

Forum Minireview

Progress in Allergy Signal Research on Mast Cells: Up-Regulation of Histamine Signal-Related Gene Expression in Allergy Model Rats

Hiroyuki Fukui^{1,*}¹Department of Molecular Pharmacology, Institute of Health Biosciences, The University of Tokushima Graduate School,
1-78-1 Shomachi, Tokushima 770-8505, Japan

Received November 5, 2007; Accepted December 18, 2007

Abstract. Brown Norway allergy model rats sensitized to toluene 2,4-diisocyanate (TDI) were developed. Histamine H₁ receptor mRNA level was elevated in nasal mucosa of allergy model rats after the provocation with TDI, which was followed by H₁-receptor up-regulation. Elevation of histamine H₁ receptor mRNA was partially suppressed by *d*-chlorpheniramine and olopatadine, antihistamines. Histamine induced increase in histamine H₁ receptor gene expression in vitro, and the protein kinase C- δ isoform was suggested to mediate the gene expression. On the other hand, elevation of histamine H₁ receptor mRNA was completely suppressed by dexamethasone in allergy model rats. Provocation with TDI also induced mRNA elevation of histidine decarboxylase, a sole histamine-forming enzyme, followed by the increase of both HDC activity and histamine content in nasal mucosa of allergy model rats. HDC mRNA elevation and increase in both HDC activity and histamine level were almost completely suppressed by dexamethasone. These observations suggest that histamine H₁ receptor up-regulation and increase in histamine level play an important role in allergy through the regulation of histamine signaling.

Keywords: allergic disease, histamine H₁ receptor, histidine decarboxylase, antihistamine, dexamethasone, allergy

Introduction

Histamine is the principal mediator in allergy. Symptoms of allergy are supposed to be greatly influenced by changing the extent of histamine signaling. Histamine signaling is regulated either by expression level of histamine H₁ receptors (H₁Rs) or by histamine level synthesized by histidine decarboxylase (HDC). Elevation of H₁R mRNA was observed in nasal mucosa of allergic rhinitis patients (1, 2) and in primary cultured cells from nasal mucosa of allergic rhinitis patients (3). Elevation of HDC mRNA was also observed in nasal mucosa of allergic rhinitis patients (4). Elevations of H₁R mRNA and HDC mRNA are supposed to be followed by the up-regulation of H₁Rs and increase in HDC activity as well as histamine level, respectively,

leading worsening of allergy symptoms.

In order to elucidate patho-physiological mechanisms of allergic diseases, studies using allergy model animals are indispensable. Various allergy model animals with increased IgE level have been developed (5, 6). These models are beneficial because increased IgE signaling is quite natural in allergy. Their manipulations, however, are so complicated that these are not suitable for studies at the molecular level. So a simple and reproducible animal model of allergic disease was highly required. Toluene 2,4-diisocyanate (TDI), an ingredient of plastic, is suspected as a causative reagent for occupational asthma in plastic factories (7). Previously we developed allergic pathogenesis in guinea pigs (8, 9) and Brown Norway rats (10, 11), and studies of allergy at the molecular level have been achieved using these animal models.

In the present mini-review, increase in gene expression of H₁R and HDC is described using Brown Norway allergy model rats. Suppression of H₁R and HDC gene

*Corresponding author. hfukui@ph.tokushima-u.ac.jp
Published online in J-STAGE
doi: 10.1254/jphs.FM0070184

expression by dexamethasone and antihistamines is also described as targets of therapeutics for allergic diseases.

Methods

Preparation of allergy model rats and assessment of nasal allergy behaviors

Six-week-old male Brown Norway rats were sensitized with TDI (Wako Chemical Co., Tokyo) (Fig. 1A) as described previously (10, 11). Briefly, 10 μ l of a 10% solution of TDI in ethyl acetate was painted bilaterally on the nasal vestibules once a day for five consecutive days (Fig. 1B). This sensitization procedure was then repeated after a 2-day interval. Nine days after the second sensitization, provocation was performed by applying 10 μ l of 10% TDI solution to the nasal

vestibules. Nasal allergy behaviors were assessed using symptom scores as described previously (8). Briefly, the scores were assessed by number of sneezes and extent of watery rhinorrhea on a scale ranging from zero to three during a 10-min period just after TDI provocation (Table 1). Animal experiments were performed as per guidelines approved by the Ethical Committee for Animal Studies, School of Medicine, The University of Tokushima.

