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CHANGE IN CATECHOLAMINE CONTENT OF *TETRAHYMENA PYRIFORMIS* W DURING GROWTH

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In 1966, Janakidevi *et al.* (1) discovered that the ciliated protozoan, *Tetrahymena pyriformis* contained catecholamines (CA) and readily converted radioactive tyrosine, phenylalanine and dihydroxyphenylalanine to CA. In our laboratory it was discovered that this protozoan contained the system catalysing the oxidation of biogenic monoamines and diamine (2). It was also found that in cultures of *Tetrahymena* monoamine oxidase activity decreased in the exponential growth phase (EGP) and increased in the stationary growth phase (SGP) (3). The present work was on the change of the endogenous CA level and the rate of conversion of tyrosine to CA during growth of this protozoan.

Tetrahymena pyriformis W was cultured as reported previously (3). After culture for various periods, cells were treated with 0.4 N perchloric acid following the procedure of Janakidevi *et al.* (1). CA was isolated by adsorption onto alumina (active neutral, Woelm) (4) and was measured fluorometrically, using adrenaline as a standard (5). CA levels during growth of cultures are expressed as total CA.

To measure the rate of conversion of tyrosine into CA, 50 m μ Ci/ml of tyrosine-¹⁴C (405 mCi/mM, Daiichi Pure Chemicals) were added to cultures at various stages of growth. After further incubation for 4 hr, radioactive CA was extracted as described above. An aliquot was dissolved in Bray's solution (6) and radioactivity was measured in a liquid scintillation counter.

The CA contents at various times during growth are summarized in Table I. The content in the early EGP was 0.39 μ g/10⁸ cells, which is in good agreement with the value of Blum *et al.* (7). The content in the SGP decreased to about one twentieth that in the EGP, therefore the endogenous CA content was higher in rapidly growing *Tetrahymena*

TABLE I. Content and radioactivity of catecholamines during growth of cultures of *Tetrahymena*

Time (hr)	Catecholamines		
	Cell number (cells/ml)	Content ($\mu\text{g}/10^8$ cells)	Radioactivity (cpm/ 10^8 cells)
24	2.1×10^3	0.39	$1,178 \times 10^2$
48	17.8 "	0.19	180 "
72	79.4 "	0.04	20 "
96	112.3 "	0.02	3 "
120	121.1 "	0.02	3 "

Radioactive tyrosine was added after various periods of culture after which incubation was continued for 4 hr. Total cultivation time is indicated.

cells than in slowly growing ones. We found previously (8) that, when transferred to fresh medium containing CA, growth of *Tetrahymena* in the EGP was slightly accelerated, while that in the SGP was inhibited. These results indicate that endogenous CA can regulate the growth rate of the protozoan. The possibility that CA regulates growth is supported by the findings of Wurtman *et al.* (9) who stated that during the estrous cycle in rats, the endogenous adrenaline level of the uterus rose significantly in the estrus period, and of Rudzik *et al.* (10) who reported that stilbesterol caused a parallel increase in the wt. and adrenaline content of the uterus.

The rate of conversion of tyrosine to CA increased abruptly in the early EGP, whereas it decreased in the SGP. The relationship between the rate of this conversion and the growth of the protozoan cannot be clarified until details of the biochemical properties of the enzyme systems involved in CA synthesis in *Tetrahymena* are known.

It should be mentioned here, that the level of cyclic adenosine 3', 5'-monophosphate in *Tetrahymena* increased rapidly in the early EGP and later decreased, as was recently observed by the authors, the report of which is now under preparation.

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