Supplementary Materials: Mutation-Structure-Function Relationship Based Integrated Strategy Reveals the Potential Impact of Deleterious Missense Mutations in Autophagy Related Proteins on Hepatocellular Carcinoma (HCC): A Comprehensive Informatics Approach

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**Figure S1.** Visualization of the protein-protein interaction sub network of LC3A (A), LC3B (B), BECN1 (C) and SCD1 (D) proteins using PinSnps tool. Illustration created using Cytoscape, an open source platform for visualizing molecular interaction networks. The connection between two nodes is indicated by an edge.
Figure S2. (A) Cartoon representation of the top modeled 3D structure of BECN1 built from I-TASSER server; (B) The Ramachandran plot calculation of the psi/phi angle distribution of BECN1 model using PROCHECK validation server showing the stereochemical quality of the structure. The most favored regions are marked as (A, B, L). The additional allowed regions are marked as [a, b, l, p]. All non-glycine and proline residues are shown as filled black squares, whereas, glycines (non-end) are shown as filled black triangles. Disallowed residues are colored red.

Figure S3. (A) Cartoon representation of the top modeled 3D structure of SCD1 built from I-TASSER server; (B) The Ramachandran plot calculation of the psi/phi angle distribution of SCD1 model using PROCHECK validation server showing the stereochemical quality of the structure. The most favored regions are marked as (A, B, L). The additional allowed regions are marked as [a, b, l, p]. All non-glycine and proline residues are shown as filled black squares, whereas, glycines (non-end) are shown as filled black triangles. Disallowed residues are colored red.
Figure S4. Normal mode analysis of WT (A) and mutant LC3B (Y113C) (B) protein. Detailed profiles of mobility (B-factors), eigenvalues and deformability have been shown.
Figure S5. Normal mode analysis of WT (A) and mutant BECN1 (I403T) (B) protein. Detailed profiles of mobility (B-factors), eigenvalues and deformability have been shown.
Figure S6. Normal mode analysis of WT (A) and mutant SCD1 (Y218C) (B) protein. Detailed profiles of mobility (B-factors), eigenvalues and deformability have been shown.
Figure S7. Normal mode analysis of WT (A) and mutant SCD1 (R126S) (B) protein. Detailed profiles of mobility (B-factors), eigenvalues and deformability have been shown.
**Table S1.** Evaluating the effect of the amino acid mutations in LC3A by HOPE sever. The distinct pattern and possible consequences of native and mutant amino acid on protein structure based on size, charge, and hydrophobicity have been shown.

| WT Amino Acid | Position | M amino Acid | Characteristics and Possible Consequences |
|---------------|----------|--------------|------------------------------------------|
| Arg           | 24       | Cys          | The mutant (M) residue is smaller and more hydrophobic than the wild-type (WT) residue. The WT residue was +vely charged, the M residue is neutral. The WT residue forms a salt bridge with Asp at position 25. The difference in charge will disturb the ionic interactions. |
| Lys           | 49       | Ala          | The M residue is smaller and more hydrophobic than the WT residue. The WT residue was +vely charged, the M residue is neutral. The WT residue forms a salt bridge with: Asp at position 48. The difference in charge will disturb the ionic interactions. |
| Lys           | 51       | Ala          | The WT residue forms a hydrogen bond with Asp at position 4 and Thr at position 50. The size difference between WT and M residue makes that the new residue is not in the correct position to make the same hydrogen bond as the original WT residue did. The difference in hydrophobicity will affect hydrogen bond formation. The WT residue forms a salt bridge with Asp at position 4, Glu at position 19 and Glu at position 36. The difference in charge will disturb the ionic interactions. |
The M residue is smaller than the WT residue. This will cause a probable loss of external interactions.

The M residue is bigger than the WT residue. The WT residue is a Pro. Pro is known to be very rigid and therefore induce a special backbone conformation which might be required at this position. The mutation can disturb this special conformation. The residue is located on the surface of the protein; mutation of this residue can disturb interactions with other molecules or other parts of the protein.

The M residue is smaller than the WT residue. The WT residue is a Pro. Pros are known to be very rigid and therefore induce a special backbone conformation which might be required at this position. The mutation can disturb this special conformation.

