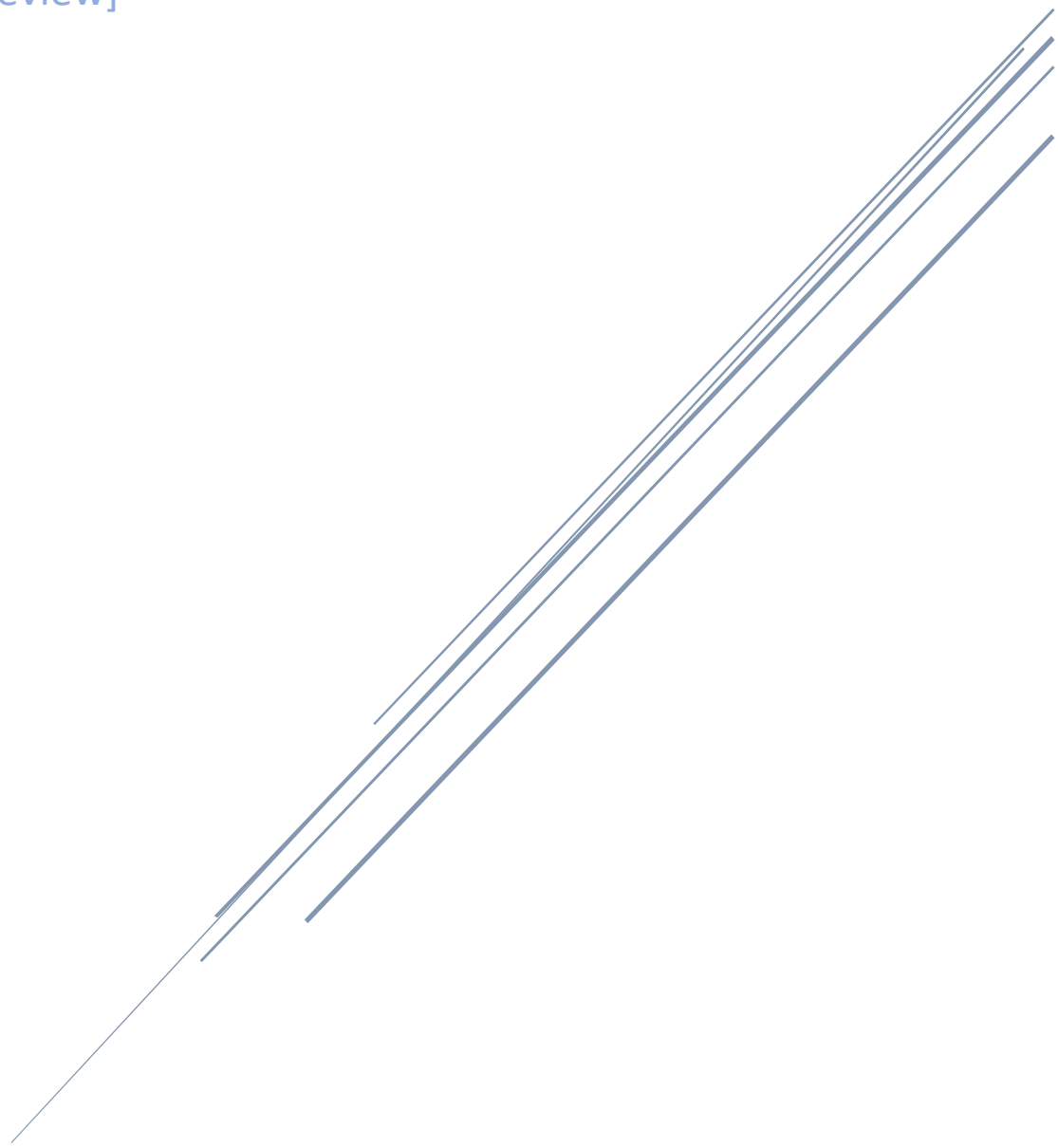
SYSTEMATIC CLASSIFICATION AND APPLIED RESEARCH OF PEPTIDES

[Review]



DilunBio Peptide Synthesizer
www.peptidescientific.com.

