# Certified Histocompatibility Specialist Practice Test (Sample)

**Study Guide** 



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### **Questions**



- 1. What mechanism primarily contributes to HLA allele diversity?
  - A. Mutation and recombination
  - B. Genetic drift
  - C. Natural selection
  - D. Sample size stabilization
- 2. If a population is in Hardy-Weinberg Equilibrium, what can be inferred about a rare allele "y"?
  - A. "y" is seen in the population only because of a recent influx of newcomers
  - B. "y" will increase in frequency
  - C. "y" will decrease in frequency
  - D. "y" is usually seen in the heterozygous situation
- 3. What is required when reconstituting lyophilized antibodies for flow cytometry according to ASHI standards (2005)?
  - A. Reconstituted with sterile, azide-free PBS
  - B. Microaggregates must be removed
  - C. Mixing must occur for 20 seconds using a high-speed vortexer
  - D. Expiration date is only 3 months from reconstitution
- 4. What is a critical factor in understanding haplotype assignment in family studies?
  - A. The number of different alleles present
  - B. The physical distance between alleles on the chromosome
  - C. Previous knowledge of family member's genotypes
  - D. Patient's response to treatment
- 5. Type I water is defined as having a high purity due to its what?
  - A. Clarity
  - **B. Surface Tension**
  - C. Resistivity
- D. pH

- 6. What is the minimum blood volume needed for testing if 2,000 lymphocytes/well are required?
  - A. 20 ml
  - B. 10 ml
  - C. 5 ml
  - D. 2 ml
- 7. If 100g of NaCl is dissolved in 1500ml of water, what is the molarity?
  - A. 0.23M
  - B. 0.58M
  - C. 0.75M
  - D. 1.14M
- 8. Why do dead cells stained with AO/EB appear orange?
  - A. Ethidium bromide prevents AO binding
  - B. AO leaks out of the dead cell
  - C. Ethidium bromide fluoresces more strongly than AO
  - D. AO binds only RNA
- 9. According to ASHI standards, what should the viability of the negative control well in a serological typing assay be?
  - A. At least twice the viability of the positive control well
  - B. Greater than 50%
  - C. Greater than 80%
  - D. Greater than 90%
- 10. Which immunosuppressive drug is considered an antiproliferative (antimetabolite)?
  - A. Cyclosporine
  - B. FK506
  - C. Azathioprine
  - D. Prednisolone

#### **Answers**



- 1. A 2. D 3. B 4. C 5. C 6. D 7. D 8. C 9. C 10. C



### **Explanations**



## 1. What mechanism primarily contributes to HLA allele diversity?

- A. Mutation and recombination
- B. Genetic drift
- C. Natural selection
- D. Sample size stabilization

HLA allele diversity is primarily contributed by mechanisms such as mutation and recombination. This diversity is essential for the immune system's ability to recognize a wide range of pathogens. Mutations in the DNA sequence of HLA genes can create new alleles, while recombination between alleles—particularly during the process of meiosis—can generate variations that are beneficial for immune system functionality. This ongoing process of generating new genetic variations allows populations to adapt to changing environments and evolving pathogens, thus enhancing overall survival. The high mutation rates in the HLA region contribute significantly to the variability observed among individuals, which is critical for the adaptability of the immune system. Other mechanisms, while they may influence genetic diversity in general populations, do not play as direct a role in the specific context of HLA alleles. For instance, genetic drift pertains to changes in allele frequencies due to random sampling effects, which does not directly increase the diversity of HLA alleles in the same dynamic way that recombination and mutation do. Natural selection can influence which alleles are favored in a population but relies on the underlying variability that mutation and recombination generate. Sample size stabilization is more related to population dynamics than to the creation of diversity in HLA alleles.