The M residue is smaller and more hydrophobic than the WT residue. The WT residue was +vely charged, the M residue is neutral. The WT residue forms a salt bridge with: Asp at position 48. The difference in charge will disturb the ionic interactions.
The WT residue was neutral, the M residue is +vely charged. The mutation introduces a charge at this position; this can cause repulsion between the M residue and neighboring residues. The residue is located on the surface of the protein; mutation of this residue can disturb interactions with other molecules or other parts of the protein.

The residue is buried in the core of a domain. The differences between the WT and M residue might disturb the core structure of this domain. The mutation will cause an empty space in the core of the protein.

The mutation will cause an empty space and loss of hydrophobic interactions in the core of the protein. The WT residue is predicted to be located in its preferred secondary structure, a β-strand. The M residue prefers to be in another secondary structure; therefore the local conformation will be slightly destabilized.

The WT residue forms a hydrogen bond with Glu at position 102. The size difference between WT and M residue makes that the new residue is not in the correct position to make the same hydrogen bond as the original WT residue did. The WT residue is involved in a multimer contact. The mutation introduces a bigger residue at this position; this can disturb the multimeric interactions. M residue will cause slight destabilization in local conformation. The mutation introduces a charge at this position; this can cause repulsion between the M residue and neighboring residues. The residue is located on the surface of the protein; mutation of this residue can disturb interactions with other molecules or other parts of the protein.
The WT residue is predicted (by the PHDsec DAS-server) to be located in an α-helix. Pro disrupts an α-helix when not located at one of the first 3 positions of that helix. In case of the mutation at hand, the helix will be disturbed and this can have severe effects on the structure of the protein.

The WT residue forms a hydrogen bond with Asp at position 106, and a salt bridge with Lys at position 8 and Arg at position 16. The difference in charge will disturb the ionic interaction made by the original, WT residue.

The WT residue forms a salt bridge with Arg at position 16. The difference in charge will disturb the ionic interaction made by the original, WT residue. The residue is located on the surface of the protein; mutation of this residue can disturb interactions with other molecules or other parts of the protein.
The WT residue is a Gly, the most flexible of all residues. This flexibility might be necessary for the protein's function. Mutation of this Gly can abolish this function. Residues in the vicinity of the mutated residue are annotated in the UniProt database as being a binding site. The mutation could affect the local structure and as a consequence affect this binding site.

Table S2. Evaluating the effect of the amino acid mutations in LC3B by HOPE sever. The distinct pattern and possible consequences of native and Mutant amino acid on protein structure based on size, charge, and hydrophobicity have been shown.

| WT Amino Acid | Position | M amino Acid | Characteristics and Possible Consequences |
|---------------|----------|--------------|------------------------------------------|
| Arg           | 11       | Cys          | The M residue is smaller and more hydrophobic than the WT residue. The WT residue was +vely charged, the M residue is neutral. The WT residue forms a hydrogen bond and salt bridge with Asp at position 19. The difference in hydrophobicity will affect hydrogen bond formation. The difference in charge will disturb the ionic interaction made by the original, WT residue. |
| Arg           | 24       | Gln          | The M residue is smaller than the WT residue. The WT residue was +vely charged, the M residue is neutral. The WT residue forms a salt bridge with Glu at position 100. The difference in charge will disturb the ionic interaction made by the original, WT residue. |
The M residue is bigger than the WT residue. The WT residue is a Pro. Pro is known to be very rigid and therefore induce a special backbone conformation which might be required at this position. The mutation can disturb this special conformation. The residue is buried in the core of a domain. The differences between the WT and M residue might disturb the core structure of this domain.

The M residue is smaller than the WT residue. The mutation will cause an empty space in the core of the protein.

The M residue is smaller and more hydrophobic than the WT residue. The WT residue was +vely charged, the M residue is neutral. The WT residue forms a hydrogen bond with Leu at position 44 and a salt bridge with Glu at position 41. The difference in charge will disturb the ionic interaction made by the original, WT residue.

The M residue is bigger and more hydrophobic than the WT residue. The WT residue is a Pro. The WT residue is a Gly, the most flexible of all residues. This flexibility might be necessary for the protein's function. Mutation of this Gly can abolish this function. The residue is located on the surface of the protein; mutation of this residue can disturb interactions with other molecules or other parts of the protein. The torsion angles for this residue are unusual. Only Gly is flexible enough to make these torsion angles, mutation into another residue will force the local backbone into an incorrect conformation and will disturb the local structure.
The M residue is smaller and more hydrophobic than the WT residue. The WT residue was +vely charged, the M residue is neutral. The charge of the WT residue is lost by this mutation. This can cause loss of interactions with other molecules.

