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This Week's Citation Classic™

Coyle J T & Henry D. Catecholamines in fetal and newborn rat brain.
J. Neurochemistry **21**:61-7, 1973.
[Laboratory of Clinical Science, National Institute of Mental Health, Bethesda, MD]

This paper described a sensitive and specific radiometric enzymatic assay for catecholamines in brain extracts. The method was applied to fetal rat brain and demonstrated the early developmental appearance of functioning catecholamine neurons. [The *SCI*® indicates that this paper has been cited in over 750 publications since 1973.]

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"This study was a product of my collaboration with David P. Henry when we were both research associates in the Laboratory of Clinical Science at the National Institute of Mental Health. David was working in the laboratory of Irwin Kopin, and I was in the laboratory of Julius Axelrod. David was involved in a project designed to measure catecholamines in serum under a variety of physiologic and pathologic conditions. My studies were directed at characterizing the development of brain noradrenergic and dopaminergic neuronal systems by quantitative neurochemical methods. Although my studies had previously demonstrated that the synthetic enzymes for catecholamines were present in the rat brain as early as 15 days of gestation, it was not possible with the existing and rather insensitive fluorometric techniques to reliably quantify catecholamine levels in rat brain before birth.

"To measure the catecholamines in the fetal brain, the radiometric enzymatic assay being developed by David was modified so that it was applicable to brain extracts. The remarkable sensitivity of the assay resulted from the availability at the time of [^{3}H]-S-adenosyl methionine (SAMe) of high specific radioactivity (4.5 mCi/μmol). The assay's considerable specificity derived from the use of the partially purified enzyme, catechol-O-methyltransferase (COMT). COMT catalyzes the inactivation of catecholamines by transferring a methyl group from SAMe to one of the ring hydroxyl groups on the catecholamine. A particularly attractive feature of the assay was that it allowed the separate determination of norepinephrine and dopamine without reliance on cumbersome procedures, such as thin-layer chromatography.

"The study demonstrated that both norepinephrine and dopamine were detectable in the fetal rat brain as early as 15 days of gestation, when the brain weighed less than two percent of that of the adult. Furthermore, pharmacologic manipulations revealed that the catecholamines in the fetal rat brain behaved in a fashion similar to those in adult brain, thereby indicating that the neurotransmitters were localized in a dynamic, functionally relevant pool. This study provided the first quantitative evidence of the remarkably early formation and functional activity of catecholaminergic neurons in the developing brain, a conclusion that was supported by subsequent histochemical and immunocytochemical studies.

"The reason for the frequent citation of this report may be that it was the first description of a sensitive, specific, and relatively simple method for measuring catecholamines in brain tissue. The overarching strategies involved in the assay—use of a partially purified methyltransferase, [^{3}H]-S-adenosyl-L-methionine, and differential organic extraction techniques—were exploited by other members of the Laboratory of Clinical Science to develop assays for dopamine-beta-hydroxylase, serotonin, tryptamine, and tyramine. Radiometric enzymatic assays for catecholamines have recently been eclipsed by the development of techniques coupling high performance liquid chromatography with electrochemical detection.[1] This latter method is less expensive and more rapid than, but as sensitive as, the radiometric enzymatic assay.[2]

"It is noteworthy that this article was selected as a *Citation Classic* this year when Julius has formally retired from the National Institute of Mental Health. This study by two young postdoctoral fellows directly issued from the conceptual approaches and the ambience of collaborative interactions that characterized Julie's laboratory."

1. **Keller R, Oke A, Mefford I & Adams R N.** Liquid chromatographic analysis of catecholamines: routine assay for regional brain mapping. *Life Sci.* 19:995-1003, 1976. (Cited 405 times.)
2. **Zaczek R & Coyle J T.** Rapid and simple method for measuring biogenic amines and metabolites in brain homogenates by HPLC-electrochemical detection. *J. Neural Transm.* 53:1-5, 1982.

