

Mechanisms of Benefit with Aspirin Therapy in Aspirin-Exacerbated Respiratory Disease



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KEYWORDS

- Aspirin-exacerbated respiratory disease (AERD) • Aspirin desensitization
- Mechanisms of action • Cysteinyl leukotrienes (CysLTs) • Cytokines
- Interleukin 4 (IL-4) • Signal transducer and activator of transcription 6 (STAT6)
- Prostaglandin D2

KEY POINTS

- Aspirin-exacerbated respiratory disease (AERD) is characterized by severe, persistent asthma, hyperplastic eosinophilic sinusitis with nasal polyps, and intolerance to aspirin and other nonsteroidal anti-inflammatory drugs.
- Aspirin desensitization is an effective therapeutic option in carefully selected patients; however, the mechanisms behind the effects of aspirin desensitization remain poorly understood.
- AERD is associated with an overexpression of cysteinyl leukotrienes (CysLTs), with marked upregulation of CysLT receptors.
- Despite increased knowledge about the pathophysiology underlying AERD, the mechanisms behind the therapeutic effects of aspirin desensitization remain poorly understood.
- Recent studies suggest that the clinical benefits of aspirin desensitization may occur through direct inhibition of tyrosine kinases and the signal transducer and activator of transcription 6 pathway, with resultant inhibition of interleukin 4 production.
- A reduction in prostaglandin D2, as a consequence of aspirin desensitization, may also produce clinical benefit in AERD by precluding recruitment of PGD2 responsive effector cells to the airways.

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INTRODUCTION

Aspirin was first synthesized by Felix Hoffman in 1897 and was marketed by Bayer as an anti-inflammatory drug in 1899.¹ Idiosyncratic reactions to aspirin were reported shortly after the drug's development, although Widal and colleagues² was the first to describe aspirin challenges and desensitization for patients with the syndrome of asthma, nasal polyposis, and aspirin intolerance in 1922. Later, following works by Samter and Beers,³ the condition was termed Samter triad because of the association of these 3 overlapping conditions, although aspirin-exacerbated respiratory disease (AERD) is now the preferred nomenclature. Stevenson and colleagues⁴ reported the successful treatment of AERD with aspirin desensitization followed by daily aspirin therapy. Their study demonstrated significant clinical improvement in nasal symptoms and requirement for nasal steroids following aspirin desensitization.⁵ Symptom improvement in AERD has been reported to occur as early as 4 weeks after initiation of aspirin therapy, and long-term benefits include considerable reductions in the number of sinus infections and surgeries, hospitalizations for asthma, and use of systemic steroids.^{6,7} Despite these data, which support the clinical benefit, the mechanisms of aspirin therapy remain the least understood aspect of the disease.

Central to AERD is the dysregulation of endogenous inflammatory and anti-inflammatory mediators produced from the metabolism of arachidonic acid (also known as eicosatetraenoic acid).⁸ Abnormalities in these eicosanoid mediators (including leukotrienes [LTs], prostaglandins [PGs], lipoxins [LXs], and their respective receptors) have all been implicated in the pathogenesis of AERD.⁹ Evidence reveals that baseline expression of CysLTs is increased in patients with AERD and these levels further increase in response to cyclooxygenase-1 (COX-1) inhibitor challenge.^{10–16} Concomitantly, in tissue mast cells (MC) and eosinophils, there is an increased expression of LTC₄ synthase (LTC₄ S), the rate limiting enzyme in CysLT synthesis.^{17,18} CysLT₁ receptor expression is likewise elevated at baseline.¹⁹ Reduction in the synthesis and under expression of PGE₂, an important inhibitor of 5-LO and thus leukotriene production, also contributes to AERD pathogenesis.²⁰

Recent studies suggest that the AERD phenotype may result from a contribution of the cytokine interleukin 4 (IL-4), particularly in the presence of IFN- γ .^{21–24} Aspirin desensitization and continued therapy leads to an immediate improvement in the dysregulation of arachidonic acid metabolism. During desensitization, urine LTE₄ (uLTE₄) levels initially increase following aspirin ingestion but subsequently decrease to basal levels with continued treatment.²⁵ CysLT₁ receptor expression is also decreased following desensitization.¹⁹ Concomitantly, there is reduction of IL-4 induced expression of leukotrienes, which may occur through direct inhibition of the IL-4-activated signal transducer and activator of transcription 6 (STAT6) pathway.^{21,24,26,27} A reduction in prostaglandin D₂, as a consequence of aspirin desensitization, may also produce clinical benefit in AERD by precluding recruitment of PGD₂ responsive effector cells to the airways.^{28,29} These findings give insight into the mechanism of aspirin desensitization and are the subject of this review.

