another H_2-receptor blocker, metiamide[5], as well as PGE_2 can induce equal potentiation in both organs. This suggests a common mechanism for both compounds. The first explanation is that PGE_2 may have a blocking effect on H_2-receptors. This seems unlikely since PGE_2 does not inhibit the relaxation (unpublished observation) induced by histamine in the cat tracheal muscle, which has been shown to be mediated by H_2-receptors[6].

It has previously been shown that histamine may stimulate the gastric acid secretion[7] and heart muscle[8] simultaneously with an increase of cellular cyclic AMP level. Both effects have been found to be blocked by burimamide. It seems likely that the stimulation of H_2-receptors in both perfused organs can produce the vasodilator effect by the increase of second messenger system, cyclic AMP. This speculation has been based upon findings obtained with theophylline, which has been described as a potent inhibitor of phosphodiesterase[9]. Addition of theophylline to the perfusion medium causes an inhibition of the pressor effect of histamine in both organs. This effect is probably due to the accumulation of cyclic AMP which represents the stimulation of H_2-receptors. Since theophylline antagonizes the potentiating action of PGE_2 on histamine responses in both organs, it is highly possible that PGE_2 and theophylline influence phosphodiesterase in opposite directions. Another possibility should be taken into consideration that PGE_2 may inhibit adenyl cyclase activity and consequently cause an inhibition of cyclic AMP in the cellular level. This point is still under investigation.

Zusammenfassung. PGE_2 verstärkt an isoliert perfundierten Kaninchennieren und Meerschweinchenlungen die Wirkung von Histamin auf den Perfusionsdruck. Gleiches wurde auch mit Metiamid, einem H_2-Rezeptorblocker, beobachtet. Anhand dieser Befunde wird die mögliche Rolle von c-AMP bei dieser Verstärkerwirkung diskutiert.

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The Metabolism of Phenylethylamine and *O*-Methylated Derivatives by Monoamine Oxidase

Phenylethylamine (PEA) and paramethoxyphenylethylamine (PMPEA) are sympathomimetic agents similar in structure to the catecholamines. Both compounds cross the blood-brain barrier and can cause central effects. PEA induces a strong, amphetamine-like central stimulation[1], while PMPEA administration results in a short-lasting catatonic state[2]. Similar catatonic responses have also been observed following the administration of 3,4-dimethyoxyphenylethylamine (3,4 DMPEA)[3]. Pretreatment with monoamine oxidase (MAO) inhibitors potentiates the depletion of central monoamines by PEA[4] and prolonges the effects of PMPEA on monosynaptic spinal reflexes[5]. Thus it is likely that PEA, and PMPEA, are metabolized in vivo by MAO, the enzyme responsible for the intraneuronal inactivation of biogenic amines. In this study we have compared the effects of PEA and PMPEA, as well as those of the dimethoxy derivatives (2,3 and 3,4 DMPEA) on brain MAO activity in vitro.

Methods of procedure. The metabolism of phenylethylamine and its derivatives was studied indirectly, by measurement of MAO activity in vitro in the presence of these agents. A water homogenate of rat brain stem (pons and medulla) was the enzyme source for these in vitro studies. MAO was measured by a modification of the micro method of McCaman[6], using C^{14} tyramine as substrate. For kinetic analysis, enzyme activities were measured at several substrate concentrations in the presence of varying concentrations of the compound tested. MAO activity was expressed as millimoles of substrate deaminated/g protein/h. Data were plotted by the method of Lineweaver-Burk[7] to give values for K_m and V_{max}. Competitive inhibitions is observed where K_m, but not V_{max}, is changed as compared to controls. In cases of competitive inhibition, the concentration of the inhibitor necessary to produce half maximal inhibition (K_i) was calculated from the slope of the Lineweaver-Burk curves. For non-competitive inhibition approximate K_i values have been calculated.

Results. The interactions of PEA and PMPEA with rat brain MAO are described in Figures 1A and 1B, respectively. Although both compounds inhibit brain MAO with tyramine as substrate, the nature of the inhibition differs. PEA is a non-competitive inhibitor (Figure 1A), whereas PMPEA inhibits in a competitive manner (Figure 1B). Qualitatively and quantitatively similar results were obtained with serotonin and dopamine as substrates. The concentration of PMPEA required to give half-maximal inhibition (K_i) is 18.0 μM. For PEA a series of K_i values are obtained, in the range of 87–142 μM.

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Like PMPEA, the dimethoxy derivatives are competitive inhibitors of rat brain MAO (Figures 2A and 2B). The K_i value for the 2,3 DMPEA is approximately 20% of that observed for the 3,4 DMPEA. The competitive nature of this inhibition is further verified by the constancy of the K_i values, irrespective of the slope from which these values are calculated.

