was measured at 480 nm with the excitation light at 356 nm using an Aminco-Bowman spectrophotofluorometer[9]. The DOPA formed enzymatically was calculated from the value of internal standard by the following equation. $[F(L) - F(D)]/[F(D + IS) - F(D)] \times 10$ mμmoles, where F(L) = reading of L-tyrosine incubation, F(D) = reading of D-tyrosine incubation, and F(D + IS) = reading of D-tyrosine plus DOPA (internal standard, 10 mμmoles) incubation.

In this procedure, DOPA is isolated specifically. The blank value was less than 1 mμmole of DOPA, when homogenate containing 33 mg (wet weight) of adrenal glands or caudate nucleus was used. Overall recovery of internal standard DOPA was 40 ± 1 (S.D.) % (n = 10), and constant. Limit of the sensitivity was about 1 mμmole DOPA formed enzymatically.

It was found in later experiments that the first Florisil column could be omitted. In this case, the reaction was stopped with 50 μl of 50% trichloroacetic acid. The incubation mixture was centrifuged. The precipitate was washed with 1 ml of water and recentrifuged. The combined supernatant was passed through an Amberlite CG-120-Na^+ (Type I, 0.6 × 4.0 cm) column. The column was washed with 5 ml of water. Subsequent procedures were the same as described above. This method gave higher blank value, but the recovery of DOPA was 60%, which was reproducible.

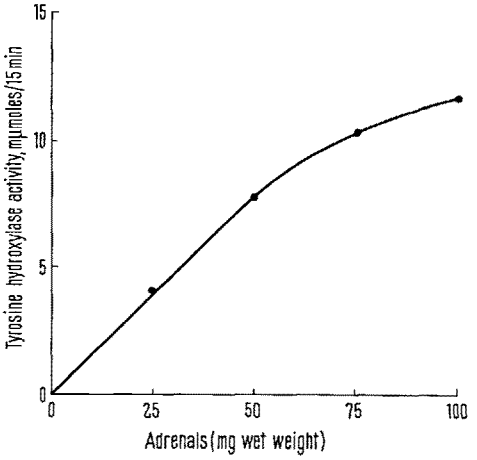
As shown in the Figure, the reaction rate measured by using sucrose homogenate was linear up to 50 mg of rabbit adrenals. Tyrosine hydroxylase activity in adrenal glands and brain were shown in the Table. This fluorescence assay could easily be applied to the measurement of the activity of purified adrenal tyrosine hydroxylase. In one experiment, 12.2 mμmoles DOPA were found by the fluorometry, and 12.3 mμmoles by the radioassay[1] in which DOPA-C^{14} was measured from L-tyrosine-C^{14}. This showed that the appearance of DOPA with this fluorometric procedure is essentially the same as calculated from the radioassay.

Although fluorescence assay is less sensitive than radioassay, the enzyme activity in homogenate of such tissues as adrenal glands or brain can be measured exactly. Fluorescence assay has some advantages. Besides the convenience that a labelled substrate and a liquid scintillation spectrometer are dispensable, separate measurement of tyrosine concentration in the homogenate is not necessary for the calculation[10].

Tyrosine hydroxylase activity in homogenates of rabbit organs measured by the fluorescence assay

Organs	Activity (mean ± S.D.) mμmoles/g tissue per h
Adrenal gland (6)	431 ± 21
Brain	
Caudate nucleus (5)	284 ± 97
Brain stem (1)	68

No. of organs in brackets.



Tyrosine hydroxylase activity as a function of enzyme amount. Sucrose homogenate of rabbit adrenal glands was used as enzyme. Incubation was for 15 min at 30 °C. The DOPA formed was isolated and assayed spectrofluorometrically as described in the text.

Zusammenfassung. Es wird eine Fluoreszenzmethode zur Bestimmung der Tyrosin-Hydroxylase-Aktivität von Homogenat beschrieben, die auf der Spektrofluorometrie der DOPA-Bildung beruht. Die Tyrosin-Hydroxylase-Aktivität von Homogenat der Nebenniere und des Gehirns (Nucleus caudatus) wurde mit dieser Methode gemessen.

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CONGRESSUS

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The fourth EUCHEM Conference on Stereochemistry will be held at the Bürgenstock, near Lucerne (Switzerland). The number of participants will be limited. Inquiries and applications (no special forms are required) should be addressed before 31 December 1968 to the Chairman, Prof. A. Kjaer, Institute of Organic Chemistry, Technical University of Denmark, Bygning 201, Lyngby (Denmark).