DESENSITIZATION EFFECTS ON PRODUCTS OF ARACHIDONIC ACID METABOLISM

LTE₄ is the most stable of the CysLTs and mediates many of the principal aspects of AERD, including bronchial constriction, hyperresponsiveness, eosinophilia, and increased vascular permeability.³⁰ Nasser and colleagues²⁵ studied the effects of aspirin desensitization on uLTE₄ concentrations in 9 patients with AERD. uLTE₄ levels

were measured before and then at specific time intervals for 9 hours after aspirin administration. Comparison was made between the following:

1. An initial dose of aspirin (mean of approximately 90 mg), which resulted in a 15% reduction in FEV₁ (forced expiratory volume in first second of expiration);
2. 600-mg aspirin 24 hours after desensitization; and
3. The 600-mg maintenance dose at least 2 months after desensitization.

Before desensitization, the initial dose of aspirin resulted in a sevenfold increase in uLTE₄ from baseline levels at 3 hours after ingestion. The comparable increases were threefold after initial desensitization and twofold with maintenance aspirin dosing (Fig. 1). Overall, the aspirin provoked increase in uLTE₄ production was reduced by 82% and 86% after initial desensitization and with continued maintenance therapy, respectively. The maximum reduction in FEV₁ was also improved from 15.3% ± 3.9% after initial aspirin dosing, to 3.3% ± 2.4% after initial desensitization and 7.4% ± 4.5% with continued therapy (Fig. 2). Additional studies also showed that continued aspirin therapy after successful desensitization results in a 20-fold reduction in bronchial responsiveness to inhaled LTE₄.^{31,32} Together these findings demonstrate that in patients with AERD, aspirin desensitization results in both decreased production of LTE₄ and reduced airway responsiveness to LTE₄.

LTB₄, which is metabolized from LTA₄ via LTA₄ hydrolase, is a potent chemotactic factor for eosinophils and neutrophils and contributes to the chronic inflammation seen in AERD.⁸ Juergens and colleagues³³ studied the release of LTB₄ and LTC₄ from calcium ionophore-stimulated peripheral blood monocytes from patients with AERD and normal volunteers before and after desensitization. Consistent with previous studies, patients with AERD had increased baseline levels of TXB₂, LTB₄, and LTC₄. After aspirin desensitization, TXB₂ was almost completely suppressed in both groups. LTB₄ release was reduced by 42% in the AERD group. Clinically, 8 of the 10 patients with AERD were taking prednisone before aspirin challenge. After desensitization, prednisone dosing was reduced, with an 85% reduction in average steroid dose (10.4 ± 2.2 mg/d to 1.6 ± 2.8 mg/d).

One of the major COX products of mast cells, PGD₂, as well as its metabolite, 9 alpha,11 Beta-PGF₂, is higher at baseline in patients with AERD than in aspirin-tolerant

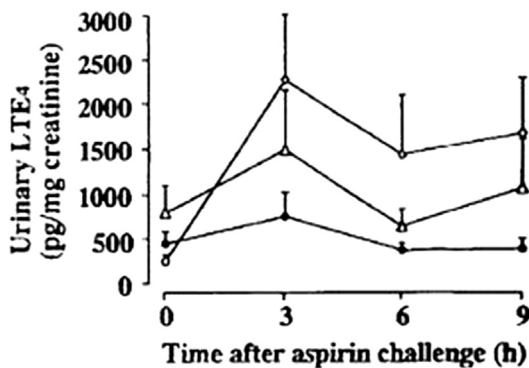


Fig. 1. uLTE₄ levels in 5 patients for 9 hours after challenge at the threshold dose of aspirin (○) and with 600 mg aspirin at 24 hours (△) and at 9 (3.2) months after aspirin desensitization (●). Each point is mean ± SEM. (From Nasser S, Patel M, Bell G, et al. The effect of aspirin desensitization on urinary leukotriene E₄ concentrations in aspirin-sensitive asthma. *Am J Respir Crit Care Med* 1995;151:1329; with permission.)

