

Plasma acetylsalicylic acid and salicylic acid levels during aspirin provocation in aspirin-sensitive subjects

Dahlén B, Boréus LO, Anderson P, Andersson R, Zetterström O. Plasma acetylsalicylic acid and salicylic acid levels during aspirin provocation in aspirin-sensitive subjects.

Allergy 1994; 49: 43–49. © Munksgaard 1994.

The ability of aspirin and other nonsteroidal anti-inflammatory drugs (NSAIDs) to inhibit the cyclo-oxygenase which catalyzes formation of prostaglandins appears to be central to the mechanisms involved in aspirin sensitivity. We have investigated whether the plasma levels of acetylsalicylic acid (ASA) and its main metabolite salicylic acid (SA) at the time of intolerance reactions correspond with the concentrations required for enzyme inhibition *in vitro*. Twelve aspirin-sensitive and 15 aspirin-tolerant subjects were followed during provocation with aspirin. ASA and SA concentrations in plasma were determined by HPLC. After oral provocation (up to 460 mg cumulative dose), the levels of ASA and SA in plasma were equivalent in aspirin-sensitive and aspirin-tolerant subjects. For the aspirin-sensitive subjects, at the time of adverse reaction, the concentration range was 2.9–33.3 μM for ASA and 18.1–245 μM for SA. Oral provocation with sodium salicylate yielding 10-fold higher SA levels did not elicit intolerance reactions. Statistically significantly lower levels of ASA and SA ($P \leq 0.01$) evoked airway obstruction, as compared with merely extrapulmonary symptoms. Bronchial absorption of aspirin was found after inhalation of lysine-aspirin and was comparable in asthmatic and nonasthmatic subjects. In three aspirin-sensitive subjects who developed airway obstruction, the plasma levels for ASA and SA were 0.9–2.6 μM and 0.0–6.7 μM , respectively. In conclusion, the plasma levels of ASA reached at the time of a positive reaction are of the magnitude known to inhibit cyclo-oxygenases. Neither differences in bioavailability of ASA nor the formation of SA seems to contribute to the aspirin-elicited reactions.

B. Dahlén¹, L. O. Boréus², P. Anderson², R. Andersson³, O. Zetterström¹

¹ Asthma and Allergy Division, Department of Thoracic Medicine, Karolinska Hospital,

² Department of Clinical Pharmacology, Karolinska Hospital, and ³ Department of Pharmacology³, Karolinska Institute, Stockholm, Sweden

Key words: acetylsalicylic acid; aspirin challenge test; aspirin-induced asthma; bronchial provocation test; drug concentrations; lysine-aspirin inhalation; salicylic acid.

Dr Barbro Dahlén
Department of Thoracic Medicine
Karolinska Hospital
S-104 01 Stockholm
Sweden

Accepted for publication 26 March 1993

Since 1902, acetylsalicylic acid (aspirin, ASA) has been known to cause unexpected intolerance reactions (21). Aspirin-induced hypersensitivity reactions include airways obstruction, naso-ocular symptoms, urticaria, angioedema, and gastrointestinal symptoms. These symptoms may occur in isolation or in any combination. Aspirin-induced bronchospasm typically afflicts patients with chronic asthma associated with rhinitis with or without polyps (the classic triad (15)). On the other hand, urticaria and angioedema mainly develop in subjects with a history of chronic urticaria (19). Especially the syndrome of aspirin-induced asthma (AIA) may involve severe, long-lasting, and sometimes fatal airway obstruction. The prevalence of AIA is estimated to be between 2.5 and 15% in adult asthmatic patients (19).

Although many hypotheses have been proposed to account for either aspirin-sensitivity in respiratory and cutaneous organs, or the underlying disease in these locations, the pathogenetic mechanism behind this intriguing syndrome remains unknown (2, 3, 7, 21, 26). However, aspirin-sensitive patients are intolerant of all the nonsteroidal anti-inflammatory drugs (NSAIDs) which act by inhibition of the cyclo-oxygenase enzyme, thereby abrogating formation of prostaglandins and thromboxane (19, 21). After desensitization with repeated doses of aspirin, cross-desensitization to the same type of NSAIDs also occurs (14). The likelihood of different NSAIDs to provoke bronchoconstriction in sensitive asthmatic patients is related to their individual potencies as inhibitors of cyclo-oxygenase *in vitro* (22), and evidence of cyclo-oxygenase inhibition *in vivo* has been

demonstrated during nasal and oral aspirin provocation (13, 21). Although it seems obvious that the cyclo-oxygenase inhibition somehow triggers the intolerance reactions, it remains a mystery why only certain persons develop this intolerance.

