

Microbiological Examination of Nonsterile Products: Microbial Enumeration Tests and (62) *Microbiological Examination of Nonsterile Products: Tests for Specified Microorganisms* are typically appropriate for use.

(611) ALCOHOL DETERMINATION

PROCEDURES

Method I—Distillation Method

Method I is to be used for the determination of alcohol, unless otherwise specified in the individual monograph. It is suitable for examining most fluid extracts and tinctures, provided the capacity of the distilling flask is sufficient (commonly two to four times the volume of the liquid to be heated) and the rate of distillation is such that clear distillates are produced.

Cloudy distillates may be clarified by agitation with talc, or with calcium carbonate, and filtered, after which the temperature of the filtrate is adjusted and the alcohol content determined from the specific gravity. During all manipulations, take precautions to minimize the loss of alcohol by evaporation.

Treat liquids that froth to a troublesome extent during distillation by rendering them strongly acidic with phosphoric, sulfuric, or tannic acid, or treat with a slight excess of calcium chloride solution or with a small amount of paraffin or silicone oil before starting the distillation.

Prevent bumping during distillation by adding porous chips of insoluble material such as silicon carbide, or beads.

For liquids presumed to contain 30% of alcohol or less: By means of a pipet, transfer to a suitable distilling apparatus not less than 25 mL of the liquid in which the alcohol is to be determined, and note the temperature at which the volume was measured. Add an equal volume of water, distill, and collect a volume of distillate about 2 mL less than the volume taken of the original test liquid, adjust to the temperature at which the original test liquid was measured, add sufficient water to measure exactly the original volume of the test liquid, and mix. The distillate is clear or not more than slightly cloudy, and does not contain more than traces of volatile substances other than alcohol and water. Determine the specific gravity of the liquid at 25°, as directed under *Specific Gravity* (841), using this result to ascertain the percentage, by volume, of C₂H₅OH contained in the liquid examined by reference to the *Alcoholometric Table* in the section *Reference Tables*.

For liquids presumed to contain more than 30% of alcohol: Proceed as directed in the foregoing paragraph, except to do the following: dilute the specimen with about twice its volume of water, collect a volume of distillate about 2 mL less than twice the volume of the original test liquid, bring to the temperature at which the original liquid was measured, add sufficient water to measure exactly twice the original volume of the test liquid, mix, and determine its specific gravity. The proportion of C₂H₅OH, by volume, in this distillate, as ascertained from its specific gravity, equals one-half that in the liquid examined.

Special Treatment

Volatile acids and bases: Render preparations containing volatile bases slightly acidic with diluted sulfuric acid before distilling. If volatile acids are present, render the preparation slightly alkaline with sodium hydroxide TS.

Glycerin: To liquids that contain glycerin add sufficient water so that the residue, after distillation, contains not less than 50% of water.

Iodine: Treat all solutions containing free iodine with powdered zinc before the distillation, or decolorize with just sufficient sodium thiosulfate solution (1 in 10), followed by a few drops of sodium hydroxide TS.

Other volatile substances: Spirits, elixirs, tinctures, and similar preparations that contain appreciable proportions of volatile materials other than alcohol and water, such as volatile oils, chloroform, ether, camphor, etc., require special treatment, as follows:

For liquids presumed to contain 50% of alcohol or less: Mix 25 mL of the specimen under examination, accurately measured, with about an equal volume of water in a separator. Saturate this mixture with sodium chloride, then add 25 mL of solvent hexane, and shake the mixture to extract the interfering volatile ingredients. Draw off the separated, lower layer into a second separator, and repeat the extraction twice with two further 25-mL portions of solvent hexane. Extract the combined solvent hexane solutions with three 10-mL portions of a saturated solution of sodium chloride. Combine the saline solutions, and distill in the usual manner, collecting a volume of distillate having a simple ratio to the volume of the original specimen.

For liquids presumed to contain more than 50% of alcohol: Adjust the specimen under examination to a concentration of approximately 25% of alcohol by diluting it with water, then proceed as directed in *For liquids presumed to contain 50% of alcohol or less*, beginning with "Saturate this mixture with sodium chloride."

In preparing *Collodion* or *Flexible Collodion* for distillation, use water in place of the saturated solution of sodium chloride directed above.

If volatile oils are present in small proportions only, and a cloudy distillate is obtained, the solvent hexane treatment not having been employed, the distillate may be clarified and rendered suitable for the specific gravity determination by shaking it with about one-fifth its volume of solvent hexane, or by filtering it through a thin layer of talc.

Method II—Gas Chromatographic Method

Use *Method IIa* when *Method II* is specified in the individual monograph. For a discussion of the principles upon which it is based, see *Gas Chromatography* under *Chromatography* (621).