Systematic Classification and Applied Research of Peptides

Beijing Dilun Biotechnology Co., Ltd.

Specializes in R&D and manufacturing of peptide synthesis equipment, providing comprehensive technical solutions and services.

Web: www.peptidescientific.com Email: duxiaoru@dilunbio.com

Peptides are compounds formed by multiple amino acids linked via peptide bonds, with molecular weights intermediate between individual amino acids (monomers) and proteins (macromolecules). They serve as crucial functional intermediates bridging the two. Owing to their structural diversity, high biological activity, and design flexibility, peptides have become central subjects of research in modern biomedicine, materials science, precision medicine, cosmetics, and functional foods, among other fields. Given the vast differences in the types, numbers, sequences, and spatial configurations of amino acids constituting peptides, they exhibit an exceptionally rich variety of structures and properties. Systematic classification is essential for advancing peptide research and applications. This review summarizes common classification schemes and applications of peptides.

1.Classification Based on Amino Acid Composition:

1.1 Quantitative Classification:

Oligopeptides: Typically consist of 2–10 amino acids, also known as small peptides or low-molecular-weight peptides, with a molecular weight usually less than 1000 Da.

Polypeptides: Generally refer to chains consisting of 10 to approximately 50 amino acids. Polypeptides within this range often retain significant biological activity while offering greater design flexibility and a certain degree of metabolic stability compared to proteins.

Proteins: Usually refer to large molecular chains composed of more than 50 amino acids, characterized by complex higher-order structures and diverse functionalities.

1.2 Classification Based on Configuration:

Homomeric peptides: Composed of the same type of amino acids, such as peptides consisting entirely of L-amino acids.

Heteromeric peptides: Contain amino acids of different configurations (e.g., a mixture of L- and D-amino acids) or incorporate non-natural amino acids. Such peptides often exhibit unique biological activities and are of significant interest in drug development.

2.Classification Based on Molecular Structural Conformation

2.1 Linear Peptides:

The most common form of peptides, wherein amino acids are linked in a linear chain via peptide bonds, with free amino and carboxyl termini. The vast majority of natural and synthetic peptides fall into this category. Examples include glutathione (a tripeptide) and bradykinin (a nonapeptide), both of which exist in a linear form.

2.2 Cyclic Peptides:

These peptides form a closed-ring structure, most commonly via an amide bond between the N-terminus and C-terminus of the peptide backbone. Cyclization can also be achieved through covalent bonds between amino acid side chains (e.g., disulfide bond formation by cysteine residues) or between a side chain and a backbone terminus. This unique structural configuration confers enhanced metabolic stability (resistance to enzymatic degradation) and conformational rigidity, which facilitates the formation of specific spatial conformations and bioactive sites. These properties make cyclic peptides promising candidates for drug design.

Naturally occurring cyclic peptides include:

• Gramicidin S: A canonical backbone-cyclized decapeptide composed of two identical pentapeptide segments linked in a reverse orientation. It is a classic antimicrobial cyclic peptide produced by Bacillus brevisvia nonribosomal synthesis.

During its biosynthesis, the light enzyme activates and thioesterifies L-phenylalanine, which is subsequently converted to D-phenylalanine, while the heavy enzyme activates and thioesterifies proline, valine, ornithine, and leucine. The two pentapeptide segments are then cyclized through specific steps. Gramicidin S disrupts the integrity of bacterial cell membranes and exhibits potent activity against Gram-positive bacteria, holding significant importance in antimicrobial research.

• Cyclosporine A: A backbone-cyclized undecapeptide with complex structural modifications. It acts as a calcineurin inhibitor: upon entering cells, it forms a complex that binds to and inhibits calcineurin, thereby suppressing the transcription of cytokines such as IL-2, IFN-γ, and TNF-α and interfering with T-cell activation. As a representative immunosuppressant, it is widely used in organ transplantation to prevent rejection and in the treatment of certain immune-related disorders.