The M residue is smaller and more hydrophobic than the WT residue. The WT residue was +vely charged, the M residue is neutral. The charge of the WT residue is lost by this mutation. This can cause loss of interactions with other molecules. Any hydrogen bond that could be made by the WT residue to other monomers will be lost now and affect the multimeric contacts.

The M residue is smaller than the WT residue. The WT residue was +vely charged, the M residue is neutral. The charge of the WT residue is lost by this mutation. This can cause loss of interactions with other molecules. Any hydrogen bond that could be made by the WT residue to other monomers will be lost now and affect the multimeric contacts.

The M residue is bigger and less hydrophobic than the WT residue. The WT residue was neutral, the M residue is -vely charged. The mutation introduces a charge at this position; this can cause repulsion between the M residue and neighboring residues.
The M residue is smaller and more hydrophobic than the WT residue. The WT residue was positively charged, the M residue is neutral. The charge of the WT residue is lost by this mutation. The residue is buried in the core of a domain. The differences between the WT and M residue might disturb the core structure of this domain.

The M residue is more hydrophobic than the WT residue. The WT residue forms a hydrogen bond with Ser at position 90. The difference in hydrophobicity will affect hydrogen bond formation.

The WT residue was negatively charged, the M residue is neutral. The WT residue forms a hydrogen bond with Lys at position 8 and Asp at position 106. The WT residue forms a salt bridge with Lys at position 8 and Arg at position 16. The difference in charge will disturb the ionic interaction made by the original, WT residue.

The M residue is smaller and more hydrophobic than the WT residue. The WT residue was positively charged, the M residue is neutral. The mutation introduces a Gly at this position. Glys are very flexible and can disturb the required rigidity of the protein at this position. The WT residue forms a hydrogen bond with Arg at position 16, Phe at position 108 and Tyr at position 110. The difference in hydrophobicity will affect hydrogen bond formation. The WT residue forms a salt bridge with Lys at position 8 and Arg at position 16.
The M residue is smaller than the WT residue. The M residue is more hydrophobic than the WT residue. This will cause a possible loss of external interactions. The mutation will cause loss of hydrogen bonds in the core of the protein and as a result disturb correct folding.

The M residue is smaller than the WT residue. The mutation will cause an empty space in the core of the protein.

The M residue is smaller and more hydrophobic than the WT residue. The WT residue was -vely charged, the M residue is neutral. The WT residue forms a salt bridge with Lys at position 42. The difference in charge will disturb the ionic interaction made by the original, WT residue. The mutation converts the WT residue in a residue that does not prefer $\alpha$-helices as secondary structure.

The WT residue is a Gly, the most flexible of all residues. This flexibility might be necessary for the protein's function. Mutation of this Gly can abolish this function. The WT and M amino acids differ in size. The M residue is bigger than the WT residue. The residue is located on the surface of the protein; mutation of this residue can disturb interactions with other molecules or other parts of the protein. The torsion angles for this residue are unusual. Only Gly is flexible enough to make these torsion angles, mutation into another residue will force the local backbone into an incorrect conformation and will disturb the local structure.
Table S3. Evaluating the effect of the amino acid mutations in BECN1 by HOPE sever. The distinct pattern and possible consequences of native and Mutant amino acid on protein structure based on size, charge, and hydrophobicity have been shown.

