

**Protein Modeling C - Protein Modeling - Camas Invitational 2020 - Camas C-Invite - 12-12-2020**

The following is both the Written Exam and the Jmol Exploration for the Protein Modeling Event. The 1st half of the test is the Written Exam while the 2nd half is the Jmol Exploration.

**1. (1.00 pts)** Proteins are a \_\_\_\_\_ composed of \_\_\_\_\_.

- ☐ A) Polymer, Hydrocarbons
- ☒ B) Polymer, Amino Acids
- ☐ C) Lattice, Amino Acids
- ☐ D) Compound, Charged Particles
- ☐ E) Lattice, Nucleotides
- ☐ F) Polymer, Nitrogenous Bases

**2. (1.00 pts)** Which of the following Amino Acids are classified as polar?

- ☐ A) Phenylalanine
- ☐ B) Glycine
- ☒ C) Threonine
- ☐ D) Valine
- ☐ E) Isoleucine
- ☐ F) Proline

**3. (1.00 pts)** What is the charge of Glycine at pH 7

- ☐ A) -2
- ☐ B) -1
- ☒ C) 0
- ☐ D) 1
- ☐ E) 2

**4. (1.00 pts)** What is the charge of Isoleucine at pH 11?

- ☐ A) -2
- ☒ B) -1
- ☐ C) 0
- ☐ D) +1
- ☐ E) +2

**5. (1.00 pts)** What is the charge of Aspartic Acid at pH 11?

- ☒ A) -2
- ☐ B) -1
- ☐ C) 0
- ☐ D) +1
- ☐ E) +2

**6. (1.00 pts)** Which of the following amino acids do not have a chiral center?

- ☐ A) Proline
- ☐ B) Aspartic Acid
- ☐ C) Alanine
- ☒ D) Glycine
- ☐ E) Threonine
- ☐ F) Phenylalanine

**7. (1.00 pts)** The driving force behind Protein Secondary Structure is \_\_\_\_\_.

- ☐ A) Intermolecular Hydrogen Bonds
- ☒ B) Intramolecular Hydrogen Bonds
- ☐ C) Hydrophobic / Hydrophilic Interactions
- ☐ D) Electrostatic Forces

**8. (1.00 pts)** Which of the following Amino Acids is most likely to appear on the exterior surface of the protein?

- ☐ A) V
- ☒ B) K
- ☐ C) I
- ☐ D) P
- ☐ E) G
- ☐ F) F

**9. (1.00 pts)** Which of the following are methods to determine protein structure?

(Mark **ALL** correct answers)

- ☐ A) Flow Cytometry
- ☒ B) Nuclear Magnetic Resonance Spectroscopy
- ☐ C) Column Chromatography
- ☒ D) X-Ray Diffraction Crystallography
- ☐ E) Gel Electrophoresis
- ☐ F) Phase Contrast Microscopy

**10. (1.00 pts)** Which of the following Amino Acids are least likely to be found in an Alpha Helix

- ☐ A) Isoleucine

- ☒ B) Proline
- ☐ C) Valine
- ☐ D) Tryptophan

11. (1.00 pts) Which of the following mutations would cause the greatest impact to a protein's function?

- ☒ A) T ---> V at a key catalytic site
- ☐ B) D ---> E at a salt bridge
- ☐ C) P ---> Y in the interior of the protein
- ☐ D) S ---> T at a metal ion binding pocket
- ☐ E) I ---> L in the middle of an alpha helix
- ☐ F) A --> G on the exterior of the protein

12. (1.00 pts) What does CRISPR stand for

- ☐ A) Customizable Regulatory Inhibitor Strong Peptide Region
- ☐ B) Centered Region Interspersed Short Parasympathetic Repeats
- ☒ C) Clustered Regularly Interspaced Short Palindromic Repeats
- ☐ D) Collaborative Reversible Interwoven Semiregular Pathogenic Repeats
- ☐ E) Centered Reversible Identifiable Standard Peptide Repeats
- ☐ F) Clustered Regularly Interwoven Standard Palindromic Regions

13. (1.00 pts) CRISPR Cas9 was first discovered in which organism?

- ☐ A) *Pandion haliaetus*
- ☐ B) *Pasteurella multocida*
- ☐ C) *Escherichia coli*
- ☐ D) *Saccharomyces Cerevisiae*
- ☐ E) *Homo sapiens*
- ☒ F) *Streptococcus pyogenes*

14. (1.00 pts) The CRISPR Cas9 system, in its native host, is most analogous to which biological system.

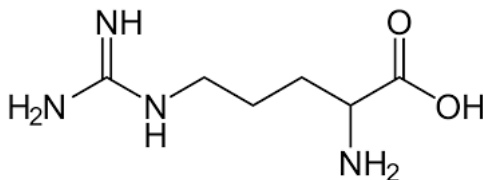
- ☒ A) Adaptive Immune System
- ☐ B) Innate Immune System
- ☐ C) Digestive System
- ☐ D) Respiratory System
- ☐ E) Circulatory System
- ☐ F) Endocrine System

15. (1.00 pts) Wild Type Cas9 is classified as what type of protein.

