. b) Barnase nullified at residue 59, electrostatic surface at isopotential of +1 kT/e. c) Overlap of wildtype (transparent yellow) and nullified barnase (green) surfaces. a,b,c) The red square encompasses the main difference in isopotential surface due to nullification. d) Wildtype barnase (blue) in complex with barstar (transparent yellow).

complementarity and therefore increases affinity. If more than 10% of amino acids exceed the prediction threshold P, it is likely the case that no amino acids contributed a significant decrease to electrostatic complementarity, or that the data contains a large amount of noise. In either case, we ignore the prediction threshold and predict that there are no amino acids that reduce complementarity. Similarly, if 10% of amino acids are less than p, then we predict that no amino acids increase electrostatic complementarity.

2) Nullification and DiffBond Mutation Prediction: Electrostatic complementarity predictions in nullification graphs can be analyzed in conjunction with bond formation data by cross-referencing influential amino acids with bonds formed. Based on nullification, we can predict whether mutating an amino acid will increase or decrease electrostatic complementarity. Similarly, knowing what intermolecular bonds can be formed by an amino acid can help predict changes in complementarity when mutated. For example, mutating one end of an ionic bond to an uncharged or like-charged residue would cause the pair to lose the attraction. Although predictions from nullification and intermolecular bond prediction have not yet been shown to be related, both methods predict behaviors in electrostatic complementarity. Predictions from two separate sources can help point to significant mutation candidates.

D. Data Set Construction

DiffBond was designed to identify electrostatic influences and bonds and, with VASP-E, form predictions of mutations that would either increase or decrease affinity for proteinprotein interaction. Because of this design, we validate Diff-Bond using several families of protein for which specific bond formations that are highly involved in protein electrostatics are well documented. The three-finger toxin family (pdb: 1yi5, 4hqp, 2qc1, 1kc4), barnase-barstar complex (pdb: 1brs), rap1A-RAF complex (pdb: 1c1y), and smad2-smad4 complex (pdb: 1u7v) were selected for validating DiffBond because they have all been extensively studied for specific electrostatically influential amino acids and bonds that affect binding affinity to their corresponding binding partners. 1) Barnase-Barstar: Barnase is an extracellular RNase of Bacillus amyloliquefaciens that is often co-expressed with its inhibitor barstar; without the concurrent expression of barnase in complex with barstar, barnase can be lethal to the cell [21]. Barstar inhibits barnase by forming a tight complex with many intermolecular steric and electrostatic interactions at binding site residues [22]–[24]. As a result, mutating residues involved in these intermolecular interactions often results in enhanced or diminished electrostatic complementarity between barnase and barstar.

2) *Rap1a-Raf:* Ras is a family of GTPase that transmits signals via protein-protein interaction to regulate many biological systems, like cell cycle progression, cell division, apoptosis, lipid metabolism, DNA synthesis, and cytoskeletal organization [25]. While little is known about ras structure in complex with its effector ligands, rap1a has a similar structure to ras; it has an almost identical binding interface, and binds competitively to ras effectors like raf, an oncogene involved in ERK 1/2 signaling [26], [27]. Like ras, the binding interface of rap1a-raf consists of a few crucial intermolecular bond interactions whose mutations alter binding affinity [26], [28], [29].

3) Smad2-Smad4: Smads is a family of structurally similar proteins that act as main signal transducers for TGF-B receptors, a super family of proteins that help regulate cell development and growth [30], [31]. R-Smad proteins, like Smad2, direct the TGF-B signaling while Smad4 help mediate the formation of the heterometic complex between R-Smads and Smad4 [30]. This dataset uses the trimer consisting of one Smad4 and two Smad2 subunits whose binding interface are well studied for electrostatic interactions and mutation effect [32].

4) Three-finger Toxin Family: Three-finger toxins are a protein superfamily consisting of many small and structurally similar toxin proteins from elapid snake venom [33], [34]. Their distinct structure consist of three beta strand loops emanating from a cysteine rich core, which facilitates interaction with many receptor or channel proteins; neurotoxin members of the family, such as α -bungarotoxin [35]–[37] and

Total	Ionic Bond	Hydrogen Bond	Salt Bridge
True Positive	14	35	6
False Positive	2	5	1
False Negative	3	12	6
True Negative	Unknown	Unknown	Unknown
Precision	87.5%	87.5%	85.7%
Recall	82.4%	74.5%	50.0%

TABLE I

PRECISION AND RECALL OF THE BOND LIST FOR PREDICTING THE FORMATION OF BONDS.

 α -cobratoxin [38] interact with neuronal and muscle nicotinic acetylcholine receptors (nAChRs) while other members can interact with muscarinic acetylcholine receptors (mAChRs) or different neuronal nAChR subtypes [34]. The interaction between many members of this toxin family and nAChRs are well studied with known interactions across the interface.

The protein complexes formed by barnase-barstar, rap1a-raf, and smad2-smad4 are commonly used protein complexes for studying electrostatic interaction. They are comprehensively tested and reviewed in literature on both sides of the interface, and provide clear descriptions of bond formation between residues and altered binding affinity due to mutation. Although some three-finger toxin members are not as well studied as the three protein complexes mentioned prior, α -bungarotoxin and α -cobratoxin are both studied extensively in mutation testing.

III. RESULTS

DiffBond is the first method for identifying ionic bonds and salt bridges, and it uses HBPlus, one of the only methods for identifying hydrogen bonds. Consequently, DiffBond cannot be compared to HBPlus and there are no other existing methods it can be compared against for identifying salt bridges and ionic bonds. Instead, we compare predictions from DiffBond to experimental data.

A. Bond Prediction Validation

We validate the DiffBond bond prediction method by comparing bond predictions with known interactions published in experimental findings. Intermolecular bond formations were gathered from published journal papers rather than from a database because we not only want to confirm the existence of a bond formed between a pair of amino acids, but we also want to attribute a reason for bond formation that came from experts in biology through mutation testing or in-depth crystallographic analysis. By collecting the predictions as a set of ionic bonds, hydrogen bonds, and salt bridges, we can measure the prediction performance of predicted bonds on each type of bond separately.

We start by counting true positives (TPs), false positives (FPs), true negatives (TNs), and false negatives (FNs). A bond prediction is defined as a TP if our findings predict that a bond forms between an amino acid pair and the literature agrees for the same specific amino acid pair. Similarly, hydrogen bonds are TPs only if experimental findings conclude a hydrogen bond exists between the pair. Finally, salt bridges are TPs only if studies either state the bond contains both a hydrogen bond and electrostatic interaction or explicitly states a salt bridge exists. FPs are bond predictions that were found but are not considered to form the predicted bond by experimental findings. TNs are bond predictions that we predicted would not occur and that experimental findings agree would not occur. FNs are bonds that DiffBond did not predict would occur, but experimental findings found those bonds to form.

We cannot fully count TNs because no studies specifically discuss and analyze the electrostatic influence of every amino acid in a protein. However, we can evaluate the prediction accuracy of DiffBond without TNs; we compute precision and recall to verify accuracy. Precision is the fraction of correctly predicted bonds among all bonds verified in experimental findings, and recall (sensitivity) is the fraction of correctly predicted bonds among all true interactions.

When searching literature for intermolecular interactions, if a bond was generated by our bond list but was not mentioned in literature, we considered this prediction to be a FP. This strict criteria verifies that the precision we report is a lower limit, with the possibility that any unmentioned bond predictions may be validated in the future.

The precision and recall for individual protein complexes and the cumulative accuracy statistics are reported in Table I. Although the total number of ionic predictions were low at 16, ionic bond prediction exhibited high precision and recall at 87.5% and 82.4% respectively. Hydrogen bond predictions showed both strong precision and recall at 87.5% and 74.5% over a large set of predictions at n=40. Salt bridge prediction had a high precision but a low recall at 85.7% and 50% respectively, over a small sample size of n=7.

Of the 3 false negatives from ionic bond prediction, all 3 pairs were correctly predicted by expanding distance threshold to 7.5Å instead of 5Å. Similarly, of the 6 false negatives for salt bridge prediction, 3 of the 6 were correctly predicted by a distance threshold of 5Å instead of our chosen threshold of 4Å. Although hydrogen bond prediction had 12 false negatives, HBPlus uses many more constraints, including an additional minimum distance cutoff that may create false negatives. However, we did not measure a larger threshold range for hydrogen bonds to determine whether distance played a large role in lowering recall.

B. Nullification Graph Predictions

Using VASP-E, we performed a nullification over each amino acid of protein complexes and calculated the volume difference between wildtype and nullified surfaces. The volume difference for each nullification in the barnase-barstar complex can be seen in Fig. 4, represented by the colored lines.



Fig. 4. Volume difference between wildtype and mutant barnase-barstar complex when nullifying barnase amino acids at k = +/-1, +/-3, +/-5, and +/-7. Significant residue nullifications are those that surpass either the upper or lower threshold.

We present all predictions from nullification in Supplemental Table S2 alongside predictions from the bond list.

IV. DISCUSSION

We have presented DiffBond, a method for identifying significant bonds in protein-protein interactions and predicting influential amino acids for mutation testing. There are few tools that perform an in-depth search of intermolecular bonds while attributing a biochemical reason to the interaction. To our knowledge, none have paired this search with a volumetric electrostatic analysis to further inform possible mutation testing. In experimental settings, the design of mutational studies can be difficult because there are many amino acids to consider, and picking a misguided mutation candidate can be time consuming, expensive and unproductive. DiffBond was designed to guide mutation testing by gathering additional structural and electrostatic information that may not be apparent to support experimental designs.

Scanning for salt bridges, ionic bonds, and hydrogen bonds is a novel approach introduced in this paper and has demonstrated promising capability in identifying intermolecular bonds in protein complexes with high precision and recall. Bond prediction was able to predict 14 ionic bonds and their paired partners out of 17 bonds known in literature. Similarly, salt bridge prediction maintained a high precision. Recall for salt bridge prediction was slightly lower which was expected due to the very strict distance cutoff when defining a salt bridge. Hydrogen bond prediction also demonstrated high precision and recall. When using textbook defined measurements for each type of interaction, we found that these definitions are strong predictors for each bond type.

One criteria in collecting data that reduced precision was that any bond predictions that had no mention in literature were considered false positives. This supports that our precision statistic is a lower limit and that future studies may verify bonds that we considered false predictions. To our knowledge, no experimental results have established that predictions we considered false positive do not occur, and so we present it as an open prediction.

Furthermore, we expect DiffBond parameters like distance threshold or bond list interpretation to vary on a case-bycase basis. The assumption for a 4Å distance threshold for salt bridges, or a 5Å distance threshold for ionic bonds may not always hold true; low resolution structures can introduce a margin of error for amino acid spatial placement. Even with high resolution structures, we cannot always account for side chain flexibility especially when residues are not sequestered. Some studies may be interested in gathering more data at the cost of precision. For example, a study may be interested in finding more salt bridges, but the conventional salt bridge definition yields few residues due to its strict distance criteria for a good salt bridge geometry. Expanding the distance threshold means fewer true salt bridges are missed, but more false positives are identified. Incorporation of dynamic information might also be helpful in improving predictions of bond locations generally, and will be considered in future work.

