Dissimilar Effects of L and D Amino Acids on the Growth of Tetrahymena

Zsuzsa Darvas, Y. Nozawa and G. Csaba

Received September 8, 1987

KEY WORDS: tetrahymena; cell growth; amino acid isomers; receptors.

Of the L and D configurations of four amino acids (phenylalanine, valine, tryptophan, tyrosine) tested for influence on the growth rate of Tetrahymena, only L-tyrosine was able to induce imprinting in Tetrahymena pyriformis Zeuthen. D-valine stimulated the division of T. pyriformis NT-1, but failed to induce imprinting. The experiments have substantiated the selectivity of the amino acid receptors of T. pyriformis, and the extraordinary imprinting potential of tyrosine as well, as judged by its influence on the growth rate.

INTRODUCTION

Hormones and hormone receptors are both products of evolution. Our earlier studies (1,2,4) have strongly suggested that the hormone (signal molecule) played the leading role in both hormone and hormone receptor evolution, inasmuch as at the lower levels of phylogenesis the presence of a signal molecule induced the transformation of non-specific membrane patterns to specific receptors, and amplification of the latter by interaction with the signal molecule accounted for persistence of the receptor function over the further course of evolution. The establishment of a hormone-receptor relationship is also promoted by the great frequency of the membrane patterns associated with feeding in the unicellular organism. We have presented experimental evidence (5) of Lenhoff’s hypothesis (7,8) that the amino acid hormone receptors of the hydra arose from simple nutrient (amino acid) receptors.

Hormonal imprinting, too, plays a decisive role in receptor development. The primary interaction with the signal molecule amplifies the membrane pattern involved...
in the interaction, and accounts thereby for an increased binding capacity and responsiveness of the cell later in life (1,2,4). The experimental observations strongly suggest that the molecules capable of imprinting transform more readily to hormones than those which lack the imprinting potential (6).

The fundamental characteristic of a receptor is its specificity. With this fact in mind, we investigated whether the L and D amino acids acted on the unicellular Tetrahymena similarly or dissimilarly in respect of the induction of imprinting.

MATERIALS AND METHODS

Two-day cultures of T.pyriformis Zeuthen and T.pyriformis NT-1, grown in enriched proteose-peptone medium at 28 and 39°C, respectively, were used. Amino acid treatment was performed with the D and L isomers of valine, phenylalanine, tryptophan and tyrosine, all added to the media of mass cultures at $10^{-8}$ M concentration, in a water bath, under shaking. Pretreatment (imprinting) lasted 24 hr, and the second exposure (during which cell proliferation was recorded) lasted 20 hr. Each experiment was performed in two replicates.

Scheme of the Experiment

The two-day mass cultures were subdivided into two groups, of which the control groups (C) were not treated either for the first or the second time (indicated in the Figures as C/ or /C). The experimental groups (A) were pretreated with amino acids for 24 hr, were returned to plain growth medium for 48 hr and were finally assigned to two subgroups, treated or untreated at the second time. Therefore, the first series (C/C) was not treated either on the first or on the second occasion, the second series (C/A) was treated only on the second occasion, the third series only on the first occasion (A/C), and the fourth series on both occasions (A/A).

The cell counts were established at 0 and 20 hr by haemocytometric counting, in that first the dead cells were counted by the trypan blue test, and secondly all cells, after fixation in 4% formalin (in PBS). The difference between total and dead cell counts was regarded as the live cell count (number of cells/ml), which was related to the control as 100 for assessment of the percental growth rate. The inter-group differences were analyzed for significance with Student's $t$-test.

RESULTS

The T.pyriformis Zeuthen and T.pyriformis NT-1 strains responded differently to the applied amino acid treatments. The Zeuthen strain responded exclusively to L-tyrosine by a significantly increased rate of multiplication after two exposures (imprinting + reexposure), and showed no appreciable response to D-tyrosine or to either the L or D variant of the other amino acids (Fig. 1). The strain NT-1 responded to D-valine by a significantly increased growth rate after only a single exposure, and this response persisted longer than 48 h (Fig. 2). Reexposure to D-valine did not