Bronchodilator activity of bitter tastants in human tissue

To the Editor:

We read with interest the article by Deshpande *et al.*[1] on bitter taste receptors (TAS2Rs) in the lungs and the expression of TAS2Rs on human airway smooth muscle (ASM). The authors describe experiments in which TAS2R agonists, such as chloroquine, evoked relaxation of mouse tracheal rings that was threefold greater in magnitude than that elicited by β-adrenergic receptor agonists[1] (the current gold-standard bronchodilator therapy for asthma and chronic obstructive pulmonary disease[2]). We sought to investigate the uncharacteristically weak bronchodilator activity of isoproterenol in human tissue.

After repeating key experiments from the paper by Deshpande *et al.*[1] in our laboratories in the UK and in the laboratories of our collaborator in the US (B.J.C.), we obtained data that is consistent with a body of literature[3] and clinical practice that suggests that isoproterenol is a potent and efficacious relaxant of human airway smooth muscle, eliciting a relaxation 4 log fold (10,000×) more potent than chloroquine, although both agents had similar efficacy (**Fig. 1a,b**). This difference in potency was apparent when we precontracted the human airway smooth muscle to 60–80% of the maximum attainable contraction with either 10 μM histamine (**Fig. 1a**) or 1 μM carbachol (**Fig. 1b**). We obtained comparable results in studies using airway smooth muscle preparations from guinea pigs, a species in which relaxation to β-agonists is elicited through the β_2-adrenergic receptor (M.G.B., N.D., M.A.B. and B.J.C., unpublished observations).

Several explanations could be proffered as to why there is a discrepancy between our results and those obtained by Deshpande *et al.*[1] When performing *in vitro* tissue bath experiments to investigate the relaxant properties of agonists on ASM, standard practice is to precontract the tissue to establish a baseline level of bronchoconstrictor tone. The tone induced would normally be submaximal to avoid functional antagonism of the potential relaxant response[4]. However, the concentration of acetylcholine used by Deshpande *et al.*[1] (0.1 mM) is supramaximal, which could functionally antagonize the relaxant response to isoproterenol, thereby making it appear smaller than expected. When we repeated these experiments under conditions of supramaximal tension (0.1 mM ACh), we also found that the relaxation to isoproterenol was compromised (60% of the maximal relaxation evoked by papaverine instead of 100%; **Fig. 1c,d**).

We should emphasize that we do not question the author's conclusions that airway smooth muscle from humans and mice express bitter taste receptors. This is potentially a physiologically important observation. We would respectfully suggest, however, that our data and the published literature cast doubt on the claim that TAS2R agonists are more effective bronchodilators than currently prescribed β_2-adrenergic receptor agonists.

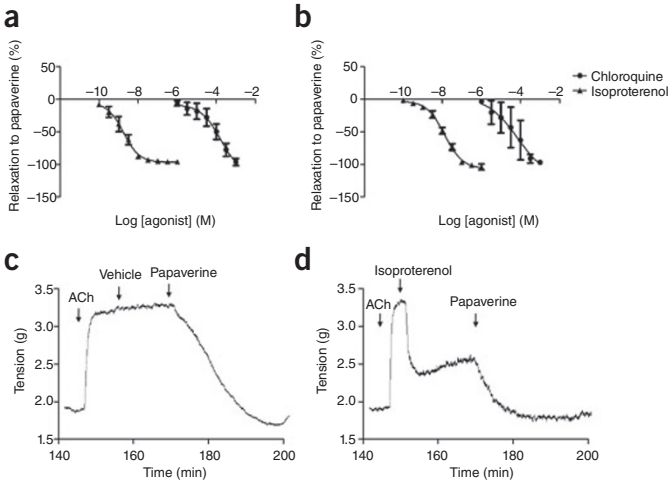

Figure 1 Comparison of chloroquine- and isoprenaline-induced relaxation of isolated human airway smooth muscle. (**a**,**b**) Relaxant responses of airway tissue induced by increasing concentrations of isoproterenol or chloroquine in tissues that were precontracted with histamine (**a**) or carbachol (**b**). Maximal tissue relaxation was assessed by treatment with papaverine (100 μM). Data shown are from experiments using samples from two separate lung samples and are expressed as the percentage of maximum relaxation to papaverine (100 μm). tension. (**c**,**d**) Representative traces from an experiment done on bronchial strips that were pre-contracted with acetylcholine (ACh; 0.1 mM) and then treated with vehicle (**c**) or 10 μM isoproterenol (**d**). Maximal tissue relaxation was induced by papaverine (100 μM). See **Supplementary Methods** for additional details.

Note: Supplementary information is available on the Nature Medicine *website.*

COMPETING FINANCIAL INTERESTS

The authors declare no competing financial interests.

Maria G Belvisi[1], Nicole Dale[1], Mark A Birrell[1] & Brendan J Canning[2]

[1]Respiratory Pharmacology Group, Pharmacology and Toxicology Section, National Heart and Lung Institute Faculty of Medicine, Imperial College London, London, UK. [2]Department of Medicine, Division of Allergy and Clinical Immunology, The Johns Hopkins Medical Institutions, Baltimore, Maryland, USA.

e-mail: m.belvisi@imperial.ac.uk

1. Deshpande, D.A. *et al. Nat. Med.* **16**, 1299–1304 (2010).
2. Bateman, E.D. *et al. Eur. Respir. J.* **31**, 143–178 (2008).
3. de Jongste, J.C. *et al. Am. Rev. Respir. Dis.* **138**, 321–326 (1988).
4. Sarria, B. *et al. Am. J. Physiol. Lung Cell. Mol. Physiol.* **283**, L1125–L1132 (2002).

Deshpande *et al.* reply:

Belvisi *et al.*[1] show in human bronchi that the bitter taste receptor (TAS2R) agonist chloroquine evokes marked relaxation; this is in agreement with our findings in mouse airways[2]. They note, however, an equivalent efficacy (degree of maximal relaxation) with the β-agonist isoproterenol in human bronchi[1]. In our paper, we carried out the vast majority of intact airway physiology experiments in mice, in which we found that bitter tastants had a greater efficacy compared with isoproterenol[2]. We never stated that TAS2R agonists were more potent than β-agonists, and we note that the differences in the potency (the EC_{50}) of isoproterenol and chloroquine are comparable in our study[2] and that of Belvisi *et al.*[1]. In our paper, we specifically noted that the threefold greater efficacy of TAS2R agonists compared to the β-agonist isoproterenol referred to mouse airways[2]. With the limited number of human airways that we studied, we were not in a position to provide quantitative efficacy data in human airways. We have now examined additional