While the pharmacokinetics of ASA is well characterized in healthy volunteers (for review, see ref. 9), plasma concentrations of ASA have not been studied in aspirin-sensitive subjects after ingestion of provocative doses of aspirin. One study has included measurement of plasma salicylic acid (SA), the main metabolite of ASA (17). Because relatively low doses of ASA (10–300 mg) are sufficient to elicit an intolerance reaction, measurement of plasma ASA in aspirin-sensitive subjects requires a method which is more sensitive than that used in clinical practice for diagnosis of intoxication. In this study we used a highly sensitive HPLC method to measure plasma ASA and SA levels during aspirin provocation in aspirin-sensitive subjects with either respiratory or cutaneous manifestations, as well as in aspirin-tolerant patients.

First, it was considered to be pertinent to determine whether disturbances in absorption or subsequent hydrolysis of ASA into SA may contribute to the aspirin-syndrome. ASA is normally rapidly hydrolyzed to salicylic acid (SA) in the gut and liver and in erythrocytes, and only minute amounts of ASA reach the circulation (9). Therefore, we also evaluated the clinical significance of the amounts of SA generated from ASA. Secondly, to evaluate further the hypothesis that aspirin-induced reactions may be linked to inhibition of the cyclo-oxygenase enzyme, we wanted to investigate whether the concentrations of ASA and SA that actually occur at the time of intolerance reaction are sufficient to inhibit the enzyme in question (10). Because bronchoprovocation with lysine-aspirin does not elicit extrapulmonary responses (8, 12), it was of interest to find out whether measurable ASA and SA levels could be demonstrated at all.

Material and methods

Plasma levels of ASA and SA were followed during oral or bronchial provocations with cumulative doses of acetylsalicylic acid in 27 subjects. The subjects were recruited from patients referred to the asthma and allergy clinic at the Karolinska Hospital. Consecutive patients were selected for provocation on the basis of possible aspirin-sensitivity. The subjects had either a positive history of a suspected adverse reaction to aspirin or other NSAIDs, or the typical clinical findings of aspirin-asthma, which include prominent nasal symptoms (watery rhinitis, nasal congestion, polyposis, loss of ability to smell). Ten subjects underwent oral aspirin provocation; nine

were challenged by inhalation of aspirin, and eight were challenged by both routes. In addition, four of the aspirin-sensitive asthmatic subjects were challenged with increasing oral doses of sodium salicylate. The study was approved by the local ethics committee, and the subjects gave informed consent.

Provocations were conducted in the hospital, the procedures being essentially as described (8). Monitoring of clinical symptoms included inspection of the nasal mucosa, and physical examination of the eyes, skin, and lungs. Pulmonary function was followed by measurement of forced expiratory flow volume during 1 s (FEV_1) by a Vitalograf®, and expressed as percent change from base line. The oral provocations involved ingestion of increasing doses of acetylsalicylic acid every 45 min. After intake of a placebo capsule containing lactose, incremental doses (10, 50, 100, and 300 mg) of ASA were successively administered. The provocation was stopped at the appearance of a "positive" reaction, or when the highest dose of ASA had been reached. A positive reaction in the airways was defined as a fall in $FEV_1 \geq 20\%$ from base line, or as a decline $\geq 15\%$ if combined with apparent development of rhonchi. Extrapulmonary positive reactions were naso-ocular responses (profound nasal congestion, rhinorrhea, and ocular injection), flush, urticaria, and angioedema. With this protocol for oral challenge, reactions may occasionally develop later and become severe (8). Therefore, a highly trained, experienced physician was always present and resuscitative equipment readily available.

The bronchoprovocation with ASA was performed by inhalation of a solution of lysine-ASA (Aspisol®, Horby Bayer AG, Germany), the concentration of ASA being $25 \text{ mg} \times \text{ml}^{-1}$, from a jet nebulizer (Aiolos, Medicinsk Teknik AB, Sweden). First 1 ml of the diluent was inhaled. Thereafter, provided FEV_1 did not change by more than 10% from prechallenge base line, incremental doses of the lysine-ASA solution were administered by inhalation of 1, 3, 9, and 27 tidal breaths and finally 1 ml of the solution. The interval between doses was 30 min. Including the final dose of lysine-ASA, a maximum of 75 mg of ASA could have been inhaled.

Analyses of plasma ASA and SA

For the analyses of plasma ASA and SA concentrations, blood samples of 5 ml were drawn before the start of the provocations and after ingestion of the placebo capsule or after inhalation of the diluent. Thereafter blood was drawn at 45 min after each oral dose or at 30 min after each inhaled dose during the oral and bronchial provocations, respectively. In case of a positive reaction, blood was drawn at a time when the symptoms had developed. The blood

samples were collected in heparinized tubes containing 5 mg sodium fluoride/ml. The samples were cooled in an ice bath and immediately centrifuged, transferred to polypropylene tubes, and frozen in a dry ice/ethanol bath. The samples were stored at -70°C and analyzed within 4 weeks. ASA and SA were extracted from acidified plasma to diisopropyl ether and determined by reversed-phase HPLC using a PRP-1 5 μ column. The mobile phase was 34% acetonitrile in 0.1 M H_3PO_4 with pH 2.2 and a flow rate of 1.2 ml/min and UV-detection at 229 nm. The lower limit of sensitivity was 0.1 $\mu\text{g/ml}$ for both compounds.

