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Inhibitory effect of a novel quinolinone derivative, TA-270, on asthmatic inflammatory responses in sensitized guinea pigs

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Abstract

TA-270 (4-hydroxy-1-methyl-3-octyloxy-7-sinapinoylamino-2(1*H*)-quinolinone), a novel quinolinone derivative, was designed as an antioxidant to scavenge reactive oxygen species. Here, we investigated the effects of TA-270, in comparison with several antiasthmatic drugs, on asthmatic responses as induced by ovalbumin in sensitized guinea pigs. When orally administered 1 h before and 3 h after the antigen challenge, TA-270 at 10 mg/kg and higher doses significantly inhibited both immediate and late responses in airway resistance induced by the antigen. The inhibitory effects were comparable to or superior, at least under the present experimental conditions, to those of several clinically used antiasthmatic drugs. Furthermore, TA-270, in a dose-dependent manner, reduced accumulation of pulmonary inflammatory cells, especially eosinophils, and significantly reversed the airway hyperresponsiveness to acetylcholine 24 h after the antigen challenge. These results suggest that TA-270 may be of therapeutic use for bronchial asthma.

Keywords: TA-270; Asthmatic response; Bronchoalveolar eosinophilia; Airway hyperresponsiveness

1. Introduction

Bronchial asthma is an atopic disease characterized by bronchoconstriction, an influx of inflammatory cells into the airway, and bronchial hyperresponsiveness. Many asthmatic patients show an immediate response that is accompanied by the bronchoconstriction in the airway by provocation of an inhaled allergen, but the bronchoconstriction usually returns to the basal level within a few hours. However, approximately 60% of patients frequently show a late bronchoconstriction response again 4–12 h after provocation (Booij-Noord et al., 1971). The immediate-airway response in association with the constriction of airway smooth muscle cells may be mediated by several mediators, such as histamine and peptide leukotrienes, which are released from inflammatory cells, especially mast cells (Holgate and Kay, 1985). The late response is associated with bronchial hyperresponsiveness to a wide variety of stimuli (Cartier et al., 1982). Although the mechanisms underlying the late-airway response are still poorly understood, an influx of inflammatory cells, especially eosinophils, into the bronchial lumen has been suggested to be one of the important events in the induction of the late response (Barnes et al., 1998). Actually, eosinophils and other inflammatory cells can release several mediators that may cause bronchoconstriction and hyperresponsiveness. Thus, agents that can improve these events, including bronchial hyperresponsiveness and infiltration of eosinophils, may be a great benefit for the therapy of bronchial asthma.

TA-270 (4-hydroxy-1-methyl-3-octyloxy-7-sinapinoylamino-2(1*H*)-quinolinone) was designed as one of the antioxidants that potently scavenges reactive oxygen species. The reactive oxygen species has been shown to be involved in inflammatory diseases including bronchial asthma (Barnes and Belvisi, 1993; Repine et al., 1997). We therefore examined the effects of TA-270, in compari-

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son with authorized antiasthmatic drugs, on antigen-induced immediate- and late-airway responses, accumulation of pulmonary inflammatory cells, and airway hyperresponsiveness in sensitized guinea pigs.

2. Material and methods

2.1. Animals

Male Hartley guinea pigs (Kyudo, Kumamoto, Japan; weight range 350–500 g and age range 4–6 weeks) were housed in an air-conditioned room at 18 ± 2°C with 55 ± 10% humidity and a 12 h light–dark cycle. The animals were given food and water ad libitum.

2.2. Immediate- and late-phase airway responses in actively sensitized guinea pigs

The guinea pigs were sensitized with 1% ovalbumin aerosol for 10 min once a day for 8 consecutive days. The aerosol was generated with an ultra-sonic nebulizer (NE-U12, OMRON, Tokyo, Japan). One week after the last sensitization, an antigen provocation was performed with the inhalation of 2% ovalbumin for 1 min for the immediate-phase airway response and for 5 min for the late response. Pyrilamine maleate (10 mg/kg, i.p.), a histamine H_1 receptor antagonist, was given 30 min before an ovalbumin challenge to protect the animals from anaphylactic shock. Methyrapone (10 mg/kg, i.p.), a cortisol synthesis inhibitor, was given 24 and 1 h before the antigen challenge. A specific resistance in the airway (sR_{aw}) as induced by the antigen inhalation was measured with a double-flow plethysmograph technique (Pennock et al., 1979) using non-invasive respiratory measurements (Pulmos-1, M.I.P.S., Osaka, Japan). The sR_{aw} was monitored before and at the indicated time (1 min, 2, 4, 5, 6, 7, 8 and 23–24 h) after the ovalbumin challenge. In contrast to the immediate response, which was sharp and peaked at around 1 min, there was not an obvious peak in the late phase as shown later (Fig. 2). Henceforth, the immediate response was defined as an increase in sR_{aw} at 1 min and the late response as an increase in area under the curve (AUC) for sR_{aw} between 4 and 8 h after antigen challenge.