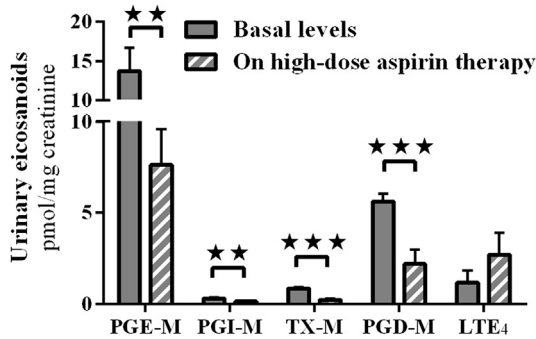


Fig. 2. Effect of high-dose aspirin therapy on urinary eicosanoid levels in patients with AERD. Urinary eicosanoid levels of patients with AERD are shown at baseline before aspirin and after 650 mg of twice-daily aspirin therapy for 8 weeks. Data are expressed as means $\pm$ SEMs. ** $P < .01$ and *** $P < .001$. (From Cahill KN, Bensko JC, Boyce JA, et al. Prostaglandin D₂: a dominant mediator of aspirin-exacerbated respiratory disease. *J Allergy Clin Immunol* 2015;135(1):245–52; with permission.)

asthmatic patients and healthy control subjects. Interestingly, these levels increase further in patients with AERD following ASA challenge, indicating mast cell activation.^{28,29,34} PGD₂ is a potent chemotactic for T_H2 cells, innate helper lymphoid cells, basophils, and eosinophils, all of which express the D prostanoid 2/chemokine receptor homologous molecule (DP2/CRTH2) receptor.³⁵ PGD₂, through DP2/CRTH2, may promote the persistent eosinophilic inflammation of the respiratory tract that characterizes AERD.^{28,29} Additionally, PGD₂ can induce bronchoconstriction through T prostanoid receptors, further contributing to the AERD phenotype.²⁹

Recent studies suggest that CysLTs may induce the release of the structural cell-derived cytokines thymic stromal lymphopoietin (TSLP) and interleukin-33 (IL-33). TSLP can act directly on mast cells, promoting the release of mast-cell products, including PGD₂. TSLP and IL-33 are strongly expressed by nasal polypoid tissue in AERD.^{29,36} Further, TSLP messenger RNA (mRNA) expression from nasal polypoid tissue in AERD correlates with urinary PGD₂ levels in these subjects.³⁷

Cahill and colleagues²⁸ examined prostaglandin generation in patients with AERD undergoing therapeutic desensitization. They compared basal urinary prostaglandin metabolite and LTE₄ levels in patients with AERD before aspirin desensitization, after desensitization, and following 8 weeks of maintenance aspirin therapy with 1300 mg of aspirin per day. They found that daily aspirin therapy for 8 weeks led to a striking decrease in prostaglandin metabolite (M) levels as follows: PGE-M (8.7 ± 2.1 pmol/mg Cr, $P < .01$), PGI-M (0.14 ± 0.03 pmol/mg Cr, $P < .01$), TX-M (0.2 ± 0.1 pmol/mg Cr, $P < .001$), and PGD-M (2.2 ± 0.8 pmol/mg Cr, $P < .001$). In contrast, there was no change in LTE₄ levels (2.7 ± 1.2 pmol/mg Cr, $P = .24$) (see Fig. 2). The authors postulate that the therapeutic benefit of high-dose aspirin in AERD may be, at least in part, the result of a reduction in these bronchoconstrictive prostaglandins. According to the investigators, the unexpected reduction in urinary PGE-M levels (8.7 ± 2.1 pmol/mg Cr, $P < .01$) following 8 weeks of aspirin therapy may reflect the ability of high-dose aspirin to not only interfere with COX-1, but also to interfere with COX-2, which is responsible for most renal PGE₂ synthesis.

Interestingly, the subjects with AERD who were unable to tolerate aspirin challenge secondary to severe reactions not only demonstrated higher levels of CysLTs but also experienced a marked increase in urinary PGD₂, during their reaction, which

correlated with the severity of airflow obstruction. In contrast, patients who were successfully desensitized and subsequently treated with high-dose aspirin displayed a steep reduction in urinary PGD₂ as well as an associated reduction in tissue eosinophils (0.31 ± 0.06 – $0.93 \pm 0.19 \times 10^3/\mu\text{L}$, $P < .01$) and a modest reduction in tissue basophils. These data suggest that the aspirin therapy may lead to a reduction in PGD₂ generation, thereby precluding recruitment of PGD₂ responsive effector cells to the airways. Although the mechanism by which aspirin contributes to this decline has not been fully elucidated, it may be the result of aspirin-induced inhibition of IL-4 expression. As IL-4 is an activator of PGD₂ synthase, a reduction in IL-4 may subsequently lead to diminished production of PGD₂. The impact of aspirin therapy on IL-4 expression is discussed later in this article.