Similar to scanning for intermolecular bonds, VASP-E and the use of nullification presents another method for identifying mutation candidates and mutation prediction. In conjunction, nullification and intermolecular bonds offer two perspectives on electrostatic interactions that can compliment each other without necessarily being related. If both methods point to a residue for mutation prediction, we can consider two hypotheses: First, mutating this residue may affect binding affinity when mutated, either increasing electrostatic complementarity if the peak points upward or decreasing electrostatic complementarity if the peak points downward. Second, mutating this residue to one that no longer forms the predicted bond may break the bond. For example, mutating from a charged acidic residue like lysine to an uncharged residue like glycine likely means any predicted ionic interactions before are no longer possible.

Outside of selecting for high quality mutation candidates,

the additional information provided by these methods offers high value to researchers in experimental design. Pointing out possible interactions and predicting electrostatic complementarity can inform how researchers design a mutation; for example, they can test each of the hypotheses above with mutations to different residues.

DiffBond has the potential to be extended to different applications. Although we have not assumed any correlation between mutation predictions from nullification and from intermolecular bond list, we can assimilate other predictor techniques to begin an artificial reasoning process. Nullification as a first step implies change in electrostatic complementarity by mutation, but does not provide a biochemical reason. The intermolecular bond list interprets three possible interactions when explaining nullification peaks. Adding more prediction techniques for different interactions can form a decision tree of reasoning for mutation prediction.

ACKNOWLEDGEMENTS

This work was funded in part by NIH Grant R01GM123131 to Brian Y. Chen, Julie M. Miwa, and Talulla Palumbo. This work was also funded in part by NIH Grant DA043567 to Julie M. Miwa.

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SUPPLEMENTAL MATERIALS

TABLE S1: Bond predictions and validation by literature. [22]–[24], [26], [28], [29], [32], [35]–[38]