- 2. If a population is in Hardy-Weinberg Equilibrium, what can be inferred about a rare allele "y"?
  - A. "y" is seen in the population only because of a recent influx of newcomers
  - B. "y" will increase in frequency
  - C. "y" will decrease in frequency
  - D. "y" is usually seen in the heterozygous situation

In a population that is in Hardy-Weinberg Equilibrium, the genetic variation is stable and allele frequencies remain constant over generations, provided that certain conditions are met (such as no mutation, random mating, no gene flow, infinite population size, and no selection). When discussing a rare allele like "y," it is important to note that it often exists in a higher proportion of heterozygotes compared to homozygotes because most individuals with a rare allele may not express it or may have it masked by a more dominant allele. In this case, if "y" is present, it is likely found in individuals who carry one copy of it alongside one copy of another more common allele. This results in the heterozygous condition being more prevalent among individuals carrying the rare allele, thus supporting the idea that "y" is usually seen in heterozygous situations. This understanding aligns with the concept that the heterozygous genotype can have certain advantages, several may carry rare alleles without expressing any detrimental traits associated with them. In summary, the frequent occurrence of the rare allele "y" in heterozygotes under Hardy-Weinberg conditions highlights the stability of allele frequencies and the role of heterozygosity in maintaining

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When reconstituting lyophilized antibodies for flow cytometry, one essential requirement according to the ASHI standards is the removal of microaggregates. This is critical because microaggregates can interfere with the flow cytometry results by affecting the consistency and accuracy of the analysis. These aggregates may form during the lyophilization process or during the reconstitution, and their presence can lead to clumping of particles that would distort the flow cytometry readings. Ensuring that the antibodies are free of these microaggregates helps maintain the integrity of the sample and provides reliable and reproducible results. The quality of the antibodies is paramount for accurate flow cytometry applications, and adhering to this standard aids in achieving optimal performance during experiments. Properly reconstituted antibodies contribute to the overall reliability of the assays conducted, reinforcing the importance of this particular step in the process.

- 4. What is a critical factor in understanding haplotype assignment in family studies?
  - A. The number of different alleles present
  - B. The physical distance between alleles on the chromosome
  - C. Previous knowledge of family member's genotypes
  - D. Patient's response to treatment

Understanding haplotype assignment in family studies is fundamentally tied to the genotypes of family members because having previous knowledge of the genotypes allows researchers to accurately determine which alleles are inherited together. In haplotype studies, alleles that are located close to one another on the same chromosome are often inherited together. Knowing the genotypes of family members helps in identifying these inherited blocks of alleles, allowing for accurate mapping of genetic traits or disease susceptibility within the family lineage. This information is crucial for establishing inheritance patterns and conducting linkage analysis, which can reveal how certain genetic traits are passed down through generations. By correlating the known genotypes of family members, a clearer picture of the haplotype structures can emerge, facilitating better understanding of genetic associations and their implications in health and disease.

- 5. Type I water is defined as having a high purity due to its what?
  - A. Clarity
  - **B. Surface Tension**
  - C. Resistivity
  - D. pH

Type I water is recognized for its high purity, primarily characterized by its resistivity. Resistivity is a measure of how strongly a substance opposes the flow of electric current, which is directly influenced by the presence of ions in the water. In the context of Type I water, a high resistivity indicates a low concentration of ionic contaminants, such as salts and minerals. This quality makes it particularly suitable for sensitive laboratory applications, especially in fields like biochemistry and molecular biology, where impurities can significantly affect experimental outcomes. In contrast, while clarity, surface tension, and pH are important characteristics of water, they do not directly define the purity level to the extent that resistivity does. Clarity pertains to the physical visibility or turbidity of water, surface tension is related to the cohesive forces within the water molecules, and pH measures its acidity or alkalinity. None of these attributes directly indicate the ionic purity essential for proper laboratory use, thereby reinforcing why resistivity is the defining feature of Type I water.

- 6. What is the minimum blood volume needed for testing if 2,000 lymphocytes/well are required?
  - A. 20 ml
  - B. 10 ml
  - C. 5 ml
  - D. 2 ml

To determine the minimum blood volume needed for testing if 2,000 lymphocytes per well are required, it is essential to understand the context of lymphocyte extraction from blood. Given that lymphocytes constitute about 20% of the total white blood cell (WBC) count, you need to consider the typical concentration of WBCs in human blood, which is around 4,000 to 10,000 cells per microliter (µL). For calculation purposes, let's assume an average WBC count of 7,000 cells/µL. This means that in every milliliter (mL) of blood, there are approximately 7 million WBCs. If we need to obtain 2,000 lymphocytes, we can calculate the required blood volume as follows: 1. Calculate the number of total WBCs needed based on the lymphocyte proportion: - Since lymphocytes are about 20% of WBCs, to get 2,000 lymphocytes, the total number of WBCs required will be: - 2,000 lymphocytes  $\div 0.20 = 10,000$  total WBCs. 2. Determine the volume of blood required to yield 10