Pretreatment with antihistamines and dexamethasone

Both *d*-chlorpheniramine and olopatadine were administered orally 1 h before TDI provocation at a dose of 30 mg/kg, and pretreatment of dexamethasone was performed intraperitoneally 24 h before TDI provocation at a dose of 1.0 mg/kg.

Determination of H1R mRNA and HDC mRNA by real-time quantitative reverse transcriptase polymerase chain reaction (real-time PCR)

H1R mRNA and HDC mRNA were determined as described previously (10, 11). Briefly, total RNA was isolated from rat nasal mucosa using TRIzol Reagent (Invitrogen Corp., Carlsbad, CA, USA). RNA samples were reverse-transcribed to cDNA. TaqMan primers and probe were designed using Primer Express software (Perkin Elmer Applied Biosystems, Foster City, CA, USA). The sequences of the primers and probes have been stated in Table 2. The transcripts were utilized for a 40-cycle 3-step PCR using the GeneAmp 5700 Sequence Detection System (Perkin Elmer Applied Biosystems). For the quantification of gene expression, GAPDH mRNA was used as an endogenous control.

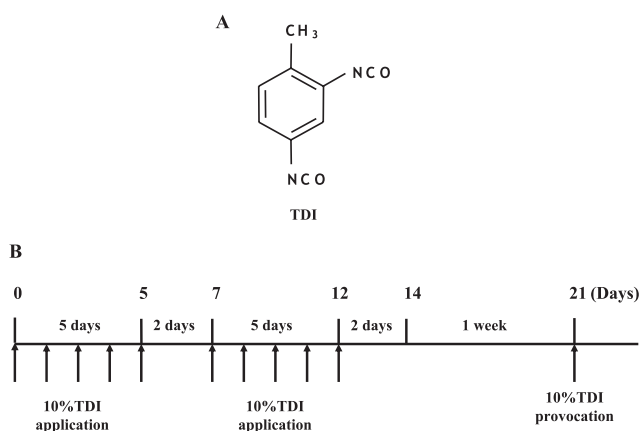


Fig. 1. Chemical structure of toluene 2,4-diisocyanate (TDI) (A) and schedule for preparation of TDI-sensitized allergy model rats (B). Each arrow indicates application of 10% TDI into rat vestibule once a day.

Table 1. Nasal score criteria of Brown-Norway allergy model rat sensitized with toluene 2,4-diisocyanate (TDI)

Score	0	1	2	3
Sneezing	(-)	1–4	5–11	>12
Watery rhinorrhea	(-)	at the nostril	intermediate	dropping discharge

Nasal scores were assessed by counting the above-mentioned two parameters for 10 min just after provocation with TDI.

Table 2. Primer and probe sequences used for real-time RT-PCR

Primer/probe name		Sequence
H1R mRNA	Sense primer	5'-CAG AGG ATC AGA TGT TAG GTG ATA GC-3'
	Anti sense primer	5'-AGC GGA GCC TCT TCC AAG TAA-3'
	Probe	FAM-CTT CTC TCG AAC GGA CTC AGA TAC CAC C-TAMRA
HDC mRNA	Sense primer	5'-GCA GCA AGG AAG AAC AAA ATC C-3'
	Anti sense primer	5'-CAA CAA GAC GAG CGT TCA GAG A-3'
	Probe	FAM-AAAGCGCATGAGCCCAATGCTGCTGAT-TAMRA

Radioligand binding assay

H1R was determined by [^{3} H]mepyramine (NEN Life Science Products, Boston, MA, USA) binding assay as described previously (10). In brief, nasal mucosa was homogenized in 10 volumes of ice-cold 50 mM Na₂/K-phosphate buffer (37.8 mM Na₂HPO₄, 12.2 mM KH₂PO₄, pH 7.4) and then centrifuged at 1800 rpm for 30 min at 4°C. The pellet was resuspended in ice-cold 50 mM Na₂/K-phosphate buffer and served as membrane sample for radioligand binding assay. Membranes were incubated with 4 nM of [^{3} H]mepyramine in the absence or presence of 10 μ M triprolidine for 60 min at 25°C in a final volume of 500 μ l. Reaction was terminated by rapid vacuum filtration through Whatman GF/B filters (Maidstone, UK) presoaked with 1% polyethylenimine. Filters were then soaked in 10 ml of Aquasol-2 (Packard Instrument Inc., Meriden, CT, USA), kept overnight in a dark place, and the radioactivity trapped on the filters was counted in a liquid scintillation counter. Specific binding was defined as the radioactivity bound after subtraction of non-specific binding as defined by 10 μ M triprolidine. Protein concentration was determined by the BCA protein assay reagent (Sigma, St. Louis, MO, USA) using BSA as a standard.