IYI5 Jonic Bond F ASP27 / B LYS34 N 7.5A F ARG33 / A GLU149 N F ARG33 / A ASP194 N 10A F ARG36 / A ASP194 N N F ARG37 / A ASP194 N 10A F ARG36 / A ASP144 N N F ARG37 / A ASP85 N F ARG36 / A ASP155 N F ARG37 / A ASP85 N F F ARG38 / A ASP85 N F F ARG38 / B GLU163 N F F LYS49 / B GLU163 N F F ARG68 / B GLU110 N F Hydrogen Bond F ARG86 / B ASP108 N F F ASP27 / T R185 Y Salt Bridge F ASP27 / T R185 Y F S Y F ASP27 / T R185 Y Salt Bridge F ASP27 / T R185 Y Y F ASP27 / T R185 Y Missed None Y A ARG36 / A ASP193 N Y 1 UYS38 / A GLU185 Y Y Y Y Y	Protein/PDB	Bond Type/Distance	Interacting Amino Acid Pair	Shown by Literature
SA F ASP27 / B LYS34 N 7.5A F ARG33 / A GLU199 N F ARG37 / A GLU19 N F ARG37 / A ASP194 N F ASP8 / A ARG148 N F ARG33 / A ASP150 N F ARG37 / A ASP160 N F LYS39 / B GLU163 N F ARG68 / B GLU110 N F ASP27 / A TYR185 Y F ASP27 / A TYR185 Y F ASP27 / TYR185 Y F ASP27 / TYR185 Y F ASP27 / TYR185 Y Missed None 7.5A I ASP30 / A ARG182 Y 7.5A I ASP30 / A ARG182 Y 7.5A I ARG26 / A ASP137 N 10A I ARG36 / A ASP137 N 11YS38 / A GLU185 Y N 10A I ARG36 / A ASP137 N 11YS38 / A GLU185 Y N 11YS38 /	1YI5	Ionic Bond		
4HQP F ARG33 / A GLU149 N 10A F ARG33 / A ASP194 N 10A F ASP8 / A ASP194 N F ASP8 / A ASP194 N N F ASP3 / A GLU190 N F F ARG3 / A GLU190 N F F KASB / A GLU163 N F F XS35 / A SEN80 N F F ARG68 / B ASP108 N F F ARG68 / B ASP108 N F F ASP27 / TYR185 Y F Salt Bridge None None Missed None N 7.5A I ARG36 / A ASP193 N 10A I ARG36 / A ASP193 N 11X7S38 / B GLU185 N N 10A I ARG36 / A ASP193 N 11X7S38 / B GLU185 N N 10A I ARG36 / A ASP17 N 10A I A		5A	F ASP27 / B LYS34	Ν
4HQP IoA F ARG33 / A ASP194 F ARG36 / A ASG148 F ASG8 / A ASG148 N F ASG8 / A ASG148 N F ARG33 / A GLU190 N F LYS33 / B GLU163 N F LYS33 / B GLU163 N F LYS33 / B GLU163 N F ARG68 / B ASP108 N F ARG68 / B ASP108 N F ASG28 / A TYR155 Salt Bridge N F ASG28 / A ASP108 N F ASG28 / A TYR155 F ASP27 / TYR185 F ASP27 / A GLU185 F ASP27 / A GLU185 F ASP27 / A GLU185 F ASP27 / A ASP193 N I LYS38 / A GLU185 F ASP27 / A ASP194 F ASP27 / A GLU185 F ASP27 / A ASP194 F ASP27 / A GLU185 F ASP27 / A ASP194 F ASP27 / A GLU185 F ASP27 / A GLU185 F ASP27 / A GLU185 F ASP27 / A ASP194 F ASP20 / A ASP194 F ASP27 / A ASP194 F ASP20 / A ASP194 F		7.5A	F ARG33 / A GLU149	Ν
IOA F ARG36 / A ASP194 N IOA F AR98 / A ARG148 N F ASP8 / A LYS180 N F ARG33 / A AGU190 N F ARG33 / A AGU190 N F LYS35 / B GLU163 N F LYS35 / B GLU163 N F LYS35 / B GLU163 N F LYS35 / B GLU100 N F LYS35 / A SER186 Y F ARG68 / B GLU110 N F ARG68 / B GLU110 N F ASP27 / A TYR185 Y F ASP27 / TYR185 Y ARG36 / A ASP193 N None None 4HQP Ionic Bond 5A I ASP30 / A ARG182 Y 7.5A I ARG36 / A ASP87 10A I ARG36 / A ASP87 10A I ARG36 / A GLU185 I LYS38 / A GLU185 Y I HS68 / A GLU185 Y I HYdrogen Bond I VAL40 / A PHE183 I LYS70 / A IVR184 Y I LYS70 /			F ARG33 / A ASP194	Ν
IOA F ASP8 / A ARG148 N F ARG33 / A LYS180 N F ARG33 / A LYS180 N F ARG33 / A GLU190 N F ARG33 / A GLU163 N F LYS35 / B GLU163 N F LYS35 / B GLU163 N F LYS49 / B GLU163 N F LYS35 / B GLU163 N F ARG68 / B GLU110 N F ARG68 / B GLU110 N F ARG68 / B GLU110 N F ASP27 / TYR185 Y Salt Bridge F ASP27 / TYR185 None None 4HQP Ionic Bond 5A I ASP30 / A ARG182 Y 1 ARG36 / A ASP193 N 10A I ASP30 / A ARG182 Y 1 LYS38 / A GLU185 N 10A I ARG36 / A ASP193 N 10A I ARG36 / A GLU185 N 1 ARG36 / A ASP191 N N 1 LYS38 / B GLU185 N N 1 ARG36 / A AGU181 N N 1 LYS38 / A GLU185			F ARG36 / A ASP194	Ν
4HQP Ionic Bond F ARG3 / A ASP85 F ARG33 / A GLU190 F LXS35 / B GLU163 N F ARG68 / B ASP108 F ARG68 / B GLU110 N F ARG68 / B GLU110 N F ARG68 / B GLU110 N F ARG68 / B GLU110 N F ARG7 / A TYR185 Salt Bridge N 4HQP Ionic Bond 5 5 A I ASP30 / A ARG182 I ARG36 / A ASP193 I LYS38 / A GLU185 None N 4HQP Ionic Bond 5 5 A I ASP30 / A ARG182 I ARG36 / A ASP193 I LYS38 / A GLU185 N N 10A I ARG36 / A ASP87 I ARG36 / A ASP87 N N N 10A I ARG36 / A ASP87 I LYS38 / B GLU185 N N 10A I ARG36 / A ASP87 I ARG36 / A ASP87 N N 10A I ARG36 / A ASP87 I ARG36 / A AGU185 N N 10A I ARG36 / A ASP87 I ARG36 / A AGU185 N N 10A I ARG36 / A ASP87 I ARG36 / A AGU185 N N 10A I ARG36 / TASP87 I ARG36 / A AGU185 N N 11YS38 / A GLU185 N N N 12YS38 / A GLU185 N N N 14RG36 / A AGU185 N Y N 14RG36 / A AGU185 N Y N 14RG36 / A AGU185 N Y N 14RG36 / TRP145 N Hydrogen Bond ARG36 / TRP		10A	F ASP8 / A ARG148	Ν
4IIQP Ionic Bond F ARG33 / A ASP85 N F LXS35 / B GLU163 N F LXS35 / B GLU163 N F LXS49 / B GLU163 N F AKG68 / B ASP108 N F ARG68 / B ASP108 N F ARG68 / B ASP108 N F ARG68 / B ASP108 N F ARG68 / B ASP108 N F ARG68 / B ASP108 N F ARG68 / B ASP108 N F ARG68 / B ASP107 / A TYR185 Y F ASP27 / TYR185 N Salt Bridge F ASP27 / A TYR185 N N Missed None N N 7.5A I ASP30 / A ARG182 Y N 1LYS38 / A GLU185 N N N 10A I ARG36 / A ASP87 N N 10A I ARG36 / A ASP87 N N 1 LYS38 / B ASP160 N N N 1 LYS38 / B ASP160 N N N 1 LYS38 / A GLU185 Y N N 1 LYS38 / A GLU185 Y N N			F ASP8 / A LYS180	Ν
4HQP F ARG33 / A GLU190 N F LYS35 / B GLU163 N F LYS49 / B GLU163 N F ARG68 / B ASP108 N F ARG68 / B GLU110 N F ARG68 / B GLU110 N F ARG68 / A SER186 Y F ASP27 / A TYR185 Y F ASP27 / TYR185 Y I ARG36 / A ASP193 N I LYS38 / A GLU185 N I LYS38 / B GLU185 N I ARG36 / A ASP187 N I ARG36 / A ASP187 N I LYS38 / B GLU185 N I LYS38 / B GLU185 N I LYS38 / A GLU185 Y I HIS68 / A GLU185 Y			F ARG33 / A ASP85	Ν
Hydrogen BondF LYS35 / B GLU163NF ARG68 / B ASP108NF ARG68 / B ASP108NF ARG68 / B GLU110NF ARG68 / B GLU110NF ARG68 / B GLU110NF ASP27 / A TYR185YF LYS35 / A SER186YF Salt BridgeF ASP27 / TYR185MissedNoneJHQPIonic Bond5AI ASP30 / A ARG182YI LYS38 / A GLU185YY7.5AI ARG36 / A ASP19310AI ARG36 / A ASP17NI LYS38 / A GLU185NI LYS38 / A GLU185YI ASP30 / A ARG182Hydrogen BondI VAL40 / A PHE183YI ASP30 / A ARG182YI ASP30 / A ARG36 / B ASP152NoneA ARG36 / B			F ARG33 / A GLU190	Ν
Hydrogen BondF LYS49 / B GLU163N F ARG68 / B ASP108N F ARG68 / B ASP108N F ARG68 / B GLU110Hydrogen BondF ASP27 / A TYR185Y F ASP27 / A TYR185Y F ASP27 / TYR185Salt Bridge MissedF ASP27 / J B LYS34N None4HQPIonic Bond- 5A- I ARG36 / A ASP193N I I LYS38 / A GLU1857.5AI ASP30 / A ARG182 I LYS38 / A GLU185Y F N I LYS38 / A GLU185N N10AI ARG36 / A ASP87N I LYS38 / B GLU185N N10AI ARG36 / A ASP87N I LYS38 / B GLU185N NHydrogen BondI LYS38 / B GLU185N I LYS38 / B GLU185N NHydrogen BondI VAL0 / A PHE183 I LYS30 / A ATYR184Y I LYS38 / A GLU185Y S A I LYS30 / A ARG182QC1Ionic BondI ASP30 / A ATYR184 I LYS30 / A ARG182 A RG36 / TYR91Y Hydrogen BondARG36 / TYR91Hydrogen BondHydrogen BondJYS38 / A GLU185Y Salt BridgeI ASP30 / A ARG182 A RG36 / TYR91MissedA ARG36 / B ASP152 A ARG36 / TRP145 LYS38 / A GLU185N S alt Bridge2QC1Ionic Bond 5A 7.5ANoneIOAA ARG36 / B ASP152 A ARG36 / B ASP200 A ARG36 / B ASP200 A ARG36 / B ASP200 N A ARG36 / B ASP200 NN N A ARG36 / B ASP200 N A AG36 / B ASP200 N			F LYS35 / B GLU163	Ν
Hydrogen BondF ARG68 / B ASP108NHydrogen BondF ASP27 / A TYR185YF LYS35 / A SER186YF LYS35 / A SER186YF ASP27 / TYR185YF ASP27 / B LYS34NMissedNoneJames ConstructionJames Const			F LYS49 / B GLU163	Ν
Hydrogen BondF ARG68 / B GLU110NF ASP27 / A TYR185YF LYS35 / A SER186YF ASP27 / TYR185YSalt BridgeF ASP27 / B LYS34NMissedNoneJan BondJan Bond5AI ARG36 / A ASP193NTLYS38 / A GLU185Y7.5AI ARG36 / A ASP87N10AI ARG36 / A ASP87N10AI ARG36 / A AGU185Y11YS38 / B GLU185NN10AI ARG36 / A AGU185Y11YS38 / B ASP160NI11YS38 / B ASP160N11YS38 / B ASP160N11YS38 / B GLU185Y11YS38 / B GLU185Y11YS38 / A GLU129N11YS38 / A GLU125<			F ARG68 / B ASP108	Ν
Hydrogen BondF ASP27 / A TYR185YF LYS35 / A SER186YF ASP27 / TYR185NSalt BridgeF ASP27 / B LYS34NMissedNone4HQPIonic Bond5AI ASP30 / A ARG182YI ARG36 / A ASP193NI LYS38 / A GLU185Y7.5AI ARG36 / A ASP8710AI ARG36 / A ASP8710AI ARG36 / A ASP8710AI ARG36 / A GLU185YI LYS38 / B GLU18510AI ARG36 / A GLU185YI LYS38 / B ASP160NI GLU41 / A ARG182NI LYS70 / A GLU185YI LYS38 / A GLU185YI LYS38 / A GLU185YI LYS38 / A GLU185YI LYS70 / A GLU185YI LYS38 / A GLU185YI LYS38 / A GLU185YI LYS38 / A GLU185YI LYS38 / A AGU185YI LYS38 / A AGU185YI ASP30 / A XRG182YMissedAG36 / TRP145LYS38/GLU185Salt BridgeZQC1Ionic Bond5ANone7.5AA ARG36 / B ASP152NA LYS52 / B GLU129NA ARG36 / B ASP152NA ARG36 / B ASP89NA ARG36 / B ASP89N <t< th=""><th></th><th></th><th>F ARG68 / B GLU110</th><th>Ν</th></t<>			F ARG68 / B GLU110	Ν
4HQP Ionic Bond F ASP27 / TYR185 Y 5A I ASP30 / A ARG182 Y 1 ARG36 / A ASP193 N 1 LYS38 / A GLU185 Y 7.5A I ARG25 / A GLU185 N 1 LYS38 / A GLU185 N N 10A I ARG36 / A ASP17 N 1 LYS38 / A GLU185 N N 10A I ARG36 / A ASP87 N 10A I ARG36 / A ASP87 N 1 GLU41 / A ARG182 N N 1 HIS68 / A GLU185 N I 1 UYS38 / B ASP160 N I 1 LYS38 / B AGLU185 Y I 1 HIS68 / A GLU185 Y I 1 HIS68 / A GLU185 Y I 1 LYS70 / A GLU185 Y I Hydrogen Bond I VAL40 / A PHE183 Y I LYS38 / A GLU185 Y I Missed ARG36 / TRP145 Hydrogen Bond ARG36 / TRP145 Hydrogen Bond ARG36 / TRP145 LYS38/GLU185		Hydrogen Bond	F ASP27 / A TYR185	Y
4HQP Ionic Bond F ASP27 / TYR185 N 5A F ASP27 / B LYS34 N 5A I ASP30 / A ARG182 Y 1 ARG36 / A ASP193 N I 1 LYS38 / A GLU185 Y Y 7.5A I ARG36 / A ASP193 N 1 LYS38 / A GLU185 Y Y 7.5A I ARG36 / A ASP87 N 10A I ARG36 / A ASP87 N 1 LYS38 / B GLU185 N I 10A I ARG36 / A ASP87 N 1 LYS38 / B GLU185 N I 1 HIS68 / A GLU185 Y I 1 HIS68 / A GLU185 Y I 1 HIS68 / A GLU185 Y I Hubrogen Bond I VAL40 / A PHE183 Y I LYS38 / A GLU185 Y Y Salt Bridge I ASP30 / A ATYR184 Y I LYS38 / A GLU185 Y Y Salt Bridge I ASP30 / A ARG182 Y Missed A ARG36 / TYR91 Hydrogen Bond <tr< th=""><th></th><th></th><th>F LYS35 / A SER186</th><th>Y</th></tr<>			F LYS35 / A SER186	Y
Salt Bridge MissedF ASP27 / B LYS34 NoneN4HQPIonic BondI ARG36 / A ASP193 I LYS38 / A GLU185 T,5AI ASP30 / A ARG182 I ARG36 / A ASP193 NY7.5AI ASC3 / A CLU185 I LYS38 / A GLU185 I LYS38 / B GLU185 I GLU185N10AI ARG36 / A ASP87 I ARG36 / A ASP87 I LYS38 / B GLU185 I GLU1751 I LYS38 / B GLU185 I GLU185 I GLU14 / A ARG182 I HIS68 / A GLU185 I Salt Bridge400I VAL40 / A PHE183 I ASP30 / A ARG182 I SS30 / A TYR184 I LYS38 / A GLU185 I Salt Bridge401I Onic Bond SA T,5A5ANone A ARG36 / TRP145 LYS38/GLU1855ANone T,5A7.5AA ARG36 / B ASP152 A ARG36 / B ASP152 N A ARG36 / B ASP200 N A ARG36 / B ASP20			F ASP27 / TYR185	
HissedNone4HQPIonic Bond-5AI ASP30 / A ARG182YI ARG36 / A ASP133NI LYS38 / A GLU185Y7.5AI ARG25 / A GLU185N10AI ARG36 / A ASP87N10AI ARG36 / A ASP87NI LYS38 / B GLU151NI LYS38 / B ASP160NI LYS38 / B AGL0185Y10AI ARG36 / A GLU151NI LYS38 / B GLU18510AI GLU41 / A ARG182NNI GLU41 / A ARG182NI LYS70 / A GLU185YHydrogen BondI VAL40 / A PHE183YI LYS38 / A GLU185YSalt BridgeI ASP30 / A ARG182YMissedARG36 / TXP91Hydrogen BondLYS38/GLU185Salt BridgeARG36 / TRP145Hydrogen BondLYS38/GLU185Salt BridgeI ASP30I ASP30 / A ARG182YMissedA ARG36 / B ASP152NA ARG36 / B ASP152NA ASP30 / B LYS145NOAA ASP30 / B LYS145NA ARG36 / B ASP200NA ARG36 / B ASP200<		Salt Bridge	F ASP27 / B LYS34	Ν
