Classification Systems:

In the middle of the eighteenth century**,** Linnaeus developed the first and the most desirable system of classification called ***natural classification***. based largely on anatomical characteristics.

Natural classification: arranges organisms into groups whose members share many characteristics and reflects as much as possible the biological nature of organisms.

There are two general ways in which classification systems can be constructed. Organisms can be grouped together based on overall similarity to form a ***phenetic system*** or they can be grouped based on probable evolutionary relationships to produce a ***phylogenetic system***.

Phenetic Classification:

Phenetic Classification, one type of natural classification , that groups organisms together based on the mutual similarity of their phenotypic characteristics**.** Although phenetic studies can reveal possible evolutionary relationships, they are not dependent on phylogenetic analysis.

Phylogenetic Classification:

Following the publication in 1859 of Darwin's on the origin of species, biologists began trying to develop ***phylogenetic*** or ***phyletic classification* systems .**

These are systems based on evolutionary relationships rather than general resemblance (the term **phylogeny** [Greek *phylon,* tribe or race, and *genesis,* generation or origin] refers to the evolutionary development of a species).

This has proven difficult for prokaryotes and other microorganisms, primarily because of the lack of a good fossil record. The direct comparison of genetic material and gene products such as RNA and proteins overcomes many of these problems.

Numerical Taxonomy:

The development of computers has made possible the quantitative approach known as ***numerical taxonomy*.**

Peter H. A. Sneath and Robert Sokal have defined numerical taxonomy as: the grouping by numerical methods of taxonomic units into taxa on the basis of their character states. Information about the properties of organisms is converted into a form suitable for numerical analysis and then compared by means of a computer**.**

3- Identification

It is the practical use of classification criteria to distinguish certain organisms from others.

Identification simply involves the comparison of an 'unknown' object (e.g., a newly isolated bacterium) with all similar bacteria that are already known. If the 'unknown' bacteria matches up with a 'known' then the former has been identified; if not, it may be considered to be a 'new' species, variety, or strain and, when adequately described, is added to the list of known bacteria. In practice this act of comparison is normally carried out not between two actual objects but between the 'unknown' *isolate* and a recorded description of previously discovered micro-organisms.

Many characteristics are used in classifying and identifying microorganisms have been divided into two groups: *classical* and *molecular*.

1-Classical Characteristics:

Classical approaches to taxonomy make use of *morphological*, *physiological*, *biochemical*, *ecological*, and *genetic characteristics*. These characteristics have been employed in microbial taxonomy.

Table 1   Some Physiological and Metabolic Characteristics Used in Classification and Identification

Carbon and nitrogen sources
Cell wall constituents
Energy sources
Fermentation products
General nutritional type
Growth temperature optimum and range

2- Molecular Characteristics:

Some of the most powerful approaches to taxonomy are through the study of proteins and nucleic acids. Because these are either direct gene products or the genes themselves, comparisons of proteins and nucleic acids yield considerable information about true relatedness. The more recent molecular approaches have become increasingly important in prokaryotic taxonomy are:

1- Comparison of Proteins
2- Nucleic Acid Base Composition
3- Nucleic Acid Hybridization
4- Nucleic Acid Sequencing

1- Comparison of Proteins

The amino acid sequences of proteins are direct reflections of *mRNA* sequences and therefore closely related to the structures of the genes coding for their synthesis. For this reason, comparisons of proteins from different microorganisms are very useful taxonomically.

The sequences of proteins with dissimilar functions often change at different rates; some sequences change quite rapidly, whereas others are very stable. If the sequences of proteins with the same function are similar, the organisms possessing them are probably closely related. The sequences of *cytochromes* and other *electron transport proteins*, *histones*, *heat-shock proteins*, *transcription and translation proteins*, and a variety of *metabolic enzymes* have been used in taxonomic studies.

2- Nucleic Acid Base Composition

The **G + C** content of many microorganisms has been determined. G + C content data are taxonomically valuable in at least two ways:

First, they can confirm a taxonomic scheme developed using other data. If organisms in the same taxon are too dissimilar in G + C content, the taxon probably should be divided.

Second, G + C content appears to be useful in characterizing prokaryotic genera since the variation within a genus is usually less than 10% even though the content may vary greatly between genera.