| WT Amino Acid | Position | M Amino Acid | Characteristics and Possible Consequences |
|---------------|----------|--------------|------------------------------------------|
| Val           | 14       | Glu          | The M residue is bigger and less hydrophobic than the WT residue. The WT residue is predicted (by the PHDsec DAS-server) to be located in its preferred secondary structure, a β-strand. The M residue prefers to be in another secondary structure; therefore the local conformation will be slightly destabilized. The mutation introduces a charge; this can cause repulsion of ligands or other residues with the same charge. |
| Ala           | 44       | Pro          | The M residue is bigger than the WT residue. This might lead to bumps. |
| Arg           | 74       | Cys          | The mutation introduces a more hydrophobic residue at this position. This can result in loss of hydrogen bonds and/or disturb correct folding. The charge of the WT residue will be lost, this can cause loss of interactions with other molecules or residues. |
| Leu           | 112      | Arg          | In both the PDB-file and in the PISA-assembly, this residue was found to be involved in a multimer contact. The mutation introduces a bigger residue at this position, this can disturb the multimeric interactions. The mutation introduces a less hydrophobic residue. Sometimes, hydrophobicity is important for multimerisation and therefore this mutation could affect the multimer contacts. The residue is located on the surface of the protein; mutation of this residue can disturb interactions with other molecules or other parts of the protein. The mutation introduces a charge at this position, this can cause repulsion between the M residue and neighboring residues. |
The WT residue forms a hydrogen bond with: Val at position 14, Val at position 14. The size difference between WT and M residue makes that the new residue is not in the correct position to make the same hydrogen bond as the original WT residue did. The difference in hydrophobicity will affect hydrogen bond formation. In both the PDB-file and in the PISA-assembly, this residue was found to be involved in a multimer contact. The mutation introduces a bigger residue at this position, this can disturb the multimeric interactions. The mutation introduces a less hydrophobic residue. Sometimes, hydrophobicity is important for multimerisation and therefore this mutation could affect the multimer contacts.

In both the PDB-file and in the PISA-assembly, this residue was found to be involved in a multimer contact. The mutation introduces a smaller residue at this position. The new residue might be too small to make multimer contacts.

The WT residue forms a hydrogen bond with: Asp at position 124. The size difference between WT and M residue makes that the new residue is not in the correct position to make the same hydrogen bond as the original WT residue did. The difference in hydrophobicity will affect hydrogen bond formation. In both the PDB-file and in the PISA-assembly, this residue was found to be involved in a multimer contact. The mutation introduces a bigger residue at this position; this can disturb the multimeric interactions. A more hydrophobic residue is introduced here. Any hydrogen bond that could be made by the WT residue to other monomers will be lost now and affect the multimeric contacts.

The charge of the WT residue will be lost; this can cause loss of interactions with other molecules or residues. The mutation introduces a more hydrophobic residue at this position. This can result in loss of hydrogen bonds and/or disturb correct folding.
The WT residue is predicted (by the PHDsec DAS-server) to be located in an α-helix. The mutation converts the WT residue in a residue that does not prefer α-helices as secondary structure. The residue is located on the surface of the protein, mutation of this residue can disturb interactions with other molecules or other parts of the protein.

In the PDB file used for this analysis the mutated residue is involved in a multimer contact. The mutation introduces a smaller residue at this position. The new residue might be too small to make multimer contacts. A more hydrophobic residue is introduced here. Any hydrogen bond that could be made by the WT residue to other monomers will be lost now and affect the multimeric contacts.

The WT residue forms a salt bridge with: Glu at position 167. The difference in charge will disturb the ionic interaction made by the original, WT residue.
Leu 194

Pro

The WT residue is predicted (by the PHDsec DAS-server) to be located in an α-helix. Pro disrupts an α-helix when not located at one of the first 3 positions of that helix. In case of the mutation at hand, the helix will be disturbed and this can have severe effects on the structure of the protein.

Leu 198

Pro

The WT residue is predicted (by the PHDsec DAS-server) to be located in an α-helix. Pro disrupts an α-helix when not located at one of the first 3 positions of that helix. In case of the mutation at hand, the helix will be disturbed and this can have severe effects on the structure of the protein.

Arg 205

Cys

The WT residue forms a salt bridge with: Glu at position 202. The difference in charge will disturb the ionic interaction made by the original, WT residue. In both the PDB-file and in the PISA-assembly, this residue was found to be involved in a multimer contact. The mutation introduces a smaller residue at this position. The new residue might be too small to make multimer contacts. A more hydrophobic residue is introduced here. Any hydrogen bond that could be made by the WT residue to other monomers will be lost now and affect the multimeric contacts.
The WT residue is predicted (by the PHDsec DAS-server) to be located in an α-helix. Pro disrupts an α-helix when not located at one of the first 3 positions of that helix. In case of the mutation at hand, the helix will be disturbed and this can have severe effects on the structure of the protein.

According to the PISA-database, the mutated residue is involved in a multimer contact. The PISA-database contains protein assemblies that are highly likely to be biologically relevant. The mutation introduces a smaller residue at this position. The new residue might be too small to make multimer contacts. A more hydrophobic residue is introduced here. Any hydrogen bond that could be made by the WT residue to other monomers will be lost now and affect the multimeric contacts.

