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Serotonin 5-HT₂ receptor activation prevents allergic asthma in a mouse model

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Am J Physiol Lung Cell Mol Physiol 308: L191–L198, 2015. First published November 21, 2014; doi:10.1152/ajplung.00138.2013.—Asthma is an inflammatory disease of the lung characterized by airways hyper-responsiveness (AHR), inflammation, and mucus hyperproduction. Current mainstream therapies include bronchodilators that relieve bronchoconstriction and inhaled glucocorticoids to reduce inflammation. The small molecule hormone and neurotransmitter serotonin has long been known to be involved in inflammatory processes; however, its precise role in asthma is unknown. We have previously established that activation of serotonin 5-hydroxytryptamine (5-HT)₂A receptors has potent anti-inflammatory activity in primary cultures of vascular and gut tissues and in the whole animal in vasculature and gut tissues. The 5-HT₂A receptor agonist, (R)-2,5-dimethoxy-4-iodoamphetamine [(R)-DOI] is especially potent. In this work, we have examined the effect of (R)-DOI in an established mouse model of allergic asthma. In the ovalbumin mouse model of allergic inflammation, we demonstrate that inhalation of (R)-DOI prevents the development of many key features of allergic asthma, including AHR, mucus hyperproduction, airways inflammation, and pulmonary eosinophil recruitment. Our results highlight a likely role of the 5-HT₂ receptors in allergic airways disease and suggest that 5-HT₂A receptor agonists may represent an effective and novel small molecule-based therapy for asthma.

serotonin; inflammation; 5-HT₂ receptor; 5-HT₂A receptor; asthma; DOI

SEROTONIN [5-hydroxytryptophan (5-HT)] is a ubiquitous, small hormone molecule, present in nearly all eukaryotes, that mediates a wide spectrum of physiological processes. In mammals, it exerts its action through 14 different receptor subtypes that comprise seven distinct families (5-HT₁–7) (34). All but one family, the ligand-gated 5-HT₃ receptor ion channel, are G-protein-coupled receptors (34). The 5-HT₂A receptor is known primarily for its role in mediating complex cognitive behaviors within the central nervous system and for mediating physiological processes, such as vasoconstriction, in the periphery (32, 34). Interestingly, the 5-HT₂A receptor is the primary target of classic hallucinogenic drugs, such as lysergic acid diethylamide, which produces intoxicating effects. Although 5-HT₂A receptor mRNA is expressed at higher levels in immune-related tissues, such as spleen, thymus, and peripheral-circulating lymphocytes, compared with other serotonin receptor subtypes (i.e., 5-HT₁A, 5-HT₁D, 5-HT₂C, 5-HT₄, 5-HT₅A, and 5-HT₅B) (42), its precise role in inflammatory processes is not well defined. With regard to the potential role of serotonin in asthma, 5-HT₂A receptors are functionally expressed in activated CD4⁺ T cells, alveolar macrophages, eosinophils, and lung epithelial and smooth muscle cells (8, 20, 21, 23, 30). In fact, migration of eosinophils in allergic asthma has been shown recently to be dependent on 5-HT₂A receptor activation (21), and 5-HT₂ receptors have been implicated in platelet function relevant to allergic asthma (13).

We reported recently that 5-HT₂A receptor agonists potently inhibit inflammation in vitro (53). The anti-inflammatory effects of one particular 5-HT₂A receptor agonist, (R)-2,5-dimethoxy-4-iodoamphetamine [(R)-DOI], is extremely potent, with an EC₅₀ of ~15 pM. Through activation of the 5-HT₂A receptor, (R)-DOI blocks the expression and activation of proinflammatory markers, including expression of chemokines (e.g., monocyte chemotactic protein-1 [MCP-1]), cellular adhesion molecules (ICAM1 and VCAM1), cytokines (e.g., IL-6), nitric oxide synthase, and activation/nuclear translocation of NF-κB, in a variety of cell types, including primary aortic smooth muscle cells (53). We have translated these in vitro findings to a whole animal model of inflammation by demonstrating that (R)-DOI, also through 5-HT₂A receptor activation, has potent anti-inflammatory effects when administered before systemically administered TNF-α. These effects are most pronounced in the vasculature and the gut, where preadministration of (R)-DOI blocks TNF-α-induced increases in the proinflammatory gene and protein expression, including circulating IL-6 (33).