Statistical evaluation

Statistical hypotheses were tested by Student's *t*-test for independent variables, and a *P* value of less than 0.05 was considered to be significant. Results are expressed as means $\pm$ SD.

Results

Plasma levels of ASA and its rapidly formed metabolite, SA, were followed during cumulative oral aspirin provocations in 18 subjects with suspected aspirin intolerance. Although comparatively low doses of aspirin were administered (the cumulative dose being between 10 and 460 mg), increasing plasma levels of both ASA and SA could be detected during the course of provocation. Nine of the subjects developed clinically evident adverse reactions (Table 1), whereas nine were tolerant of the highest dose of aspirin administered. As illustrated in Fig. 1, the plasma levels of ASA and SA did not differ between the aspirin-sensitive and the aspirin-tolerant subjects. The two groups were comparable with respect to sex, history and age; that is, each

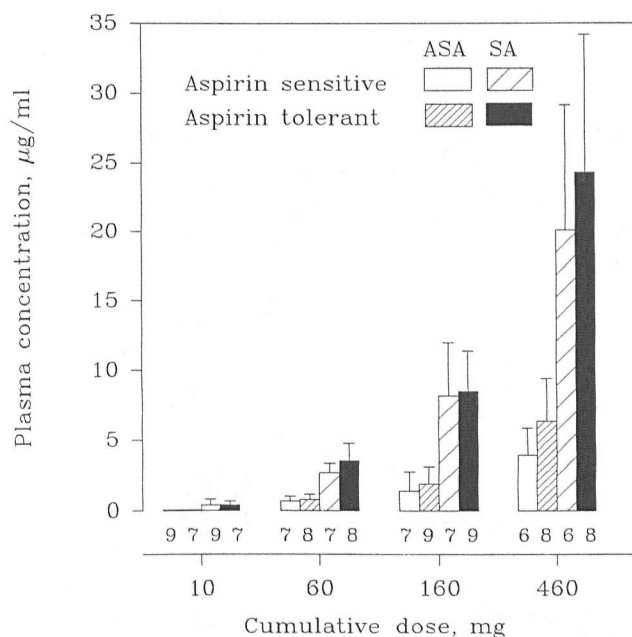


Fig. 1. Plasma levels of ASA and SA (means $\pm$ SD) in aspirin-sensitive and aspirin-tolerant subjects after ingestion of single doses of 10, 50, 100, and 300 mg of ASA at 45-min intervals. Number of subjects is shown below each bar. Blood samples were collected before start of provocation and at 45 min after each dose. After 10-mg dose, ASA levels were below detection limit in 12/16 subjects and just above this limit in four subjects. There was no statistically significant difference in plasma ASA or plasma SA levels between the two groups.

group comprised four men and five women; four were asthmatic and five had rhinitis, polyposis, urticaria, and/or angioedema; and the mean ages were 44 ± 16 ($\pm$ SD) and 48 ± 10 ($\pm$ SD) years in the aspirin-sensitive group and in the aspirin-tolerant group, respectively.

Fig. 2 shows a pronounced airway obstruction evoked by 160 mg of aspirin orally in one of the

Table 1. Aspirin-sensitive patients: characteristics and results

Subject	Age	History	Regular treatment	Cumulative aspirin dose		Plasma level $\mu\text{g/ml}$		Reaction
				Oral; mg	Inhaled; no breaths	ASA	SA	
MS	57	Asthma	IB, IS, OB, OT	160		1.10	11.4	FEV ₁ -48% congestion
LV	47	Asthma	IB	90		0.53	4.40	FEV ₁ -15%, g-i
BS	69	Asthma	IB, IS, OT	460		1.50	9.00	FEV ₁ -17%, rhinitis/congestion
RTN	44	Asthma	IB, IS	60		0.61	2.50	FEV ₁ -33%
CR	23	Angioedema	-	460		3.15	21.6	Angioedema
KS	26	Angioedema	-	460		5.50	12.4	Angioedema
AB	22	Urticaria	-	460		5.42	18.4	Urticaria
IS	46	Rhinitis	-	460		2.10	33.8	Rhinitis/congestion, flush
KB	40	Rhinitis	NS	460		6.00	25.6	Rhinitis/congestion
AB	20	Asthma	IB, IS, NS		40 ($\approx$ 30 mg)	0.21	0.42	FEV ₁ -38%
MIN	40	Asthma	IB, IS, NS		40 ($\approx$ 30 mg)	0.46	0.92	FEV ₁ -18%, rhonchi cough
TN	44	Asthma	NS		7 ($\approx$ 5 mg)	0.16	b.d. (0.09)	FEV ₁ -15%, rhonchi cough, PEFR - 20%

IB=inhaled β -agonist; IS=inhaled corticosteroid; OB=oral β -agonist; OT=oral theophylline; NS=nasal corticosteroid; b.d.=below detection limit; g-i=gastrointestinal symptoms; PEFR=peak expiratory flow rate.

