A comparative study of trace amine-derived responses of guinea pig isolated ileum

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The recently identified trace amine receptors (TARs) interact with the endogenous trace amines, 2-phenylethylamine (2-PEA), tyramine (TYR), tryptamine (TRP), and octopamine (OCT) and related molecules (amphetamine, AMPH), (Borowski et al 2001; Wolinsky et al 2007). Moreover, one of the group of investigators reported expression of the TAR mRNA in human small intestine (Borowski et al 2001); the function of the receptors remains to be determined. Therefore, the present experiments were carried out to characterize the responses of TAs in guinea pig isolated ileum.

Male guinea pigs (Dunkin Hartley strain, 300–1000g) were killed by a blow to the head and exanguinated. 2 cm lengths of ileum (10 cm from caecum) were suspended in Krebs-Henseleit solution gassed with 5% CO_2 in O_2 (pH 7.4) at 37 ^oC (Broadley et al 1985). Longitudinal contractions were recorded isometrically with a load of 0.5g using a force–displacement transducer connected to a computer data acquisition system (AD Instruments Powerlab Chart 5). Following 60 min equilibration and washes every 15 min, cumulative–concentration response curves (CRCs) were obtained to TAs (2-PEA, TYR, TRP, OCT and AMPH, 1μM – 10mM), in absence and presence of atropine (1 μM, muscarinic antagonist) or combination of the alpha- and β-adrenoceptor antagonists, phentolamine (1 μM) and propranolol (1 μM). Contractions were expressed as % of maximal contraction to acetyl-β-methylcholine (1 μM, MCh). Maximum (E_{Max}) and concentration of agonist eliciting a half-maximal response (EC_{50} value) were estimated by nonlinear regression analysis of the CRCs according to logistic equation. Responses are expressed as means±SEM and EC_{50} as –log EC_{50} ±SEM.

There was a concentration-dependent increase in contraction for all the amines, followed except for TYR by a relaxation. The rank order of potency, with pEC_{50} values in parentheses, was AMPH (4.62±0.12, n=4) > TYR (3.68±0.09, n=6) > TRP (3.57±0.15, n=4) > 2-PEA (3.16±0.05, n=4) > OCT (2.12±0.07, n=4). One-way ANOVA, followed by Bonferrroni's *post-hoc* test showed a significant difference between OCT and the other four compounds ($P < 0.001$ in all cases) and between 2-PEA and AMP ($P < 0.05$). The E_{Max} followed the order AMPH (123±12, n=4), TYR (78±2, n=6), TRP (52±3, n=4), 2-PEA (59±2, n=4) and OCT (36±2, n=4), with significant differences between each agonist. The combination of phentolamine and propranolol did not alter the EC_{50} or E_{Max} for any of the amines. However, atropine had a partial effect on potency of TYR ($P < 0.05$) and OCT ($P < 0.01$).

The biphasic response implies the existence of two distinct post-synaptic receptor populations and/or desensitisation at highest concentrations of TAs. Actions of TAs are mediated by non-adrenergic and non-cholinergic mechanisms in the ileum, possibly through TARs.

MAA was funded by Wellcome Trust (Research Fellowship). The study was also supported by The Welsh School of Pharmacy, Cardiff University.

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