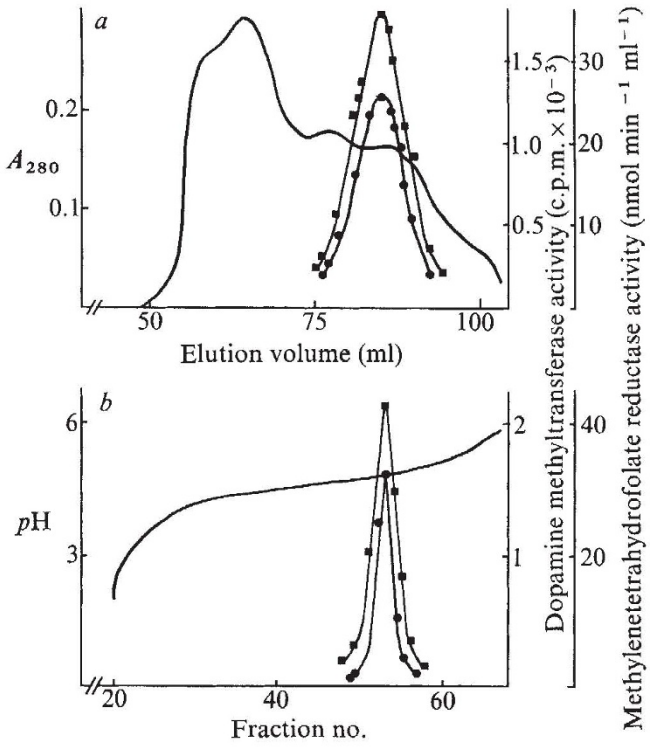

Fig. 2 *a*, Isolation of methylenetetrahydrofolate reductase by gel filtration on Sephadex G-200. 10 mg methylenetetrahydrofolate reductase (specific activity, 0.1 U mg^{-1}) purified from pig liver by the method of Kutzbach and Stokstad[11] was applied to a Sephadex G-200 column (1.5×95 cm) equilibrated in 10 mM potassium phosphate buffer, *p*H 7.5 containing 1 mM EDTA and 1 mM mercaptoethanol. Fractions were assayed for protein (A_{280}) and for methylenetetrahydrofolate reductase (●) and dopamine methyltransferase (■) activities as described in the text. *b*, Isoelectric focusing of methylenetetrahydrofolate reductase. 10 mg methylenetetrahydrofolate reductase was electrofocused at 4 °C in a 110 ml LKB column using ampholine, *p*H 3.5–5.0, in a 20–60% glycerol gradient. When electrofocusing was complete the column was drained and 2 ml fractions were collected. The *p*H value and the activities of methylenetetrahydrofolate reductase (●) and dopamine methyltransferase (■) of the fractions were measured. The recovery of enzyme activity was 50–60%.

isoelectric point of 4.8 (Fig. 2*b*). A similar result was obtained when dopamine methyltransferase, partially purified from bovine brain by the method of Laduron[4], was assayed by isoelectric focusing.

Active fractions from the gel filtration column were pooled and concentrated and used to investigate the effects of several additives on enzyme activity. The assay procedure for methylenetetrahydrofolate reductase routinely includes the addition of 2 μM FAD which produces a fourfold stimulation of the rate observed in the absence of the flavin coenzyme[11] and, correspondingly, the activity of dopamine methyltransferase was shown to be significantly increased in the presence of FAD (Table 1). 3,4-Dihydroxyphenylacetic acid (DOPAC) has been reported to inhibit the transferase[12]. We observed that 10^{-2} M DOPAC inhibited both the transferase and methylenetetrahydrofolate reductase activities to similar extents (Table 1).

Methylenetetrahydrofolate reductase is the initial enzyme in the pathway leading to the synthesis of S-adenosyl methionine (see Fig. 1). The reductase is allosterically inhibited by S-adenosyl methionine and this inhibition is relieved by S-adenosyl homocysteine[11]. As shown in Table 1, 1.6×10^{-4} M S-adenosyl methionine inhibited both the reductase and the transferase to similar extents and this inhibition was indeed reversed by S-adenosyl homocysteine.

The evidence presented here therefore supports the hypothesis that methylenetetrahydrofolate reductase and dopamine methyltransferase represent two activities exhibited by a single enzyme species. It is noteworthy that the regional distribution of these two activities in brain have previously been shown to be similar[13,14]. The mechanism of 1-carbon transfer to biogenic amines by the reductase *in vitro* would seem to involve oxidation of MTHF to methylenetetrahydrofolate and subsequent non-enzymic dissociation of formaldehyde from the folate derivative (Fig. 1). It is, at present, uncertain whether mediation in the synthesis of tetrahydroisoquinoline and tetrahydro-β-carboline alkaloids represents a physiological activity of this enzyme. Both the equilibrium of the reaction[11] and the NADPH–NADP$^+$ ratio in the cytosol[15] would favour MTHF synthesis rather than the reverse reaction involving formaldehyde release. Furthermore, free formaldehyde may also bind to tissue protein[16] or be oxidised by formaldehyde dehydrogenase[17]. The feedback inhibition of methylenetetrahydrofolate reductase by S-adenosyl methionine also supports the concept that the physiological function of this enzyme is in the ultimate synthesis of S-adenosyl methionine rather than the metabolism of biogenic amines. Since the formation of pharmacologically active alkaloids[18] from MTHF and amines may be detected *in vitro*, however, it would seem to be important to investigate their possible formation *in vivo*.

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Errata

In the Matters Arising contribution "Development of visual acuity and the sensitive period" by J. T. Flynn, D. I. Hamasaki, T. E. Flynn and M. Barricks (*Nature*, **257**, 337; 1975) the author of the original article quoted in ref. 1 should be D. N. Freeman and not R. D. Freeman.

In the article "Superior growth of the right gonad in human foetuses" by U. Mittwoch and D. Kirk (*Nature*, **257**, 791; 1975) the second sentence of the legend to Fig. 1 should begin: Bars represent the means of the . . . and not as printed.