Future bronchodilator therapy: a bitter pill to swallow?

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Bronchodilation via phosphodiesterase (the enzyme that breaks down cAMP) mechanisms including receptor internalization, uncoupling of the ASM cells, caused by a number of potential molecular mechanisms. However, this either have not made it to clinical trial, required high doses, had little effect in patients, or had a high incidence of side effects. Recent data suggests that a novel bronchodilator target exists, the bitter taste receptor TAS2R. Two recent studies [An SS, Wang WC, Koziol-White CJ, Ahn K, Lee DY, Kurten RC, Panettieri RA Jr, Liggert SB. Am J Physiol Lung Cell Mol Physiol 303: L304–L311, 2012; Pulkkinnen V, Manson ML, Säfholm J, Adner M, Dahlén SE. Am J Physiol Lung Cell Mol Physiol, doi:10.1152/ajplung.00205.2012.] provide new understanding of the signaling pathways utilized by TAS2Rs to mediate their bronchodilatory effects and how TAS2R-mediated signaling is affected by β-agonist signaling desensitization. As our understanding of TAS2Rs and their agonists increases, they move closer to a viable therapeutic option; however, further definition is still required and questions remain to be answered. This editorial focus discusses these studies within the context of existing literature and raises questions and challenges for the future development of bitter (better?) therapies for asthma.

airway smooth muscle; asthma; bitter taste receptors; bronchodilation

β-AGONISTS ARE A GOLD STANDARD asthma therapeutic, causing bronchodilation via β2-adrenoceptor (β2-AR) signaling. β-Agonists bind β2-ARs on airway smooth muscle (ASM) cells. β2-ARs are G protein-coupled receptors (GPCRs) that signal via Gαs to adenylyl cyclase (AC), increasing cAMP, activating protein kinase A (PKA), and resulting in myosin light chain kinase phosphorylation, ASM cell relaxation, and bronchodilation (Fig. 1). However, β-agonist tachyphylaxis is a major clinical issue. β-Agonist tachyphylaxis is thought to be primarily accounted for by β2-AR-mediated signaling desensitization in ASM cells, caused by a number of potential molecular mechanisms including receptor internalization, uncoupling of the GPCR from the G protein, reduced AC activity, and increased phosphodiesterase (the enzyme that breaks down cAMP) activity (7). A recent study published in the American Journal of Physiology (2) provides intriguing data showing that TAS2R agonists could be of therapeutic potential even when β2-AR desensitization has occurred.

TAS2Rs are bitter taste receptors, originally identified on the tongue and presumed to have evolved to stimulate signals to avoid ingestion of plant toxins. Like β2-ARs, they are GPCRs and were identified on ASM cells in 2010 (3). Their activation by TAS2R agonists (chloroquine, denatonium, saccharin, and quinine) relaxes both airway smooth muscle cells in culture and intact mouse airways to a similar extent as the β-agonist isoproterenol, making them an attractive novel bronchodilator target for asthma therapy. However, as the name “bitter taste receptor” suggests, they may prove a “bitter pill to swallow”! The signaling responsible for the relaxation is an area of discussion and a second study recently published in the American Journal of Physiology presents data suggesting different agonists may utilize different pathways (9). Defining these pathways will be crucial to the development of specific therapeutic targets in the future.

β2-AR-mediated signaling is sensitive to both homologous desensitization (in response to pretreatment with its own agonist) and heterologous desensitization (in response to pretreatment with an agonist that utilizes a different receptor but the same signaling). Taking this into consideration and the fact that β-agonists and TAS2R agonists cause the same physiological function, An et al. (2) wished to determine whether TAS2R agonists were efficacious in the presence of β2-AR homologous desensitization. An et al. show that treatment of ASM cells with albuterol (β-agonist)-induced β2-AR desensitization as signified by a reduced decrease in cell stiffness in response to isoproterenol. However, the TAS2R agonist chloroquine evoked identical decreases in cell stiffness irrespective of β2-AR desensitization by albuterol treatment. They also confirmed this in intact airways. This lack of heterologous TAS2R desensitization raises the possibility that TAS2Rs may represent a potential bronchodilator therapy in patients unresponsive to β-agonists. It would be interesting to know whether the reverse was true and whether β-agonists are sensitive to TAS2R agonist pretreatment.

It seems likely that the lack of effect of β2-AR desensitization on TAS2R efficacy is due to differential signaling downstream of the different receptors. β2-AR signaling is well defined as outlined above and in Fig. 1; however, the signaling downstream of TAS2R agonists is becoming a controversial area. Originally, Deshpande et al. (3) showed that saccharin- and chloroquine-mediated ASM relaxation was ablated by the large-conductance Ca2+-activated potassium (K+)(BKCa) channel antagonists charybdotoxin and iberiotoxin and suggested that relaxation was caused by calcium-dependent opening of BKCa channels and resultant membrane hyperpolarization. However, this finding is under debate (1, 10), and in the present issue Pulkkinen et al. (9) also suggest agonist effects may not be straightforward. Using isolated guinea pig trachea,

