intermediate between that seen in the absence of phosphate and with an IHP/Hb_4 ratio of 1. Similar reversal of organic phosphate effects has previously been observed in the case of –SH reactivity[11]. The situation at these very high levels of phosphate is no doubt complex and will not be considered further.

Cat haemoglobin B was used as a control in the present experiment because it has repeatedly been found to be unresponsive to any organic phosphate[7,11,12]. The reason for this insensitivity is that the β-chain amino terminals of this haemoglobin are acetylated[13]. The negative result with cat haemoglobin B in the present experiment was therefore expected. In addition, this lack of response indicates that the binding site of organic phosphates on methaemoglobin includes some of the same loci as the binding site on deoxyhaemoglobin. As was noted previously[12], this is not unlikely as the β-chain amino terminals reside on the surface of the molecule in both the liganded and unliganded conformations. Careful examination of Fig. 1 reveals that the cat haemoglobin has a substantially greater peroxidase activity than human haemoglobin. The meaning of this finding is uncertain, but it may be related to the fact that the oxygen affinity of cat haemoglobin is much lower than that of human haemoglobin[14].

Rein *et al.* have previously reported ESR spectra which demonstrate that ATP can markedly increase the high spin component of human methaemoglobin at acid *p*H (ref. 15). As the effects of ATP, 2,3-DPG, and IHP have generally been found to be qualitatively identical, there is little reason to doubt that IHP can have the same effect. In fact, Perutz has already suggested that this is the case[4]. The results presented here support such an interpretation if the protein with the high-spin configuration does indeed have greater catalytic activity, as suggested by Williams[6].

Although our results are consistent with Perutz's discussion regarding the interaction of IHP with methaemoglobin, they do not prove that his interpretation is correct as they do not provide direct evidence for the postulated change to a deoxy-conformation. Furthermore, it is difficult, if not impossible, to predict the magnitude of the effect that IHP should have on the peroxidase activity of methaemoglobin based on Perutz's model, so the agreement found here must remain strictly qualitative.

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Configuration of Bicuculline, a GABA Antagonist

We are grateful to Gilardi[1] for drawing attention to our use[2] of the incorrect configuration of bicuculline. We have in fact, in more recent publications[3,4], used the correct configuration as determined by optical rotatory dispersion and circular dichroism measurements[5]. The energy calculations of the protonated bicuculline cation by Gilardi[1] seem to be consistent with those made using other methods[4,6].

The substance illustrated incorrectly as bicuculline by us[2,7] and others[8,9] is the alkaloid adlumidine, which has yet to be evaluated as a GABA antagonist. In other publications[6,10] bicuculline has been incorrectly illustrated as its mirror image, (−)-bicuculline, which is inactive as a GABA antagonist.

Our original suggestion[11] regarding the structural similarities between GABA and bicuculline remains consistent with all of the available data which now include studies of a wide variety of GABA analogues of restricted conformation.

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