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Sterilization of ninhydrin

Ninhydrin is used in large quantities for amino acid analyzers. Although it is stable as a solid, solutions of it become unusable after about a month at room temperature. We are constructing a miniature SPACKMAN-MOORE-STEIN amino acid analyzer for use on the surface of Mars, and are therefore concerned with the stability of ninhydrin under the sterilization conditions for the Viking Mars mission. This involves five periods of heating to 135° for 40 h followed by three periods of heating to 135° for 60 h[1].

Heating ninhydrin dissolved in the standard methyl cellosolve buffer solution at 135° in an evacuated sealed tube causes the solution to turn wine red and form a precipitate. After filtration, the solution formed additional precipitate which clogged the lines of the analyzer when mixed with the effluent from the column. Heating solid ninhydrin to 135° for several hours results in a wine-red liquid. The m.p. of ninhydrin[2] is 241° (dec.). Apparently the ninhydrin is partially dehydrated, and a concentrated aqueous solution is formed. After completing the sterilization cycle, this ninhydrin was dissolved in the methyl cellosolve buffer and a white precipitate was filtered off. This solution gave nearly the same color yields as regular ninhydrin at 570 nm, but there was a high background at 440 nm.

Reduced ninhydrin (hydrindantin dihydrate) did not melt in a sealed evacuated tube under the sterilization conditions, but it turned brown and would not dissolve in the methyl cellosolve buffer.

When anhydrous ninhydrin in a sealed evacuated tube was put through the sterilization cycle, it did not discolor nor form a precipitate on solution. The color yields were the same as the standard ninhydrin preparation. The base lines at 570 and 440 nm were normal. These results show that anhydrous ninhydrin can be used as a reagent in an analyzer for detecting amino acids in Mars soil samples.

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