The WT residue forms a salt bridge with: Asp at position 262. The difference in charge will disturb the ionic interaction made by the original, WT residue. The charge of the WT residue is lost by this mutation. This can cause loss of interactions with other molecules.

The mutation introduces a more hydrophobic residue at this position. This can result in loss of hydrogen bonds and/or disturb correct folding.
The residue is buried in the core of a domain. The differences between the WT and M residue might disturb the core structure of this domain. The mutation will cause an empty space in the core of the protein.

The WT residue forms a hydrogen bond with: Glu at position 422, Val at position 298. The size difference between WT and M residue makes that the new residue is not in the correct position to make the same hydrogen bond as the original WT residue did. The WT residue forms a salt bridge with: Glu at position 422. The difference in charge will disturb the ionic interaction made by the original, WT residue.

The WT residue forms a hydrogen bond with: Glu at position 422, Val at position 298. The size difference between WT and M residue makes that the new residue is not in the correct position to make the same hydrogen bond as the original WT residue did. The difference in hydrophobicity will affect hydrogen bond formation. The WT residue forms a salt bridge with: Glu at position 422. The difference in charge will disturb the ionic interaction made by the original, WT residue.

The residue is buried in the core of a domain. The differences between the WT and M residue might disturb the core structure of this domain. The mutation will cause an empty space in the core of the protein.
The M residue is bigger than the WT residue. The WT residue was buried in the core of the protein. The M residue is bigger and probably will not fit.

The WT residue forms a hydrogen bond with: Arg at position 289, Arg at position 289. The size difference between WT and M residue makes that the new residue is not in the correct position to make the same hydrogen bond as the original WT residue did. The WT residue forms a salt bridge with: Arg at position 289. The difference in charge will disturb the ionic interaction made by the original, WT residue. The residue is located on the surface of the protein, mutation of this residue can disturb interactions with other molecules or other parts of the protein.

The mutation converts the WT residue in a residue that does not prefer \( \alpha \)-helices as secondary structure. The residue is buried in the core of a domain. The differences between the WT and M residue might disturb the core structure of this domain.

The M residue is smaller than the WT residue. This will cause a possible loss of external interactions.
The M residue is smaller than the WT residue. The M residue is more hydrophobic than the WT residue. This will cause a possible loss of external interactions.

The residue is located on the surface of the protein; mutation of this residue can disturb interactions with other molecules or other parts of the protein. The local conformation will be slightly destabilized.

The residue is buried in the core of a domain. The differences between the WT and M residue might disturb the core structure of this domain. The WT residue is a Pro. Pros are known to be very rigid and therefore induce a special backbone conformation which might be required at this position. The mutation can disturb this special conformation.

The M residue is smaller and more hydrophobic than the WT residue. The WT residue is predicted (by the PHDsec DAS-server) to be located in its preferred secondary structure, a β-strand. The M residue prefers to be in another secondary structure; therefore the local conformation will be slightly destabilized.
The M residue is smaller and more hydrophobic than the WT residue. The WT residue is predicted (by the PHDsec DAS-server) to be located in its preferred secondary structure, a β-strand. The M residue prefers to be in another secondary structure; therefore the local conformation will be slightly destabilized.

The WT residue was buried in the core of the protein. The M residue is bigger and probably will not fit. The hydrophobicity of the WT and M residue differs. The mutation will cause loss of hydrophobic interactions in the core of the protein.

The WT residue forms a hydrogen bond with Gly at position 355. The size difference between WT and M residue makes that the new residue is not in the correct position to make the same hydrogen bond as the original WT residue did. The difference in hydrophobicity will affect hydrogen bond formation. The mutation introduces a charge at this position, this can cause repulsion between the M residue and neighboring residues.

The M residue introduces a charge in a buried residue which can lead to protein folding problems. The WT residue was buried in the core of the protein. The M residue is bigger and probably will not fit.
The residue is buried in the core of a domain. The differences between the WT and M residue might disturb the core structure of this domain. The M residue is smaller and less hydrophobic than the WT residue.

The M residue is smaller and less hydrophobic than the WT residue. The residue is buried in the core of a domain. The differences between the WT and M residue might disturb the core structure of this domain. The mutation will cause an empty space in the core of the protein.

