## Attempts at Measuring Water Contents in the Brain Tissue with <sup>3</sup>H<sub>2</sub>O

-with a special reference to sample procedures in liquid scintillation counting-

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In measuring the increment rate of water contents in brain tissues by 3H<sub>2</sub>O-liquid scintillation counter we attempted to compare with those reported by various investigators who determine the water content by weighing the fresh and dried samples. We studied several sample procedures for the liquid scintillation counter, and carried out a comparative study of water contents in edematous and normal brains by using those sample procedures. As for the sample procedures, approximately 50 mg of brain tissue is first prepared, put into 1 ml-disposable syringe, and eluted in a centrifuge tube containing 2.5 ml methyl alcohol. Then it is subjected to supersonication (with supersonication bacteria homogenizer T-A-4201) for 3 minutes to prepare brain tissue homogenate. The homogenate thus obtaind is then mixed in a vial with 15 ml scintillator previously prepared and radioactive counts are taken.

Following this, using various scinitillators such as 1) one liter of toluene +5g ppo +300mg popop (simple scintillator); 2) simple scintillator+ Triton $\times 100$  (2V/IV); 3) simple scintillator+ Cab-O-Sil (3 $\sim 5\%$ ); homogenates are again homogenized in Soluen 100 and eluate was sub-

jected to radioactivity counting with simple scintillator 4), simple sintillator+Triton  $\times 100$  5), and simple scintillator+Cab-O-Sil 6), to determin the efficiency, figures of merit (F.M.) and reproducibility of each scintillator. As a result the most suitable scintillator is found to be the suspension method using Cab-O-Sil.

After these preliminary experiments we entered into actual comparative observation on the increment rate of water contents of the brain tissues in edematous brain. For the animals we used rats and prepared cold induced edema in the right parietal region. Immediately,  $50\mu a/0.3$  ml of  $^3H_2O$  and 0.3 ml of 2.5% Evans blue were injected into the femoral vein of animal.

As a result it has been demonstrated that counts on the edematous side is increased by about 5% as compared to those on the contralateral side.

These findings seem to indicate that in measuring the increment rate of water contents in brain tissues the most suitable method will be to homogenize the brain tissue by supersonication and use the suspension method using Cab-O-Sil as the scintillator for taking counts of radioactivity.