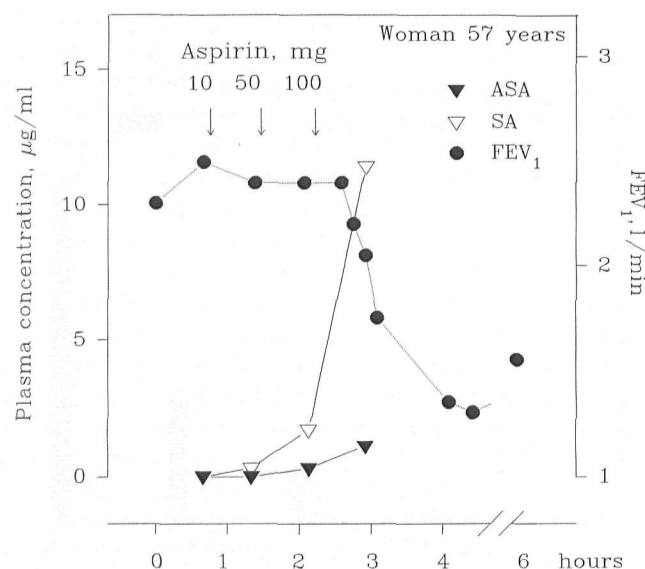


Fig. 2. Time course of oral aspirin provocation in one sensitive asthmatic patient. Airway obstruction started to develop after cumulative dose of 160 mg. Thirty minutes after onset of symptoms, plasma concentrations of ASA and SA were 1.1 and 11.4 $\mu\text{g/ml}$, respectively. FEV_1 declined by 48% of the base-line value, and rescue treatment was given.

asthmatic subjects. The fall in FEV_1 (48%) was associated with a striking rise in plasma SA levels, whereas the rise in ASA levels was comparatively modest. For the aspirin-sensitive subjects as a group, mean plasma levels after oral challenge were 2.9 ± 2.2 ($\pm$ SD) $\mu\text{g} \times \text{ml}^{-1}$ of ASA and 15.5 ± 10.3 ($\pm$ SD) $\mu\text{g} \times \text{ml}^{-1}$ of SA (the concentration range being 0.5–6.0 $\mu\text{g} \times \text{ml}^{-1}$ for ASA and 2.5–33.8 $\mu\text{g} \times \text{ml}^{-1}$ for SA, respectively) at the time of adverse reaction. Five of these patients had typical extrapulmonary symptoms in response to the provocation, i.e., profuse rhinorrhea, flush, urticaria, and angioedema, but did not show an airway reaction, whereas four of the patients responded with airway obstruction. The levels of ASA and SA (i.e., the doses; see Fig. 1) were significantly lower in the group which developed bronchoconstriction ($P \leq 0.01$), as shown in Fig. 3.

Four aspirin-sensitive asthmatic patients underwent cumulative oral provocation with sodium salicylate (1440 mg cumulative dose). Despite plasma SA-levels amounting to 1 log order of magnitude above the plasma SA-levels measured during aspirin provocation in sensitive patients, sodium salicylate failed to evoke adverse reactions in this group of patients (Fig. 4). As expected, ASA was not detected in plasma.

Bronchial provocations with inhaled lysine-ASA were performed in 17 subjects, eight of whom were asthmatic, and nine subjects with normal airways. Three of the asthmatic patients developed a signif-

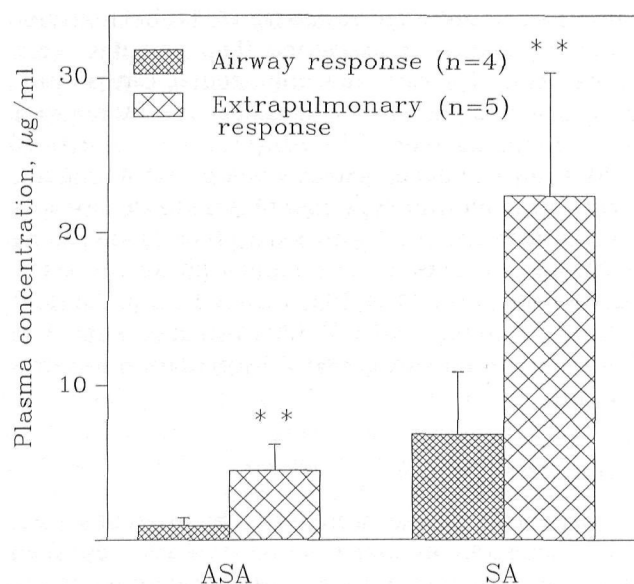


Fig. 3. Four aspirin-intolerant patients developed airway obstruction, whereas five acquired typical extrapulmonary symptoms. Airway responses occurred at significantly lower plasma levels of ASA and SA (0.74 ± 0.3 and 6.09 ± 4.7 $\mu\text{g/ml}$, respectively) than did extrapulmonary symptoms (3.95 ± 1.9 and 20.1 ± 9 $\mu\text{g/ml}$ for ASA and SA).

