6505-890-2010 (P. D. No. 3)

4.4 Tests.

4.4.1 Promethazine hydrochloride assay in finished preparation. Use low actinic glassware throughout test.

Transfer an accurately measured sample of 25 cc to a 250-ml separator. Add 5 cc of purified water and 15 cc of 28 percent ammonium hydroxide. Completely extract the promethazine base with 40 cc portions of chloroform (about 6 extractions usually required), collecting the extractions in a second separator. Extract the ipecac with a 20 cc portion of 10 percent hydrochloric acid, followed by five (5) 15 cc portions of 1 percent hydrochloric acid. Wash the combined acid extracts with 25 cc of chloroform and add the chloroform washings to the main chloroform extract. (Discard the aqueous washings). Filter the combined chloroform extracts through a pledget of cotton previously wetted with chloroform, into a beaker and wash the separatory funnel and pledget of cotton with several small portions of chloroform. Evaporate the combined chloroform extracts on a water bath with the aid of a current of air to a volume of about 5-10 cc. Discontinue heating and continue evaporation with the aid of a current of air, to dryness. Add 50 cc of purified water and warm on a steam bath to dissolve the residue. Transfer the aqueous solution to a 500 cc volumetric flask with the aid of warm purified water. Cool to room temperature and dilute to the mark with purified water. Mix thoroughly, filter through a medium-porosity sintered glass filter rejecting the first few cc portion of filtrate, and collect the subsequent filtrate in a dry glass stoppered flask. (Reserve a portion of this filtrate for the preparation of the solution (for the identification test.). Determine the absorbancy of the clear filtrate, and a solution of Promethazine Hydrochloride U.S.P. Reference Standard, diluted to the same concentration as that of the sample, at 298 mu in a Beckman DU spectrophotometer, using a 1-cm quartz cell, with purified water as the reference liquid. Calculate percent of required amount of promethazine hydrochloride.

4.4.2 Identification.

a. Transfer exactly 10 cc of the clear filtrate obtained in 4.4.1 to a 100-cc volumetric flask. Dilute to the mark with purified water and mix thoroughly. Determine the absorbance at 249 mu with a Beckman DU spectrophotometer, using a 1 cm, quartz cell and purified water as the reference liquid.

Calculation: $\frac{10 \times A249}{A298}$ = ratio of absorbancies

b. Transfer 50 cc of sample to a 250 cc separatory funnel, add 50 cc of chloroform and 10 cc of anhydrous methanol. Stopper and shake vigorously for 5 minutes. Allow the two phases to separate and transfer the bottom layer to a 200 cc beaker. Evaporate to dryness on a steam bath with the aid of a current of air. Add 5 cc of 3.5 percent anhydrous methanol picric acid solution and heat on a steam bath for 5 minutes. Cool in an ice bath until yellow crystals appear.