DESENSITIZATION CHANGES IN CYSTEINYL LEUKOTRIENE RECEPTOR EXPRESSION

In 2002, Sousa and colleagues¹⁹ examined the expression of CysLT₁ and LTB₄ receptors in inflammatory leukocytes from nasal biopsies of patients with AERD. Aspirin desensitization was associated with a significant reduction in cells expressing the CysLT₁ R, which was seen as early as 2 weeks and persisted through follow-up at 6 months. Although this finding may partially explain the clinical benefit of aspirin desensitization, the study did not postulate further as to why the CysLT₁ R was downregulated with continued aspirin therapy. As is discussed in the following section, this reduction in CysLT₁ R, expression likely results from aspirin-induced changes in IL-4 production.

INTERLEUKIN-4 ALTERATIONS FOLLOWING ASPIRIN DESENSITIZATION

The cascade of inflammation in AERD involves the stimulatory signaling of various cytokines such as IL-5 promoting eosinophil development, differentiation, and survival, and IL-13—enhancing bronchial smooth muscle proliferation and inhibition of PGE₂ biosynthesis.⁸ More recently, research has focused on the role of IL-4 in the development of AERD.

Hsieh and colleagues³² studied the priming effects of IL-4 on the in vitro production of histamine, CysLTs, and PGs from human cord blood–derived MC. CysLT release was increased by 27-fold and PGD₂ release was increased by 6-fold when these MCs were passively stimulated with IL-4 in addition to stem cell factor (SCF). The addition of IL-3 or IL-5 to the 4/SCF-primed cells led to a further increase in CysLT production by sixfold and fourfold, respectively. This increase was found to occur as a result of IL-4 induction of LTC₄ S. Additional evidence published by Early and colleagues³⁸ showed the compounded effects of IL-4 on CysLT receptor expression. Using quantitative polymerase chain reaction (PCR) analysis of mRNA and receptor proteins extracted from immune cells, the investigators were able to show that IL-4 stimulation resulted in significant increases in both CysLT₁R and CysLT₂R on T and B lymphocytes ($P < .01$) and CysLT₂R on eosinophils ($P < .05$). CysLT₂R mRNA was also increased in monocytes, T cells, and B cells after IFN- γ stimulation. The mechanisms of these effects have been shown to occur through IL-4/STAT6-dependent pathways, which are also important in the IL-4-induced suppression of PG production.^{39–41} Combined results from these studies imply a likely role for IL-4 in the pathogenesis of AERD.

The pharmacologic effects of aspirin have been extended to include the inhibition of IL-4. Cianferoni and colleagues²⁶ examined the effect of aspirin on the expression of various cytokines, including IL-4, IL-13, IL-2, and IFN- γ in purified human CD4⁺ T cells. Experiments showed that therapeutic concentrations of aspirin significantly inhibited IL-4 secretion in peripheral blood T cells by $47\% \pm 2.4\%$ ($P < .05$) and that this inhibition occurred without significant aspirin-induced apoptosis to account for

the change. Using multiple NSAIDs, the investigators showed that IL-4 suppression occurred independently of COX inhibition. They identified aspirin inhibition of IL-4 gene expression via salicylate-targeted regions in the IL-4 promoter as the possible mechanism of action, although the exact pathway was not defined. Aspirin did not have any significant effect on the levels of IL-2, IL-13, or IFN- γ expression.

Expanding on these findings, several groups have now identified aspirin inhibition of IL-4-induced STAT6 activation as the possible mechanism associated with the therapeutic benefits of aspirin desensitization. Using a murine model, Perez and colleagues⁴² showed that sodium salicylate (NaSal) and aspirin blocked the ability of IL-4 to induce DNA-binding activity of STAT6 in a dose-dependent manner (Fig. 3). They showed that aspirin inhibits IL-4-dependent induction of CD23 expression on human peripheral blood mononuclear cells. These findings were confirmed in a more recent study published by Steinke and colleagues.²⁷ Using THP-1 human mononuclear cells, they showed that aspirin inhibited IL-4-induced CysLT₁ R and F_{C ϵ} RI α mRNA expression in a dose-dependent fashion. Similar results occurred with ketorolac but not NaSal. These investigators also reported the presence of STAT6-binding sites within the promoters for the CysLT₁ R and LTC₄ S, which are engaged by IL-4 nuclear extracts and were effectively inhibited by aspirin.