4HQP Ionic Bond I ASP30 / A ARG182 Y 5A I ARG36 / A ASP193 N I LYS38 / A GLU185 Y 7.5A I ARG36 / A ASP87 N 10A I ARG36 / A ASP87 N I LYS38 / B GLU185 N N I LYS38 / B ASP160 N N I LYS38 / B ASP160 N N I HIS68 / A GLU185 Y I HIS68 / A GLU185 Y I HIS68 / A GLU185 Y I HIS68 / A GLU185 Y I HIS68 / A GLU185 Y I HIS68 / A GLU185 Y I LYS70 / A GLU185 Y I LYS70 / A GLU185 Y Hydrogen Bond I VAL40 / A PHE183 Y I LYS70 / A AG182 Y Missed AG36 / TYR91 Hydrogen Bond AG36 / TRP145 Hydrogen Bond LYS38/GLU185 Salt Bridge I XSS2 / B GLU129 N A LYSS2 / B GLU129		Missed	None	
4HQPIonic Bond 5AI ASP30 / A ARG182 I ARG36 / A ASP193 N I LYS38 / A GLU185 N I LYS38 / A GLU185 N I LYS38 / A GLU185 N I ARG36 / A ASP87 N I ARG36 / A ASP87 N I ARG36 / A GLU151 N I LYS38 / B GLU151 N I LYS38 / B ASP160 N I GLU41 / A ARG182 N I HIS68 / A GLU185 Y I LYS70 / A GLU185 Y I LYS70 / A GLU185 Y I LYS38 / A GLU129 N A ARG36 / B ASP152 N A ARG36 / B ASP152 N A ARG36 / B ASP200 N A ARG36 / B ASP200 				
4HQP Ionic Bond Y 5A I ASP30 / A ARG182 Y I ARG36 / A ASP193 N I LYS38 / A GLU185 Y 7.5A I ARG36 / A ASP87 N 10A I ARG36 / A ASP87 N 10A I ARG36 / A ASP87 N I LYS38 / B GLU185 N N I UQA I ARG36 / A ASP87 N I ARG36 / A ASP87 N I ARG36 / A GLU185 I0A I ARG36 / A ASP87 N I LYS38 / B GLU185 N I LYS38 / B GLU185 I GLU41 / A ARG182 N I HIS68 / A GLU185 Y I HIS68 / A GLU185 Y I HYS00 / A TYR184 Y I LYS30 / A TYR184 Y I ASP30 / A ARG182 Y Missed ARG36 / TXP91 Hydrogen Bond ARG36 / TXP145 Hydrogen Bond ARG36 / TXP145 LYS38/GLU185 Salt Bridge I ZQC1 Ionic Bond I XLYS52 / B GLU129 IOA ARG36 / TXP145 Hydrogen B				
SA I ASP30 / A ARG182 Y I ARG36 / A ASP193 N I LYS38 / A GLU185 Y 7.5A I ARG25 / A GLU185 N 10A I ARG36 / A ASP87 N I LYS38 / B GLU185 N I I0A I ARG36 / A ASP87 N I GLU41 / A ASP87 N I I GLU41 / A ARG182 N N I GLU41 / A ARG182 N I Hydrogen Bond I VAL40 / A PHE183 Y I ASP30 / A TYR184 Y I LYS38 / A GLU185 Y Y Hydrogen Bond I VAL40 / A PHE183 Y I ASP30 / A TYR184 Y Y I LYS38 / A GLU185 Y Y Missed ARG36 / TYR91 Hydrogen Bond ARG36 / TRP145 Hydrogen Bond ARG36 / TRP145 LYS38/GLU185 Salt Bridge Salt Bridge 2QC1 Ionic Bond - 5A None - 7.5A A ARG36 / B ASP152 N A ARG36 / B ASP30 N A ARG36 / B ASP89	4HQP	Ionic Bond		
2QC1 I ARG36 / A ASP193 N I ARG36 / A ASP193 N I ARG25 / A GLU185 N I ARG36 / A ASP87 N I0A I ARG36 / A ASP87 I0A I ARG36 / A ASP87 I ARG36 / A GLU185 N I0A I ARG36 / A ASP87 N I ARG36 / A GLU185 N I LYS38 / B ASP160 N I LYS38 / A GLU185 Y I HIS68 / A GLU185 Y I HIS68 / A GLU185 Y I ASP30 / A ARG182 Y I ASP30 / A ARG182 Y Missed ARG36 / TRP145 Hydrogen Bond ARG36 / TRP145 Hydrogen Bond LYS38/GLU185 Salt Bridge ZQC1 Ionic Bond 5A None 7.5A A ARG36 / B ASP152 N A ARG36 / B ASP152 <		5A	I ASP30 / A ARG182	Y
2QC1 Ionic Bond I APG25 / A GLU185 N 10A I ARG36 / A ASP87 N 10A I ARG36 / A ASP87 N 10A I ARG36 / A GLU151 N 10A I ARG36 / A ASP87 N 1 ARG36 / A ASP87 N N 1 LYS38 / B ASP160 N N I LYS38 / B ASP160 N N I LYS38 / A GLU185 Y Y I HIS68 / A GLU185 Y Y I HY70 / A GLU185 Y Y I VAL40 / A PHE183 Y Y I ASP30 / A TYR184 Y Y I LYS38 / A GLU185 Y Y Missed ARG36 / TYR91 Hydrogen Bond ARG36 / TRP145 Hydrogen Bond LYS38/GLU185 I LYS32 / B GLU125 N A LYS52 / B GLU129 N 10A A ASP30 / B LYS145 N A ARG36 / B ASP89			I ARG36 / A ASP193	Ν
2QC1 Ionic Bond I ARG36 / A GLU185 N IOA I ARG36 / A ASP87 N IOA I ARG36 / A ASP87 N I ARG36 / A GLU151 N N I LYS38 / B ASP160 N I GLU41 / A ARG182 N I GLU41 / A ARG182 N I HIS68 / A GLU185 Y I HIS68 / A GLU185 Y I HIS68 / A GLU185 Y I VYS70 / A GLU185 Y I VY Y Hydrogen Bond I VYS70 / A GLU185 Y Y I LYS38 / B GLU185 Y Y Y Salt Bridge I ASP30 / A ATK184 Y Y Missed ARG36 / TYR91 Hydrogen Bond Hydrogen Bond ARG36 / TRP145 Hydrogen Bond LYS38/GLU185 Salt Bridge JOA A ARG36 / B ASP152 N A ARG36 / B ASP152 N IOA A ARG36 / B ASP39 N A ARG36 / B ASP89 N A ARG36 / B ASP89 N A ARG36 / B ASP200 N A GLU41 / B LYS145 N N N			I LYS38 / A GLU185	Y
2QC1 Ioi I LYS38 / B GLU185 N 10A I ARG36 / A ASP87 N I ARG36 / A GLU151 N I LYS38 / B ASP160 N I GLU41 / A ARG182 N I HIS68 / A GLU185 Y I LYS70 / A GLU185 Y Salt Bridge I ASP30 / A TYR184 Y I LYS38 / A GLU185 Y I LYS38 / A GLU185 Y Salt Bridge Missed ARG36 / TRP145 LYS38/GLU185 Salt Bridge ZQC1 Ionic Bond 5A None 7.5A A ARG36 / B ASP152 N A LYS52 / B GLU129 N A ARG36 / B ASP89 I 0A A ASP30 / B LYS145 A A		7.5A	I ARG25 / A GLU185	Ν
2QC1 IoiA I ARG36 / A ASP87 N I ARG36 / A GLU151 N I ARG36 / A GLU151 N I LYS38 / B ASP160 N I GLU41 / A ARG182 N I HIS68 / A GLU185 Y I HIS68 / A GLU185 Y Hydrogen Bond I VAL40 / A PHE183 Y Y I LYS70 / A GLU185 Y Y Y I ASP30 / A TYR184 Y Y Y Salt Bridge I ASP30 / A TYR184 Y Y Missed ARG36 / TRP145 Hydrogen Bond Hydrogen Bond ARG36 / TRP145 Hydrogen Bond LYS38/GLU185 Salt Bridge Jonic Bond Salt Bridge Salt Bridge None 7.5A A ARG36 / B ASP152 N N I0A A ASP30 / B LYS145 N N I0A A ASP30 / B LYS145 N A ARG36 / B ASP89 N A ARG36 / B ASP200 N A ARG36 / B ASP200 N A ARG36 / B ASP200 N			I LYS38 / B GLU185	Ν
2QC1Ionic BondI ARG36 / A GLU151NI LYS38 / B ASP160NI GLU41 / A ARG182NI HIS68 / A GLU185YI HIS68 / A GLU185YI HIS68 / A GLU185YI VS70 / A GLU185YI VAL40 / A PHE183YI ASP30 / A TYR184YI LYS38 / A GLU185YSalt BridgeI ASP30 / A ARG182MissedARG36 / TYR91Hydrogen BondLYS38/GLU185Salt BridgeI ASP30 / A ARG182MissedARG36 / TRP145Hydrogen BondLYS38/GLU185Salt BridgeI ASP30 / A ARG182MissedARG36 / B ASP152NA LYS52 / B GLU12910AA ASP30 / B LYS145A ARG36 / B ASP89NA ARG36 / B ASP89NA ARG36 / B ASP200NA GLU41 / B LYS145N		10A	I ARG36 / A ASP87	Ν
2QC1 Ionic Bond I LYS38 / B ASP160 N I GLU41 / A ARG182 N I GLU41 / A ARG182 N I HIS68 / A GLU185 Y I HIS68 / A GLU185 Y I HYS70 / A GLU185 Y I Y I VAL40 / A PHE183 Y Y Y I ASP30 / A TYR184 Y Y Y I LYS38 / A GLU185 Y Y Y Salt Bridge I ASP30 / A ARG182 Y Y Missed ARG36 / TR91 Hydrogen Bond Hydrogen Bond ARG36 / TRP145 Hydrogen Bond HYS38/GLU185 Salt Bridge 2QC1 Ionic Bond Salt Bridge Salt Bridge 10A A ARG36 / B ASP152 N N 10A A ASP30 / B LYS145 N N A ARG36 / B ASP9 N A ARG36 / B ASP89 N A ARG36 / B ASP200 N A ARG36 / B ASP200 N A GLU41 / B LYS145 N N N N			I ARG36 / A GLU151	Ν
I GLU41 / A ARG182NI HIS68 / A GLU185YI HIS68 / A GLU185YI HIS68 / A GLU185YI LYS70 / A GLU185YI LYS70 / A GLU185YI VAL40 / A PHE183YI ASP30 / A TYR184YI LYS38 / A GLU185YSalt BridgeI ASP30 / A ARG182MissedARG36 / TYR91Hydrogen BondHydrogen BondARG36 / TRP145Hydrogen BondLYS38/GLU185Salt BridgeSalt BridgeI ASP30 / A ARG182MissedARG36 / TRP145Hydrogen BondLYS38/GLU185Salt BridgeI ASP30 / B LYS145IOAA ASP30 / B LYS145A ARG36 / B ASP200NA ARG36 / B ASP200NA ARG36 / B ASP200NA GLU41 / B LYS145N			I LYS38 / B ASP160	Ν
I HIS68 / A GLU185YI HIS68 / A GLU185YI HIS68 / A GLU189NI LYS70 / A GLU185YI LYS70 / A GLU185YI VAL40 / A PHE183YI ASP30 / A TYR184YI LYS38 / A GLU185YSalt BridgeI ASP30 / A ARG182MissedARG36 / TYR91ARG36 / TRP145Hydrogen BondLYS38/GLU185Salt BridgeZQC1Ionic Bond5ANone7.5AA ARG36 / B ASP152NA LYS52 / B GLU12910AA ASP30 / B LYS145A ARG36 / B ASP89A ARG36 / B ASP200NA ARG36 / B ASP200NA ARG36 / B ASP200NA GLU41 / B LYS145N			I GLU41 / A ARG182	Ν
I HIS68 / A GLU189NHydrogen BondI LYS70 / A GLU185YI VAL40 / A PHE183YI ASP30 / A TYR184YI LYS38 / A GLU185YSalt BridgeI ASP30 / A ARG182MissedARG36 / TYR91Hydrogen BondARG36 / TRP145Hydrogen BondLYS38/GLU185Salt BridgeARG36 / TRP145MissedNone7.5AA ARG36 / B ASP15210AA ASP30 / B LYS145NA ARG36 / B ASP200A ARG36 / B ASP200NA ARG36 / B ASP200NA ARG36 / B ASP200NA ARG36 / B ASP200N			I HIS68 / A GLU185	Y
Hydrogen BondI LYS70 / A GLU185YI VAL40 / A PHE183YI ASP30 / A TYR184YI LYS38 / A GLU185YI LYS38 / A GLU185YSalt BridgeI ASP30 / A ARG182MissedARG36 / TYR91Hydrogen BondARG36 / TRP145Hydrogen BondLYS38/GLU185Salt BridgeSalt BridgeLYS38/GLU185Salt BridgeIARG36 / B ASP152N10AA ARG36 / B ASP152NA ARG36 / B ASP89A ARG36 / B ASP200NA ARG36 / B ASP200NA ARG36 / B ASP200NA GLU41 / B LYS145N			I HIS68 / A GLU189	Ν
Hydrogen BondI VAL40 / A PHE183Y I ASP30 / A TYR184Y I ASP30 / A TYR184Y I LYS38 / A GLU185YSalt BridgeI ASP30 / A ARG182Y ARG36 / TYR91Hydrogen Bond Hydrogen Bond ARG36 / TRP1452QC1Ionic BondSalt Bridge5ANone-7.5AA ARG36 / B ASP152N10AA ASP30 / B LYS145N10AA ARG36 / B ASP200NA ARG36 / B ASP200N			I LYS70 / A GLU185	Y
Salt BridgeI ASP30 / A TYR184YI LYS38 / A GLU185YI ASP30 / A ARG182YARG36 / TYR91Hydrogen BondARG36 / TRP145Hydrogen BondLYS38/GLU185Salt BridgeSANone7.5AA ARG36 / B ASP15210AA ASP30 / B LYS145ARG36 / B ASP200NA ARG36 / B ASP200NA ARG36 / B ASP200NA ARG36 / B ASP200N		Hydrogen Bond	I VAL40 / A PHE183	Y
Salt Bridge MissedI LYS38 / A GLU185 I ASP30 / A ARG182 ARG36 / TRP1 Hydrogen Bond Hydrogen Bond Hydrogen Bond Salt Bridge2QC1Ionic Bond 5A 7.5ANone A ARG36 / B ASP152 A LYS52 / B GLU129 I 0ANone A ARG36 / B ASP152 A ARG36 / B ASP89 A ARG36 / B ASP89 A ARG36 / B ASP200 N A GLU41 / B LYS145N			I ASP30 / A TYR184	Y
Salt Bridge MissedI ASP30 / A ARG182 ARG36 / TYR91 ARG36 / TRP145 LYS38/GLU185Y2QC1Ionic Bond 5A 7.5ANone A ARG36 / B ASP152 B GLU129N10AA ARG36 / B ASP152 A ARG36 / B ASP89 A ARG36 / B ASP89 A ARG36 / B ASP200 A GLU41 / B LYS145N			I LYS38 / A GLU185	Y
MissedARG36 / TYR91 ARG36 / TRP145 LYS38/GLU185Hydrogen Bond Hydrogen Bond Salt Bridge2QC1Ionic Bond 5ANone 7.5ANone A ARG36 / B ASP152N10AA ASP30 / B LYS145N10AA ARG36 / B ASP89 A ARG36 / B ASP200NA ARG36 / B ASP200NA ARG36 / B ASP200NA GLU41 / B LYS145N		Salt Bridge	I ASP30 / A ARG182	Y
2QC1Ionic Bond 5A 7.5AHydrogen Bond Salt Bridge10AA ARG36 / B ASP152 A ARG36 / B ASP152N10AA ARG36 / B ASP89 A ARG36 / B ASP89 A ARG36 / B ASP200 N A GLU41 / B LYS145N		Missed	ARG36 / TYR91	Hydrogen Bond
2QC1Ionic BondSalt Bridge5ANone7.5AA ARG36 / B ASP152NA LYS52 / B GLU12910AA ASP30 / B LYS145NA ARG36 / B ASP89A ARG36 / B ASP200NA GLU41 / B LYS145			ARG36 / TRP145	Hydrogen Bond
2QC1Ionic BondNone5ANone7.5AA ARG36 / B ASP152NA LYS52 / B GLU129N10AA ASP30 / B LYS145NA ARG36 / B ASP89NA ARG36 / B ASP200NA GLU41 / B LYS145N			LYS38/GLU185	Salt Bridge
2QC1Ionic Bond 5ANone5ANone7.5AA ARG36 / B ASP152NA LYS52 / B GLU129N10AA ASP30 / B LYS145NA ARG36 / B ASP89NA ARG36 / B ASP200NA GLU41 / B LYS145N				
2QC1Ionic BondNone5ANone7.5AA ARG36 / B ASP152NA LYS52 / B GLU129N10AA ASP30 / B LYS145NA ARG36 / B ASP89NA ARG36 / B ASP200NA GLU41 / B LYS145N				
5ANone7.5AA ARG36 / B ASP152NA LYS52 / B GLU129N10AA ASP30 / B LYS145NA ARG36 / B ASP89NA ARG36 / B ASP200NA GLU41 / B LYS145N	2QC1	Ionic Bond		
7.5AA ARG36 / B ASP152NA LYS52 / B GLU129N10AA ASP30 / B LYS145NA ARG36 / B ASP89NA ARG36 / B ASP200NA GLU41 / B LYS145N		5A	None	
A LYS52 / B GLU129 N 10A A ASP30 / B LYS145 N A ARG36 / B ASP89 N A ARG36 / B ASP200 N A GLU41 / B LYS145 N		7.5A	A ARG36 / B ASP152	N
10AA ASP30 / B LYS145NA ARG36 / B ASP89NA ARG36 / B ASP200NA GLU41 / B LYS145N			A LYS52 / B GLU129	N
A ARG36 / B ASP89NA ARG36 / B ASP200NA GLU41 / B LYS145N		10A	A ASP30 / B LYS145	N
A ARG36 / B ASP200 N A GLU41 / B LYS145 N			A ARG36 / B ASP89	N
A GLU41 / B LYS145 N			A ARG36 / B ASP200	N
			A GLU41 / B LYS145	N