In an effort to extend our findings to the potential use of (R)-DOI as a therapeutic in inflammatory airways disease, herein, we examine the ability of (R)-DOI to block the key features of allergic asthma in the well-established mouse model of ovalbumin (OVA)-induced allergic asthma. In this model, mice are sensitized and challenged with inhaled chicken OVA peptide to induce a phenotype resembling human asthma, including airways hyper-responsiveness (AHR) in response to methacholine (MeCh), mucus hyperproduction, and pulmonary inflammation characterized by eosinophilia (5). We show here that inhaled (R)-DOI blocks AHR, recruitment of eosinophils to the lung, mucus hyperproduction, and inflammatory airway remodeling. We speculate that 5-HT₂ receptor agonism may represent a novel therapeutic strategy for asthma.

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Measurement of total IgE and OVA-specific IgE. Whole blood was taken via cardiac puncture by a 23-gauge needle on protocol day 28. Whole blood was placed into plasma separator tubes coated in lithium heparin (Becton Dickinson, Franklin Lakes, NJ). Plasma was isolated from whole blood following the manufacturer’s protocols. Total mouse IgE in the isolated plasma was determined using the ELISA MAX Deluxe kit (Cat. No. 432404) and OVA-specific IgE was determined using LEGEND MAX Mouse OVA Specific IgE ELISA Kit (Cat. No. 439807), purchased from BioLegend (San Diego, CA).

Cytokine and chemokine analysis by qRT-PCR. lungs were harvested 48 h after the final OVA exposure, and expression levels of cytokines were determined using reverse transcription and quantitative real-time PCR (qRT-PCR). For all lung tissues, RNA was extracted with TRIzol reagent, purchased from Life Technologies (Carlsbad, CA), following the manufacturer’s instructions. RNA was processed into first-strand cDNA using the ImProm-II cDNA synthesis kit (Promega, Madison, WI), following the manufacturer’s instructions. The input cDNA for each reaction was 500 ng total RNA. Cytokine and chemokine mRNA expression, examined by probe-based qRT-PCR, included the following: II-4, II-5, II-6, II-10, II-13, Tnfα, Mcp-1, and granulocyte macrophage colony-stimulating factor (Gm-csf). Primers were designed to be compatible with the Universal ProbeLibrary system using the Universal ProbeLibrary Assay Design Center (Roche Diagnostics, Indianapolis, IN) and synthesized by Integrated DNA Technologies (Coralville, IA). Primer sequences used in this study are as listed: II-4 forward 5'-cagccgctatggtagtcggg-3' and reverse 5'-cagctgctcttccttgttt-3'; II-5 forward 5'-acatgggaacccaggt-3' and reverse 5'-caccatcgagcagctcg-3'; II-6 forward 5'-ttcaagcttcatcgaacagg-3' and reverse 5'-tgcttgccattcctcctc-3'; II-10 forward 5'-cagcgacaggtctcctag-3' and reverse 5'-gtctgctgtgctttgtt-3'; II-13 forward 5'-cttcgcacccagcacttt-3' and reverse 5'-gtctgctgtgcttttgtt-3'; Tnfα forward 5'-tctctccattccttgtttg-3' and reverse 5'-gtctgctgtgcttttgtt-3'; Mcp-1 forward 5'-tcaagtgccttgctcctct-3' and reverse 5'-gtctgctgtgcttttgtt-3' and reverse 5'-agctgctgtgcttttgtt-3' and reverse 5'-gctgctgtgcttttgtt-3' and reverse 5'-gctgctgtgcttttgtt-3' and reverse 5'-gctgctgtgcttttgtt-3'. Probes were used from the Universal ProbeLibrary (Roche Diagnostics, Indianapolis, IN) and are listed with the following universal probe numbers: U2, U97, U78, U41, U17, U49, U22, and U79 for II-4, II-5, II-6, II-10, II-13, Tnfα, Mcp-1, and Gm-csf, respectively. Quantification of gene expression (see Fig. 6) was performed on a Roche LightCycler 480 Instrument II LC (Roche Diagnostics). Gene-expression levels were calculated using the comparative threshold cycle method and normalized to internal Gapdh expression, as determined using the Mouse GAPD Gene Assay (Cat. no. 0504621001; Roche Diagnostics) in multiplex format.

Statistics. All statistical analysis was performed using GraphPad Prism (GraphPad Software, La Jolla, CA). See figure legends for specific data.