For example :
Staphylococcus has a G + C content of 30 to 38%, whereas
Micrococcus DNA has 64 to 75% G + C; yet these two genera of
gram-positive cocci have many other features in common.

Table 2 : Representative G + C Contents of some bacteria

| Bacteria	Percent G + C
Bacillus	32–62
Clostridium	21–54
Escherichia	48–52
Neisseria	47–54
Nitrobacter	60–62

3- Nucleic Acid Hybridization

The similarity between genomes can be compared more directly by use of
nucleic acid hybridization studies. If a mixture of single stranded DNA
formed by heating dsDNA is cooled and held at a temperature about 25°C
below the *Tm*, strands with complementary base sequences will
reassociate to form stable dsDNA, whereas noncomplementary strands
will remain single.

In one of the more widely used hybridization techniques, nitrocellulose
filters with bound nonradioactive DNA strands are incubated at the
appropriate temperature with single-stranded DNA fragments made
radioactive with 32P or 14C. After radioactive fragments are allowed to
hybridize with the membrane-bound ss- DNA, the membrane is washed
to remove any nonhybridized ssDNA and its radioactivity is measured.
The quantity of radioactivity bound to the filter reflects the amount of
hybridization and thus the similarity of the DNA sequences.

Two strains whose DNAs show at least 70% relatedness under optimal
hybridization conditions and less than a 5% difference in *Tm* often are
considered members of the same species.

If DNA molecules are very different in sequence, they will not form a
stable, detectable hybrid. Therefore DNA-DNA hybridization is used to
study only closely related microorganisms. More distantly related
organisms are compared by carrying out DNA-RNA hybridization
experiments using radioactive ribosomal or transfer RNA.
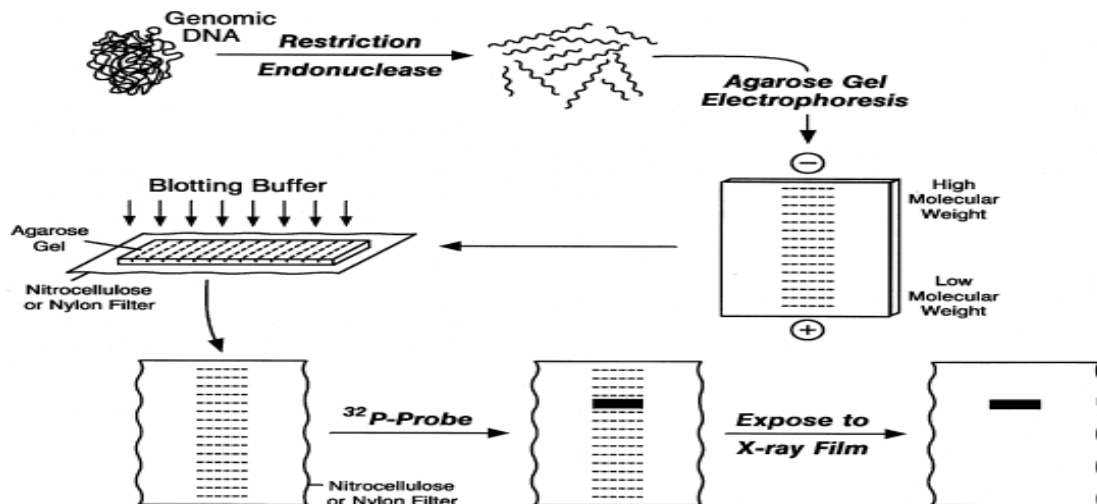
4- Nucleic Acid Sequencing

Genome structures can be directly compared only by sequencing DNA and RNA. Techniques for rapidly sequencing both DNA and RNA are now available, thus far RNA sequencing has been used more extensively in microbial taxonomy.

The rRNAs are almost ideal for studies of microbial evolution and relatedness since they are essential to a critical organelle found in all microorganisms. Most attention has been given to sequences of the 5S and 16S rRNAs isolated from the 50S and 30S subunits, respectively, of prokaryotic ribosomes.

Bergey's Manual of Systematic Bacteriology:

In 1923, **David Bergey**, professor of bacteriology at the University of Pennsylvania, and four colleagues published a classification of bacteria that could be used for identification of bacterial species, the *Bergey's Manual of Determinative Bacteriology* (The first edition), a more detailed work that contains descriptions of all prokaryotic species currently identified. This manual is now in its ninth edition.