H1R promoter assay

HeLa cells cultured in 12-well culture plates were co-transfected with Human H1R reporter plasmid (pH1R) and the internal control plasmid pRL-MPK containing cDNA encoding mouse pyruvate kinase and Rluc (Promega, Madison, WI, USA) by the ratio of 100:1 using PolyFect transfection reagent (Qiagen, Tokyo) according to manufacturer's instructions. After 5 h, the medium was replaced with 1 ml of serum free medium and starved for 24 h. The cells were stimulated with appropriate reagents for the indicated time in the same medium. Where appropriate, antagonists were added 30 min prior to histamine stimulation. After stimulation, cells were washed twice with 500 μ l of ice-cold phosphate-buffered saline and lysed with 100 μ l of passive lysis buffer (Promega). The lysate was frozen for at least 3 h at -85°C and then thawed at room temperature. The luciferase activity was determined by the dual-luciferase reporter assay system (Promega) as per the manufacturer's protocol. Luminescence was measured by photoluminescence reader BLR 302 (Aloka, Tokyo). The measurement was integrated over 20 s with no delay.

Determination of HDC activity and histamine content

Samples for the determination of HDC activity and histamine content were prepared by the method of Yamada et al. (12). HDC activity was determined by the method of Watanabe et al. (13). Histamine content was

measured using a high-performance liquid chromatography with a cation exchanger and an automated o-phthalaldehyde fluorometric detection system in accordance with the method of Yamatodani et al. (14).

Statistical analyses

The results are presented as mean $\pm$ S.E.M. All *P* values were determined by using one-way ANOVA and Fisher's paired least-significant difference test. *P* values less than 0.01 were considered significant.

Results

Increase in allergic symptom after provocation and their suppression by therapeutics for allergy

Severe nasal swelling and watery rhinorrhea was observed in Brown Norway allergy model rats after the provocation with TDI (Fig. 2, right panel). The number of sneezes was also highly increased. Nasal scores based on number of sneezes and watery rhinorrhea increased remarkably compared with the control animals (Fig. 3).

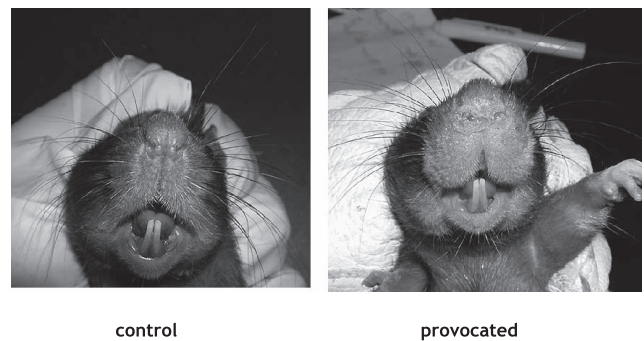


Fig. 2. Nasal swelling and rhinorrhea of Brown-Norway allergy model rat after provocation with 10% toluene 2,4-diisocyanate. Control (left panel) and allergy model rat (right panel).

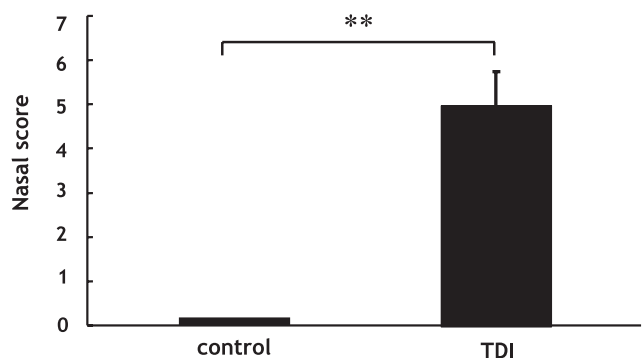


Fig. 3. Increase in nasal score of Brown-Norway allergy model rats after provocation. Score was assessed for 10 min just after provocation according to Table 1. Data each represent a mean $\pm$ S.E.M. ***P* < 0.01, *n* = 4.