The WT residue is a Gly, the most flexible of all residues. This flexibility might be necessary for the protein's function. Mutation of this Gly can abolish this function. The torsion angles for this residue are unusual. Only Gly is flexible enough to make these torsion angles, mutation into another residue will force the local backbone into an incorrect conformation and will disturb the local structure.

The WT residue forms a hydrogen bond with: Ile at position 415. The size difference between WT and M residue makes that the new residue is not in the correct position to make the same hydrogen bond as the original WT residue did. The mutation will cause an empty space in the core of the protein.
The WT residue forms a hydrogen bond with Ile at position 415. The size difference between WT and M residue makes that the new residue is not in the correct position to make the same hydrogen bond as the original WT residue did. The mutation will cause an empty space in the core of the protein.

The M residue is smaller than the WT residue. The mutation will cause an empty space in the core of the protein. The residue is buried in the core of a domain. The differences between the WT and M residue might disturb the core structure of this domain.

**Table S4.** Evaluating the effect of the amino acid mutations in SCD1 by HOPE sever. The distinct pattern and possible consequences of native and Mutant amino acid on protein structure based on size, charge, and hydrophobicity have been shown.

| WT Amino Acid | Position | M Amino Acid | Characteristics and Possible Consequences |
|---------------|----------|--------------|------------------------------------------|
| Tyr           | 41       | Cys          | The M residue is smaller and more hydrophobic than the WT residue. This will cause a possible loss of external interactions. The mutation introduces a more hydrophobic residue at this position. This can result in loss of hydrogen bonds and/or disturb correct folding. |
The M residue is bigger and less hydrophobic than the WT residue. The WT residue was neutral; the M residue is positively charged. The WT residue was buried in the core of the protein. The M residue is bigger and probably will not fit. The WT residue is a Pro. Pros are known to be very rigid and therefore induce a special backbone conformation which might be required at this position. The mutation can disturb this special conformation. Hydrophobic interactions, either in the core of the protein or on the surface, will be lost.

The M residue is smaller and less hydrophobic than the WT residue. The WT residue was neutral, the M residue is negatively charged. The mutation introduces a charge at this position; this can cause repulsion between the M residue and neighboring residues.

The M residue is smaller and more hydrophobic than the WT residue. The WT residue was negatively charged, the M residue is neutral. The mutated residue is not in direct contact with a ligand; however, the mutation could affect the local stability which in turn could affect the ligand-contacts made by one of the neighboring residues. The mutation introduces a Gly at this position. Glys are very flexible and can disturb the required rigidity of the protein at this position.

The M residue is smaller and more hydrophobic than the WT residue. This will cause a possible loss of external interactions. The mutated residue is located in a domain that is important for the activity of the protein and in contact with another domain that is also important for the activity. The interaction between these domains could be disturbed by the mutation, which might affect the function of the protein.
The M residue is bigger and less hydrophobic than the WT residue. The WT residue was neutral; the M residue is +vely charged. The WT residue is a Gly, the most flexible of all residues. This flexibility might be necessary for the protein's function. Mutation of this Gly can abolish this function. The torsion angles for this residue are unusual. Only Gly is flexible enough to make these torsion angles, mutation into another residue will force the local backbone into an incorrect conformation and will disturb the local structure.

The M residue is bigger and more hydrophobic than the WT residue. The mutation will cause loss of hydrogen bonds in the core of the protein and as a result disturb correct folding. The WT residue was buried in the core of the protein. The M residue is bigger and probably will not fit.

The M residue is smaller than the WT residue. The mutated residue is located in a domain that is important for the activity of the protein and in contact with another domain that is also important for the activity. The interaction between these domains could be disturbed by the mutation, which might affect the function of the protein.

The M residue is bigger and less hydrophobic than the WT residue. The WT residue was neutral; the M residue is -vely charged. The WT residue is a Gly, the most flexible of all residues. This flexibility might be necessary for the protein's function. Mutation of this Gly can abolish this function. The WT residue was buried in the core of the protein. The M residue is bigger and probably will not fit.
The M residue is smaller and more hydrophobic than the WT residue. The WT residue was +vely charged; the M residue is neutral. The mutated residue is not in contact with a metal; however, one of the neighboring residues does make a metal-contact that might be affected by the mutation in its vicinity. The mutated residue is not in direct contact with a ligand; however, the mutation could affect the local stability which in turn could affect the ligand-contacts made by one of the neighboring residues. The WT residue forms a hydrogen bond with: Glu at position 295, Thr at position 321, and Asn at position 318. The WT residue forms a salt bridge with: Glu at position 295, Asp at position 307 and Asp at position 325. The mutation will cause loss of hydrogen bonds in the core of the protein and as a result disturb correct folding.