Katial and colleagues²¹ reported the effects of aspirin desensitization on novel inflammatory sputum biomarkers in patients with AERD. In addition to IL-4, these investigators studied changes in sputum tryptase, matrix metalloproteinase 9 (MMP-9), tissue inhibitors of metalloproteinases 1 (TIMP-1) and FMS-like tyrosine kinase 3 ligand. Traditional measures such as symptom scores, exhaled nitric oxide (FeNO), and lung function were also assessed. Twenty-one patients were enrolled in the study,

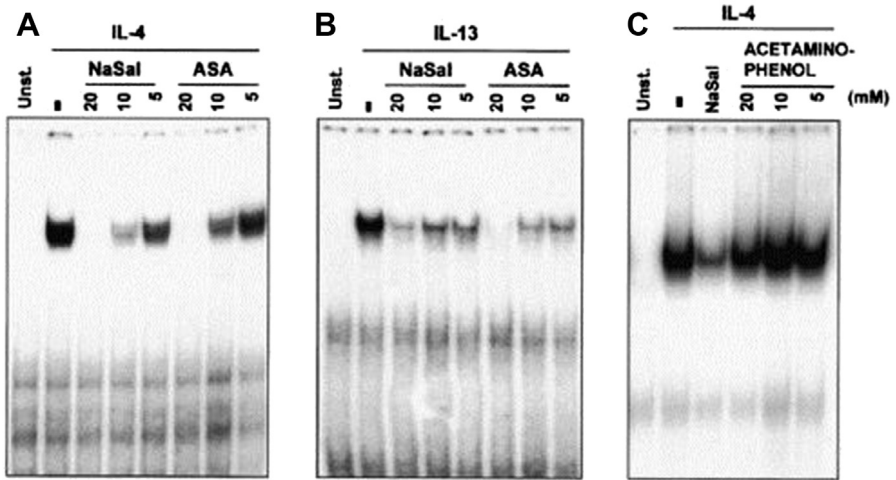


Fig. 3. Salicylates, but not acetaminophen, inhibit STAT6 activation. (A) M12 cells were cultured with the indicated concentrations of NaSal and aspirin for 1 hour before stimulation with IL-4 (10 ng/mL) for 30 minutes. STAT6 DNA-binding activity in cell extracts was analyzed by electrophoretic mobility shift assay (EMSA) using the IFN- γ activation site sequence contained in the C ϵ promoter. (B) U937 cells were cultured with NaSal or ASA for 1 hour, and then stimulated for 30 minutes with IL-13 (400 ng/mL). STAT6 activation was then analyzed by EMSA. (C) M12 cells were cultured with nothing, NaSal (20 mM), or the indicated concentrations of acetaminophen. IL-4 was then added, and STAT6 binding to DNA was analyzed by EMSA. (From Perez-GM, Melo M, Keegan A, et al. Aspirin and salicylates inhibit the IL-4 and IL-13 induced activation of STAT6. *J Immunol* 2002;168:1429; with permission.)

with 16 completing successful sputum induction. Six months after desensitization, while on high-dose aspirin, sputum IL-4 levels decreased 94.7% (95% confidence interval [CI], 67%–99.2%) relative to baseline (baseline mean 28.1 pg/mL, 6-month mean 1.5 pg/mL; $P = .004$) (Fig. 4). This change was accompanied with a similar decrease in MMP-9 after 6 months. This suggests that the reported increases in CysLTs may be regulated by IL-4–dependent pathways, which are then downregulated by daily aspirin treatment after desensitization.