Table 3. *d*-Chlorpheniramine and dexamethasone suppress nasal symptoms in allergy model rats

	Control	TDI control	TDI + <i>d</i> -chlorpheniramine	TDI + dexamethasone
Nasal score	0	5.0 ± 0.3	2.2 ± 0.4**	3.5 ± 0.5**

The values are the mean ± S.E.M. ** $P < 0.01$ vs TDI-sensitized group.

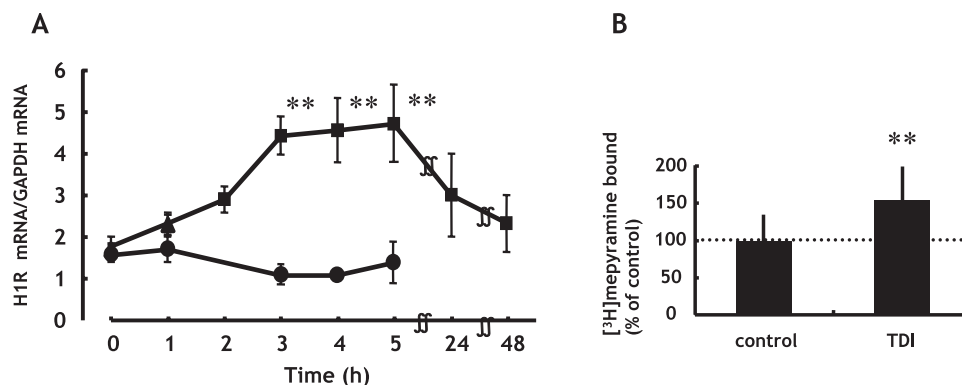


Fig. 4. Elevation of histamine H₁ receptor mRNA (A) and H₁ receptor up-regulation (B) in nasal mucosa of Brown-Norway allergy model rats after provocation. In panel A, mRNA levels after provocation and control are represented with closed squares and closed circles, respectively. Data each represent a mean ± S.E.M. ** $P < 0.01$ vs control, $n = 4$.

Increase in nasal score was significantly suppressed by the administration of *d*-chlorpheniramine prior to TDI provocation (Table 3). Although pretreatment of dexamethasone also suppressed the elevation of symptom score, the extent of suppression was smaller than *d*-chlorpheniramine. The score assessment was achieved for 10 min after the provocation, and the blockade by antihistamine of H₁R signaling seemed to suppress the symptom more strongly.

Elevation of H1R mRNA and H1R protein level

Elevation of H₁R mRNA was induced after the provocation with TDI (Fig. 4A), and this led to H₁R up-regulation (Fig. 4B). It is well known that H₁R is desensitized after repetitive stimulation of H₁R, and down-regulation of H₁R is induced as a final step of desensitization (15–17). Therefore expression level of H₁R is the result of up-regulation due to H₁R gene expression and down-regulation due to the desensitization. H₁R up-regulation actually overcame the down-regulation in Brown Norway allergy model rats.

H₁R mRNA up-regulation was partially but significantly suppressed by *d*-chlorpheniramine and olopatadine, antihistamines, in allergy model rats (Fig. 5). This data suggested that H₁R mRNA up-regulation is mediated through H₁R signaling and some other factors may also be involved in this up-regulation. We previously demonstrated the H₁R-mediated elevation of H₁R mRNA in HeLa cells. As shown in Fig. 6A, Ro-31-

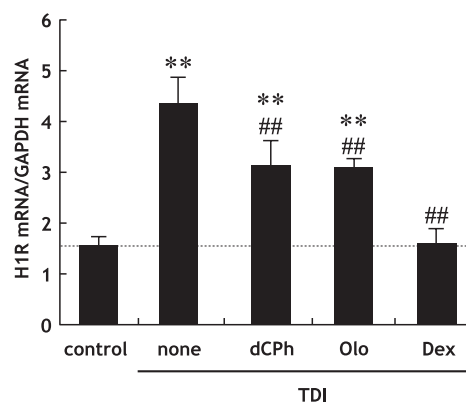


Fig. 5. Suppression of histamine H₁ receptor mRNA elevation by antihistamines (*d*-chlorpheniramine: dCPh, 30 mg/kg, i.p.; olopatadine: Olo, 30 mg/kg, i.p.) and dexamethasone (Dex, 1 mg/kg i.p.) in nasal mucosa of Brown-Norway allergy model rats after provocation. Data each represent a mean ± S.E.M. ** $P < 0.01$ vs control, $n = 4$ and ## $P < 0.01$ vs none, $n = 4$.