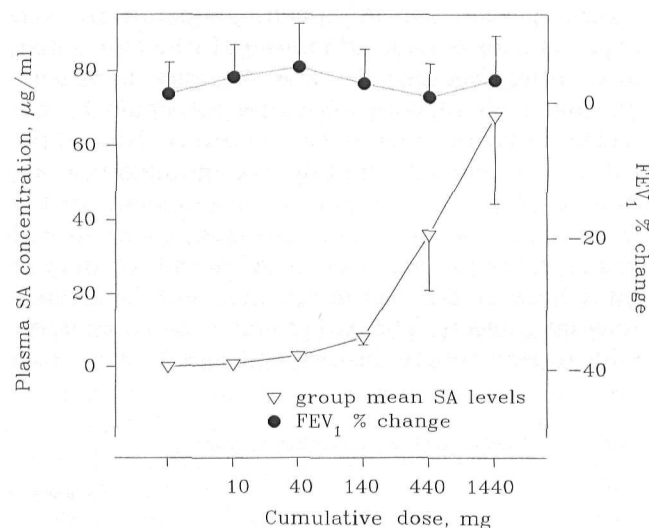


Fig. 4. SA plasma levels during cumulative oral provocation with sodium salicylate in four aspirin-sensitive asthmatic patients. All four tolerated drug despite SA plasma levels exceeding by 10-fold SA levels measured at occurrence of airway obstruction during aspirin provocation in same group of patients.

icant airway response without extrapulmonary symptoms (Table 1), while the remainder did not develop any signs of an adverse reaction in response to the highest dose of ASA administered (i.e., about 60 breaths of a 25 mg $\times$ ml $^{-1}$ solution). The bronchial provocations resulted in small but detectable amounts of both ASA and SA in plasma. For the

aspirin-tolerant subjects, these levels did not differ between asthmatic and nonasthmatic subjects, as illustrated in Fig. 5. Even in the three aspirin-sensitive asthmatic patients, who inhaled less of the lysine-ASA solution, bronchial absorption of ASA and SA was shown to occur. At the time of a bronchoconstrictive response, mean $\pm$ SD plasma levels were $0.28 \pm 0.16 \mu\text{g} \times \text{ml}^{-1}$ and $0.45 \pm 0.46 \mu\text{g} \times \text{ml}^{-1}$ for ASA and SA, respectively (Fig. 6), levels which were lower than after the oral challenges. For comparison, the plasma ASA and SA levels measured at the time of airway obstruction after oral aspirin provocation are shown in Fig. 6.

Discussion

It is not known why certain persons exhibit intolerance to ASA and other NSAIDs. The present investigation was intended to determine whether or not differences in pharmacokinetics of ASA could contribute to the propensity for reacting adversely. This possibility had not been assessed previously. Since the plasma levels of ASA and SA after standardized cumulative oral provocation with ASA were identical in aspirin-sensitive and aspirin-tolerant subjects, the findings refute the hypothesis of differences in the rate of absorption or subsequent hydrolysis of ASA into SA as an important feature in the pathogenesis of ASA-intolerance. Interestingly, in their classic paper of 1967, Samter & Beers (15) mentioned a series of *in vitro* experiments which indicated that the rate of deacetylation and plasma

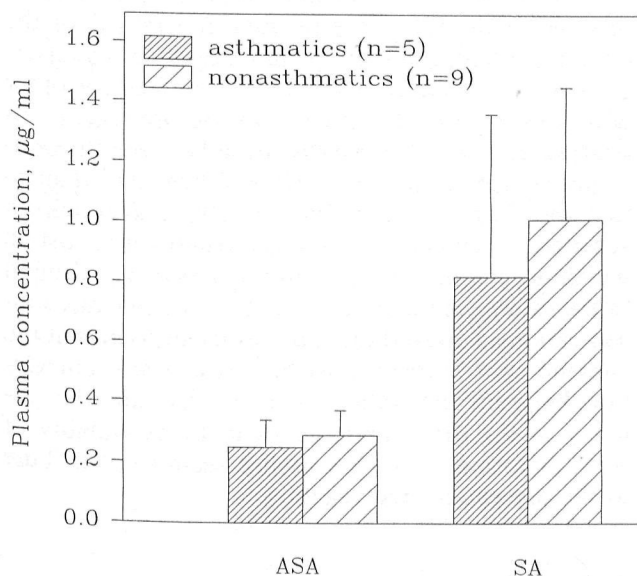


Fig. 5. ASA and SA plasma levels after inhalation of 1 ml of 25-mg/ml aspirin solution in five aspirin-tolerant asthmatic subjects (0.25 ± 0.09 and $0.82 \pm 0.54 \mu\text{g}/\text{ml}$) and nine subjects with normal airways (0.29 ± 0.08 and $1.01 \pm 0.44 \mu\text{g}/\text{ml}$).