More recently, Katial and colleagues⁴³ investigated the mechanism underlying IL-4 suppression by aspirin therapy. They evaluated the in vitro effects of nonsteroidal anti-inflammatory drugs on the activation of peripheral blood CD4+ T cells through STAT6 signaling. Peripheral blood mononuclear cells from 11 aspirin-sensitive asthmatic individuals, 10 aspirin-tolerant asthmatic individuals, and 10 healthy controls were stimulated with anti-CD3 antibody and IL-4, both in the presence and absence of NSAIDs. Subsequently, the expression of phosphorylated STAT6 (pSTAT6), pSTAT4, and IL-4 were measured in CD4+ T lymphocytes by flow cytometry. They found that stimulation with the combination of anti-CD3 and IL-4 induced pSTAT6 in CD4+ T cells from all subjects; however, this induction was significantly higher in aspirin-sensitive asthmatic individuals as compared with controls. Aspirin, indomethacin, and to a lesser degree the weak COX-1 inhibitor sodium salicylate, inhibited pSTAT in all subjects in a dose-dependent manner (Fig. 5). These studies suggest that aspirin desensitization may be effective, in part, by the ability of COX-1 inhibitors to mitigate STAT6 signaling, thus blocking the biologic activities of IL-4 with subsequent downregulation of CysLT production and CysLT₁R expression.

EFFECT OF ASPIRIN DESENSITIZATION ON LEUKOCYTE-DERIVED CYTOKINES

Studies addressing the cytokine milieu in AERD have demonstrated a predominantly T_H2-like profile, which is not surprising given the fundamental role of eosinophils in the

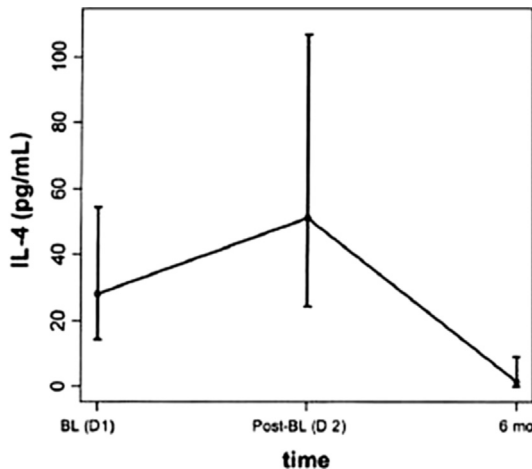


Fig. 4. IL-4 means over time, with 95% CIs, based on the linear mixed model fit. IL-4 was modeled on the natural log scale, and estimated and CI end points were then inverted back for presentation, resulting in longer upper bars than lower bars. Note that CIs are relevant for fixed time points only and do not indicate variability of estimated for differences between time points because repeated-measures data were involved. BL, baseline. (From Katial R, Strand M, Prasertsuntarasai T, et al. The effect of aspirin desensitization on novel biomarkers in aspirin-exacerbated respiratory diseases. *J Allergy Clin Immunol* 2010;126:741; with permission.)