Protem/PDB Bond Type/Distance Interacting Amino Acid Pair Shown by Literature Hydrogen Bond A GLU41 / B HIS186 N A ARG36 / B CYS192 Y A ARG36 / B CYS192 Y A ARG36 / B TYR190 Y A ARG36 / B ARG149 Y A ARG36 / B TYR148 Y A VAL40 / B PHE189 Y A HIS68 / B SER191 N None None Barnase/Barstar Ionic Bond 5A A ARG59 / D GLU76 Y 7.5A A LYS27 / D GLU80 N A ARG59 / D ASP39 Y A GLU60 / D HIS17 N A ARG37 / D ASP39 Y A LYS27 / D ASP39 10A A LYS27 / D ASP39 Y A LYS27 / D ASP35 Y A LYS27 / D ASP35 10A A LYS27 / D ASP39 Y	Table S1 continued from previous page					
A GLU41 / B HIS186NHydrogen BondA ASP30 / B TYR190YA ARG36 / B CYS192YA ARG36 / B CYS192YA ARG36 / B ARG149YA LYS38 / B SER191YA LYS38 / B SER191NA LYS70 / B CYS192NSalt BridgeNoneMissedNoneSalt BridgeNone7.5AA ARG59 / D GLU76YA GLU60 / D ASP35Y7.5AA LYS70 / D GLU80NA ARG59 / D GLU80NA ARG59 / D GLU80NA ARG59 / D ASP35Y7.5AA LYS27 / D GLU80A ARG59 / D ASP39YA GLU60 / D HIS17NA ARG59 / D ASP39YA ARG59 / D ASP39YA ARG59 / D ASP35YA ARG58 / D ASP35YA ARG58 / D ASP35YA ARG83 / D ASP35YA ARG	Protein/PDB	Bond Type/Distance	Interacting Amino Acid Pair	Shown by Literature		
Hydrogen BondA ASP30 / B TYR190YA ARG36 / B CYS192YA ARG36 / B THR148YA ARG36 / B THR148YA ARG36 / B ARG149YA LYS38 / B SER191YA VAL40 / B PHE189YA VAL40 / B PHE189YA LYS70 / B CYS192NSalt BridgeNoneMissedNoneBarnase/BarstarIonic Bond5AA ARG59 / D GLU76YA ARG59 / D ASP35YA HIS102 / D ASP39Y7.5AA LYS27 / D GLU80NA ARG59 / D ASP39YA ARG59 / D ASP35YA ARG37 / D ASP35 <td< td=""><td></td><th></th><td>A GLU41 / B HIS186</td><td>N</td></td<>			A GLU41 / B HIS186	N		
A ARG36 / B CYS192 Y A ARG36 / B THR148 Y A ARG36 / B ARG149 Y A LYS38 / B SER191 Y A VAL40 / B PHE189 Y A HIS68 / B SER191 N A LYS70 / B CYS192 N None Missed None Barnase/Barstar Ionic Bond 5A A ARG59 / D GLU76 Y A ARG59 / D ASP35 Y 7.5A A ARG59 / D GLU76 Y A HIS102 / D ASP39 Y 7.5A A LYS27 / D GLU80 N A ARG59 / D ASP39 Y A GLU60 / D HIS17 N A ARG87 / D ASP39 Y 10A A LYS27 / D GLU46 N A LYS27 / D GLU46 N A LYS27 / D ASP39 Y 10A A LYS27 / D ASP39 Y A ARG87 / D ASP35 Y A ARG59 / D ASP35 N A HIS102 / D ASP35 N A HIS102 / D ASP35 N A ARG59 / D ASP35 Y A ARG59 / D GLU76 Y A ARG59 / D ASP35 Y A ARG59 / D ASP39 Y A ARG57 / D ASP39 Hydrogen Bond + Electrostat ARG57/ASP39 Hydrogen Bond + Electrostat ARG57/ASP39 Hydrogen Bond + Electrostat ARS7/ASP39 Hydrogen Bond + Electrostat ARS7/ASP39 Hydrogen Bond + Electros		Hydrogen Bond	A ASP30 / B TYR190	Y		
A ARG36 / B THR148YA ARG36 / B ARG149YA LYS38 / B SER191YA VAL40 / B PHE189YA VAL40 / B PHE189YA VAL40 / B PHE189YA LYS70 / B CYS192NSalt BridgeNoneMissedNoneSalt BridgeNoneNoneYA LYS70 / B CYS192NSalt BridgeNoneJoneYA LYS70 / B CYS192NSalt BridgeNoneNoneYA ARG59 / D GLU76YA ARG59 / D ASP35YA LYS27 / D GLU80NA ARG59 / D GLU80NA ARG59 / D GLU80NA ARG59 / D GLU80NA ARG59 / D ASP39YA ARG37 / D ASP39YA ARG83 / D ASP39YA LYS27 / D THRE42YA ARG59 / D GLU46NA LYS27 / D THRE42YA ARG59 / D ASP35YA ARG3 / D ASP35YA ARG3 / D ASP35YA ARG3 / D ASP35NA GLU60 / D ASP35NA GLU60 / D ASP35NA ARG3 / D ASP39YA ARG3 / D ASP39YA ARG3 / D ASP39YA ARG37 / D AS			A ARG36 / B CYS192	Y		
A ARG36 / B ARG149YA LYS38 / B SER191YA VAL40 / B PHE189YA HIS68 / B SER191NA LYS70 / B CYS192NNoneNoneBarnase/BarstarIonic Bond5AA ARG59 / D GLU76YA ARG59 / D ASP35Y7.5AA LYS27 / D GLU80NA ARG59 / D ASP39Y7.5AA LYS27 / D GLU80A ARG59 / D ASP39YA GLU60 / D HIS17NA ARG87 / D ASP39YA ARG87 / D ASP39YA ARG59 / D ASP39YA ARG59 / D ASP39YA ARG59 / D ASP39YA ARG87 / D ASP39YA ARG87 / D ASP35NA ARG59 / D ASP35NA ARG59 / D ASP35YA ARG59 / D ASP35YA ARG59 / D GLU76YA ARG37 / D ASP35YA ARG59 / D ASP35NA ARG59 / D GLU76YA ARG37 / D ASP35NA ARG59 / D GLU76YA ARG37 / D ASP39YA ARG37 / D ASP39YA ARG37 / D ASP35NA ARG37 / D ASP39YA ARG37 / D ASP39YA ARG37 / D ASP39 <td></td> <th></th> <td>A ARG36 / B THR148</td> <td>Y</td>			A ARG36 / B THR148	Y		
Barnase/BarstarIonic Bond SAIY A HIS68 / B SER191 NoneY A HIS68 / B SER191 N A LYS70 / B CYS192 NBarnase/BarstarIonic Bond SAY A ARG59 / D GLU76 A ARG59 / D GLU76 Y A ARG59 / D GLU76 Y A ARG59 / D ASP35 Y A HIS102 / D ASP39 Y 7.5AY A ARG59 / D GLU80 A ARG59 / D GLU80 N A ARG59 / D ASP39 Y A GLU60 / D HIS17 N A ARG87 / D ASP39 Y A ARG87 / D ASP35 N A ARG59 / D GLU46 N A LYS27 / D ASP35 Y A ARG59 / D ASP35 N A AGG3 / D ASP35 N A ARG63 / D ASP35 N A ARG63 / D ASP35 N A ARG63 / D ASP35 N A ARG63 / D ASP39 Y A ARG83 / D DYR29 Y A ARG83 / D DYR29 Y A ARG83 / D DYR29 Y A ARG83 / D DYR29 Y A ARG83 / D DSP39 Y A ARG87 / D ASP39 Y A ARG87 /			A ARG36 / B ARG149	Y		
A VAL40 / B PHE189Y A HIS68 / B SER191N N A LYS70 / B CYS192NBarnase/BarstarIonic BondCYS192N5AA ARG59 / D GLU76Y A ARG59 / D ASP35Y A ARG59 / D ASP39Y7.5AA LYS27 / D GLU80NA ARG59 / D GLU80N A ARG59 / D GLU80NA ARG59 / D GLU80N A ARG59 / D GLU80NA ARG59 / D GLU80N A ARG59 / D GLU80NA ARG59 / D GLU80N A ARG87 / D ASP39Y10AA LYS27 / D ASP39Y10AA LYS27 / D ASP39YA ARG63 / D ASP35NA ARG63 / D ASP35NA ARG68 / D ASP35YA ARG68 / D ASP35NA ARG69 / D GLU76YA ARG59 / D GLU76YA ARG63 / D ASP35NA GLU60 / D LEU34YA ARG83 / D ASP39YA ARG83 / D ASP39YA ARG83 / D ASP39YA ARG83 / D ASP35NA GLU60 / D LEU34YA ARG87 / D ASP39YA ARG87 / D ASP39 <td></td> <th></th> <td>A LYS38 / B SER191</td> <td>Y</td>			A LYS38 / B SER191	Y		
Barnase/BarstarIonic Bond 5AA RG59 / D GLU76 A ARG59 / D GLU76 A ARG59 / D ASP35 Y A HIS102 / D ASP39 YNoneBarnase/BarstarIonic Bond 5AA ARG59 / D GLU76 A ARG59 / D ASP35 Y A HIS102 / D ASP39 YY Y7.5AA LYS27 / D GLU80 A ARG59 / D GLU80 N A ARG59 / D GLU80 A ARG59 / D GLU80 N A ARG59 / D GLU80 A ARG59 / D ASP39 YN N10AA ARG57 / D ASP39 A ARG87 / D ASP39 Y A ARG87 / D ASP39 YY Y Y10AA LYS27 / D ASP39 A ARG87 / D ASP39 Y A ARG59 / D GLU46 A LYS27 / D THRE42 Y A ARG59 / D GLU76 Y A ARG59 / D ASP35 N A AIS102 / D ASP35 YY Y Y A ARG59 / D GLU76 Y A ARG83 / D ASP39 Y A ARG83 / D ASP39 YHydrogen BondA LYS27 / D THRE42 Y A ARG59 / D GLU76 Y A ARG83 / D ASP39 Y A ARG87 / D ASP39 Hydrogen Bond + Electrostat ASN84 / TYR29Hydrogen Bond + Electrostat ASN84 / TYR29Hydrogen Bond + Electrostat ASN84 / TYR29			A VAL40 / B PHE189	Y		
Salt Bridge MissedNoneNBarnase/BarstarIonic Bond 5AAARG59 / D GLU76 A ARG59 / D ASP35 A HIS102 / D ASP39 YY7.5AA LYS27 / D GLU80 A ARG59 / D GLU80 A ARG59 / D GLU80 A ARG59 / D ASP39 YN7.5AA LYS27 / D GLU80 A ARG59 / D ASP39 YN7.5AA LYS27 / D GLU80 A ARG59 / D ASP39 YN8ARG59 / D ASP39 A GLU60 / D HIS17 A ARG83 / D ASP39 YN9A ARG87 / D ASP39 YY10AA LYS27 / D GLU46 A LYS27 / D ASP35 YN9A ARG87 / D ASP39 YY10AA LYS27 / D THRE42 YY9A ARG59 / D GLU76 YY9A ARG59 / D GLU76 YY9A ARG59 / D GLU76 YY9A ARG83 / D ASP35 YN9A ARG59 / D GLU76 YY9A ARG83 / D ASP39 YY9A ARG59 / D GLU76 YY9A ARG83 / D ASP39 YY9A ARG83 / D ASP39 YY9A ARG87 / D ASP39 Y<			A HIS68 / B SER191	N		
Salt Bridge MissedNone NoneBarnase/BarstarIonic Bond5AA ARG59 / D GLU76YA ARG59 / D ASP35YA HIS102 / D ASP39YYYA LYS27 / D GLU80NA ARG59 / D GLU80NA ARG59 / D ASP39YYYA GLU60 / D HIS17NA ARG57 / D ASP39YA GLU60 / D HIS17NA ARG57 / D ASP39YA LYS27 / D ASP39YA ARG57 / D ASP35NA HIS102 / D ASP35A HIS102 / D ASP35YA ARG59 / D GLU76YA ARG59 / D GLU76YA ARG37 / D ASP39YA ARG37 / D ASP39YA ARG37 / D ASP35NA GLU60 / D LEU34YA ARG37 / D ASP39YA ARG37 /			A LYS70 / B CYS192	N		
MissedNoneBarnase/BarstarIonic Bond5AA ARG59 / D GLU76YA ARG59 / D ASP35YA HIS102 / D ASP39YA LYS27 / D GLU80NA ARG59 / D ASP39YA GLU60 / D HIS17NA ARG87 / D ASP39A GLU60 / D HIS17NA ARG87 / D ASP39YA LYS27 / D ASP39YA LYS39 / D GLU46NA LYS27 / D ASP35YA LYS27 / D ASP35YA LYS39 / D GLU46NA LYS62 / D ASP35YA ARG59 / D GLU76YA ARG59 / D GLU76YA ARG59 / D GLU76YA ARG83 / D ASP39YA ARG83 / D ASP35YA ARG83 / D ASP35YA ARG83 / D ASP35YA ARG83 / D ASP39YA ARG87 / D ASP39YA ARG87/ASP39Hydrogen Bond + ElectrostatARG87/ASP39 <t< th=""><th></th><th>Salt Bridge</th><th>None</th><th></th></t<>		Salt Bridge	None			
Barnase/BarstarIonic BondA ARG59 / D GLU76Y A ARG59 / D ASP35Y A A HIS102 / D ASP39Y Y A A HIS102 / D ASP39Y A A A RG59 / D GLU80N N A A ARG59 / D GLU80N N A ARG59 / D ASP39Y Y A A GLU60 / D HIS17N N A ARG83 / D ASP39Y A A ARG83 / D ASP39Y A A ARG87 / D ASP35N A A HIS102 / D ASP35N A A HIS102 / D ASP35N A A ARG59 / D GLU46N A LYS27 / D THRE42Y A ARG59 / D GLU76Y A ARG87 / D ASP35N A A GLU60 / D LEU34Y A ARG83 / D TYR29Y A ARG83 / D GLY43Y A ARG83 / D GLY43Y A ARG87 / D ASP39Y A ARG87 / D ASP39Y A ARG87 / D ASP39Y A ARG87 / D ASP35N A ARG87 / D ASP39Y A ARG87 / D ASP39		Missed	None			
Barnase/BarstarIonic BondA ARG59 / D GLU76Y5AA ARG59 / D ASP35YA ARG59 / D ASP39Y7.5AA HIS102 / D ASP39Y7.5AA ARG59 / D GLU80NA ARG59 / D GLU80NA ARG59 / D GLU80NA ARG59 / D ASP39YA GLU60 / D HIS17NA ARG83 / D ASP39YA ARG87 / D ASP39Y10AA LYS27 / D ASP39YA ARG87 / D ASP39YA ARG87 / D ASP35NA HIS102 / D ASP35A Hydrogen BondA LYS27 / D THRE42YA ARG59 / D ASP35YA ARG59 / D ASP35YA ARG83 / D ASP35YA ARG83 / D ASP35YA ARG83 / D ASP35A GLU60 / D LEU34YA ARG83 / D ASP39YA ARG87 / D ASP39Y						
Datitise barsalTotal bold5AA ARG59 / D GLU76Y5AA ARG59 / D ASP35YA HIS102 / D ASP39Y7.5AA LYS27 / D GLU80NA ARG59 / D GLU80NA ARG59 / D ASP39YA GLU60 / D HIS17NA ARG83 / D ASP39YA GL060 / D HIS17NA ARG87 / D ASP39Y10AA LYS27 / D ASP39YA ARG87 / D ASP39YA ARG87 / D ASP35YA LYS27 / D ASP35YA LYS27 / D ASP35YA ARG59 / D ASP35YA ARG83 / D ASP39YA ARG83 / D ASP39YA ARG83 / D ASP39YA ARG83 / D ASP39YA ARG87 / D ASP39Y <td>Barnaco/Barctar</td> <th>Ionia Bond</th> <td></td> <td></td>	Barnaco/Barctar	Ionia Bond				
JA A ARG59 / D ASP35 Y A ARG59 / D ASP35 Y A HIS102 / D ASP35 Y A HIS102 / D ASP39 Y A LYS27 / D GLU80 N A ARG59 / D ASP39 Y A GLU60 / D HIS17 N A ARG83 / D ASP39 Y A GLU60 / D HIS17 N A ARG87 / D ASP39 Y A ARG87 / D ASP35 N A HIS102 / D ASP35 Y A ARG59 / D GLU76 Y A ARG59 / D GLU76 Y A GLU60 / D LEU34 Y A ARG83 / D ASP39 Y A ARG87 / D ASP39 Y A ARG87 / D ASP39 Y A ARG87 / D ASP39 Y	Darnase/Darstar		A APG50 / D CLU76	v		
A ARG09 / D ASP39 1 A HIS102 / D ASP39 Y A ARG59 / D GLU80 N A ARG59 / D GLU80 N A ARG59 / D ASP39 Y A GLU60 / D HIS17 N A ARG83 / D ASP39 Y A ARG87 / D ASP39 Y A LYS27 / D THRE42 Y A ARG59 / D GLU76 Y A ARG59 / D GLU76 Y A ARG83 / D TYR29 Y A ARG83 / D ASP39 Y A ARG83 / D GLV73 Y A HIS102 / D		JA	A ABG50 / D ASD25			