• Conotoxins: These are toxins derived from the venom glands of marine cone snails (Conusspecies), consisting of 7 to 41 or more amino acid residues. Many conotoxins exhibit di- or multicyclic structures, typically stabilized by two or more disulfide bonds, with C-terminal amidation also commonly contributing to structural stability. They specifically target various receptor subtypes of acetylcholine and other neurotransmitters, as well as sodium, potassium, and calcium channels, demonstrating high biological potency. For instance, some conotoxins produce potent analgesic effects by acting on specific receptors in the nervous system, making them highly valuable in neurobiological research and drug development.

• Microcystins: A class of characteristic monocyclic heptapeptides that often incorporate non-proteinogenic amino acids. The cyclic structure is crucial for their bioactivity, particularly hepatotoxicity. Produced primarily by cyanobacteria, microcystins can be released during algal blooms in eutrophic water bodies, posing significant risks to aquatic life and human health.

3.Classification Based on Origin and Production Method

3.1 Natural Peptides

A broad class of peptides widely present in nature, which are naturally synthesized by living organisms (including plants, animals, microorganisms, and humans) and serve as key bioactive molecules performing various physiological functions. Examples include: thyrotropin-releasing hormone (TRH, a tripeptide) secreted by the human hypothalamus; melittin (26 amino acids, with antimicrobial and hemolytic activity) from bee venom; classic peptide hormones such as oxytocin (a nonapeptide); insulin (composed of 51 amino acids, classified as a small protein hormone); neuroactive peptides including substance P (a neurotransmitter, 11 amino acids) and enkephalins (involved in pain relief, 5 amino acids); host defense peptides such as cecropin (35–37 amino acids, antibacterial) and defensins (approximately 30–50 amino acids, with both antimicrobial and immunomodulatory functions); as well as structurally and functionally diverse immunomodulatory peptides. These naturally occurring peptides play indispensable roles in numerous core biological processes—including growth and development, metabolism, neural signaling, immune response, defense mechanisms, and resistance to pathogenic infections.

3.2 Synthetic Peptides

Chemically Synthetic Peptides: Produced artificially via solid-phase peptide synthesis (SPPS) or liquid-phase peptide synthesis. A key advantage of this approach is the precise control over the amino acid sequence and length of the peptide chain, making it one of the primary methods in drug discovery and peptide research.

Recombinant Peptides: Specific peptides produced using genetic engineering techniques, expressed in microbial systems (e.g., E. coli, yeast) or cell cultures. This method is suitable for longer peptides, naturally occurring sequences, or peptides requiring specific modifications, such as recombinant insulin.

Enzymatically Hydrolyzed Peptides: Products obtained by hydrolyzing protein sources (e.g., soy, milk, or collagen) using proteases, resulting in a mixture of

low-molecular-weight peptides and amino acids with diverse sequences. These are widely used in functional foods, cosmetic ingredients, and nutritional supplements—examples include hydrolyzed whey peptide and collagen peptide.

4. Classification Based on Function and Biological Activity (A Core Classification)

4.1 Peptide Hormones

Peptide hormones are a class of signaling molecules synthesized and secreted by endocrine glands or specialized cells. As crucial regulators of the endocrine system, they are transported via the bloodstream to distant target cells. There, they bind to specific membrane receptors, initiate intracellular signal transduction, and precisely modulate a wide range of physiological processes—including growth, development, metabolism, reproduction, and the maintenance of homeostasis.

Representative members include:

• Ghrelin: Acts as an endogenous ligand for the growth hormone secretagogue receptor (GHSR), effectively stimulating pulsatile secretion of growth hormone from the anterior pituitary. This promotes linear bone growth (prior to epiphyseal closure) and systemic protein anabolism.

• Insulin: A classic protein hormone composed of 51 amino acids, secreted by pancreatic β-cells. It activates the insulin receptor (InsR)-mediated tyrosine kinase signaling pathway, promoting glucose uptake and utilization in peripheral tissues (such as skeletal muscle and adipose tissue), enhancing hepatic glycogen synthesis, and inhibiting gluconeogenesis. It is the only hormone in the body capable of significantly lowering blood glucose levels.