8220, a nonspecific PKC inhibitor, completely inhibited histamine-induced up-regulation of H₁R gene expression. Similar results were obtained in the H₁R promoter assay also (Fig. 6B). These data indicate that the PKC pathway is involved in H₁R-mediated H₁R gene expression. Many PKC isoforms have been reported. Some are involved in desensitization of various receptors. The PKC- α isoform was reported to engage desensitization of α_1 -adrenoceptors (18) and μ -opioid receptors (19). Recently, we observed that an inhibitor of the PKC- δ

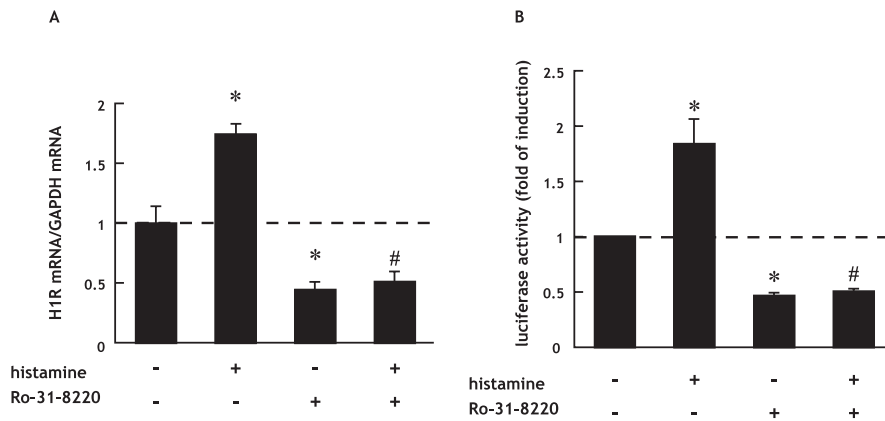


Fig. 6. Effect of PKC inhibitor on H1R mRNA expression (A) and H1R promoter activity (B). A: HeLa cells were pretreated with Ro-31-8220 1 h prior to histamine (10 μ M) treatment. The abundance of H1R mRNA was determined by quantitative real-time RT-PCR 3 h after histamine treatment and was normalized by GAPDH mRNA levels. The data are each presented as the mean $\pm$ S.E.M. from four independent assays in triplicate. * P <0.05 vs control, # P <0.05 vs histamine. B: HeLa cells were co-transfected with pH1R and pRL-MPK vector as described in Methods and treated with Ro-31-8220 30 min prior to histamine treatment. After 4 h of histamine stimulation, luciferase activity was determined by the dual-luciferase assay system. Data are each expressed as fold of the control luminescence $\pm$ S.E.M. from four independent assays in triplicate. * P <0.05 vs control, # P <0.05 vs histamine.

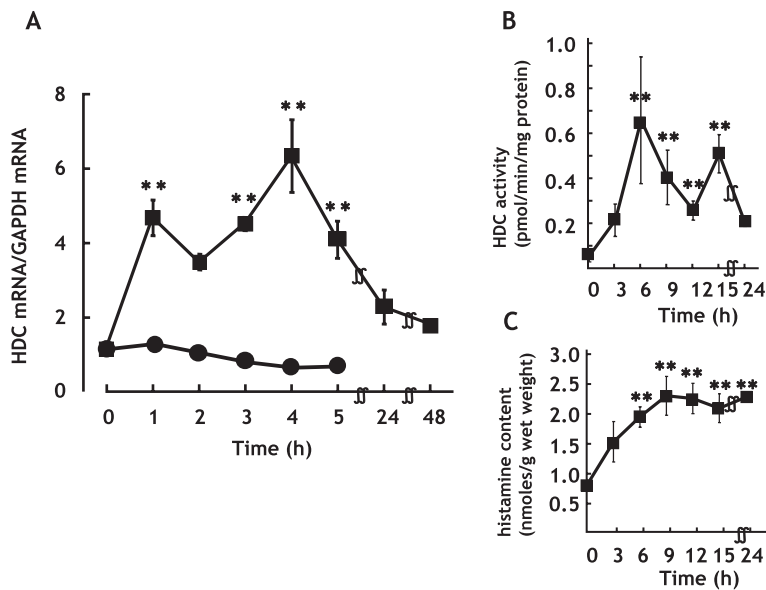


Fig. 7. Elevation of histidine decarboxylase (HDC) mRNA (A), HDC activity (B), and histamine level (C) in nasal mucosa of Brown-Norway allergy model rats after provocation with 10% toluene 2,4-diisocyanate. In panel A, mRNA levels after provocation with TDI and control are represented by closed squares and closed circles, respectively. In panels B and C, HDC activity and histamine level are represented by closed circles. Data each represent a mean $\pm$ S.E.M. ** P <0.01 vs control in panel A, n = 4 and ** P <0.01 vs values at 0 h, n = 4 in panels B and C.