RESULTS

Pulmonary administration of (R)-DOI is effective in preventing AHR in a mouse model of allergic asthma. Following OVA sensitization and challenge, we measured airways resistance by two different methods in separate groups of mice. For the first method, we used the forced oscillation technique and in the second method, whole-body plethysmography in awake, freely moving mice. As expected, mice receiving only OVA develop significant AHR in both methods (Fig. 1). Mice pretreated with inhaled (R)-DOI at 0.01 (Fig. 1, A and B) or 1.0 mg/kg (Fig. 1, A and C), before OVA challenge, display airways responsiveness not significantly different from naive, as measured by either method.

(R)-DOI prevents pulmonary inflammation and mucus hyperproduction. Histopathological analysis of lung sections from the different treatment groups demonstrated that as ex-
pected, OVA mice develop significant pulmonary inflammation and mucus. Animals treated with (R)-DOI (1.0 mg/kg) before OVA exposure exhibit very little peribronchial inflammation or mucus. Mice treated with two orders of magnitude less (R)-DOI (0.01 mg/kg) demonstrate significantly reduced inflammation and mucus production compared with the OVA-only-exposed lungs (Figs. 2 and 3).

(R)-DOI reduces pulmonary inflammation and BALF eosinophilia. Pulmonary inflammation is a common feature of asthma and is partly responsible for increased AHR (15). To associate (R)-DOI treatment and decreased AHR, as well as normal-appearing histological results with lack of inflammation, we performed cell-differential counts on BALF cell populations for each mouse in each group. As expected, OVA induced a significant increase in the total number of cells recovered in the BALF compared with naïve and (R)-DOI-treated animals. A large fraction of the BALF cellularity was due to elevated numbers of eosinophils (Fig. 4). Total BALF cell numbers and eosinophil numbers for naïve, 0.01 mg/kg DOI/H11001 OVA, and 1.0 mg/kg DOI/H11001 OVA were significantly lower than the OVA-only mice (Fig. 4). Although the eosinophil numbers for the (R)-DOI-treated mice were greater than

Fig. 1. (R)-2,5-Dimethoxy-4-iodoamphetamine [(R)-DOI] prevents the development of airways hyperresponsiveness (AHR). A: in forced oscillation-resistance measurements (flexiVent; SCIREQ), naïve mice and those treated nose only with 0.01 and 1.0 mg/kg (R)-DOI during the sensitization process exhibited significantly different resistances from the ovalbumin (OVA)-only-treated group at 50 mg/ml methacholine (MeCh) and were not significantly different from naïve, saline-treated mice [resistance = average of the fractional difference (Δ) of the value measured vs. the individual baseline values]. B and C: results from whole-body plethysmography experiments in awake, freely moving mice are consistent with the forced oscillation results: pretreatment with (B) 0.01 mg/kg (R)-DOI nose only and (C) 1.0 mg/kg (R)-DOI nose only significantly reduced the development of airways resistance. In all figure panels, DOI represents (R)-DOI at the indicated dose. *P < 0.05 OVA vs. Naïve, #P < 0.05 OVA vs. (R)-DOI (1.0 mg/kg) + OVA, and ^P < 0.05 OVA vs. (R)-DOI (0.01 mg/kg) + OVA; n = 5–9 animals/treatment group; error bars represent ± SE; 2-way ANOVA with Bonferroni post hoc test. Peak enhanced pause maximum (PenH Max) values represent baseline-normalized values.

Fig. 2. OVA-induced lung inflammation and mucus hyperproduction are inhibited by nose-only (R)-DOI. Representative sections of airways (4 μm) stained with the periodic acid-Schiff (PAS) technique are shown in this figure to highlight mucus (bright pink color). Saline-treated animals have normal airway morphology and no mucus or inflammation (A and E). OVA-alone-treated animals have thickened airways with a significant amount of mucus present (B), as well as peribronchial inflammation (F; arrows indicate inflammatory cells). Animals pretreated with (R)-DOI (1.0 mg/kg and 0.01 mg/kg nose only) demonstrate normal airway morphology, with little to no detectable mucus or inflammation (C and G and D and H, respectively). A–D, 40× objective; E–H, 10× objective.
as expected from our previous studies in different inflammatory models (Fig. 6).

