

## Histamine Receptor Bearing Suppressor Cells in Nickel Sensitivity

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**Abstract.** Histamine receptor bearing suppressor lymphocytes regulating blast transformation are present in the peripheral blood of nickel-sensitive subjects but not in nickel-unresponsive subjects. Removal of these cells caused a 63% enhancement of the nickel-induced blast transformation of 8 nickel-sensitive subjects, but no significant enhancement was seen in 8 healthy control subjects. Soluble histamine had a significant suppressive effect on the nickel-induced blast transformation in the allergic subjects. The inhibitory effect was mediated through histamine receptor type 2, since the receptor type 2 antagonist cimetidine had an enhancing effect (33%) on the blast transformation which was not noticed with the receptor type 1 antagonist prometasine hydrochloride.

**Key words:** Histamine receptors; Suppressor cells; Nickel sensitivity

Histamine is an important mediator in immediate hypersensitivity, but it plays a regulatory role in delayed hypersensitivity, too. Soluble histamine itself has an inhibitory effect on blast transformation (3). T lymphocytes have been found to produce a histamine-induced suppressor factor (4). Antigen-specific suppressor cells bearing histamine receptors have been found to develop during allergy desensitization in man (5).

### MATERIALS AND METHODS

#### Subjects

Thirteen nickel-allergic subjects were tested for the histamine sensitivity of the nickel-induced blast transformation. Eight nickel-allergic and 8 healthy subjects were tested for the presence of histamine receptor bearing lymphocytes. The cells of 8 nickel-allergic subjects and 2 healthy controls were studied with histamine receptor type 1 and type 2 antagonists. The diagnosis of nickel allergy was based on a positive skin chamber test (Finn Chamber<sup>®</sup>, Epitest Ltd, Helsinki, Finland) with 2.5% nickel sulphate in petrolatum T.

#### Stimulants and immunomodulators

Nickel sulphate (Merck, Darmstadt, West Germany) was used at final concentrations of 1.25 and 6.25  $\mu\text{g/ml}$ . Histamine dihydrochloride (Sigma, St. Louis, USA) was used at final concentrations of 0.1 and 0.5  $\text{ng/ml}$ . Histamine receptor type 1 antagonist prometasine hydrochloride (Phenergan<sup>®</sup>, Rhône-Poulenc, Birkerød, Denmark) was used in a final concentration of 0.1  $\text{ng/ml}$ , and histamine receptor type 2 antagonist cimetidine (Tagamet<sup>®</sup>, Lääke Oy, Turku, Finland) was used at a final concentration of 0.1  $\text{ng/ml}$ . The stimulants were diluted in RPMI-1640.

#### Separation of lymphocytes bearing histamine receptors

A modified method of antigen-coated dishes (2) was used. Bacteriological Petri dishes were coated with histamine-rabbit serum albumin (hist-RSA) conjugate, which was prepared as described by Weinstein et al. (8). Control dishes were prepared using RSA without histamine. The specificity of the dishes was checked by FITC-conjugated histamine. The cells attached to the hist-RSA coated plates contained 60–70% histamine receptor bearing cells and those attached to the control dishes 4–6%.

#### Cell cultures

The peripheral blood mononuclear cells were separated from venous blood samples by the Bøyum method (1). The lymphocyte blast transformation reactions were performed as described earlier (6). In addition, macrophages of 50  $\mu\text{l}$  cell suspension were allowed to attach (2 h, +37°C) to the culture well bottoms in the tests done with cells, with and without histamine receptors, and the wells were washed three times with Hanks' balanced salt solution before culturing the wells.

#### Statistical methods

The Wilcoxon test for pair differences was used.

### RESULTS

#### Sensitivity of nickel-induced blast transformation to histamine and histamine receptor type 1 and type 2 antagonists

Histamine had a clearly inhibiting effect on the nickel-induced blast transformation reaction. The mean of the nickel sulphate reaction without histamine was  $7280 \pm 1848$  cpm in the contact sensitivity patients. In the presence of 0.5  $\text{ng/ml}$  of histamine, it was  $4051 \pm 1848$  cpm ( $p < 0.01$ ), and with 0.1  $\text{ng/ml}$  of histamine, the mean stimulation was  $4160 \pm 797$  cpm ( $p < 0.01$ ). The background of these tests was  $940 \pm 313$  cpm.

The histamine receptor type 2 antagonist cimetidine had a marked ( $p < 0.025$ ) enhancing effect on the nickel-induced blast transformation, whereas the receptor type 1 antagonist prometasine hydrochloride had no enhancing effect. The results are summarized in Table 1.