isoform completely suppressed H1R-mediated H1R gene expression (unpublished data). PKC- α and PKC- δ isoforms seem to exert opposite regulatory functions in receptor signaling. On the other hand, H1R up-regulation was completely suppressed by dexamethasone (Fig. 5). There are many reports about suppression of various gene expressions by dexamethasone (20, 21). Genes related to inflammation seem to be suppressed by dexamethasone. Dexamethasone did not show any strong advantageous effect on symptoms just after TDI provocation (Table 2), but thought to induce long-term suppression of H1R signaling through suppression of H1R expression.

Dexamethasone suppresses TDI-induced elevation of HDC mRNA, HDC activity and histamine level in allergic rats

Elevation of HDC mRNA was also induced in nasal mucosa of Brown Norway allergy model rats after provocation with TDI (Fig. 7A). Increase in HDC activity and histamine level also followed (Fig. 7: B and C). Increase in histamine level in nasal mucosa is thought to increase histamine signaling and worsen allergy symptoms. Elevation of HDC mRNA, HDC activity, and histamine level were all suppressed strongly by dexamethasone (Fig. 8). Long-term suppression of histamine signaling seems to be exerted through

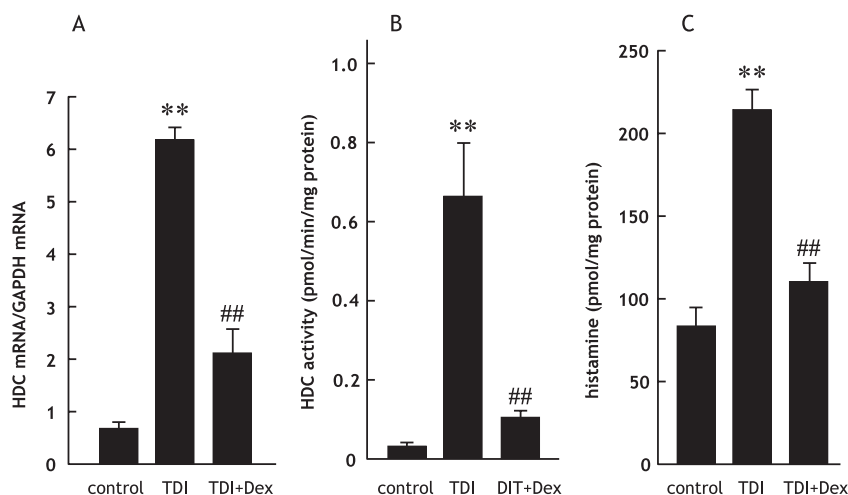


Fig. 8. Dexamethasone suppresses the elevation of histidine decarboxylase (HDC) mRNA (A), HDC activity (B), and histamine level (C) in nasal mucosa of Brown Norway allergy model rats after provocation with 10% toluene 2,4-diisocyanate (TDI). Dexamethasone (Dex) at a dose of 1 mg/kg was administered (intraperitoneally) 24 h prior to TDI provocation. HDC mRNA, HDC activity, and histamine were determined after 4, 6, and 12 h, respectively, of TDI provocation. Data each represent a mean $\pm$ S.E.M. ** $P < 0.01$ vs control, $n = 4$ and ## $P < 0.01$ vs TDI, $n = 4$.

lowering not only the H1R expression level but also the histamine level.

Concluding remarks

Levels of H1R mRNA and HDC mRNA were highly elevated in nasal mucosa of Brown Norway allergy model rats after the provocation with TDI (Figs. 4A and 8A). Up-regulation of H1R and increase in HDC activity and histamine level also followed these increases (Figs. 4B, 8B, and 8C). Increase in gene expression of H1R and HDC is suggested to play an important role in

increasing histamine signaling and worsening allergy symptoms (Fig. 9). Antihistamines are used to suppress immediate allergy symptoms by blocking H1R signaling. Although the effect was partial, antihistamines also suppressed H1R mRNA elevation. The data indicate that the therapeutic effect of antihistamines is long lasting. Dexamethasone completely suppressed elevation of both H1R mRNA and HDC mRNA. Pretreatment of dexamethasone could maintain H1R and histamine at lower levels. The effect of dexamethasone on histamine signal-related gene expression is thought long lasting, compared with antihistamines.