The M residue is smaller and more hydrophobic the WT residue. The size differences between the WT and M residue disturb the interaction with the metal-ion; "ZN". The WT residue forms a hydrogen bond with His at position 120, His at position 157, His at position 157 and His at position 301. The difference in hydrophobicity will affect hydrogen bond formation.

The M residue is smaller and more hydrophobic the WT residue. The WT residue was +vely charged; the M residue is neutral. The WT residue forms a hydrogen bond with Asp at position 204, Glu at position 208 and Asp at position 204. The difference in hydrophobicity will affect hydrogen bond formation. The WT residue forms a salt bridge with Asp at position 204 and Glu at position 208. The difference in charge will disturb the ionic interaction made by the original, WT residue.

The M residue is smaller the WT residue. The WT residue was +vely charged; the M residue is neutral. The WT residue forms a hydrogen bond with Asp at position 204, Glu at position 208 and Asp at position 204. The difference in hydrophobicity will affect hydrogen bond formation. The WT residue forms a salt bridge with Asp at position 204 and Glu at position 208. The difference in charge will disturb the ionic interaction made by the original, WT residue.
The M residue is bigger and more hydrophobic than the WT residue. The WT residue was +vely; the M residue is neutral. The residue is located on the surface of the protein; mutation of this residue can disturb interactions with other molecules or other parts of the protein.

The M residue is smaller than the WT residue. The WT residue was +vely charged; the M residue is neutral. The WT residue forms a salt bridge with Glu at position 208. The difference in charge will disturb the ionic interaction made by the original, WT residue.

The M residue is bigger and more hydrophobic than the WT residue. The WT residue was +vely; the M residue is neutral. The residue is located on the surface of the protein; mutation of this residue can disturb interactions with other molecules or other parts of the protein.

The M residue is smaller and less hydrophobic than the WT residue. The mutation might cause loss of hydrophobic interactions with other molecules on the surface of the protein.
The M residue is smaller and more hydrophobic than the WT residue. The WT residue forms a hydrogen bond with Glu at position 206. The mutation will cause loss of hydrogen bonds in the core of the protein and as a result disturb correct folding.

The M residue is smaller and more hydrophobic than the WT residue. The WT residue was positively charged; the M residue is neutral. The WT residue forms a salt bridge with Glu at position 70. The difference in charge will disturb the ionic interaction made by the original, WT residue. The difference in hydrophobicity will affect hydrogen bond formation.

The M residue is bigger and more hydrophobic than the WT residue. The WT residue forms a hydrogen bond with Ala at position 192. The residue is buried in the core of a domain. The differences between the WT and M residue might disturb the core structure of this domain. The mutation will cause loss of hydrogen bonds in the core of the protein and as a result disturb correct folding.

The M residue is smaller than the WT residue. This will cause a possible loss of external interactions.
The M residue is bigger than the WT residue. The WT residue was neutral; the M residue is +vely. The mutated residue is not in direct contact with a ligand; however, the mutation could affect the local stability which in turn could affect the ligand-contacts made by one of the neighboring residues. The WT residue forms a hydrogen bond with Val at position 150. The size difference between WT and M residue makes that the new residue is not in the correct position to make the same hydrogen bond as the original WT residue did. The WT residue was buried in the core of the protein. The M residue is bigger and probably will not fit.

The M residue is smaller and less hydrophobic than the WT residue. The WT residue forms a hydrogen bond with Gln at position 147. The difference in hydrophobicity will affect hydrogen bond formation.

The M residue is smaller and more hydrophobic than the WT residue. The WT residue forms a hydrogen bond with Gln at position 147. The difference in hydrophobicity will affect hydrogen bond formation.

The M residue is smaller and less hydrophobic than the WT residue. The mutation might cause loss of hydrophobic interactions with other molecules on the surface of the protein.
The M residue is bigger and more hydrophobic than the WT residue. The WT residue forms a hydrogen bond with Phe at position 227 and Thr at position 250. The mutation will cause loss of hydrogen bonds in the core of the protein and as a result disturb correct folding.