DISCUSSION

To determine if serotonin 5-HT$_2$ receptor activation with (R)-DOI is an effective mechanism to treat a pathological inflammatory disease, we investigated the effects of the highly selective 5-HT$_2$ receptor agonist (R)-DOI in a mouse model of allergic asthma. By building upon our earlier in vitro and in vivo studies, we demonstrate here that inhaled (R)-DOI has potent anti-inflammatory effects and blocks the development of allergic asthma in the OVA mouse model. Importantly, we have already established that the anti-inflammatory effects of (R)-DOI in vitro and in vivo are mediated through activation of the serotonin 5-HT$_{2A}$ receptor subtype (33, 53). Here, we tested two different doses of (R)-DOI. The 1.0-mg/kg dose is in the range of that used in typical behavioral experiments (41). The very low dose of 0.01 mg/kg was chosen to test the super potency of (R)-DOI, predicted by our previous cellular studies (53). Anti-inflammatory effects of this very low dose were also observed in our recent in vivo study examining the ability of (R)-DOI to block the effects of systemic administration of TNF-α (33). Because activation of the 5-HT$_{2A}$ receptor subtype and not the 5-HT$_{2C}$ receptor subtype was found to be necessary for the anti-inflammatory effects of (R)-DOI in our previous studies, we hypothesized that the effects of (R)-DOI against allergic asthma were also mediated through 5-HT$_{2A}$ receptor activation. Although we were not able to validate this here, we have confirmed the presence of 5-HT$_{2A}$ receptor mRNA on whole-lung tissue (33). Furthermore, the expression of 5-HT$_{2A}$ receptors has been reported in airway smooth muscle cells (2) and alveolar macrophages (30), and although naïve T cells do not express high levels of the 5-HT$_{2A}$ receptor, activated T cells do express high levels of 5-HT$_{2A}$

those of naïve mice, they were not significantly different. There is a trend for a decrease in the neutrophil numbers in (R)-DOI-treated mice compared with OVA-treated mice; however, the difference was not significant.

(R)-DOI does not alter lung leak or plasma IgE levels. Increased protein content of the BALF is a hallmark of asthma and the OVA model (46). Analysis of BALF total protein by BCA assay from different treatment groups revealed a significant increase between naïve and OVA groups but showed no difference between mice treated with (R)-DOI + OVA and those animals that were treated with OVA only (Fig. 5). The OVA model characteristically produces increased serum levels of IgE and OVA-specific IgE (19, 28, 56); therefore, we tested the effects of (R)-DOI on total IgE and OVA-specific IgE. In both cases, we measured a significant increase between naïve and OVA-treated groups. (R)-DOI treatment, however, had no effect on either total IgE or OVA-specific IgE as induced by OVA (Fig. 5).

(R)-DOI suppresses expression of genes involved in the T cell and innate-immune cell response. A panel of cytokines and chemokines typically involved in asthma and the OVA model (II-4, II-5, II-6, II-10, II-13, Tnfa, Mcp-1, and Gm-csf) was examined in the lungs by qRT-PCR (6, 10, 17, 29, 36, 45, 49).

There were, as anticipated, significant increases in mRNA for II-4, II-5, II-10, II-13, Mcp-1, and Gm-csf with OVA treatment compared with naïve mice. There was a trend that did not reach significance for II-6 and Tnfa expression. (R)-DOI had no effect on the expression levels of OVA-induced II-4 or II-10. Interestingly, (R)-DOI treatment significantly repressed the OVA-induced increases in mRNA expression for Mcp-1, II-13, and II-5 and completely blocked the increase in Gm-csf (Fig. 5).

Although II-6 expression was not up-regulated significantly in the OVA group compared with vehicle control, (R)-DOI did significantly reduce II-6 expression levels in OVA-treated mice,
receptor mRNA (23). We suggest that the site of therapeutic action is directly on the pulmonary tissues, including resident-activated T cell populations and/or innate-immune cells.

The major components of allergic asthma in humans include AHR, pulmonary inflammation, and mucus hyperproduction (7). In addition, eosinophils, which release cytotoxic mediators and leukotrienes, are recruited in large numbers to the lungs of asthmatic individuals (37). Eosinophil production, chemotaxis, and survival are controlled by regulated on activation, normal T cell expressed and secreted (CC chemokine ligand 5), macrophage inflammatory protein 1 \(\alpha\), eotaxins, IL-5, and GM-CSF (18, 25, 35, 43, 47). IL-5 and GM-CSF are derived from activated pulmonary epithelial cells, eosinophils themselves, and activated T-lymphocytes (1, 27, 50). IL-5 and GM-CSF are