The *o*-methylated phenylethylamine derivatives appear to be good substrates for rat brain monoamine oxidase. The calculated K_i values of 18 μM for PMPEA and 173.5 μM for 3,4 DMPEA obtained with rat brain homogenates compare favorably with published Km values of 15.1 and 278 μM for those amines with purified porcine brain MAO preparation[8]. In these studies we observed a variable decrease in K_i values with *o*-methylation, with the greatest decrease observed for PMPEA, and intermediate effects seen with the two dimethoxy isomers. Likewise, tyramine ring substitution[8] and *meta* *o*-methylation of noradrenaline as well as adrenaline[9] increase the affinity of these substrates for porcine brain MAO.

The parent amine, PEA, and the *para* *o*-methylated derivative are of pharmacological interest, in that both are sympathomimetic amines. PEA depletes central norepinephrine (NE) stores[4], whereas PMPEA depletes both NE and 5HT[10]. The 3,4 dimethoxy derivative is of biological interest, in that it is a possible abnormal metabolite in schizophrenia[3].

The 2,3 DMPEA has been included in these studies to illustrate the effects of positional ring substitution clearly seen in the large increase in the K_i value of that amine as compared to 3,4 DMPEA. 2,3 DMPEA probably is not formed in vivo, and has been found to be extremely toxic in reflex experiments in the anesthetized cat (WILLIS et al., unpublished).

In this in vitro study only the *o*-methylated derivatives have been found to be competitive inhibitors. It is suggested that PEA, PMPEA and 3,4 DMPEA can interfere with the inactivation of biogenic amines, by competing with naturally occurring substrates for MAO.

The biological half life and pharmacological effectiveness of these drugs is in part determined by the rate of their inactivation by MAO. That these amines are in fact substrates for MAO in vivo is supported by the following observation: MAO inhibition enhances the effectiveness of PEA and prolonges the duration of the PMPEA enhancement in the size of the monosynaptic reflex in the cat lumbosacral cord[11]. The transient effects of PMPEA on the size of the monosynaptic reflex are readily explainable by the rapid inactivation of that amine by MAO.

On the basis of the kinetic data obtained in this study, it is suggested that PMPEA effects on the monosynaptic reflex would differ in duration from those of the parent amine and that of the dimethoxy derivatives; reflex experiments to test this suggestion are in progress.

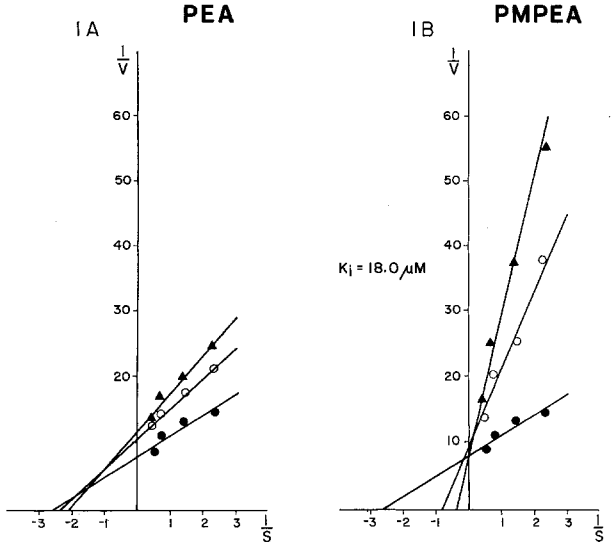

Fig. 1. Effects of PEA and PMPEA on MAO activity in vitro. Michaelis-Menten plot of MAO activity in rat brain homogenates at PEA and PMPEA concentrations of 0.05 mM (○) and 0.1 mM (▲), with 1-C^{14} tyramine as substrate.

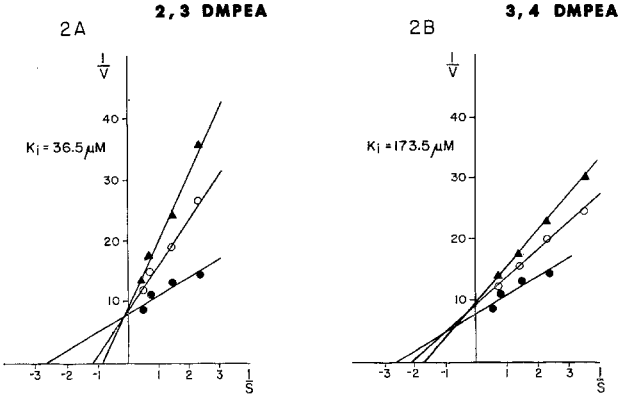

Fig. 2. Effects of 2,3 and 3,4 DMPEA on MAO activity in vitro. Michaelis-Menten plot of MAO activity in rat brain homogenates at 2.3 DMPEA and 3,4 DMPEA concentrations of 0.05 mM (○) and 0.1 mM (▲), with 1-C^{14} tyramine as substrate.

Zusammenfassung. Nachweis, dass Phenylethylamin und Paramethoxyphenyletheramin wirkungsvolle Substrate für die Monoaminoxylase im homogenisierten Rattenhirn in vitro sind, während 3,4-Dimethoxyderivat weniger wirkungsvoll als das Monoaminsubstrat ist. Die biologische Wirkung dieser Drogen scheint von der Geschwindigkeit ihrer Inaktivierung der Monoaminoxydase in vivo abhängig zu sein.

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