2.3. Antigen-induced infiltration of inflammatory cells into bronchoalveolar lavage fluid

The guinea pigs sensitized with ovalbumin and treated as described in Section 2.2 were killed with sodium pentobarbital (50 mg/kg i.p.) 24 h after the antigen challenge. The lungs of the animals were washed twice with 5 ml of saline. The lavage fluid was centrifuged (1100 rpm) for 5 min, and the cell pellet was resuspended in 1 ml of saline. The total cells were counted in a hemocytometer. The smears were made of cell suspensions and stained with May–Gruendward–Giemsa stain. Each cell per smear was counted and the total population of each cell type was calculated.

2.4. Antigen-induced airway hyperresponsiveness to acetylcholine in actively sensitized guinea pigs

The guinea pigs were sensitized with ovalbumin and treated as described in Section 2.2. Acetylcholine was spontaneously inhaled, 22–26 h after the ovalbumin challenge, with the sequential concentration of 0.0625, 0.125, 0.25, 0.5, 1, and 2 mg/ml for 1 min until a 100% increase in sR_{aw} was observed as described in Section 2.2. Airway hyperresponsiveness to acetylcholine was expressed as PC_{100}ACh, a provocative concentration of acetylcholine aerosols causing a 100% increase in the airway resistance.

2.5. Drugs and chemicals

TA-270 (4-hydroxy-1-methyl-3-octyloxy-7-sinapinoylamino-2(1*H*)-quinolinone) (Fig. 1) was synthesized by Dainippon Ink and Chemicals: the procedure of synthesis will be described in detail elsewhere. Azelastine (Isogai and Hasegawa, 1995), pranlukast (Toda et al., 1986) and seratrodast (Terao, 1989) were synthesized as previously described. Other chemicals were obtained from following sources: tranilast (Shiratori Pharmaceutical, Japan); carboxymethyl cellulose (Wako, Japan); ovalbumin (Grade V), pyrilamine maleate, and metyrapone (Sigma, USA); acetylcholine chloride (Daiichi Pharmaceutical, Japan); May–Gruenward solution and Giemsa solution (Muto Pure Chemical, Japan). All antiasthmatic drugs were suspended in a 0.5% carboxymethyl cellulose solution and were given orally in a volume of 5 ml/kg, 1 h before and 3 h after antigen challenge.

2.6. Statistical analysis

The results are expressed as the means ± S.E. unless otherwise specified. When data involved three or more groups, Dunnett's multiple test was used. Student's *t*-test was used for analyzing the difference between two groups. A significant difference was accepted when $P < 0.05$.

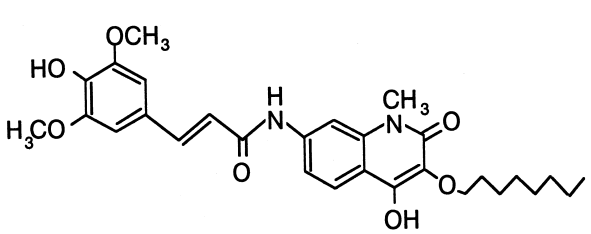

Fig. 1. Structure of TA-270 (4-hydroxy-1-methyl-3-octyloxy-7-sinapinoylamino-2(1*H*)-quinolinone).

