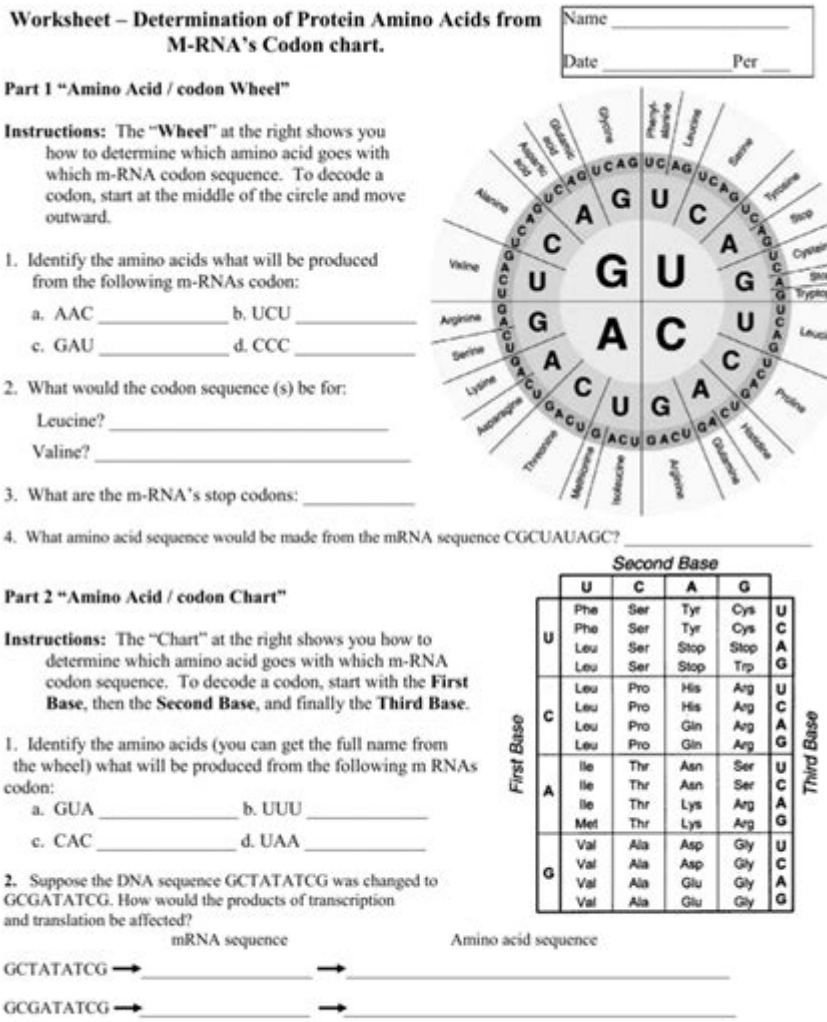
Worksheet Determination Of Protein Amino Acids



Worksheet – Determination of Protein Amino Acids from M-RNA's Codon chart.

Name _____

Date _____ Per ___

Part 1 "Amino Acid / codon Wheel"

Instructions: The "**Wheel**" at the right shows you how to determine which amino acid goes with which m-RNA codon sequence. To decode a codon, start at the middle of the circle and move outward.

1. Identify the amino acids what will be produced from the following m-RNAs codon:
   a. AAC _____ b. UCU _____
   c. GAU _____ d. CCC _____
2. What would the codon sequence (s) be for:
   Leucine? _____
   Valine? _____
3. What are the m-RNA's stop codons: _____
4. What amino acid sequence would be made from the mRNA sequence CGCUAUAGC? _____

Part 2 "Amino Acid / codon Chart"

Instructions: The "Chart" at the right shows you how to determine which amino acid goes with which m-RNA codon sequence. To decode a codon, start with the **First Base**, then the **Second Base**, and finally the **Third Base**.