• Glucagon: Secreted by pancreatic α-cells, it acts as an insulin antagonist. It elevates blood glucose levels by stimulating hepatic glycogenolysis and gluconeogenesis in response to energy demand.

• Growth Hormone-Releasing Hormone (GHRH): Produced by hypothalamic neurons, it specifically activates the GHRH receptor (GHRHR) on pituitary somatotrophs, markedly promoting the synthesis and release of growth hormone.

4.2 Antimicrobial Peptides (AMPs)

Antimicrobial peptides are a class of small-molecule peptides (e.g., cationic antimicrobial peptides) with broad-spectrum antibacterial, antiviral, and even antifungal activities. They are widely distributed across living organisms and serve as key effector molecules of the innate immune system. The primary antimicrobial mechanism involves their amphipathic structure targeting the negatively charged microbial cell membranes, leading to the formation of transmembrane pores or disruption of lipid bilayer integrity. This causes ion imbalance and leakage of intracellular contents, ultimately resulting in pathogen death. These molecules exhibit strong inhibitory effects not only against Gram-positive and Gram-negative bacteria but also against fungi, enveloped viruses (such as HIV and HSV), and even tumor cells.

• Defensins: A family of cysteine-rich cationic peptides widely found in fungi, plants, and animals. In mammals, they are primarily expressed in epithelial tissues (e.g., skin and mucosa) and myeloid immune cells (e.g., neutrophils), where they play a crucial role in defending against pathogen invasion. Defensins possess direct bactericidal activity and represent an important group of antimicrobial peptides.

• Magainins: A family of cationic antimicrobial peptides first isolated in 1987 by Zasloff et al. from the skin of the African clawed frog (Xenopus laevis). They exhibit broad-spectrum antimicrobial activity. The family consists of two closely related peptides, each 23 amino acids in length, differing by only two amino acid substitutions. Magainins are effective against various microorganisms, including Gram-negative and Gram-positive bacteria.

• Cecropins: The first animal antimicrobial peptides discovered, identified in 1980 by

Boman et al. in the hemolymph of the Cecropia moth (Hyalophora cecropia). Typically composed of 34–39 amino acid residues, they can form nearly perfect amphipathic alpha-helical structures. Subsequently, cecropin-like peptides have been isolated from silkworms, hornworms, fruit flies, and flesh flies. Cecropins demonstrate antibacterial, antifungal, antiviral, and antitumor activities, as well as immunomodulatory functions. They exhibit high efficacy against multidrug-resistant bacterial strains.

4.3 Neuropeptides

Neuropeptides are peptide molecules that facilitate communication between neurons, serving as endogenous bioactive substances primarily found in the nervous system and involved in its functions. These molecules exhibit multiple signaling roles—functioning as neurotransmitters, neurohormones, neuromodulators, and cytokines—and participate in complex physiological regulatory networks underlying various disease processes. As a central focus in contemporary neuroscience research, neuropeptides are not only widely distributed throughout the central and peripheral nervous systems but are also present in multiple organs across the body, playing key regulatory roles throughout an organism's development.

Prominent examples include:

• Hypothalamic neuropeptides: Vasopressin (VP), Thyrotropin-Releasing Hormone (TRH), Corticotropin-Releasing Hormone (CRH), Oxytocin (OT);

• Brain-gut peptides: Substance P (SP), Vasoactive Intestinal Peptide (VIP), Glucagon, Insulin, Cholecystokinin (CCK);

• Other important members: Angiotensin II, Calcitonin Gene-Related Peptide (CGRP), Calcitonin, Neuropeptide Y (NPY), Atrial Natriuretic Peptide (ANP), Brain Natriuretic Peptide (BNP), and Galanin.