Thr

The M residue is smaller and more hydrophobic than the WT residue. The mutation introduces a Gly at this position. Glys are very flexible and can disturb the required rigidity of the protein at this position.

Trp

The M residue is smaller and more hydrophobic than the WT residue. The WT residue was neutral, the M residue is +vely charged. The mutation is located within a topological domain. The mutation might disrupt this topological domain.

Ala

The M residue is bigger and less hydrophobic than the WT residue. The WT residue was neutral, the M residue is -vely charged. The residue is buried in the core of a domain. The differences between the WT and M residue might disturb the core structure of this domain.
The M residue is bigger and less hydrophobic than the WT residue. The WT residue was neutral, the M residue is +vely charged. The WT residue is a Gly, the most flexible of all residues. This flexibility might be necessary for the protein's function. Mutation of this Gly can abolish this function. The residue is located on the surface of the protein; mutation of this residue can disturb interactions with other molecules or other parts of the protein. The torsion angles for this residue are unusual. Only Gly is flexible enough to make these torsion angles, mutation into another residue will force the local backbone into an incorrect conformation and will disturb the local structure.

The M residue is smaller and more hydrophobic than the WT residue. The WT residue is annotated to be located in its preferred secondary structure, a β-strand. The M residue prefers to be in another secondary structure; therefore the local conformation will be slightly destabilized.

The M residue is smaller and more hydrophobic than the WT residue.

The M residue is bigger and more hydrophobic than the WT residue. The WT residue was +vely charged, the M residue is neutral. The mutated residue is not in contact with a metal, however, one of the neighboring residues does make a metal-contact that might be affected by the mutation in its vicinity. The WT residue forms a salt bridge with Glu at position 165. The difference in charge will disturb the ionic interaction made by the original, WT residue. The residue is located on the surface of the protein; mutation of this residue can disturb interactions with other molecules or other parts of the protein.
The M residue is smaller than the WT residue. The WT residue is located in a region annotated to form an α-helix. Pro disrupts an α-helix when not located at one of the first 3 positions of that helix. In case of the mutation at hand, the helix will be disturbed and this can have severe effects on the structure of the protein.

The M residue is smaller than the WT residue. This will cause a possible loss of external interactions.

The M residue is smaller and less hydrophobic than the WT residue. The mutation introduces a Gly at this position. Glys are very flexible and can disturb the required rigidity of the protein at this position.

The M residue is bigger than the WT residue. The residue is located on the surface of the protein; mutation of this residue can disturb interactions with other molecules or other parts of the protein.
The M residue is smaller than the WT residue. The WT residue is located in a region annotated to form an α-helix. The mutation converts the WT residue into a residue that does not prefer α-helices as secondary structure.

The M residue is bigger than the WT residue. The WT residue is a Gly, the most flexible of all residues. This flexibility might be necessary for the protein’s function. Mutation of this Gly can abolish this function. The residue is located on the surface of the protein; mutation of this residue can disturb interactions with other molecules or other parts of the protein. The torsion angles for this residue are unusual. Only Gly is flexible enough to make these torsion angles; mutation into another residue will force the local backbone into an incorrect conformation and will disturb the local structure.

The M residue is bigger and more hydrophobic than the WT residue. The residue is buried in the core of a domain. The differences between the WT and M residue might disturb the core structure of this domain. The WT residue was buried in the core of the protein. The M residue is bigger and probably will not fit.

The M residue is smaller than the WT residue. The WT residue was +vely charged, the M residue is neutral. The WT residue forms a hydrogen bond with Asp at position 325. The size difference between WT and M residue makes that the new residue is not in the correct position to make the same hydrogen bond as the original WT residue did. The WT residue forms a salt bridge with Asp at position 325 and Asp at position 335. The charge of the WT residue is lost by this mutation. This can cause loss of interactions with other molecules.
The M residue is smaller and more hydrophobic than the WT residue. The WT residue was positively charged, the M residue is neutral. The WT residue forms a hydrogen bond with Asp at position 325. The size difference between WT and M residue makes that the new residue is not in the correct position to make the same hydrogen bond as the original WT residue did. The difference in hydrophobicity will affect hydrogen bond formation. The WT residue forms a salt bridge with Asp at position 325 and Asp at position 335. The difference in charge will disturb the ionic interaction made by the original, WT residue.