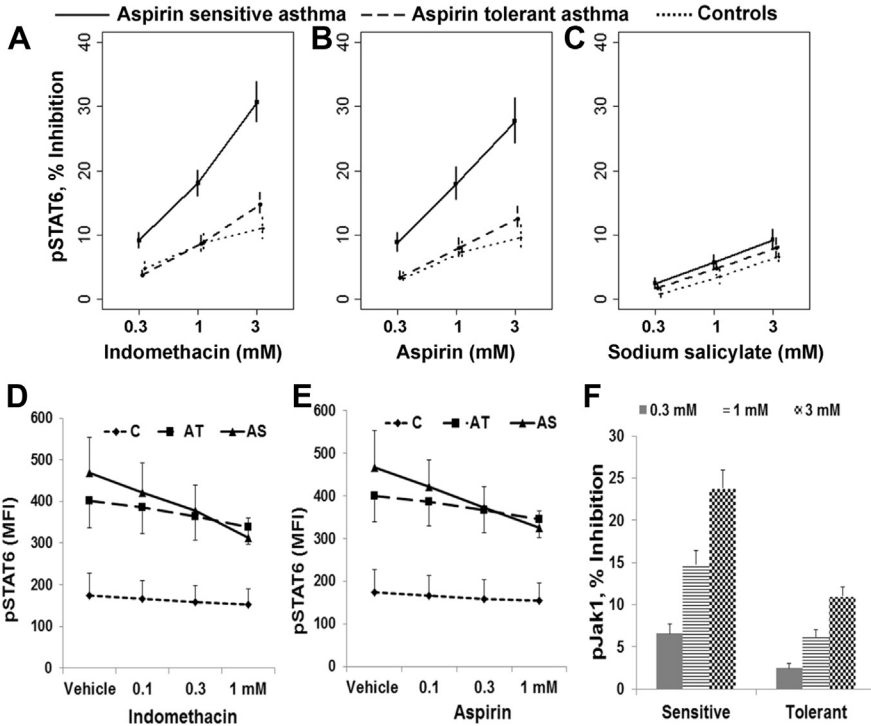


Fig. 5. (A–F) Comparison of the effect of indomethacin, aspirin, and sodium salicylate on CD4 T-cell pSTAT6 expression in aspirin-sensitive ($n = 11$) and aspirin-tolerant ($n = 10$) patients with asthma and control subjects ($n = 10$). (F) Effect of aspirin on phosphorylated Jak1 in aspirin-sensitive ($n = 11$) and aspirin-tolerant ($n = 10$) patients with asthma. $P = .01$ for 0.1 mM aspirin and $P < .01$ for other doses of aspirin (Mann-Whitney U test). (From Katial RK, Martucci M, Burnett T et al. Nonsteroidal anti-inflammatory-induced inhibition of signal transducer and activator of transcription 6 (STAT-6) phosphorylation in aspirin-exacerbated respiratory disease. *J Allergy Clin Immunol* 2016;138(2):583; with permission.)

disease. However, newer observations suggest that AERD is characterized by a mixed T_H1 and T_H2 inflammatory profile with a prominent $IFN-\gamma$ expression. Steinke and colleagues⁴⁴ highlighted this observation through their examination of the cytokine composition in nasal polyp tissue from subjects with AERD as compared with subjects with aspirin-tolerant chronic hyperplastic eosinophilic sinusitis (CHES) and healthy controls.

Through quantitative PCR of nasal polyp tissue, they found polyps from patients with AERD demonstrated variable expression of T_H2 cytokines and had significantly elevated levels of IL-4 mRNA ($P < .05$). However, in contrast to subjects with CHES, those with AERD also had profound $IFN-\gamma$ mRNA transcript expression. They performed intracellular cytokine staining and reported that most of the IL-4 and $IFN-\gamma$ staining was observed in eosinophils, indicating the eosinophilic source of these cytokines as well as their intracellular accumulation in the eosinophil granules.

Alteration of $IFN-\gamma$ expression in T lymphocytes has also been reported in relation to aspirin desensitization. Shome and colleagues⁴⁵ demonstrated that subjects with

AERD have elevated baseline levels of IFN- γ in circulating CD8+ T lymphocytes as compared with aspirin-tolerant controls. Interestingly, this pattern decreased significantly after desensitization, although neither the mechanism nor significance behind this change was elucidated.

ROLE OF PLATELETS AND TARGETED ANTIPLATELET THERAPIES IN ASPIRIN-EXACERBATED RESPIRATORY DISEASE

There is relatively new evidence that the proinflammatory actions of activated platelets may play a central role in the pathogenesis of AERD. Increased membrane expression of P-selectin is a hallmark of platelet activation and mediates platelet adhesion to leukocytes through P-selectin glycoprotein ligand 1.⁴⁶ Platelet-leukocyte aggregates are well-recognized contributors to vascular inflammation and cardiovascular disease⁴⁷; similarly, they may be effectors of AERD pathogenesis. Laidlaw and colleagues⁴⁸ found that peripheral blood and nasal polyp tissue from patients with AERD contained significantly more platelet-adherent leukocytes than aspirin-tolerant control subjects, and that 80% of the circulating eosinophils were adherent to platelets. Furthermore, these cells had increased expression of adhesion molecules such as beta integrins, implying that adherent platelets may facilitate adhesion and augment recruitment of eosinophils to peripheral tissue. An aberration in the process that controls this may contribute to the pathogenesis of AERD.

In response to activation, platelets precipitately liberate arachidonic acid from membrane lipids for conversion to eicosanoids such as TXA₂, which likewise facilitates leukocyte recruitment. PGD₂ is also generated, which signals through DP2 as a major chemoattractant for eosinophils, basophils, T_H2 cells, and innate type 2 helper cells. Thus, platelets are implicated as key mediators of leukocyte recruitment.⁴⁹

In addition to self-generation of eicosanoids, platelets can also modulate the generation of leukotrienes by inflammatory leukocytes. Leukocytes can oxidize arachidonic acid through 5-LO to form LTA₄. Although neutrophils lack LTC₄ S activity and cannot convert LTA₄ into LTC₄, platelets possess abundant LTC₄ S activity in the absence of 5-LO.⁵⁰ In vitro studies have shown that neutrophils generate LTA₄ in excess of their ability to hydrolyze it to LTB₄; however, when platelets are added to the neutrophils, the excess LTA₄ is subsequently converted to LTC₄, suggesting that there is a reciprocal transcellular mechanism by which platelets might contribute to leukotriene generation, possibly by providing LTC₄ S.^{48,51}