- ☐ A) Reductase

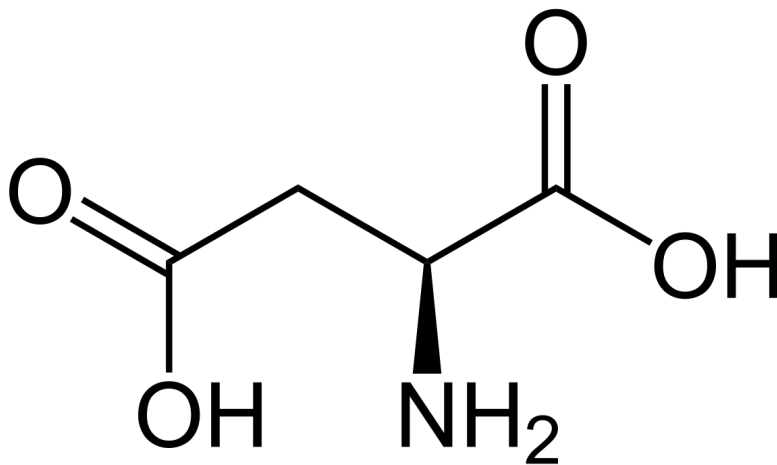
- ☐ B) Helicase
- ☐ C) Exonuclease
- ☒ D) Endonuclease
- ☐ E) Nickase
- ☐ F) Protease

16. (2.00 pts) Identify the following Amino Acid by Name (Spelling Matters), 3 Letter Abbreviation, and 1 Letter Abbreviation



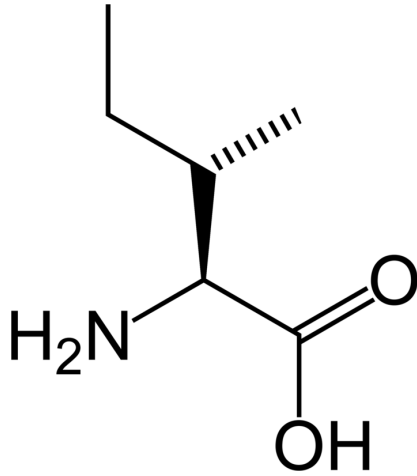
**Expected Answer:** Arginine, Arg, R

17. (2.00 pts) Identify the following Amino Acid by name (spelling matters), 3 Letter abbreviation, and 1 Letter abbreviation.



**Expected Answer:** Aspartic Acid, Asp, D

18. (2.00 pts) Identify the following Amino Acid by name (spelling matters), 3 Letter Abbreviation, and 1 Letter Abbreviation.



**Expected Answer:** Isoleucine, Ile, I

**19. (2.00 pts)** A PAM sequence is necessary for Cas9 to bind to DNA. What does PAM stand for and what is the sequence?

**Expected Answer:** Protospacer Adjacent Motif. The Sequence is NGG

**20. (2.00 pts)** The most common use of Cas9 is to knockout a particular gene through NHEJ. What does NHEJ stand for?

**Expected Answer:** Non Homologous End Joining

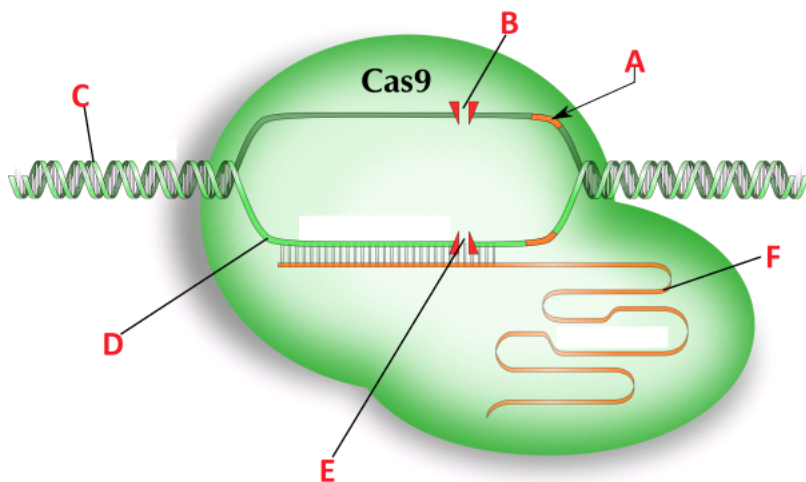
**21. (3.00 pts)**

The First Generation Base Editor, made by David Liu's Lab, is a protein conjugate of rat APOBEC1 - XTEN - dCas9. APOBEC1 is the Cytidine Deaminase and XTEN is a peptide linker. What is dCAS9 and how is it different than wild type CAS9?