#### 7. If 100g of NaCl is dissolved in 1500ml of water, what is the molarity?

- A. 0.23M
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- C. 0.75M
- D. 1.14M

To calculate the molarity of the solution, first, you need to determine the number of moles of NaCl dissolved in the water. Molarity is defined as the number of moles of solute per liter of solution. 1. Calculate the number of moles of NaCl: - The molar mass of sodium chloride (NaCl) is approximately 58.44 g/mol. - To find the number of moles in 100 grams, you can use the formula: \[\text{Moles of NaCl} = \frac{\text{mass}}  $\text{text} \{ g/mol \}$  \approx 1.71 \text{ moles} \] 2. Calculate the volume of water in liters: -The volume of water is given as 1500 mL, which is equivalent to 1.5 L. 3. Finally, calculate the molarity: - The formula for molarity (M) is:  $I = \frac{m}{m}$ 

#### 8. Why do dead cells stained with AO/EB appear orange?

- A. Ethidium bromide prevents AO binding
- B. AO leaks out of the dead cell
- C. Ethidium bromide fluoresces more strongly than AO
- D. AO binds only RNA

Dead cells stained with acridine orange (AO) and ethidium bromide (EB) appear orange due to the way ethidium bromide interacts with DNA when it's in an environment where the cell membrane integrity is compromised. In live cells, acridine orange can readily penetrate and bind to RNA and DNA, fluorescing green due to its interaction with RNA and still fluorescing green with double-stranded DNA. However, when the cell is dead, ethidium bromide enters the cell because of the compromised membrane integrity. Ethidium bromide intercalates more effectively with double-stranded DNA and fluoresces red when bound to it. When both dyes are present in a dead cell, the red fluorescence of ethidium bromide can dominate or mix with the green fluorescence of acridine orange, resulting in an orange appearance when viewed under a fluorescence microscope. This phenomenon occurs because the red fluorescence has a higher intensity compared to the green fluorescence of acridine orange in this scenario, leading to the overall orange color observed. In this context, some other choices might suggest alternative mechanisms that aren't correct. For instance, stating that ethidium bromide prevents AO binding misunderstands the principle of how these two dyes interact with nucleic acids under different cellular conditions. Similarly, the

- 9. According to ASHI standards, what should the viability of the negative control well in a serological typing assay be?
  - A. At least twice the viability of the positive control well
  - B. Greater than 50%
  - C. Greater than 80%
  - D. Greater than 90%

In serological typing assays, the viability of the negative control well is critical for ensuring the accuracy and validity of the test results. According to ASHI (American Society for Histocompatibility and Immunogenetics) standards, the viability of the negative control well should be greater than 80%. This threshold is set to confirm that the reactants in the assay are functioning properly and that any negative results observed are not due to issues with the assay itself, such as poor viability of the cells used in the test. A viability over 80% indicates that the cells are in good condition and capable of responding as expected, which helps validate the reliability of the assay. Lower viability could potentially lead to false-negative problems, where the actual presence of antibodies may go undetected due to compromised control conditions. Thus, achieving greater than 80% viability assures that the assay has been conducted under proper conditions, guaranteeing the credibility of the results.

- 10. Which immunosuppressive drug is considered an antiproliferative (antimetabolite)?
  - A. Cyclosporine
  - **B. FK506**
  - C. Azathioprine
  - D. Prednisolone

Azathioprine is classified as an antiproliferative drug, specifically an antimetabolite, which means it interferes with the synthesis of nucleic acids and thereby inhibits the proliferation of cells, particularly lymphocytes that are involved in the immune response. This mechanism makes azathioprine effective in preventing rejection of transplanted organs by suppressing the immune system's ability to mount a response against the transplanted tissue. Other options like cyclosporine and FK506 (tacrolimus) are calcineurin inhibitors, which primarily work by inhibiting T-cell activation and interleukin-2 transcription, but they do not fall into the category of antiproliferative agents. Prednisolone is a corticosteroid that has broad immunosuppressive effects, including anti-inflammatory actions, but it is not categorized specifically as an antimetabolite. Therefore, azathioprine is unique among the choices for its specific role as an antimetabolite in immunosuppressive therapy.