7.5A A $LYS27 / D GLU80$ N A ARG59 / D GLU80 N A ARG59 / D ASP39 Y A GLU60 / D HIS17 N A ARG83 / D ASP39 Y A ARG87 / D ASP35 N A HIS102 / D ASP35 N A HIS102 / D ASP35 Y A ARG59 / D GLU76 Y A ARG83 / D TYR29 Y A ARG83 / D ASP39 Y A ARG83 / D ASP39 Y A ARG83 / D ASP39 Y A ARG87 / D ASP39 Y						
A A RGS9 / D GLU80NA ARGS9 / D GLU80NA ARGS9 / D ASP39YA GLU60 / D HIS17NA ARG83 / D ASP39YA ARG87 / D ASP39YA ARG87 / D ASP39YA LYS27 / D ASP39YA LYS27 / D ASP39YA LYS27 / D ASP35NA HIS102 / D ASP35YA HIS102 / D ASP35YA ARG59 / D GLU46NA LYS27 / D THRE42YA ARG59 / D GLU76YA GLU60 / D ASP35NA GLU60 / D LEU34YA ARG83 / D TYR29YA ARG83 / D GLY43YA ARG87 / D ASP39YA HIS102 / D GLU76YMissedARG87/ASP39ARG87/ASP39Hydrogen Bond + ElectrostatASN84 / TYR29Hydrogen Bond		7 5 4	$\Delta IVS27 / D CI II20$			
A ARG59 / D GLU30NA ARG59 / D ASP39YA GUU60 / D HIS17NA ARG83 / D ASP39YA ARG87 / D ASP39YA ARG87 / D ASP39YA LYS27 / D ASP39YA LYS27 / D ASP35NA LYS62 / D ASP35NA HIS102 / D ASP35YA ARG59 / D GLU76YA ARG59 / D GLU76YA GLU60 / D ASP35NA ARG59 / D GLU76YA ARG83 / D CLU34YA ARG83 / D ASP39YA ARG83 / D GLY43YA ARG87 / D ASP39YA HIS102 / D GLY31YA HIS102 / D GLU76YMissedARG87/ASP39ASN84 / TYR29Hydrogen Bond + ElectrostatASN84 / TYR29Hydrogen Bond		7.3A	A LIS277 D GLU80			
A ARG39 / D ASF39 A GLU60 / D HIS17 N A ARG83 / D ASP39 Y A ARG87 / D ASP39 Y A ARG87 / D ASP39 Y A LYS27 / D ASP39 Y A LYS27 / D ASP35 N A HIS102 / D ASP35 Y Hydrogen Bond A LYS27 / D THRE42 Y A ARG59 / D GLU76 Y A ARG59 / D GLU76 Y A ARG59 / D GLU76 Y A ARG83 / D TYR29 Y A ARG83 / D TYR29 Y A ARG83 / D GLY43 Y A ARG83 / D GLY43 Y A ARG87 / D ASP39 Y A HIS102 / D ASP39 Y A HIS102 / D ASP39 Y A HIS102 / D GLY31 Y A ARG37/ASP39 Hydrogen Bond + Electrostat ARG87/ASP39 Hydrogen Bond + Electrostat ARG87/ASP39 Hydrogen Bond + Electrostat ASN84 / TYR29 Hydrogen Bond			A ABG50 / D ASD20			
A GLUGO / D HIST/NA ARG83 / D ASP39YA ARG87 / D ASP39YA ARG87 / D ASP39YA LYS27 / D ASP39YA LYS39 / D GLU46NA LYS62 / D ASP35NA HIS102 / D ASP35YHydrogen BondA LYS27 / D THRE42YA ARG59 / D ASP35A ARG59 / D ASP35YA ARG59 / D GLU76YA GLU60 / D ASP35NA ARG83 / D TYR29YA ARG83 / D ASP39YA ARG87 / D ASP39YA ARG87 / D ASP39YA ARG87 / D ASP39YA HIS102 / D GLY31YA HIS102 / D GLU76YMissedARG59 / D GLU76ARG83/ASP39Hydrogen Bond + ElectrostatARG87/ASP39Hydrogen Bond			A ARG397 D ASP39 A CLUG07 D $UIS17$			
A ARO35 / D ASP39IA ARG87 / D ASP39YA ARG87 / D ASP39YA LYS27 / D ASP39YA LYS39 / D GLU46NA LYS62 / D ASP35NA HIS102 / D ASP35YA ARG59 / D ASP35YA ARG59 / D ASP35YA ARG59 / D ASP35YA ARG59 / D GLU76YA GLU60 / D ASP35NA GLU60 / D ASP35NA ARG83 / D TYR29YA ARG83 / D TYR29YA ARG83 / D ASP39YA ARG83 / D ASP39YA ARG87 / D ASP39YA ARG87/ASP39Hydrogen Bond + ElectrostatARG87/ASP39Hydrogen Bond + ElectrostatASN84 / TYR29Hydrogen Bond			A OLUOU / D HIST / A ADC 82 / D ASD 20			
10AA AR08/7 D ASP39110AA LYS27 / D ASP39YA LYS39 / D GLU46NA LYS62 / D ASP35NA HIS102 / D ASP35YA ARG59 / D ASP35YA ARG59 / D ASP35YA ARG59 / D GLU76YA GLU60 / D ASP35NA GLU60 / D ASP35NA ARG83 / D TYR29YA ARG83 / D GLY43YA ARG83 / D ASP39YA ARG87 / D ASP39YA ARG87 / D ASP39YA HIS102 / D ASP39YA HIS102 / D ASP39YA ARG87/ASP39Hydrogen Bond + ElectrostatARG87/ASP39Hydrogen Bond + ElectrostatARG87/ASP39Hydrogen Bond + ElectrostatASN84 / TYR29Hydrogen Bond			A ARG83 / D ASP39	I V		
IOAA L132/7 / D ASP39IA LYS39 / D GLU46NA LYS62 / D ASP35NA HIS102 / D ASP35YA HIS102 / D ASP35YA ARG59 / D GLU76YA ARG59 / D GLU76YA GLU60 / D ASP35NA GLU60 / D ASP35NA GLU60 / D LEU34YA ARG83 / D TYR29YA ARG83 / D ASP39YA ARG83 / D ASP39YA ARG83 / D ASP39YA ARG87 / D ASP39YA ARG87 / D ASP39YA ARG87 / D ASP39YA HIS102 / D ASN33YA HIS102 / D ASP39YA HIS102 / D ASP39YA ARG87/ASP39Hydrogen Bond + ElectrostatARG87/ASP39Hydrogen Bond + ElectrostatASN84 / TYR29Hydrogen Bond		10.4	A AKU677 D ASP39			
A LYS39 / D GU046NA LYS62 / D ASP35NA LYS62 / D ASP35YA HIS102 / D ASP35YA ARG59 / D ASP35YA ARG59 / D GLU76YA GLU60 / D ASP35NA GLU60 / D ASP35NA GLU60 / D LEU34YA ARG83 / D TYR29YA ARG83 / D ASP39YA ARG83 / D ASP39YA ARG83 / D ASP39YA ARG83 / D ASP39YA ARG87 / D ASP39YA ARG87 / D ASP39YA HIS102 / D ASN33YA HIS102 / D ASP39YA HIS102 / D ASP39YA ARG87/ASP39Hydrogen Bond + ElectrostatARG87/ASP39Hydrogen Bond + ElectrostatARG87/ASP39Hydrogen Bond + ElectrostatASN84 / TYR29Hydrogen Bond		10A	A LYS20 / D CLU46	I N		
A LYS62 / D ASP35NA HIS102 / D ASP35YA HIS102 / D ASP35YA LYS27 / D THRE42YA ARG59 / D ASP35YA ARG59 / D GLU76YA GLU60 / D ASP35NA GLU60 / D ASP35NA GLU60 / D LEU34YA ARG83 / D TYR29YA ARG83 / D GLY43YA ARG83 / D GLY43YA ARG87 / D ASP39YA ARG87 / D ASP39YA HIS102 / D ASP39YA HIS102 / D ASP39YA HIS102 / D GLY31YA HIS102 / D ASP39YA HIS102 / D ASP39Hydrogen Bond + ElectrostatARG87/ASP39Hydrogen Bond + ElectrostatASN84 / TYR29Hydrogen Bond			A LISSY / D GLU40			
Hydrogen BondA HIS102 / D ASP33YA LYS27 / D THRE42YA ARG59 / D ASP35YA ARG59 / D GLU76YA GLU60 / D ASP35NA GLU60 / D LEU34YA ARG83 / D TYR29YA ARG83 / D ASP39YA ARG83 / D ASP39YA ARG83 / D ASP39YA ARG87 / D ASP39YA ARG87 / D ASP39YA HIS102 / D ASN33YA HIS102 / D GLY31YA HIS102 / D ASP39YA HIS102 / D ASP39Hydrogen Bond + ElectrostatARG87/ASP39Hydrogen Bond + ElectrostatARG87/ASP39Hydrogen BondASN84 / TYR29Hydrogen Bond			A LISO2 / D ASP35 A μ S102 / D ASP35			
Hydrogen BondA LYS2//D THRE42TA ARG59 / D ASP35YA ARG59 / D GLU76YA GLU60 / D ASP35NA GLU60 / D LEU34YA ARG83 / D TYR29YA ARG83 / D ASP39YA ARG83 / D ASP39YA ARG83 / D ASP39YA ARG87 / D ASP39YA ARG87 / D ASP39YA ARG87 / D ASP39YA HIS102 / D ASP39YA HIS102 / D GLY31YA HIS102 / D GLY31YA RG59 / D GLU76YMissedARG87/ASP39Hydrogen Bond + ElectrostatARG87/ASP39Hydrogen Bond + ElectrostatASN84 / TYR29Hydrogen Bond		Hudus con Dond	A HISTOZ / D ASP35	I V		
A ARG39 / D ASP35YA ARG59 / D GLU76YA GLU60 / D ASP35NA GLU60 / D LEU34YA ARG83 / D TYR29YA ARG83 / D ASP39YA ARG83 / D GLY43YA ARG83 / D GLY43YA ARG87 / D ASP39YA HIS102 / D GLY31YA HIS102 / D GLY31YA HIS102 / D GLU76YMissedARG83/ASP39ARG87/ASP39Hydrogen Bond + ElectrostatARG87/ASP39Hydrogen Bond + ElectrostatASN84 / TYR29Hydrogen Bond		Hydrogen Bond	A LYS277D THRE42	I V		
A ARG39 / D GLU76YA GLU60 / D ASP35NA GLU60 / D LEU34YA ARG83 / D TYR29YA ARG83 / D ASP39YA ARG83 / D GLY43YA ARG87 / D ASP39YA ARG87 / D ASP39YA HIS102 / D ASN33YA HIS102 / D GLY31YA HIS102 / D GLU76YMissedARG87/ASP39Hydrogen Bond + ElectrostatARG87/ASP39Hydrogen Bond + ElectrostatASN84 / TYR29Hydrogen Bond			A ARG59 / D ASP35	Y V		
A GLU60 / D ASP35NA GLU60 / D LEU34YA ARG83 / D TYR29YA ARG83 / D ASP39YA ARG83 / D GLY43YA ARG87 / D ASP39YA ARG87 / D ASP39YA ARG87 / D ASP39YA HIS102 / D ASN33YA HIS102 / D GLY31YA HIS102 / D ASP39YA ARG59 / D GLU76YMissedARG83/ASP39ARG87/ASP39Hydrogen Bond + ElectrostatASN84 / TYR29Hydrogen Bond			A ARG59 / D GLU/6	Y		
A GL060 / D LE034YA ARG83 / D TYR29YA ARG83 / D ASP39YA ARG83 / D GLY43YA ARG87 / D ASP39YA ARG87 / D ASP39YA HIS102 / D ASN33YA HIS102 / D GLY31YA HIS102 / D GLU76YMissedARG87/ASP39ARG87/ASP39Hydrogen Bond + ElectrostatARG87/ASP39Hydrogen Bond + ElectrostatARG87/ASP39Hydrogen Bond + Electrostat			A GLU60 / D ASP35	N		
A ARG83 / D TYR29YA ARG83 / D ASP39YA ARG83 / D ASP39YA ARG83 / D GLY43YA ARG87 / D ASP39YA ARG87 / D ASP39YA HIS102 / D ASN33YA HIS102 / D GLY31YA HIS102 / D ASP39YA HIS102 / D ASP39YA RG59 / D GLU76YMissedARG87/ASP39ARG87/ASP39Hydrogen Bond + ElectrostatARG87/ASP39Hydrogen Bond + ElectrostatASN84 / TYR29Hydrogen Bond			A GLU60 / D LEU34	Y		
A ARG83 / D ASP39YA ARG83 / D GLY43YA ARG87 / D ASP39YA ARG87 / D ASP39YA ARG87 / D ASP39YA HIS102 / D ASN33YA HIS102 / D GLY31YA HIS102 / D GLY31YA HIS102 / D ASP39YA RG59 / D GLU76YMissedARG87/ASP39ARG87/ASP39Hydrogen Bond + ElectrostatASN84 / TYR29Hydrogen Bond			A ARG83 / D TYR29	Y		
A ARG83 / D GLY43YA ARG87 / D ASP39YA ARG87 / D ASP39YA HIS102 / D ASN33YA HIS102 / D GLY31YA HIS102 / D GLY31YA HIS102 / D GLU76YMissedARG83/ASP39ARG87/ASP39Hydrogen Bond + ElectrostatARG87/ASP39Hydrogen Bond + ElectrostatASN84 / TYR29Hydrogen Bond			A ARG83 / D ASP39	Y		
A ARG8 / / D ASP39YA HIS102 / D ASN33YA HIS102 / D GLY31YA HIS102 / D GLY31YA HIS102 / D ASP39YA HIS102 / D ASP39YA RG59 / D GLU76YMissedARG83/ASP39ARG87/ASP39Hydrogen Bond + ElectrostatARG87/ASP39Hydrogen Bond + ElectrostatASN84 / TYR29Hydrogen Bond			A AKG83 / D GLY43			
A HIS102 / D ASN33YA HIS102 / D GLY31YA HIS102 / D GLY31YA HIS102 / D ASP39YA HIS102 / D ASP39YARG59 / D GLU76YMissedARG83/ASP39ARG87/ASP39Hydrogen Bond + ElectrostatASN84 / TYR29Hydrogen Bond			A ARG87 / D ASP39	Y		
Salt Bridge MissedA HIS102 / D GLY31 A HIS102 / D ASP39 ARG59 / D GLU76 ARG83/ASP39 ARG87/ASP39 ASN84 / TYR29Y Y Hydrogen Bond + Electrostat Hydrogen Bond + Electrostat Hydrogen Bond + Electrostat			A HIS102 / D ASN33			
Salt Bridge MissedA HIS102 / D ASP39 ARG59 / D GLU76 ARG83/ASP39YHydrogen Bond + Electrostat ARG87/ASP39 ASN84 / TYR29Hydrogen Bond + Electrostat Hydrogen Bond + Electrostat			A HIS102 / D GLY31			
Salt Bridge MissedARG59 / D GLU76 ARG83/ASP39 ARG87/ASP39 ASN84 / TYR29Y Hydrogen Bond + Electrostat Hydrogen Bond + Electrostat Hydrogen Bond		G H D 1	A HIS102 / D ASP39			
Missed ARG83/ASP39 Hydrogen Bond + Electrostat ARG87/ASP39 ASN84 / TYR29 Hydrogen Bond + Electrostat Hydrogen Bond + Electrostat		Salt Bridge	ARG59 / D GLU76	Y		
ARG8//ASP39 ASN84 / TYR29 Hydrogen Bond Hydrogen Bond		Missed	ARG83/ASP39	Hydrogen Bond + Electrostatic		
ASN84 / TYR29 Hydrogen Bond			ARG87/ASP39	Hydrogen Bond + Electrostatic		
			ASN84 / TYR29	Hydrogen Bond		
Kap1a/Kaf Ionic Bond	Rap1a/Raf	Ionic Bond				
- 5A A GLU3 / B LYS65 Y	•	5A	A GLU3 / B LYS65	Y		
A ASP33 / B LYS84 Y			A ASP33 / B LYS84	Y		
A GLU37 / B ARG67 Y			A GLU37 / B ARG67	Y		
A ASP38 / B ARG89 Y			A ASP38 / B ARG89	Y		
7.5A A GLU37 / B ARG59 Y		7.5A	A GLU37 / B ARG59	Ŷ		
A GLU54 / B LYS65 N			A GLU54 / B LYS65	N		
A GLU54 / B ARG67 N			A GLU54 / B ARG67	N		
10A A GLU30 / B LYS87 N		10A	A GLU30 / B LYS87	N		