3. Results

3.1. Immediate- and late-phase-airway responses in conscious guinea pigs

When actively sensitized guinea pigs were challenged with ovalbumin, biphasic airway responses were observed. Thus, the first phase (immediate phase) occurring at around 1 min after the antigen challenge showed a remarkable increase in a specific resistance in the airway (sR_{aw}). The sR_{aw} rather rapidly declined almost to the basal level about 2 h after the antigen challenge. Four hours or later, however, the sR_{aw} again gradually increased, although the maximal response was less than that in the immediate phase (Fig. 2). We examined the effect of TA-270 on the biphasic airway responses. In this experiment, TA-270 (50 mg/kg p.o.) was administered 1 h before and 3 h after the ovalbumin challenge. As shown in Fig. 2, TA-270 clearly inhibited both immediate and late responses.

In Fig. 3, we examined the dose–response effects of TA-270 on the immediate- and late-airway responses and compared these effects with those obtained by several other antiasthmatic drugs. TA-270 was effective even at 10 mg/kg in inhibiting both immediate and late responses, and the effective dose was comparable with that of seratrodast, a thromboxane A_2 receptor antagonist. All other antiasthmatic drugs including tranilast (a mast cell stabilizer), azelastine (an H_1 histamine receptor antagonist), and pranlukast (a peptide leukotriene receptor antagonist) were also effective at appropriate doses against both immediate and late responses; however, the inhibition pattern was not always identical for both responses. For example, azelastine significantly and greatly inhibited the immediate response, while it only slightly (the effect was not statistically significant) inhibited the late response (Fig. 3).

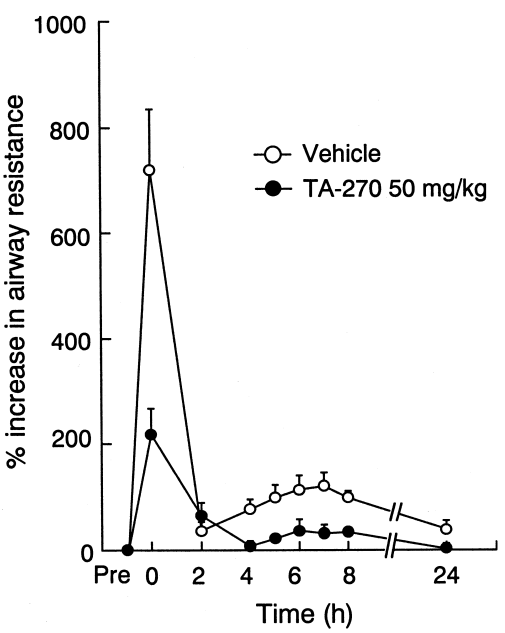

Fig. 2. Time course of the increase in airway resistance (sR_{aw}) induced by ovalbumin. TA-270 (50 mg/kg p.o.) and its vehicle were administered orally 1 h before and 3 h after an ovalbumin challenge. The results are expressed as the means ± S.E. of the percent increase in sR_{aw} over the baseline control from eight animals.