Only little attention has been paid to the long-term increase in histamine signaling through increase in histamine signal-related gene expression. To my best knowledge, no attention has been paid to development of new therapeutics for allergy by suppressing gene expression of H1R and HDC. Antihistamines actually showed some effect on H1R gene expression (Fig. 5). Although dexamethasone showed a strong effect on the two key genes (Figs. 5 and 8), use of this compound is limited due to the side effect. Therefore other medicines possessing the suppressing effect on elevation of H1R mRNA and HDC mRNA with less side-effect is worth developing. Elevation of H1R mRNA was strongly suppressed by suplatast tosilate and other natural source medicines including *Sho-seiryu-to* (A. Das et al., unpublished data) and *Kujin* (*Sophora flavescens*) (S. Dev et al., unpublished data). Long-term treatment of antihistamines also showed potent suppressing effect on H1R gene expression (M. Hatano et al., unpublished data). In the case of HDC gene expression, some antihistamines revealed to possess strong suppressing effects (A. Das et al., unpublished data). Although the mechanism of HDC gene expression is complicated and has not been extensively investigated, the molecular target of some

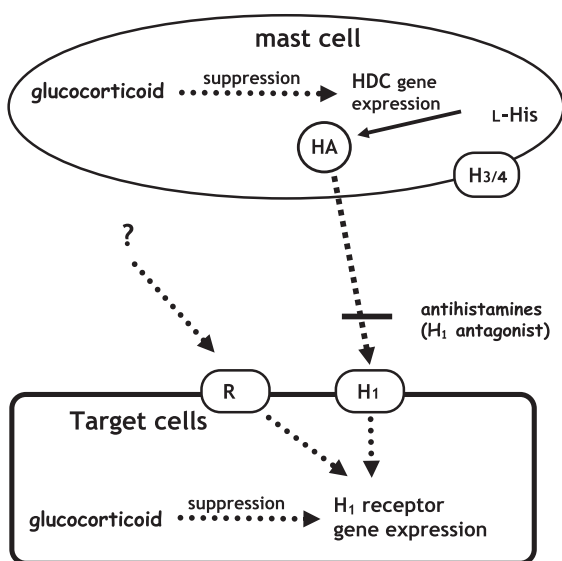


Fig. 9. Histamine synthesis by histidine decarboxylase (HDC) in mast cells and H1R-mediated signal transduction in the target cells. Dexamethasone suppresses gene expression of both HDC and H1 receptor. Antihistamines suppress not only H1 receptor-mediated patho-physiological responses but also H1 receptor gene expression.

antihistamines remains to be elucidated. Finally, targets of these medicines are quite different. On the other hand each allergic patient could show different patterns of H1R gene expression and HDC gene expression. Thus, a therapeutic method for each medicine should be developed as a tailor-made therapy.

Acknowledgments

I highly appreciate the following collaborators: A. Miyoshi, Y. Murata, Y. Kitamura, R. Mishima, K. Miyoshi, S. Yoshimura, M. Kodama, S. Horinaga, A.K. Das, K. Kawazoe, Y. Takaishi, and N. Takeda (The University of Tokushima); K. Fujimoto (Hiroshima University); and K. Maeyama (Ehime University).