Fig. 5. Total protein and IgE levels are not affected by \((R)-\text{DOI}\) (1.0 mg/ml, nose only). A: the total of protein content in the BALF, as measured by bicinchoninic acid assay, is increased significantly in the OVA-only-treated lungs compared with naïve. \((R)-\text{DOI}\) does not alter total BALF protein induced by OVA. B: total plasma IgE, as measured by ELISA, is increased significantly by OVA treatment. \((R)-\text{DOI}\), administered before OVA challenge, has no effect on total plasma IgE. C: OVA-specific plasma IgE, as measured by ELISA, is increased significantly by OVA treatment. \((R)-\text{DOI}\), administered before OVA challenge, has no effect on OVA-specific plasma IgE. ***\(P < 0.001\) vs. OVA; n.s. = no significance vs. OVA; \(n = 7–17\) animals/treatment group; error bars represent \(±\) SE; ANOVA with Tukey post hoc test.

Fig. 6. Inhaled \((R)-\text{DOI}\) (1.0 mg/kg) inhibits proinflammatory gene expression in the whole lung. Quantitative RT-PCR measurement of mRNA expression levels of several inflammatory markers is shown. OVA produces a significant increase in the mRNA levels of Il-4 (A), Il-10 (B), monocyte chemotactic protein-1 (Mcp-1; E), Il-13 (F), Il-5 (G), and granulocyte macrophage colony-stimulating factor (Gm-csf; H) compared with naïve. No significant effect of OVA was observed on Tnf \(\alpha\) (C) or Il-6 (D) expression. \((R)-\text{DOI}\) produces significant inhibition of the OVA-induced increases in the mRNA expression of Mcp-1, Il-13, Il-5, and Gm-csf. Although Il-6 levels were not statistically different between naïve and OVA groups, \((R)-\text{DOI}\) elicited a significant decrease in Il-6 expression levels when administered before OVA exposure compared with OVA alone. ***\(P < 0.0001\), **\(P < 0.01\), and *\(P < 0.05\); n.s. = no significance; \(n = 4\) animals for the Naïve group, and \(n = 10\) animals for the OVA and DOI + OVA treatment groups; error bars represent \(±\) SE; ANOVA with Tukey post hoc test.
AHR, and represses T helper cell 2 (Th2) and innate-immune phils to the lung, prevents mucus hyperproduction, blocks demonstrated in pulmonary tissues by our lab and others, the inhaled; data not shown).

response (9), or airways constriction in mice (7). We oral intoxication, as indicated by the classical head-twitch magnitude less than those necessary to produce either behav-

tion of bone marrow-derived dendritic cells, although the receptor(s) mediating these effects remain unknown (13). Con-

molecules important in the development of asthma and are increased in serum and BALF of asthmatics in the clinic (12, 44). Significantly, our data show that both genes are suppressed by administration of (R)-DOI in the OVA mouse model.

The role of eosinophils in asthma is both direct, causing bronchoconstriction and destruction to airways, and indirect by provoking degranulation of mast cells and basophils (7). We demonstrate here that (R)-DOI blocks recruitment of eosino-

phils to the lung, prevents mucus hyperproduction, blocks AHR, and represses T helper cell 2 (Th2) and innate-immune cell gene expression (e.g., II-5 and Mcp-1). We delivered (R)-DOI directly to the lung using inhalation techniques in these experiments, and it remains to be determined whether systemically injected (R)-DOI has the same or similar effects on the development of asthma. Importantly, effective levels of (R)-DOI, administered by this route (inhalation), are orders of magnitude less than those necessary to produce either behav-

ioral intoxication, as indicated by the classical head-twitch response (9), or airways constriction in mice (>10 mg/kg inhaled; data not shown).