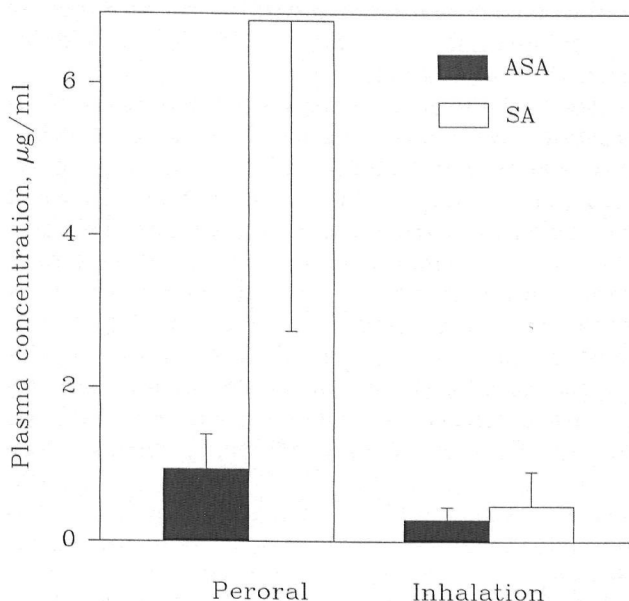


Fig. 6. ASA and SA plasma levels in four aspirin-sensitive asthmatic patients undergoing oral aspirin provocation and in three aspirin-sensitive asthmatic patients after inhalation of lysine-aspirin. Concentrations were measured when significant airway obstruction had developed.

protein binding was also normal in aspirin-sensitive patients, but the experimental data were not given.

It is reasonably well established that cyclooxygenase inhibition is the single mode of action in common for all drugs that evoke intolerance reactions in subjects with aspirin-intolerance (22). In fact, the potency of a drug as cyclo-oxygenase inhibitor is one factor which may predict the risk of intolerance reactions. However, the plasma level of aspirin or any other NSAID at the time of the intolerance reaction has not been determined previously. Therefore, it was of interest to find that the plasma levels of ASA at the time of the clinical appearance of an intolerance reaction were in the range ($0.2 \mu\text{g} \times \text{ml}^{-1}$ – $6 \mu\text{g} \times \text{ml}^{-1}$ = 1.1 – $33 \mu\text{M}$), which compares to what is known to be required for inhibition of cyclo-oxygenases by ASA in various *in vitro* systems (for review, see ref. 11). To the best of our knowledge, studies have not been performed, even in aspirin-tolerant subjects, to establish, by actual parallel measurements, the plasma levels of ASA and SA that correlate with *in vivo* inhibition of prostaglandin biosynthesis, but it seems reasonable to assume that significant inhibition of cyclo-oxygenase activity should occur when the plasma concentration of ASA is in the low micromolar range (11). Other properties of some NSAIDs, such as inhibition of neutrophil function and suppression of antigen-antibody reactions, require much higher drug concentrations; e.g., 2–3 mM for salicylates (1, 10). Therefore, the present findings lend further support

to the concept that inhibition of the cyclo-oxygenase is fundamental in the process leading to aspirin-elicited intolerance reactions.

Although the plasma levels of SA at the time of the reactions were much higher than those of ASA, provocation with sodium salicylate, yielding, on average, an additional 10 times higher plasma level of SA, failed to evoke intolerance reactions. This corroborates the clinical experience with SA and clearly demonstrates that SA is not responsible for the adverse reactions to aspirin (21). Incidentally, SA is at least 20 times less potent than ASA as a cyclo-oxygenase inhibitor *in vitro* (1, 25). It has been reported that certain salicylate derivatives, which are devoid of cyclo-oxygenase inhibiting effect in clinically used doses, are well tolerated by aspirin-sensitive subjects (4, 23). On the other hand, it is possible that sufficiently high doses (anti-rheumatic) of salicylate derivatives that are weak cyclo-oxygenase inhibitors may evoke adverse reactions in these patients, as has previously been suggested (18). Nevertheless, our own findings and those of Szczeklik et al. (22, 23) strongly support the view that clinically used doses of SA (0.5–1.5 g) yielding plasma levels of SA below $150 \mu\text{g} \times \text{ml}^{-1}$ can be tolerated by ASA-sensitive patients.

In addition, the present data may serve as a guideline for selecting the concentrations of aspirin that are relevant in experiments attempting to explore the mechanism of aspirin-intolerance. For instance, much basic research focuses on the effects of cyclo-oxygenase inhibition *in vitro* in different blood-, lavage-, and tissue cells. It is worth consideration that some *in vitro* studies (2) have used considerably higher concentrations of ASA than those presently shown to occur during the intolerance reaction.