References

- Iriyoshi N, Takeuchi N, Yuta A, Ukai A, Sasakura Y. Increased expression of histamine H₁ receptor mRNA in allergic rhinitis. *Clin Exp Allergy*. 1996;26:379–385.
- Dinh QT, Cryer A, Dinh S, Peiser C, Wu S, Springer J, et al. Transcriptional up-regulation of histamine receptor-1 in epithelial, mucus and inflammatory cells in perennial allergic rhinitis. *Clin Exp Allergy*. 2005;35:1443–1448.
- Hamano N, Terada N, Maesako K, Ikeda T, Fukuda S, Wakita J, et al. Expression of histamine receptors in nasal epithelial cells and endothelial cells – the effects of sex hormones. *Int Arch Allergy Immunol*. 1998;115:220–227.
- Hirata N, Takeuchi K, Ukai K, Sakakura Y. Expression of histidine decarboxylase messenger RNA and histamine N-methyltransferase messenger RNA in nasal allergy. *Clin Exp Allergy*. 1999;29:76–83.
- Takahashi N, Aramaki Y, Tsuchiya S. Allergic rhinitis model with Brown Norway rat and evaluation of antiallergic drugs. *J Pharmacobiodyn*. 1990;13:414–420.
- Kasper M, Sims G, Koslowski R, Kuss H, Thuemmler M, Fehrenbach H, et al. Increased surfactant protein D in rat airway goblet and Clara cells during ovalbumin-induced allergic airway inflammation. *Clin Exp Allergy*. 2002;32:1251–1258.
- Krone CA. Diisocyanates and nonoccupational disease: a review. *Arch Environ Health*. 2004;59:306–316.
- Abe Y, Takeda N, Irifune M, Ogino S, Kalubi B, Imamura I, et al. Effects of capsaicin desensitization on nasal allergy-like symptoms and histamine release in the nose induced by toluene diisocyanate in guinea pigs. *Acta Otolaryngol*. 1992;112:703–709.
- Abe Y, Ogino S, Irifune M, Imamura I, Liu YQ, Fukui H, et al. Histamine content, synthesis and degradation in nasal mucosa and lung of guinea-pigs treated with toluene diisocyanate (TDI). *Clin Exp Allergy*. 1993;23:512–517.
- Kitamura Y, Miyoshi A, Murata Y, Kalubi B, Fukui H, Takeda N. Effect of glucocorticoid on upregulation of histamine H₁ receptor mRNA in nasal mucosa of rats sensitized by exposure to toluene diisocyanate. *Acta Otolaryngol*. 2004;124:1053–1058.
- Kitamura Y, Das AK, Murata Y, Maeyama K, Dev S, Wakayama Y, et al. Dexamethasone suppresses histamine synthesis by repressing both transcription and activity of HDC in allergic rats. *Allergol Int*. 2006;55:279–286.
- Yamada M, Watanabe T, Harino S, Fukui H, Wada H. The effects of protease inhibitors on histidine decarboxylase activities and assay of enzyme in various rat tissues. *Biochim Biophys Acta*. 1980;615:458–464.
- Watanabe T, Nakamura (Fukui) H, Liang LY, Yamatodani A, Wada H. Partial purification and characterization of L-histidine decarboxylase from foetal rats. *Biochem Pharmacol*. 1979;28:1149–1155.
- Yamatodani A, Fukuda H, Wada H, Iwaeda T, Watanabe T. High-performance liquid chromatographic determination of plasma and brain histamine without previous purification of biological samples: cation-exchange chromatography coupled with post-column derivatization fluorometry. *J Chromatogr*. 1985;344:115–123.
- Smit MJ, Timmerman H, Hijzelendoorn JC, Fukui H, Leurs R. Regulation of the histamine H₁ receptor stably expressed in Chinese hamster ovary cells. *Br J Pharmacol*. 1996;117:1071–1080.
- Horio S, Ogawa M, Kawakami N, Fujimoto K, Fukui H. Identification of amino acid residues responsible for agonist-induced down-regulation of histamine H₁ receptors. *J Pharmacol Sci*. 2004;94:410–419.
- Horio S, Kato T, Ogawa M, Fujimoto K, Fukui H. Two threonine residues and two serine residues in the second and third loops are both involved in histamine H₁ receptor down-regulation. *FEBS Lett*. 2004;573:226–230.
- Alcantara-Hernandez R, Leyva-Illades D, Garcia-Sainz JA. Protein kinase C- α (1b)-adrenoceptor coimmunoprecipitation: effect of hormones and phorbol myristate acetate. *Eur J Pharmacol*. 2001;419:9–13.
- Kramer HK, Simon EJ. Role of protein kinase C (PKC) in agonist-induced mu-opioid receptor down-regulation: II. Activation and involvement of the alpha, epsilon, and zeta isoforms of PKC. *J Neurochem*. 1999;72:594–604.
- Smoak KA, Cidlowski JA. Mechanisms of glucocorticoid receptor signaling during inflammation. *Mech Ageing Dev*. 2004;125:697–706.
- Novac N, Baus D, Dostert A, Heinzl T. Competition between glucocorticoid receptor and NFkappaB for control of the human FasL promoter. *FASEB J*. 2006;20:1074–1081.