

RESOLUTION OF RACEMIC MIXTURES

Racemic mixture contains equal amounts of optically active '*d*' and '*l*' isomers. The process of the separation of a racemic mixture into optically active *d* and *l* isomers is known as resolution.

The synthesis of organic compounds in the absence of a chiral reagent or chiral catalyst results in a mixture of both the enantiomers, i.e., a racemic mixture is obtained. Since *d* and *l*-isomers have identical melting points, solubilities, and all other physical properties except the direction in which they rotate the plane of polarized light, optical isomers cannot be separated by the usual methods such as distillation, crystallization, extraction etc.

Various methods used for separation of a racemic mixture are given below :

- a) **Mechanical separation** : The first racemic mixture was separated by Pasteur in 1848. He observed that when sodium and ammonium salt of racemic tartaric acid is allowed to crystallize slowly from water solution, the crystals obtained were of two shapes, which were mirror images of each other. Pasteur was able to separate these two types of crystals by sorting under microscope and found that while a solution of one type of crystals was dextro-rotatory, the solution of second type was laevo-rotatory.

Pasteur's mechanical separation is a difficult method as most often substances do not crystallize into readily distinguishable crystals and, therefore, the separation would be difficult. Also it cannot be applied to liquid substances.

- b) **Biochemical method** : This method was also discovered by Pasteur. He found that when certain organisms such as bacteria, fungi or yeasts are allowed to grow in a solution containing racemic compound, one of the enantiomer is selectively destroyed or selectively assimilated, so that an excess of one of the active forms gets accumulated in solution. For example when penicillium galucum is allowed to grow in racemic tartaric acid or racemic ammonium tartarate, the dextro form is selectively destroyed, leaving behind laevo-tartaric acid in the solution.

This method can be applied only to a few cases because one of the enantiomers is completely destroyed in separation.

- c) **Chemical method**. This is one of the most important methods developed by Pasteur and finds greater utility than the other two methods. This method is based on the fact that when a racemic acid, for example, is allowed to react with an optically active base, two diastereoisomers are produced. Since diastereoisomers have different physical properties, they can be separated by physical methods.

If a racemic acid is allowed to react with an optically active base, two salts will be formed.



Pair of Diastereomers

These two salts are diastereoisomers but not enantiomers and hence exhibit different physical properties. The mixture of two diastereoisomes can be separated by fractional crystallisation. The salts separated can be converted into *d*-acid and *l*-acid by treating with some acid.

In a similar manner, racemic organic base can also be resolved by treating it with some optically active acid as shown below :



Pair of Diastereomers

A large number of alkaloids which are found in nature are optically active bases and can be employed for separation of racemic acids. Some of the commonly used alkaloids for resolution of dl-acids are : *l*-brucine, *l*-cinchonine and *l*-quinine.

- d) **Chromatographic method.** When a racemic mixture solution is passed through an optically active adsorbent, the two enantiomers can be separated. As the adsorbates, produced by the combination of enantiomers and adsorbents, are diastereomers and as they are not equally stable, one of them will pass through the column faster than other. The two enantiomers are then obtained from the two diastereomers by careful elutions.