4.4 Immunomodulatory Peptides

Immunomodulatory peptides are a class of peptide molecules that precisely regulate the immune response by modulating immune cell activity and cytokine secretion. They can be derived from various sources, including organisms and dietary proteins. These peptides contribute to the homeostasis of both innate and adaptive immunity through a dual regulatory mechanism (immune activation/suppression), holding significant potential for applications in anti-tumor therapy, anti-infection strategies, and the treatment of autoimmune diseases. Their core mechanisms of action include immune activation, immune suppression, and regulation of cytokine networks. Naturally occurring immunomodulatory peptides, also known as host defense peptides, are peptide-like substances produced by organisms during defensive responses against exogenous pathogens or other foreign entities. Examples include:

• Human KAMP-19: A glycine-rich peptide discovered in the human eye;

• Microcin: Secreted by Enterobacteriaceae in the gut, involved in regulating intestinal immunity;

• Placental Immunomodulatory Peptides: Derived from animal placentas, approved as a novel veterinary drug—e.g., sheep placental transfer factor;

• Thymosin: Sourced from animal thymus tissue and approved by the U.S. FDA as an immunostimulant.

4.5 Other Functional Categories

Additionally, numerous peptides exhibit a wide range of other biological functions, such as:

• Cell-Penetrating Peptides (CPPs): Short oligo-/polypeptides (typically 5–30 amino acids) capable of efficiently crossing cellular membrane barriers. They facilitate intracellular delivery of biomacromolecules (e.g., nucleic acids, proteins) and small-molecule drugs via endocytosis or membrane disturbance mechanisms, serving as core carriers in novel drug delivery systems.

• Antioxidant Peptides: Peptides that scavenge free radicals (ROS/RNS), inhibit lipid peroxidation, and repair oxidative damage. Their activity stems from sequences rich in reductive amino acids such as histidine and tyrosine, showing potential applications in delaying aging and preventing metabolic diseases.

• Enzyme-Inhibitory Peptides: Peptides that reversibly or irreversibly inhibit target enzyme activity by specifically binding to active sites or allosteric sites. Based on their mechanisms, they are classified into:

• Competitive inhibitory peptides: angiotensin-converting enzyme inhibitory peptides;

• Allosteric regulatory peptides: e.g., cathepsin inhibitory peptides;

• Suicide substrate-type inhibitory peptides: e.g.,irreversible serine protease inhibitors.

•Carrier Peptides: Peptide scaffolds primarily designed to enhance the stability, solubility, or targeting of conjugated molecules (e.g., albumin-binding peptides, transferrin receptor-binding peptides). While often lacking strong intrinsic bioactivity, they are widely used in constructing multifunctional agents such as antibody-peptide conjugates (APCs) and peptide-drug conjugates (PDCs) through chemical conjugation or other strategies.

5. Peptide Modification and Engineering

Peptide modification strategies can be divided into two categories: biologically derived post-translational modifications (PTM) and engineered modifications.

PTM represents a fundamental biological mechanism that regulates peptide conformation, stability, and signal transduction. Post-translationally modified peptides (PTM-Peptides) are natural products of enzymatic modification within living organisms and are directly involved in signaling pathways. Common types include glycosylated peptides, phosphorylated peptides, acylated peptides, and disulfide-stabilized peptides.

Engineered peptides, on the other hand, are systematically optimized through chemical strategies—such as D-amino acid substitution, incorporation of non-canonical amino acids, PEGylation, and cyclization—to improve stability, bioavailability, and pharmacological properties (e.g., the long-acting GLP-1 analog liraglutide). In modern peptide drug development, artificial modification has become a core technological approach to overcome the limitations of natural peptides.

6. Conclusion

In summary, peptides can be classified through a variety of lenses—including molecular structural conformation, origin and production methods, functional and biological activity, amino acid composition, as well as modifications and engineering—highlighting their complexity and diversity. These classification frameworks not only enhance our understanding of peptide characteristics and functions but also provide a critical theoretical foundation for their research and application across numerous fields such as biomedicine and materials science. With continuous advances in science and technology, our knowledge of peptides will keep deepening, and new classification systems and research outcomes will continue to emerge, further driving innovation and development in peptide-related disciplines.