		continueu from previous page	
Protein/PDB	Bond Type/Distance	Interacting Amino Acid Pair	Shown by Literature
		A GLU37 / B ARG89	Ν
		A ASP38 / B ARG67	Ν
		A ASP38 / B LYS84	N
		Λ Λ SP57 / B IVS84	N
		A ASIST / D ADC90	
		A ASPS//B ARG89	N
	Hydrogen Bond	A ASP33 / B LYS84	Y
		A GLU37 / B VAL69	Y
		A GLU37 / B ARG59	Y
		A ASP38 / B THR68	Y
		A ASP38 / B ARG89	Y
		A SED20 / D ADC67	I V
		A SER39 / D AROU/	1 V
		A SER39 / B ARG89	Y
		A ARG41 / B ASN64	Y
	Salt Bridge	ASP33 / B LYS84,	Y
	Missed	ASP38 / ARG89	Salt Bridge
		GLU37/ARG67	Salt Bridge
		GLU37 / ARG50	Salt Bridge
		APC41 / CLN 66	Judragen Dand
		ARG41 / GLN 00	Hydrogen Bond
1KC4	Ionic Bond		
	5A	A LYS38 / B GLU188	Y
		A HIS68 / B GLU188	Y
	7.5A	A ARG25 / B GLU188	N
	,	A ARG36 / B GLU192	N
	10.4	Λ Λ $PG26 / P CLU184$	N
	IUA	A ADC2(/ D CLU104	IN N
		A ARG36 / B GLU188	Y
	Hydrogen Bond	A TRP28 / B TYR194	N
		A ARG36 / B GLU188	Y
	Salt Bridge	None	
	Missed	Arg36 / Phe186	Hydrogen Bond
		Arg36 / Tyr 194	Hydrogen Bond
			, ,
Smad2/Smad4	Ionic	B chain with C chain	
	54	B LVS340 / C GLU288	v
	511	D = 1103407 C = 010200	I V
		D ASP493 / C AKU321	
		в ASP493 / С ARG329	Y
	7.5A	B HIS317 / C ASP304	N
		B ASP493 / C ARG330	N
		B ASP494 / C ARG329	N
		B ASP494 / C ARG330	N
		B ARG496 / C GLU281	N
		B ARG496 / C GLU326	N
		\mathbf{D} ADC407 / C CLU224	N
		D AKU49// C ULU320	
		B ASP537 / C ARG310	Y
		B HIS541 / C ASP300	N
		B ASP547 / C ARG330	N
		B ASP547 / C HIS331	N
	10A	B GLU337 / C ARG285	N
		B GLU337 / C HIS291	N
		$\begin{array}{c} \mathbf{D} \mathbf{O} = $	N
		$\frac{1}{10} \frac{1}{100} \frac{1}{$	
		D AKU302 / C GLU320	
		B LYS519 / C GLU281	N