Subjects who reacted with airway obstruction had lower plasma concentrations of ASA and SA at the time of the reaction, as compared with those who only developed other symptoms. The reason for this difference is not known. The number of subjects was limited, but should this observation be a consistent finding, it might give some clues in the search for the basic abnormality in aspirin-sensitivity. One possibility is that skin and connective tissue cells are exposed to lower levels of ASA than those of the lung. Alternatively, the target cells resident in the lungs may be more sensitive to ASA than are the target cells in the skin or connective tissue. A difference in the plasma levels at the time of pulmonary and extrapulmonary reactions may bring into question the view that a circulating cell is the common trigger of pulmonary and extrapulmonary symptoms. In opposition to this view, as also implied by the results of the aspirin-inhalation (see below), the findings suggest that the intolerance can be related to a specific dysfunction of tissue-residing cells. There is evi-

dence of mast cell activation in association with aspirin-elicited reactions (5, 16). Since mast cells are one major source of leukotrienes in the lung, it is of interest that cysteinyl-leukotrienes have been shown to be significant mediators of the airway obstruction induced by aspirin (6, 7). It has also been reported that a subset of patients with systemic mastocytosis exhibit aspirin-intolerance (20), a fact which may lend further circumstantial support to the hypothesis that tissue mast cells have altered function in subjects with aspirin-sensitivity. Of course, one possibility is that mechanisms other than inhibition of cyclo-oxygenase contribute to aspirin-induced extrapulmonary expressions such as urticaria, angioedema, and rhinitis. For instance, pretreatment with a histamine H_1 -antagonist has been reported to block the skin symptoms but not the airway obstruction after aspirin-challenge (12, 24), indicating that other mediators, such as leukotrienes, are more important in the lung.

Finally, bronchial absorption of ASA and SA did occur to the same extent in aspirin-sensitive and aspirin-tolerant asthmatic patients as well as in normal subjects. This has not been shown previously. It thus seems that airway inflammation does not influence absorption of ASA. Williams et al. (26) found no plasma salicylate after bronchoprovocation with lysine-aspirin, but they used another method for measurement. Interestingly, after bronchial provocation, the plasma levels of ASA, and in particular SA, were lower at the time of airway response than after oral challenge. This finding gives further support to the view that the airway obstruction is triggered locally in the airways.

In conclusion, the plasma concentration of ASA reached at the time of a positive reaction is of the dimension that is known to inhibit cyclo-oxygenases, but does not amount to the levels required for other effects of aspirin. Together with the previous demonstration of a positive correlation between the anti-cyclo-oxygenase activity *in vitro* of the drugs in question and their potency to induce asthmatic attacks in sensitive patients (22), two basic criteria are satisfied which are required to explain the action of a drug in terms of enzyme inhibition (10): 1) the plasma concentrations achieved must be sufficient to inhibit the enzyme, and 2) there must be a reasonable correlation between the antienzyme activity and the potency of the drug. Differences in bioavailability of ASA or formation of SA do not seem to contribute to the aspirin-elicited reactions.

Acknowledgments

We thank Ms H. Blomqvist and Ms M. Mohlin for expert technical assistance. The study was supported by grants from the Swedish Heart Lung Foundation, the Swedish Association

Against Asthma and Allergy (RmA), and the Swedish Medical Research Council (projects 14X-9071 and 522).

