**Criteria Useful in the Identification and/or Classification of Microorganisms**

1. **Morphology** – This is the study of external features (colonies and/or cells).

2. **Mode of reproduction** – Most bacteria undergo binary fission.

3. **Nutrition and Metabolism**
   **Mode of nutrition** is based on energy source and carbon source used.
   **Phototrophs** use light energy and **chemotrophs** use chemical energy.
   **Autotrophs** use inorganic carbon sources and **heterotrophs** use preformed organic compounds as their carbon sources.
   Four nutritional categories are:
   **Photoautotrophs** = Organisms that use light energy and inorganic carbon.
   **Photoheterotrophs** = Organisms that use light energy and organic carbon.
   **Chemoautotrophs** = Organisms that use chemical energy and inorganic carbon. The term lithotroph is also applied to this group.
   **Chemoheterotrophs** = Organisms that use chemical energy and organic carbon. The term chemoorganotrophs is sometimes applied to these.

   Subcategories under chemoheterotrophs include:
   **Saprotrophs =** Organisms that feed on dead or decaying materials.
   **Parasites =** Organisms that live inside other organisms and use them as nutrients**.**
   **Hypotrophs** = Organisms that grow and reproduce only when inside cells.
   Carnivores = Organisms that use meat as their source of nutrition.
   Herbivores = Organisms that use plant materials as their source of nutrition.
   Omnivores = Organisms that consume both meat and plant materials.

   **Microbial metabolism** is typically described as being either **respiratory** or **fermentative**, but many types of organisms are capable of both.
   **Respiratory organisms** typically use some type of inorganic compound (such as molecular oxygen) as their final electron acceptor.
   **Fermentative organisms** typically use some type of organic compound (such as pyruvic acid) as their final electron acceptor.

4. **Gas requirements**
   **Obligate aerobes** require oxygen for growth.
   **Obligate anaerobes** cannot tolerate exposure to oxygen.
   **Facultative anaerobes** grow well with or without oxygen available to them.
   **Microaerophils** grow best when oxygen levels are below atmospheric.
   **Be aware that obligate anaerobes are not necessarily fermentative as they may use inorganic compounds other than $O_2$ as electron acceptors.**

5. **Temperature Requirements**
   Organisms may be categorized on the basis of where they grow best as:
   **Psychrophiles** = Organisms that grow best in cold (-5 to 20° C) environments.
   **Mesophiles =** Organisms that grow best in moderate (20 to 45° C) environments.
   **Thermophiles =** Organisms that grow best in warm (45 to 60° C) environments.
   **Hyperthermophiles =** Organisms that grow best in hot (above 60° C) environments.
   Organisms can also be categorized on the basis of what temperatures they can endure or tolerate. These categories are:
   **Psychroduric** = Organisms that can endure or survive exposure to cold. Most bacteria are extremely psychroduric.
   **Thermoduric** = Organisms able to endure exposure to heat. Endospores are thermoduric.

6. **Acidity VS Alkalinity or pH Requirements**
   Most bacteria prefer neutral environments (pH close to 7.0), and **buffers** (substances that tend to resist pH change) are often added to media to keep pH levels near neutral. Substances called **pH indicators** change color in response to pH shifts, and are often added to media as a means of detecting the formation of acidic or alkaline end products.
   **Acidophiles** = Organisms that prefer acidic environments.

7. **Osmotic Pressure Requirements**
   The effective osmotic pressure or **tonicity** of an environment is influenced by the amount of solute present. Environments can be described as being:
   **Isotonic** = Environments with a solute concentration similar to protoplasm.
   **Hypotonic =** Environments with low solute content, e.g., water.
   **Hypertonic =** Environments with high solute content, e.g., sea water.

   Though hypertonic environments often inhibit microbial growth, organisms called **halophiles** grow best in high salt environments. Halophiles may be extreme/obligate or facultative depending on their salt requirements.

8. **Environmental Relationships**
   **Symbiosis** = A condition existing when two or more different types of organisms are living in a close association. Symbiotic relationships may be:
   **Pathogenic –** As exist when microorganisms infect host organisms and cause damage resulting in disease symptoms or sometimes death.
   **Parasitic –** As exist when parasites are living on or within a host and using it as nutrient.
   **Mutualistic –** As exist when both organism types forming the symbiotic relationship benefit or gain some advantage from it.

9. **Biochemical Analyses**
   Biochemical analyses often provide detailed information useful in characterizing organisms. Some sub-categories under this heading include:
   A. **Enzymatic testing** – A procedure used extensively during PUK1 identification.
   B. **Chromatography** – A method not commonly applied in our laboratory.
   C. **Serological testing** – Methods involving the use of **antibodies** (proteins made by animals) to identify or detect the presence of specific **antigens** (often bacteria, fungi, viruses, etc.), or vise-versa.
   D. **Phage typing** – A method involving the use of bacteriophages (viruses that infect bacteria) to identify specific types of bacteria.
   E. **Nucleic Acid Analysis** – Techniques used in the study of DNA and RNA include: percent base composition, nucleic acid hybridization, polymerase chain reaction (PCR), gel electrophoresis, DNA fingerprinting or RFLP and nucleotide sequencing.
   F. **Protein analysis –** Using gel electrophoresis and amino acid sequencing.

Once criteria have been set and characteristics determined, those obtained for different types of organisms can be compared using **dichotomous keys** or **cladograms**.

**Cladograms =** Branching tree-like patterns developed through cladistic analysis and indicating relationships between organisms.

**Cladistics =** A method for hypothesizing the evolutionary relationships among organisms and classifying them based on these relationships.

**Phylogenetic trees =** Complex cladograms indicating the evolutionary relationships between multiple different types of currently existing organisms and their ancestral forms.

**Phylogeny =** Evolutionary history.