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Pulkkinen et al. confirmed airway relaxation in response to TAS2R agonists thiamine, denatonium, chloroquine, and noscapine, but in contrast to Deshpande they showed that the BKCa channels were only required for denatonium- and not chloroquine-induced relaxation. Further differences between the agonists were also shown. The cyclooxygenase inhibitor indomethacin and the prostanoid receptor (EP1) antagonist ONO-8310 enhanced airway relaxation in response to denatonium and thiamine but had no effect on chloroquine-induced relaxation. Interestingly, EP1 activation decreases β2-AR-mediated relaxation via heterodimerization of the receptors. It would be interesting to determine whether heterodimerization occurs between the TAS2R and EP1 receptor. Additionally, although chloroquine was able to induce relaxation in response to a number of contractile agents including carbachol, antigen in sensitized animals challenged with ovalbumin, the thromboxane receptor agonist U-46619, and histamine, denatonium was only effective against carbachol precontraction. Together these data suggest that different signaling mechanisms may be activated downstream of different TAS2R agonists. Attempts to determine signaling pathways distinct from calcium only succeeded in ruling out protein kinase A-, protein kinase C-, protein kinase G-, and cAMP-mediated signaling. It is important that future studies are conducted to dissect the signaling pathways downstream of TAS2R in response to the different agonists. It should also be noted that the current Pulkkinen et al. paper used guinea pig trachea whereas earlier studies used mouse tissue and isolated human ASM cells. It may therefore be the case that the differences seen represent species differences and this should be taken into account during future study design. In terms of therapeutic viability of TAS2R agonists, a clear understanding of their mechanism of action will increases the opportunity to develop specific and efficacious compounds.

So what further questions need to be answered before TAS2R agonists can be used for bronchodilator therapy in preference to β2-agonists? Is it likely that suitable formulations can be found? Synthetic agents that activate bitter taste receptors and are nontoxic exist, and there are a large number of plant-derived bitter tastants with good therapeutic profiles (3), which provides encouragement that formulations could be found. Can they be provided at a suitable dose by oral or inhaled administration? In the recent An et al. (2) study the TAS2R agonist chloroquine was used at a concentration between 50 and 100 times higher than the β2-AR agonist. Similarly in the Pulkkinen study (9) TAS2R agonists were used.
at a concentration ~100 times higher than the β2-AR, suggesting that high doses of TAS2R agonist may be required. Can more potent compounds be developed and, if taken orally, is there any evidence that the vasculature will not be detrimentally affected? Clarification of the signaling mechanisms utilized downstream of TAS2R may allow more specific and potent compounds to be synthesized.

It should also be considered whether homologous desensitization of TAS2Rs occurs. That is, do TAS2Rs experience desensitization in response to their own ligand, as is the downfall of β-agonists? Robinett et al. (8) have shown that in ASM cells pretreatment with the TAS2R agonist quinine caused a decrease in subsequent peak quinine-stimulated calcium response, equivalent to a 31% TAS2R desensitization. In intact airways quinine pretreatment reduced quinine-promoted relaxation from 53 to 36%. Thus it is likely that long-term TAS2R agonist treatment would result in the same reduced efficacy seen with β-agonists? Is this TAS2R agonist specific and dependent on downstream signaling? Would combined treatment allow for lower doses of each agonist to be used and protect against signaling desensitization? Deshpande et al. (3) showed that combined isoproterenol and chloroquine induced greater murine airway relaxation than either compound alone, but how the combination treatment affects signaling desensitization and thus potential therapeutic tachyphylaxis requires further investigation.

Of further interest is the fact that β2-AR desensitization is reversible and efficacy returns upon removal of treatment (4), allowing continued use of β2-ARs as reliever therapies for acute asthma attacks. It would be important to determine whether TAS2R homologous desensitization (or any potential heterologous effects on β2-AR signaling) is reversible or whether repeated usage would continue to reduce efficacy of the compounds. If it is confirmed that different TAS2R agonists utilize different signaling mechanisms, is there the potential to cycle between agonists to reduce potential desensitization effects?

Finally, β2-AR agonists are usually given in combination with glucocorticosteroids (GCs) and there is evidence to show that GCs can reverse β2-AR desensitization via regulation of β2-AR expression (6), adenyl cyclase activity, and G-protein-coupled receptor kinase activity (5). It would be interesting to determine whether glucocorticoids have the same effect on TAS2R receptor-mediated signaling desensitization. If the case, it is intriguing to speculate that a combination of glucocorticoid, β-agonist, and TAS2R agonist may allow more effective asthma therapy than is currently available.

In conclusion, the studies by An et al. (2) and Pulkkinen et al. (9) reinforce the role of TAS2R agonists as bronchodilators and add weight to the possibility of bitter taste receptor agonists being utilized in asthma therapy. The studies show that the bronchodilatory effect of TAS2R agonists are not impeded by β2-AR desensitization and delve further into the mechanism of how TAS2R agonists induce relaxation. The studies represent a further step toward the development of a novel bronchodilator therapy. Having said that, anyone who has taken bitter taste agonists for other clinical implications, chloroquine as antimalarials for example, will know how unpleasant they can be, so inhalation of these compounds may prove a challenge!

REFERENCES