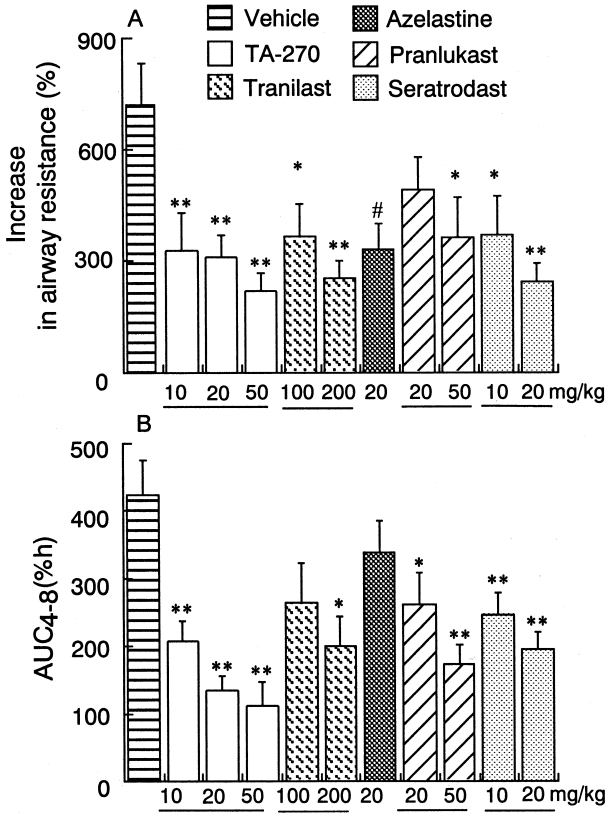

Fig. 3. The effect of TA-270 and other antiasthmatic drugs on the immediate-airway response (A) and late-airway response (B). The immediate response is expressed as a percentage increase in airway resistance (sR_{aw}) over the baseline at 1 min after the ovalbumin challenge. The late response is expressed as a percentage increase in the area under the curve of sR_{aw} between 4 and 8 h after the antigen challenge. TA-270, other drugs and their vehicle were administered both 1 h before and 3 h after the ovalbumin challenge. The results are expressed as the means ± S.E. from eight animals. $^{*}P < 0.05$, $^{**}P < 0.01$, significantly different from the vehicle by Dunnett's multiple test. $\#P < 0.05$, significantly different from the vehicle by Student's *t*-test.

3.2. Antigen-induced infiltration of inflammatory cells into bronchoalveolar lavage fluid

Ovalbumin inhalation clearly induced infiltration of several inflammatory cells including macrophages, neutrophils and eosinophils into bronchoalveolar lavage fluid 24 h after the antigen challenge (Table 1). We next examined the effect of TA-270, in comparison with other antiasthmatic drugs, on the antigen-induced infiltration of the inflammatory cells in bronchoalveolar lavage fluid.

Table 1
Effect of TA-270 and other antiasthmatic drugs on infiltration of inflammatory cells into bronchoalveolar lavage fluid

Drug	Dose (mg/kg)	Cell number ($\times 10^5$ cells)				
		Total cells	Macrophages	Neutrophils	Eosinophils	Lymphocytes
Saline inhalation	–	25.6 ± 2.7	9.1 ± 2.0	6.7 ± 1.7	7.5 ± 2.0	2.4 ± 1.0
Antigen challenge						
Vehicle	–	86.0 ± 13.6	24.7 ± 6.9	24.8 ± 8.8	32.8 ± 6.3	3.7 ± 0.6
TA-270	10	64.5 ± 10.0	12.7 ± 2.0	26.5 ± 10.0	21.5 ± 2.7	3.8 ± 0.8
	20	52.1 ± 11.0	13.0 ± 3.4	17.8 ± 6.8	18.0 ± 2.9[a]	3.2 ± 0.7
	50	48.3 ± 8.3	11.8 ± 2.7	17.4 ± 5.9	16.6 ± 3.7[a]	2.4 ± 0.6
Tranilast	100	72.7 ± 15.8	19.6 ± 5.3	24.2 ± 5.9	24.8 ± 5.9	4.2 ± 0.8
	200	73.6 ± 17.4	20.9 ± 6.7	25.9 ± 13.0	22.8 ± 4.7	4.0 ± 0.7
Azelastine	10	75.1 ± 6.8	13.6 ± 2.4	29.8 ± 4.0	28.8 ± 6.8	2.9 ± 0.5
Pranlukast	20	73.4 ± 7.7	14.0 ± 3.4	26.5 ± 5.9	29.3 ± 6.3	3.7 ± 0.3
	50	52.0 ± 7.7[a]	12.5 ± 3.0	14.6 ± 3.1	21.6 ± 4.7	3.3 ± 0.8
Seratrodast	10	81.8 ± 16.7	19.3 ± 5.0	26.3 ± 6.3	31.5 ± 6.9	4.8 ± 0.7
	20	86.4 ± 17.4	22.4 ± 4.9	28.1 ± 7.7	31.4 ± 7.3	4.5 ± 0.8

TA-270 and other drugs were administered both 1 h before and 3 h after ovalbumin challenge. Results are expressed as means ± S.E. from eight animals.
[a] $P < 0.05$, significantly different from vehicle by Dunnett's multiple test.

The TA-270 treatment significantly inhibited the accumulation of eosinophils, and it also tended to inhibit the infiltration of total cells, macrophages and neutrophils. Among the antiasthmatic drugs examined, only pranlukast showed a significant effect on the infiltration of total cells. Even though a significant effect was not observed, however, this antiasthmatic drug, like TA-270, also tended to inhibit the infiltration of macrophages and neutrophils. In other antiasthmatic drugs, with the exception of azelastine effect on macrophage infiltration, we hardly detected an inhibitory effect on the antigen-induced infiltration (Table 1).