References

1. ABRAMSON S, KORCHAK H, LUDEWIG R, et al. Modes of action of aspirin-like drugs. *Proc Natl Acad Sci USA* 1985; 82: 7227-31.
2. AMEISEN JC, CAPRON A, JOSEPH M, et al. Aspirin-sensitive asthma: abnormal platelet response to drugs inducing asthmatic attacks. *Int Arch Allergy Appl Immunol* 1985; 78: 438-48.
3. ARM JP, O'HICKEY SP, SPUR BW, LEE TH. Airway responsiveness to histamine and leukotriene E_4 in subjects with aspirin-induced asthma. *Am Rev Respir Dis* 1989; 140: 148-53.
4. BIANCO S, PETRIGNI C, FELISI R, ROBUSHI M. Tolerance of guaiacolic ester of acetylsalicylic acid by patients with aspirin-induced asthma. *Scand J Respir Dis* 1989; 60: 350-4.
5. BOSSO JV, SCHWARTZ LB, STEVENSON DD. Trypsin and histamine release during aspirin-induced respiratory reactions. *J Allergy Clin Immunol* 1991; 88: 830-7.
6. CHRISTIE PE, SMITH CM, LEE TH. The potent and selective sulfidopeptide leukotriene antagonist, SK&F 104,353, inhibits ASA-induced asthma. *Am Rev Respir Dis* 1991; 144: 957-8.
7. DAHLÉN B, KUMLIN M, MARGOLSKIE D, et al. The leukotriene-receptor antagonist MK-0679 blocks airway obstruction induced by bronchial provocation with lysine-aspirin in aspirin-sensitive asthmatics. *Eur Respir J* 1993; 6: 1018-26.
8. DAHLÉN B, ZETTERSTRÖM O. Comparison of bronchial and per oral provocation with aspirin in aspirin-sensitive asthmatics. *Eur Respir J* 1990; 3: 527-34.
9. DAVISON C. Salicylate metabolism in man. *Ann N Y Acad Sci* 1971; 179: 249-68.
10. FLOWER RJ, MONCADA S, VANE JR. Analgesic-antipyretics and anti-inflammatory agents; drugs employed in the treatment of gout. In: GILMAN AG, GOODMAN LS, RALL TW, MURAD F, eds. *The pharmacological basis of therapeutics*. New York: Macmillan, 1985: 674-715.
11. GRYGLEWSKI RJ. Screening and assessment of potency of anti-inflammatory drugs *in vitro*. In: VANE JR, FERREIRA SH, eds. *Anti-inflammatory drugs. Handbook of experimental pharmacology*. Vol. 50, part II. Berlin: Springer-Verlag, 1979: 1-43.
12. PHILLIPS GD, FOORD R, HOLGATE ST. Inhaled lysine-aspirin as a bronchoprovocation procedure in aspirin-sensitive asthma: its repeatability, absence of late-phase reaction, and the role of histamine. *J Allergy Clin Immunol* 1989; 84: 232-41.
13. PICADO C, RAMIS I, ROSELLÒ J, et al. Release of peptide leukotriene into nasal secretions after local instillation of aspirin in aspirin-sensitive asthmatic patients. *Am Rev Respir Dis* 1992; 145: 65-9.
14. PLESKOW WW, STEVENSON DD, MATHISON DA, SIMON RA, SCHATZ M, ZEIGER RS. Aspirin-sensitive rhinosinusitis/asthma: spectrum of adverse reactions to aspirin. *J Allergy Clin Immunol* 1983; 71: 574-9.
15. SAMTER M, BEERS RF. Intolerance to aspirin. Clinical studies and consideration of its pathogenesis. *Ann Intern Med* 1968; 68: 975-83.
16. SLADEK K, SZCZEKLIK A. Leukotriene (LTs) overproduction and mast cell (MC) activation during aspirin-provoked bronchoconstriction in aspirin-induced asthma. *J Allergy Clin Immunol* 1992; 89: 233 (Abstract).
17. STEVENSON DD, ARROYAVE CM, BHAT KN, TAN EM. Oral aspirin challenges in asthmatic patients: a study of plasma histamine. *Clin Allergy* 1976; 6: 493-505.
18. STEVENSON DD, HOUGHAM AJ, SCHRANK PJ, GOLDLUST B, WILSON RR. Salsalate cross-sensitivity in aspirin-sensitive patients with asthma. *J Allergy Clin Immunol* 1990; 86: 749-58.
19. STEVENSON DD, SIMON RA. Aspirin sensitivity: respiratory and cutaneous manifestations. In: MIDDLETON E Jr, REED CE, ELLIS EF, ADKINSON NF, YUNGINGER JW, eds. *Allergy, principles and practice*. St Louis, MO: Mosby, 1988: 1537-54.
20. SUTTER MC, BEAULIEU G, BIRT AR. Histamine liberation by codeine and polymyxin B in urticaria pigmentosa. *Arch Dermatol* 1962; 86: 217-21.
21. SZCZEKLIK A. Aspirin-induced asthma: an update and novel findings. In: SAMUELSSON B, DAHLÉN S-E, FRITCH J, TAYLOR W, HEDQVIST P, eds. *Advances in prostaglandin, thromboxane and leukotriene research*. New York: Raven Press, 1994: (in press).
22. SZCZEKLIK A, GRYGLEWSKI RJ, CZERNIAWSKA-MYSIK G. Relationship of inhibition of prostaglandin biosynthesis by analgesics to asthma attacks in aspirin-sensitive patients. *BMJ* 1975; 1: 67-9.
23. SZCZEKLIK A, NIZANKOWSKA E, DWORSKI R. Choline magnesium trisalicylate in patients with aspirin-induced asthma. *Eur Respir J* 1990; 3: 535-9.
24. SZCZEKLIK A, SERWONSKA M. Inhibition of idiosyncratic reactions to aspirin in asthmatic patients by clemastine. *Thorax* 1979; 34: 654-7.
25. VANE JR. Inhibition of prostaglandin synthesis as a mechanism of action of aspirin-like drugs. *Nature New Biol* 1971; 231: 232-5.
26. WILLIAMS RW, PAWLOWICZ A, DAVIES BH. *In vitro* tests for the diagnosis of aspirin-sensitive asthma. *J Allergy Clin Immunol* 1990; 86: 445-51.

This document is a scanned copy of a printed document. No warranty is given about the accuracy of the copy. Users should refer to the original published version of the material.