Supporting this theory, Laidlaw and colleagues⁴⁸ found that uLTE₄ levels strongly correlate with the percentage of platelet adherent leukocytes in the peripheral circulation. Additionally, LTC₄ S activity in blood granulocytes from patients with AERD was significantly higher than levels from aspirin-tolerant control subject granulocytes. Stripping the granulocytes of adherent platelets, resulted in a marked reduction in LTC₄ S activity, suggesting that adherent platelets likely contribute to CysLT overproduction in patients with AERD.

Indeed, aberrant platelet function may contribute to the pathogenesis of AERD and therapies which target platelet-leukocyte aggregates may represent a therapeutic option in these patients. Klinkhardt and colleagues⁵² found that platelet-leukocyte aggregates are reduced by clopidogrel. It appears that 100 mg of daily aspirin does not reduce platelet-leukocyte aggregation, albeit this dose is likewise not sufficient to reduce nasal and pulmonary symptoms or prevent nasal polyp regrowth in AERD.⁵³ It may be that the high-dose aspirin required for the treatment AERD does result in

platelet-leukocyte aggregate inhibition and contributes to the therapeutic benefit of aspirin therapy in these patients. This is an area in which further research is needed.

SUMMARY

AERD is a clinical syndrome characterized by severe, persistent asthma, hyperplastic eosinophilic sinusitis with nasal polyps, and intolerance to aspirin and other NSAIDs that preferentially inhibit COX-1. Although aspirin desensitization has proved to be of significant long-term benefit in carefully selected patients with AERD, the mechanisms behind the therapeutic effects of aspirin desensitization have not been clearly delineated. Current research suggests that the derangements in inflammatory mediators seen in AERD may in part be a result of increased production and hyperresponsiveness to IL-4. This suggestion is reinforced by evidence that aspirin may derive its therapeutic benefit through direct inhibition of the IL-4-activated STAT6 pathway. The proposed mechanism would occur from continued aspirin ingestion causing inhibition of IL-4/STAT6-mediated production of CysLT and CysLT₁ R expression, with a subsequent attenuation of airway inflammation and clinical improvement (Fig. 6). Levels of PGD₂, a potent chemotactic for T_H2 cells, are also elevated at baseline in AERD and likely contribute to the eosinophilic airway inflammation characteristic of the disease. Interestingly, urinary PGD₂ levels decline following high-dose aspirin therapy, conceivably by preventing the chemotaxis of effector cells into the tissues.

Despite these advances, the limited data published regarding long-term mechanistic effects after aspirin desensitization leave many outstanding questions. Answering these questions requires further prospective, long-term evaluations of both the cellular and signaling alterations that occur in patients undergoing desensitization. In addition, as we learn more about the varied mechanisms of aspirin therapy related to inhibition of nuclear transcription factors, we may define new avenues of research exploration. Such further investigation will continue to increase our insight and understanding of this complex disease and it is hoped provide new therapeutic options and treatments for our patients.

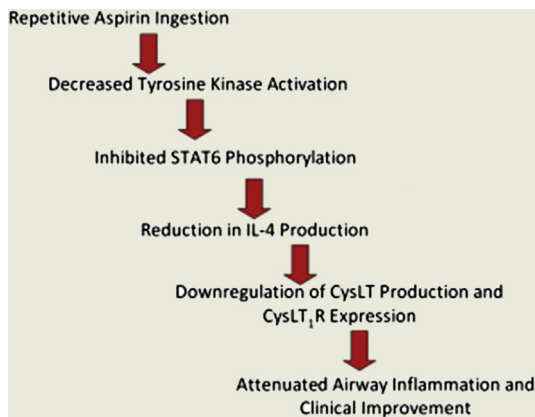


Fig. 6. Mechanism of inhibition of inflammation after aspirin desensitization. Proposed cascade of events after aspirin desensitization and continued ingestion. Signal inhibition via an IL-4/STAT6 pathway results in downregulation of CysLT production and CysLT₁ R expression, with subsequent reduction of tissue inflammation. (From Burnett T, Katial RK, Alam R. Mechanisms of aspirin desensitization. *Immunol Allergy Clin North Am* 2013;33(2):232; with permission.)

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