**Expected Answer:** dCAS9 is catalytically inactivated Cas9. Both the nuclease domains have been mutated to prevent them from cutting DNA.

22. (6.00 pts)

The following is a diagram of Cas9

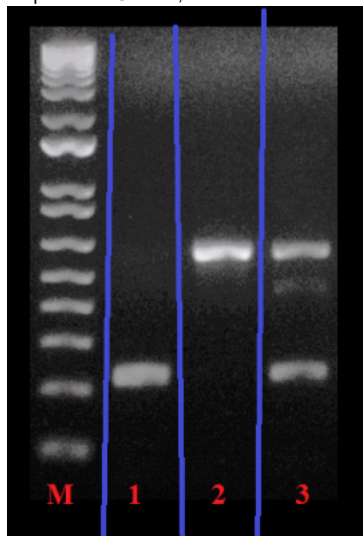


Provide a brief description for each of the six labeled sites. Make sure you indicate which label your description corresponds to. 1-3 words per description should suffice.

**Expected Answer:** A - PAM sequence - (5' NGG 3') Cas9 will bind if found, and match DNA after PAM ■ B - Cleavage site for non-target DNA, using RuvC domain ■ C - Host DNA (in viral DNA case, it binds with Cas9 of the bacteria) or Genomic DNA in gen editing ■ D - Target strand DNA - matching genomic sequence, complement to guide RNA ■ E - Cleavage site for target DNA - Cas9 using NHN domain for this cut ■ F - Guide RNA (accept tracrRNA and crRNA as long as function explained) - Guide Cas9 to specific sequence

23. (5.00 pts)

I treated a group of Human Stem Cells with Cas9 with guide RNA for Gene A in the hopes of knocking out Gene A. I then took individual cells from that group, extracted DNA, PCR amplified for Gene A, and then ran the results on an agarose gel. Here are the results. Gene A has a wild type length of 600bp while the knockout has a length of 200bp.



The Lane marked M is the ladder with each band corresponding to 100bp.

Lanes 1, 2, and 3 are DNA from three different cells.

Which cells were edited by Cas9?

Why does Lane 3 have two bands while Lane 1 and 2 only have 1 band?

**Expected Answer:** cells 1 and 3 were edited Lane 3 is heterozygous for the edit. Cas9 only knockout one copy of the gene.

This is the Jmol Exploration portion of the Protein Modeling Event. In this event you will answer question about a specific protein using information from the RCSB website and jmol. You may either use a desktop version of jmol or the browser based environment found here (<https://cbm.msoe.edu/markMyweb/jmolDesignEnvironment/#forward>). The PDB ID for our protein is: 3WDG. The link to the RCSB page is here (<https://www.rcsb.org/structure/3WDG>).

**24. (1.00 pts)** How many peptide chains are present in this File?

**Expected Answer:** 2

**25. (1.00 pts)** What is the name of Chain A?

**Expected Answer:** Uracil-DNA glycosylase

**26. (1.00 pts)** How many Amino Acids long is chain A?

**Expected Answer:** 216 (218 also accepted)

**27. (1.00 pts)** What method was used to identify this protein structure?

**Expected Answer:** X-Ray Diffraction

**28. (1.00 pts)** How Beta Strands are there is Chain B?

**Expected Answer:** 6

**29. (1.00 pts)** How many Cysteines are there in Chain A?

**Expected Answer:** 3

**30. (2.00 pts)** Do any of the Cysteines in Chain A form a disulfide bridge? If so, indicate which ones?

**Expected Answer:** They do at C97 and C214

**31. (1.00 pts)** How many Beta Strands are there in chain A?

**Expected Answer:** 6

**32. (1.00 pts)** How many Beta Sheets are present in Chain A?

**Expected Answer:** 2

**33. (1.00 pts)** Chain A's Active site has 1 amino acid of importance. What is the residue number of this amino acid and what is it's identity (Give the full name).

**Expected Answer:** #59 it is D (Aspartic Acid)



**34. (1.00 pts)** Is the N and C terminus of Chain A on the same face or on different faces of the protein?

**Expected Answer:** Same Face

**35. (1.00 pts)** What is identity of Amino Acid 15 on chain A? Give the full name (spelling matters).

**Expected Answer:** Aspartic Acid

**36. (3.00 pts)** What are the X, Y, and Z coordinates of the alpha carbon of Amino Acid 15 on chain A.

**Expected Answer:** 47.483 -24.644 63.766

**37. (5.00 pts)**

There is an instance in the sequence of Chain A where 6 amino acids are repeated in a row. What is this Amino Acid? Where is this sequence located? What is the purpose of this sequence?

**Expected Answer:** Histidine, C terminus, It is an expression / purification tag so that after expressing the protein in E. Coli you can isolate it for characterization