1. Identify the amino acids (you can get the full name from the wheel) what will be produced from the following m RNAs codon:
   a. GUA _____ b. UUU _____
   c. CAC _____ d. UAA _____
2. Suppose the DNA sequence GCTATATCG was changed to GCGATATCG. How would the products of transcription and translation be affected?

mRNA sequence Amino acid sequence

GCTATATCG → _____ → _____

GCGATATCG → _____ → _____

First Base	Second Base U	Second Base C	Second Base A	Second Base G	Third Base
U	Phe	Ser	Tyr	Cys	U
	Phe	Ser	Tyr	Cys	C
	Leu	Ser	Stop	Stop	A
	Leu	Ser	Stop	Trp	G
C	Leu	Pro	His	Arg	U
	Leu	Pro	His	Arg	C
	Leu	Pro	Gln	Arg	A
	Leu	Pro	Gln	Arg	G
A	Ile	Thr	Asn	Ser	U
	Ile	Thr	Asn	Ser	C
	Ile	Thr	Lys	Arg	A
	Met	Thr	Lys	Arg	G
G	Val	Ala	Asp	Gly	U
	Val	Ala	Asp	Gly	C
	Val	Ala	Glu	Gly	A
	Val	Ala	Glu	Gly	G

Worksheet determination of protein amino acids is an essential aspect of biochemistry and molecular biology that enables researchers to understand the composition and function of proteins. Amino acids are the building blocks of proteins, and determining their specific types and quantities is crucial for studying protein structure and function. This article will explore the various methods and techniques used in the worksheet determination of protein amino acids, including sample preparation, analytical methods, and applications.

Understanding Amino Acids and Proteins

Amino acids are organic compounds that serve as the foundation for proteins. There are 20 standard

amino acids, each with a unique side chain that influences the protein's properties and function. Proteins are vital for numerous biological processes, including enzyme catalysis, cellular signaling, and structural support.

The Importance of Amino Acid Determination

Understanding the amino acid composition of proteins offers insights into:

1. Protein Function: Different amino acids contribute to the protein's functionality in biochemical pathways.
2. Genetic Information: The sequence of amino acids is determined by the genetic code, allowing for the study of gene expression and regulation.
3. Disease Research: Alterations in amino acid sequences can lead to misfolded proteins, which are implicated in diseases such as Alzheimer’s and certain cancers.
4. Nutritional Studies: Determining amino acid profiles in food sources helps assess nutritional value and dietary requirements.

Sample Preparation for Amino Acid Analysis

The first step in the worksheet determination of protein amino acids is the preparation of protein samples. Proper sample preparation is crucial for accurate results, and it typically involves:

1. Protein Extraction

- Cell Lysis: Breaking down cell membranes using physical or chemical methods (e.g., sonication, detergents).
- Precipitation: Isolating proteins from the lysate using ammonium sulfate or organic solvents.

2. Hydrolysis of Proteins

Proteins must be hydrolyzed into their constituent amino acids for analysis. This can be done using:

- Strong Acids: Hydrochloric acid (HCl) is commonly used, and the reaction typically occurs at elevated temperatures (110°C for 24 hours).
- Enzymatic Hydrolysis: Specific enzymes can be utilized to cleave peptide bonds selectively.

3. Neutralization and Derivatization

After hydrolysis, the sample’s pH must be adjusted to neutral levels, and derivatization may be needed to enhance detection:

- Derivatization Agents: Common agents include phenyl isothiocyanate (PITC) and 9-fluorenylmethoxycarbonyl chloride (FMOC).
- Purpose: This step improves amino acid volatility and solubility for chromatographic analysis.

Analytical Techniques for Amino Acid Determination

Various analytical methods are employed for the worksheet determination of protein amino acids. Each method has its advantages and limitations, making it imperative to choose the right technique based on the sample type and required sensitivity.

1. Chromatography Techniques

Chromatography is one of the most widely used techniques for amino acid analysis. The two main types include:

- High-Performance Liquid Chromatography (HPLC):
- Method: Separates amino acids based on their charge and hydrophobicity.
- Detection: UV-visible or fluorescence detectors are commonly used.
- Advantages: High resolution and sensitivity.

- Gas Chromatography (GC):
- Method: Involves the conversion of amino acids into volatile derivatives.
- Detection: Mass spectrometry (MS) is often coupled with GC for enhanced sensitivity.
- Limitations: Not suitable for all amino acids, especially those that are non-volatile.

