Stereochemical Course of the Enzymic Synthesis of L-Tyrosine from Phenol and L-Serine Catalysed by Tyrosine Phenol Lyase from *Escherichia intermedia*

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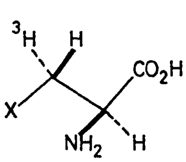
Summary The enzymic synthesis of L-tyrosine from phenol and L-serine catalysed by fibre-entrapped tyrosine phenol lyase from *Escherichia intermedia* proceeds with retention of configuration.

AMONG enzymic reactions involving β-replacement of L-serine[1] those catalysed by the pyridoxal phosphate-dependent enzyme tyrosine phenol lyase (β-tyrosinase)[2] have recently been studied.[3]

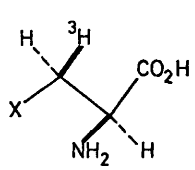
We now report on the stereochemical course of the synthesis of L-tyrosine from phenol and L-serine catalysed by fibre-entrapped tyrosine phenol lyase from *Escherichia intermedia.*

D,L-(3*R*)-[3-^{3}H;3-^{14}C]-Serine (**1**) and its (3*S*)-isomer (**4**) have been prepared from (1*R*)-[1-^{3}H]-phenaethyl alcohol and its (1*S*)-isomer, according to the procedure followed in the deuteriated series.[4] The conversion of the labelled substrates (**1**) and (**4**) into L-tyrosine by the enzyme took place in the presence of a large excess of ammonium ions which prevented deamination of L-serine. The reaction mixture consisted of 1000 ml 0·1 M phosphate buffer, pH 7·4, containing 5 mmol D,L-serine, 0·1 mmol pyridoxal phosphate, 0·1 mmol *erythro*-2,3-dihydroxy-1,4-butandithiol and 25 mmol of NH$_4$Cl, to which 50 mmol phenol were added during 24 h, at 28°.

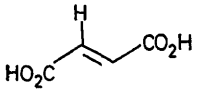
The stereochemistry of the tritium in position β of the L-tyrosine samples (**2**) and (**5**) obtained without significant



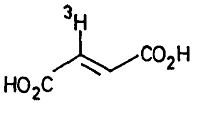
(1) X = OH
(2) X = p-OH-C$_6$H$_4$
(3) X = CO$_2$H



(4) X = OH
(5) X = p-OH-C$_6$H$_4$
(6) X = CO$_2$H



(7)



(8)

tritium loss, from the doubly labelled serine samples (**1**) and (**4**), has been determined by a recently described procedure[5] which involves conversion by ozonolysis into aspartic acid, (**3**) and (**6**), and then chemical conversion into malic acid. This is then enzymatically converted (malate hydrolyase) into fumaric acid with removal of a *pro-R* hydrogen from the β position.

The nearly complete (*ca.* 95%) loss and the high (over 90%) retention of the tritium label in the two fumarate samples (**7**) and (**8**) obtained from (**2**) and (**5**) indicate that the β-replacement had taken place with retention of configuration. The same stereochemical course has been observed in the synthesis of L-tryptophan from indole and L-serine catalysed by tryptophan synthetase.[4,6]

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