Table S1 continued from previous page

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Protein/PDB	Bond Type/Distance	Interacting Amino Acid Pair	Shown by Literature
		B GLU526 / C ARG321	Y
		B HIS528 / C GLU288	N
		B HIS530 / C GLU288	Ν
		B HIS530 / C ASP304	Ν
		B ASP537 / C ARG329	N
		B HIS541 / C ASP304	N
	Hbond	B THR338 / C GLU288	N
		B LEU533 / C THR303	Y
		B ASP537 / C ARG310	Y
		B GLU526 / C SER317	Y
		B ASP493 / C ARG321	Y
	Salt bridge	B LYS340 / C GLU288	Y
		B ASP493 / C ARG321	Y
		B ASP493 / C ARG329	Y
	Missed	LYS340 / GLU288	Hydrogen Bond
		ASP332 / ASN320	Hydrogen Bond
		HIS528 / SER318	Hydrogen Bond
		GLN534 / ASP304	Hydrogen Bond
		ASP537 / THR303	Hydrogen Bond
		ASP537 / ASP304	Hydrogen Bond

Table S1 continued from previous page

TABLE S2: Nullification predictions and agreement with bond list.

PDB ID	Nullification	Peak Direction	Agrees with Bond List?
1YI5			
	ASP27	-	Y
	ALA28	-	N
	ARG33	+	Y
	ARG36	+	Y
4HQP			
	ASP30	-	Y
	SER34	+	N
	SER35	+	N
	ARG36	+	Y
	GLY37	+	Ν
	LYS38	+	Y
	GLU56	-	Ν
	LYS70	+	Y
2QC1			
	ASP30	-	Y
	SER35	+	N
	ARG36	+	Y
1brs			
	LYS27	+	Y
	ASP54	-	Ν
	SER57	+	N
	ARG59	+	Y
	GLY65	-	N
	GLU73	-	N
	SER80	-	N

Table S2 continued from previous page			
PDB ID	Nullification	Peak Direction	Agrees with Bond List?
	ARG83	+	Y
	ARG87	+	Y
1c1y			
	GLU3	+	Y
	5	-	N
	16	-	N
	ASP33	+	Y
	36	-	N
	GLU37	+	Y
	ASP38	+	Y
	SER39	-	Y
	ARG41	-	Y
	GLU54	+	Y
	ASP57	+	Y
1KC4			
	ARG36	-	Y
	GLY37	+	N
SMAD2/SMAD4			
	VAL492	-	N
	ASP493	+	Y
	ARG496	-	Y
	ARG497	-	N
	ARG502	-	Y
	LYS519	-	N
	ASP537	+	Y
	ASP547	+	Y