Although the presence of 5-HT2A receptor mRNA has been demonstrated in pulmonary tissues by our lab and others, the role of this receptor in the lung has remained largely undefined. A few reports have suggested that the 5-HT2A receptor mediates AHR in allergic asthma (14, 40, 54). However, these studies used the antigen ketaserin, which is nonselective in rodents for 5-HT2 receptors and also has high affinity for histamine H1 and α-adrenergic receptors, to block the effects of serotonin. This makes it difficult to interpret results using ketaserin. In any case, these reports indicated that serotonin activation of 5-HT2A receptors contributes to AHR rather than preventing it. Serotonin itself has been implicated in airways inflammation in allergic asthma by acting as a critical factor to recruit inflammatory cells and prime Th2 responses by activation of bone marrow-derived dendritic cells, although the receptor(s) mediating these effects remain unknown (13). Con-

versely, blockade of serotonin receptors with a nonselective antagonist for multiple subtypes has demonstrated antiasthma effects in the OVA model (24, 40). Why then, if serotonin appears to have a proinflammatory effect in the lung, does activation of 5-HT2 receptors with (R)-DOI have an anti-

inflammatory effect? One possibility is that selective activation of 5-HT2 receptors with (R)-DOI avoids activation of other serotonin receptor types responsible for the inflammatory re-

sponse. A more likely explanation is that (R)-DOI, which has a much higher affinity for the 5-HT2 receptors than serotonin, is acting as a functionally selective ligand and recruiting anti-inflammatory effector pathways that serotonin itself does not (26, 31). Significantly, (R)-DOI has already been shown to activate different signaling pathways than serotonin at the 5-HT2A receptor in vivo (38, 39).

It is unlikely that the therapeutic mechanistic site of action of (R)-DOI is on the B cell or the antigen-presenting cell, as (R)-DOI has no effect on OVA-induced II-4 gene expression. Recent reports indicate that IgE-dependent mast cell activation, yet prevents AHR, suggests (R)-DOI is acting on activated rather than naïve T cells to block AHR through nonmast cell-dependent mechanisms. Because (R)-DOI blocks Mcp-1 and Gm-csf mRNA production, the therapeutic target may also include innate immune cells. There is also the possibility that (R)-DOI may be acting on the naïve CD4+ population; however, naïve T cells do not express high levels of 5-HT2A receptor mRNA until activated. Our data demonstrate that (R)-DOI treatment significantly inhibits the OVA-induced expression of Th2-related genes that include II-13, II-5, and Gm-csf in the lung. Interestingly, vascular (or more likely, epithelial) permeability is not improved with (R)-DOI, as total protein in the BALF is not reduced compared with OVA alone.

We propose a model, shown in Fig. 7, where the pool of 5-HT2A receptors activated by (R)-DOI that responds with

![Diagram](https://example.com/diagram.png)

**Fig. 7.** A proposed therapeutic mechanism of (R)-DOI. The presented data show that (R)-DOI has no effect on II-4 gene expression, as well as no effect on humoral IgE production. These data provide evidence that the therapeutic action of (R)-DOI is not on the B cell, the antigen-presenting cell (APC), and/or the naïve CD4+ population. Importantly, we show that (R)-DOI treatment significantly inhibits expression of T helper cell 2 (Th2)-related genes, including Mcp-1, II-13, II-5, and Gm-csf compared with asthmatic animals. Taken to-

together, we suggest that (R)-DOI exerts its therapeutic action in the OVA asthma model by activating anti-

flammatory signaling pathways through the serotonin 5-hydroxytryptamine 2A receptors on T cells and/or innate immune cells, leading to a decrease in secretion of II-5 and Gm-csf, leading to a decrease in eosinophilia recruitment; and a decrease in Mcp-1 production, leading to a decrease in Th2 polar-

ization. These changes contribute to a general decrease in both inflammation and AHR.
anti-inflammatory properties could reside on activated Th2 cells and/or innate immune cells. In this proposed model, 5-HT2A receptor activation would lead to reduced IL-5, GM-CSF, and MCP-1 secretion, in turn, decreasing eosinophil recruitment, Th2 polarization, and IL-13 production (16, 22, 48, 55). Overall, these effects would combine to reduce inflammation and AHR. The precise cellular signaling pathways, however, remain to be elucidated.

In conclusion, we have identified an important and new functional role of 5-HT2 receptors in the lung. (R)-DOI activation of serotonin 5-HT2 receptors potently prevents the development of a clinically relevant mouse model of allergic asthma at drug levels far below those necessary to invoke adverse cardiovascular or behavioral effects. Based on our previous in vitro and in vivo studies, we predict that it is the 5-HT2A receptor that is the therapeutic target of (R)-DOI in our model. Our results demonstrate that activation of 5-HT2 receptors differentially regulates Th2 signaling, innate cytokine responses, and other relevant inflammatory effector pathways and that selective activation with (R)-DOI, or perhaps other 5-HT2A agonists in its class, represents a novel, small molecule-based therapeutic strategy for the treatment of asthma.

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