2. Mass Spectrometry (MS)

Mass spectrometry is a powerful tool for amino acid analysis, capable of providing detailed molecular information:

- Method: Measures the mass-to-charge ratio of ions derived from amino acids.
- Applications: Useful for identifying and quantifying amino acids in complex mixtures.
- Advantage: High specificity and sensitivity, allowing for the detection of low-abundance amino acids.

3. Colorimetric Methods

Colorimetric assays can be employed for quick and simple amino acid determination:

- Ninhydrin Reaction: Amino acids react with ninhydrin to produce a colored complex, which can be quantified spectrophotometrically.
- Limitations: This method may not distinguish between different amino acids without further separation.

Data Analysis and Interpretation

Once the amino acids have been separated and detected, the next step involves data analysis. This includes:

1. Calibration and Quantification

- Calibration Curves: Constructing calibration curves using known amino acid standards to quantify the amounts in samples.
- Internal Standards: Using internal standards to correct for variability and improve accuracy.

2. Software and Statistical Analysis

- Software Tools: Various software programs are available for data interpretation, allowing for peak identification and quantification.
- Statistical Analysis: Employing statistical methods to assess the reliability and significance of results.

Applications of Amino Acid Determination

The worksheet determination of protein amino acids has numerous applications across various fields:

1. Biotechnology and Pharmaceuticals

- Drug Development: Understanding protein structure and function aids in designing effective therapeutics.
- Biopharmaceuticals: Quality control of recombinant proteins ensures efficacy and safety.

2. Nutritional Science

- Dietary Analysis: Assessing amino acid profiles in foods helps in formulating balanced diets.
- Supplement Development: Creating amino acid supplements tailored to specific health needs.

3. Clinical Diagnostics

- Disease Markers: Identifying abnormal amino acid levels can serve as biomarkers for certain diseases.
- Metabolic Disorders: Amino acid profiling assists in diagnosing metabolic conditions, such as phenylketonuria (PKU).

Conclusion

In conclusion, the worksheet determination of protein amino acids is a critical process that involves meticulous sample preparation and the application of advanced analytical techniques. From chromatography to mass spectrometry, each method offers unique benefits and challenges that must be considered in the context of the specific research objectives. The insights gained from amino acid analysis have profound implications in biotechnology, nutrition, and clinical diagnostics, underscoring the importance of continued research and innovation in this field. Understanding amino acids not only enhances our knowledge of protein biology but also paves the way for advancements in health and disease management.

Frequently Asked Questions

What is the purpose of a worksheet for determining protein amino acids?

The purpose of the worksheet is to systematically organize data and calculations related to the identification and quantification of amino acids in protein samples.

What techniques are commonly used to determine amino acids in proteins?

Common techniques include chromatography (such as HPLC), mass spectrometry, and colorimetric assays.

How can I prepare a sample for amino acid analysis?

Samples are typically hydrolyzed using acid or enzyme digestion, followed by filtration and dilution before analysis.

What information should be included in a worksheet for amino acid determination?

The worksheet should include sample identification, methods used, conditions of analysis, results, and any calculations performed.

What are the most common amino acids analyzed in protein studies?

Common amino acids include alanine, lysine, glutamic acid, and phenylalanine, among others.

Why is it important to measure the amino acid composition of proteins?

Measuring amino acid composition is crucial for understanding protein function, nutritional value, and quality in food and feed.

What role does pH play in the determination of amino acids?

pH can affect the ionization state of amino acids, influencing their retention time in chromatography and their detection in assays.

How do you interpret the results from an amino acid analysis worksheet?

Results are interpreted by comparing the detected amino acid concentrations against known standards or reference proteins to assess composition.

What are some common pitfalls in amino acid determination?

Common pitfalls include incomplete hydrolysis, contamination, and improper calibration of analytical instruments.

How can software tools assist in the analysis of amino acid data?

Software tools can automate data entry, perform complex calculations, generate visualizations, and facilitate statistical analysis of amino acid data.

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