Table 1. The effect of histamine receptor type 1 antagonist prometasine hydrochloride (0.1 ng/ml) and type 2 antagonist, cimetidine (0.1 ng/ml) on nickel (6.25 µg/ml) induced blast transformation (cpm ± SEM) in 8 nickel-allergic subjects and 2 healthy subjects

	Nickel-allergic subjects	p value	Healthy subjects
Unstimulated cultures	1 695 ± 648		1 134 ± 460
Cells with the receptor antagonists	1 714 ± 495		1 661 ± 641
Nickel reaction without the receptor antagonists	17 321 ± 2 227	} <0.025 NS	842 ± 222
Nickel reaction with cimetidine	22 975 ± 3 515		1 400 ± 831
Nickel reaction with prometasine hydrochloride	17 577 ± 1 953		1 118 ± 461

#### Suppressive effect of cells bearing histamine receptors

Removal of the cells bearing histamine receptors had a significantly enhancing effect (63%) on the nickel-induced blast transformation when compared with the reactions of the unseparated cells ( $p=0.005$ ). The cells bearing histamine receptors did not react to nickel. When the cells were separated on the control dishes coated with RSA, the non-attached cells showed a slight enhancement (17%) of nickel-induced blast transformation.

In the control subjects no significant cell proliferation was seen in either cell group. The results are summarized in Table II.

#### DISCUSSION

Unresponsiveness of the healthy lymphocytes to nickel does not seem to be due to suppressor cells regulating blast transformation in peripheral blood,

which confirms the previous findings where HLA-identical lymphocytes from healthy subjects were not able to suppress the nickel reaction of nickel-sensitive subjects (7). Histamine receptor bearing antigen-specific suppressor T cells have been reported to develop during allergy desensitization in man (5) and the present study shows that they are also produced in nickel sensitivity. Their role is possibly to prevent nickel-sensitive clones from causing excessive damage in the affected subject. However, separation of lymphocytes with the control dishes without histamine on them also caused a slightly increased nickel reaction of the non-attached cells. This indicates that other cells also have a regulatory role. A subpopulation of macrophages is possibly involved, since they have a non-specific ability to adhere. Ia-antigen positive macrophages or Langerhans cells are essential as nickel-presenting cells to T cells, but the present study indicates the functional heterogeneity of the

Table II. The effect of removal of histamine receptor bearing cells on the nickel (mean of 1.25 and 6.25 µg/ml) induced blast transformation reaction (cpm ± SEM) in 8 nickel-allergic and healthy subjects

The histamine receptor bearing cells were separated by a monolayer technique with histamine-RSA coated dishes: dishes coated with RSA were used as controls. cpm values of unstimulated cultures (mean 1 741 ± 615 cpm) have been subtracted

	Nickel-allergic subjects (cpm)	p value	Healthy control subjects (cpm)	p value
Nickel reaction of unseparated cells	11 429 ± 3 357	} =0.005 <0.025	506 ± 245	} NS
Nickel reactions of cells separated with histamine-RSA dishes:				
non-attached cells	18 578 ± 4 129		1 361 ± 477	
cells attached	261 ± 190	0		
Nickel reactions of cells separated with control RSA dishes:				
non-attached cells	13 317 ± 2 963		1 070 ± 432	
cells attached	227 ± 190		0	

macrophages in the regulation of nickel sensitivity.

Soluble histamine had an inhibitory effect on nickel-induced blast transformation. Even in the absence of exogenous histamine, the receptor type 2 antagonist was able to increase the nickel reaction, which suggests that the suppressive effect of histamine receptor bearing cells is not dependent on excessive amounts of histamine but is a readily expressed ability when nickel-sensitive clones begin to proliferate.

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## Clinical Evaluation and Pathomechanism of Urticaria-like Skin Eruption in Systemic Lupus erythematosus

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**Abstract.** Clinical observations of 10 SLE patients having urticaria-like skin symptoms showed that the clinical process of urticaria found in patients suffering from SLE differs from the classic one. On the basis of histological and immunohistological studies in 5 cases, a characteristic histological manifestation of leukocytoclastic vasculitis of small veins in the upper third of the cutis was found. The various urticaria-like skin symptoms in SLE are caused by immunocomplexes of various components. The continuous presence of urticaria is caused by C-activating formation of anaphylatoxin. The appearance of urticaria in SLE patients is an unfavourable precursor which has been confirmed by statistical analysis.

**Key words:** Systemic lupus erythematosus; Urticaria; Immunocomplex

Systemic lupus erythematosus (SLE) is not infrequently accompanied by urticaria or by urticaria-like manifestations (1, 2, 3, 4, 5, 8, 10). *Dubois* found urticaria in 6.9% of 520 cases, while *Harvey* found it in 7% of 105. *Estes* found a higher rate of urticaria—13%—in patients suffering from SLE. In our study the proportion was 12.5%.

The urticaria-like skin symptoms prompt the following questions: What are their clinical characteristics; to what extent do they differ from urticaria caused by histamine; are they connected functionally with SLE; what is their importance for the prognosis of the disease?

The purpose of our study was to find answers to these questions raised in the course of clinical observations of 10 SLE patients having urticaria-like skin symptoms and by histological and immunohistological analysis of biopsies made of skin symptoms.

#### MATERIALS AND METHODS

For 9 years, 10 SLE patients with urticaria-like skin symptoms (on the basis of ARA criteria) were observed (age range 24–57 yrs, average 40.5, all females). The