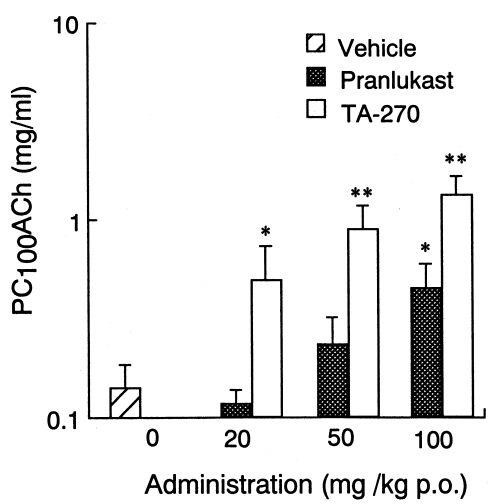

Fig. 4. The effects of TA-270 and pranlukast on airway hyperresponsiveness to acetylcholine. The results are expressed as the means ± S.E. of PC_{100}ACh, a provocative concentration of acetylcholine aerosol causing a 100% increase in the airway resistance in eight animals. TA-270 and pranlukast were administered both 1 h before and 3 h after the ovalbumin challenge. The PC_{100}ACh value in control animals was approximately 1.2 mg/ml. $^{*}P < 0.05$, $^{**}P < 0.01$, significantly different from vehicle by Dunnett's multiple test.

3.3. Antigen-induced airway hyperresponsiveness to acetylcholine

Acetylcholine inhalation causes an increase in airway resistance even in normal guinea pigs. In actively sensitized guinea pigs, the sensitivity of acetylcholine to cause an increase in airway resistance is markedly enhanced. Thus, the concentration to induce a 100% increase in the airway resistance (PC_{100}ACh) was decreased from 1.2 mg/ml in normal guinea pigs to about 0.15 mg/ml in the sensitized animals (Fig. 4). The hyperresponsiveness to acetylcholine in the sensitized guinea pigs was reversed by a pretreatment of animals with TA-270 in a dose-dependent manner. Since the inhibitory pattern against infiltration of inflammatory cells in bronchoalveolar lavage fluid was similar each other between TA-270 and pranlukast (Table 1), we also examined pranlukast effect. This drug was also effective for the reversal of hyperresponsiveness to acetylcholine, but a much higher dose was required to achieve the same effect as that obtained by TA-270 (Fig. 4).

4. Discussion

In the present study, we showed that a novel quinolinone derivative, TA-270, exerted potent inhibitory actions against immediate- and late-phase bronchoconstriction in guinea pigs sensitized with ovalbumin. The late-airway response is usually accompanied by an increase in airway responsiveness to nonspecific stimuli in human asthma (Cockcroft and Murdock, 1987; Cartier et al., 1982) and in animal asthma models (Marsh et al., 1985). In both cases, influx in the accumulation of inflammatory cells into the airway has been demonstrated (De Monchy et al., 1985). Among inflammatory cells, especially eosinophils have

been suggested to play an important role in airway hyperresponsiveness (Sanjar et al., 1990; Tarayre et al., 1990). TA-270 was also effective in inhibiting the pulmonary accumulation of eosinophils and airway hyperresponsiveness to acetylcholine in the sensitized guinea pigs.

These antiasthmatic actions of TA-270 were comparable to or superior, at least under the present experimental conditions, to those obtained by other antiasthmatic drugs including pranlukast (a peptide leukotriene receptor antagonist), seratrodast (a thromboxane A_2 receptor antagonist), tranilast (a mast cell stabilizer), and azelastine (an H_1 histamine receptor antagonist). Either seratrodast or tranilast failed to inhibit the accumulation of inflammatory cells into bronchoalveolar lavage fluid (Table 1). The failure of seratrodast and tranilast to inhibit infiltration of inflammatory cells has already been reported by others (Matsumoto et al., 1994; Tominaga et al.,1997). In the case of azelastine, although we observed an inhibitory, but not significant, effect on infiltration of macrophages (Table 1), this drug was ineffective in inhibiting the induction of the late bronchoconstriction response (Fig. 3B). Similar pharmacological action pattern of azelastine has already been described in the previous report (Nakagawa et al., 1993). In the present study, the extent of the inhibitory effect of azelastine on the infiltration of inflammatory cells was small compared with those reported in the previous studies (Chand et al., 1992; Nakagawa et al., 1993), although it has also been reported that azelastine is ineffective for the infiltration of eosinophils (Evangelista et al., 1998). The controversial results of azelastine for the infiltration of the inflammatory cells may be partly explained by the difference in the protocol for the administration of azelastine (Nakagawa et al., 1993; Evangelista et al., 1998) or by the difference in the protocol for the sensitization of the animal with antigen (Chand et al., 1992). When azelastine was administered 5 min before the antigen challenge, it was ineffective for the infiltration of the inflammatory cells (Evangelista et al., 1998). In the case where azelastine exerted the marked inhibitory effect (Nakagawa et al., 1993), it was administered twice, i.e., first administration was 1 h before and the second one was at 6 h after antigen challenge. In the present study, azelastine was administered twice, i.e., 1 h before but 3 h after antigen challenge. In another study (Chand et al., 1992) where azelastine was effective, the sensitization of animal was performed by an intraperitoneal injection of the antigen, while it was performed by an aerosol administration in the present study. The rather milder procedure for the sensitization may lead to the induction of the marked inhibitory effect of the drug. Thus, only pranlukast, as TA-270, inhibited all asthmatic responses listed above as induced by an antigen challenge. For example, both TA-270 and pranlukast were effective in inhibiting infiltration of eosinophils, although the effect of pranlukast was not significant (Table 1). The failure of the significant inhibitory effect of pranlukast in the present study might be again partly due to the difference in the protocol of drug administration like in the case of azelastine as discussed above. If this was the case, we might underestimate the inhibitory effects of pranlukast and other antiasthmatic drugs on the antigen-induced actions. Thus, it should be noted that the present study might no longer demonstrate the superiority of TA-270 to other antiasthmatic drugs.

The similarity of the inhibitory pattern of TA-270 with pranlukast suggests that TA-270 might inhibit the asthmatic responses through the mechanisms which are related to the production or action of peptide leukotrienes. Peptide leukotrienes are well-known inflammatory mediators that are released from mast cells and eosinophils and cause bronchoconstriction, plasma exudation and edema. Actually, the mediators have been demonstrated to play important roles in bronchial asthma for experimental animals and human (Tomioka et al., 1989; Rasmussen et al., 1992). In our preliminary experiments, we failed to observe any antagonistic property of TA-270 against a peptide leukotriene receptor, but instead observed inhibitory effects on leukotriene production in cultured basophilic cells.

In conclusion, TA-270 inhibited the immediate- and late-airway responses, pulmonary inflammatory cell accumulation, and airway hyperresponsiveness. These actions of TA-270 were comparable to or superior, at least under the present experimental conditions, to those of authorized antiasthmatic drugs. Thus, TA-270 may be a useful drug for therapy of bronchial asthma and other allergic diseases.

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References

Barnes, P.J., Belvisi, M.G., 1993. Nitric oxide and lung disease. Thorax 48, 1034–1043.

Barnes, P.J., Chung, K.F., Page, C.P., 1998. Inflammatory mediators of asthma: update. Pharm. Rev. 50, 515–596.

Booij-Noord, H., Orie, N.G., De Vries, K., 1971. Immediate and late bronchial obstructive reactions to inhalation of house dust and protective effects of disodium cromoglycate and prednisolone. J. Allergy Clin. Immunol. 48, 344–354.

Cartier, A., Thomson, N.C., Frith, P.A., Roberts, R., Hargreave, F.E., 1982. Allergen-induced increase in bronchial responsiveness to histamine: relationship to the late asthmatic response and change in airway caliber. J. Allergy Clin. Immunol. 70, 170–177.

Chand, N., Nolan, K., Diamantis, W., Sofia, R.D., 1992. Inhibition of aeroallergen-induced bronchial eosinophilia by azelastine in guinea pigs. Int. Arch. Allergy Immunol. 97, 229–232.

Cockcroft, D.W., Murdock, K.Y., 1987. Comparative effects of inhaled salbutamol, sodium cromoglycate, and beclomethasone dipropionate

on allergen-induced early asthmatic responses, late asthmatic responses, and increased bronchial responsiveness to histamine. J. Allergy Clin. Immunol. 79, 734–740.

De Monchy, J.G., Kauffman, H.F., Venge, P., Koeter, G.H., Jansen, H.M., Sluiter, H.J., De Vries, K., 1985. Bronchoalveolar eosinophilia during allergen-induced late asthmatic reactions. Am. Rev. Respir. Dis. 131, 373–376.

Evangelista, S., Boni, P., Castellucci, A., Perretti, F., Pretolani, M., Joseph, D., Renzetti, A.R., Subissi, A., Manzini, S., 1998. Antihistaminic and antiallergic properties of dextro-mequitamium iodide in upper and lower guinea pig airways: comparison with azelastine. Gen. Pharmacol. 30, 513–519.

Holgate, S.T., Kay, A.B., 1985. Mast cells, mediators and asthma. Clin. Allergy 15, 221–234.

Isogai, Y., Hasegawa, A., 1995. Preparation of azelastine. Jpn. Kokai Tokyo Koho. JP 07224058.

Marsh, W.R., Irvin, C.G., Murphy, K.R., Behrens, B.L., Larsen, G.L., 1985. Increases in airway reactivity to histamine and inflammatory cells in bronchoalveolar lavage after the late asthmatic response in an animal model. Am. Rev. Respir. Dis. 131, 875–879.

Matsumoto, T., Ashida, Y., Tsukuda, R., 1994. Pharmacological modulation of immediate and late airway response and leukocyte infiltration in the guinea pig. J. Pharmacol. Exp. Ther. 269, 1236–1244.

Nakagawa, N., Obata, T., Kobayashi, T., Okada, Y., Nambu, F., Terawaki, T., Furuya, T., Muryobayashi, K., Sawada, M., Aishita, H., 1993. Effect of a peptide leukotriene receptor antagonist, ONO-1078, on guinea-pig models of asthma. Eur. J. Pharmacol. 235, 211–219.

Pennock, B.E., Cox, C.P., Rogers, R.M., Cain, W.A., Wells, J.H., 1979. A noninvasive technique for measurement of changes in specific airway resistance. J. Appl. Physiol. 46, 399–406.

Rasmussen, J.B., Eriksson, L.O., Margolskee, D.J., Tagari, P., Williams, V.C., Andersson, K.E., 1992. Leukotriene D4 receptor blockade inhibits the immediate and late bronchoconstrictor responses to inhaled antigen in patients with asthma. J. Allergy. Clin. Immunol. 90, 193–201.

Repine, J.E., Bast, A., Lankhorst, I., 1997. Oxidative stress in chronic obstructive pulmonary disease. Oxidative Stress Study Group. Am. J. Respir. Crit. Care Med. 156, 341–357.

Sanjar, S., Aoki, S., Kristersson, A., Smith, D., Morley, J., 1990. Antigen challenge induces pulmonary airway eosinophil accumulation and airway hyperreactivity in sensitized guinea-pigs: the effect of anti-asthma drugs. Br. J. Pharmacol. 99, 679–686.

Tarayre, J.P., Aliaga, M., Barbara, M., Tisseyre, N., Vieu, S., Tisne-Versailles, J., 1990. Model of bronchial hyperreactivity after active anaphylactic shock in conscious guinea pigs. J. Pharmacol. Methods 23, 13–19.

Terao, S., 1989. Quinone derivatives: synthesis and structure-activity relations of a novel class of eicosanoid antagonists, AA-2414 and its analogs. Adv. Prostaglandin, Thromboxane, Leukotriene Res. 19, 651–654.

Toda, M., Arai, Y., Miyamoto, T., 1986. (Acylamino)benzodioxanes and –chromenones. Eur. Pat. Appl. EP173516.

Tominaga, T., Watanabe, A., Tsuji, J., Koda, A., Nagai, H., Kumazawa, Y., Shimada, H., 1997. Effects of TYB-2285 on the accumulation of eosinophils in the airway induced by antigen exposure in actively sensitized brown Norway rats. Gen. Pharmacol. 28, 301–303.

Tomioka, K., Garrido, R., Stevenson, J.S., Abraham, W.M., 1989. The effect of an orally active leukotriene (LT) antagonist YM-16638 on antigen-induced early and late airway responses in allergic sheep. Prostaglandins, Leukotrienes Essent. Fatty Acids 36, 43–47.