Immunohistochemical Study of Pi Class of Glutathione S-Transferase in Psoriasis and Eczematous Dermatitis

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Background: **Glutathione S-transferase(GST) is a family of isoenzymes that play an important role in protecting cells from cytotoxic and carcinogenic agents. Strong activities of GST-Pi in the epithelial tissues was observed in the upper layer of skin, gastrointestinal tract and placenta which have been exposed to exogenous chemicals.**

Objective: **This study was done to observe the distribution pattern of GST-Pi in normal, acute and chronic psoriasis or eczematous dermatitis, using paraffin-embedded human skin tissues.**

Method: **Twenty-one psoriatic and twenty-six eczematous dermatitis specimens were observed by immunohistochemical staining using an anti-rabbit GST-Pi polyclonal antibody.**

Results: **Stainining reaction for the GST is weakly to moderately stained in the normal epidermis. In the acute stage, upper layer shows weak and moderate staining in the lower epidermis of the psoriasis and eczematous dermatitis but in the chronic stage GST-Pi are noted strongly expression in upper epidermis.**

Conclusion: **Immunohistochemical staining for the GST-Pi reveals a more abundant distribution in the chronic stage rather than in the acute stage of psoriasis and eczema tous dermatitis, showing no disease specificity. Therefore it is suggested that the detoxifying capacity decreases in the acute stage of above dermatosis.**

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Key Words: Glutathione S-transferase- Pi, Eczema tous dermatitis, Psoriasis

Glutathione(GSH) is the tripeptide ι-γ- glutamyl-ι- cysteinyl-glycine. It is found in almost all mammalian cells and is usually the most abundant intracellular thiol[1]. GSH participates in biologic function as a coenzyme for several enzymes and conjugating with endogenous and foreign compounds. It is involved in amino acid transport, the formation of DNA precursors and protection of cells against oxygen toxicity with two glutathione dependent enzymes, *glutathione-S-transferase*(GST) and glutathione peroxidase(GPX)[1].

Glutathione S-transferase(GST) is one of the major enzymes, responsible for detoxification against toxic chemicals[2]. Many isoenzymes of GST can be separated by biochemical and immunologic characteristics into three distinct classes named alpha, mu, and pi[3,4]. Among GST isoenzymes, the acidic form of GST(GST-Pi in human and GST-P in rat) has been of interest to the oncologists because the carcinogenic insult would induce GST-P expression especially in the preneoplastic lesion of the murine model[5]. However in the human study the

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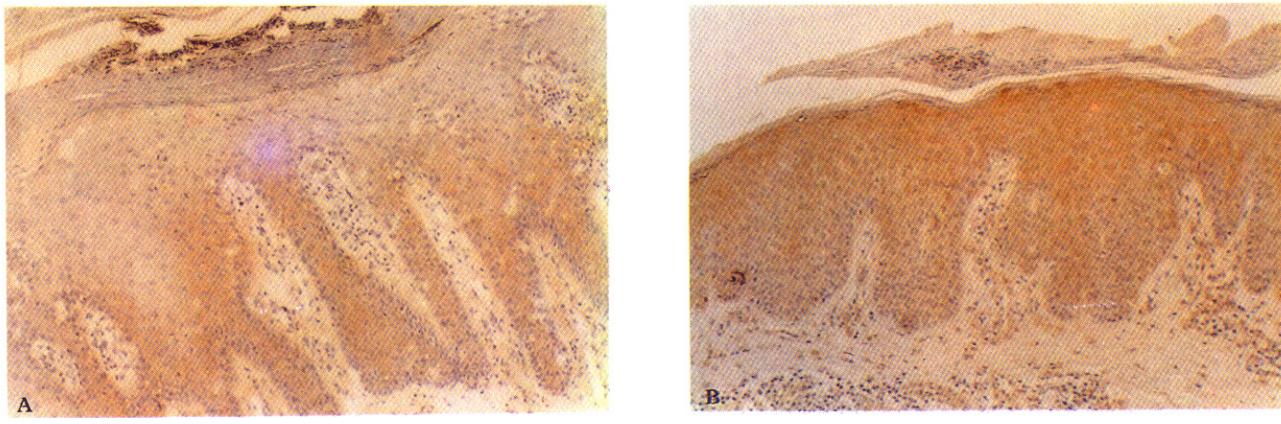

Fig. 1. Weak reactions of lower epidermis in acute psoriasis(A) and strong reaction of upper epidermis in chronic psoriasis(B) to GST-Pi antibody (ABC immunoperoxidase staining, × 100).

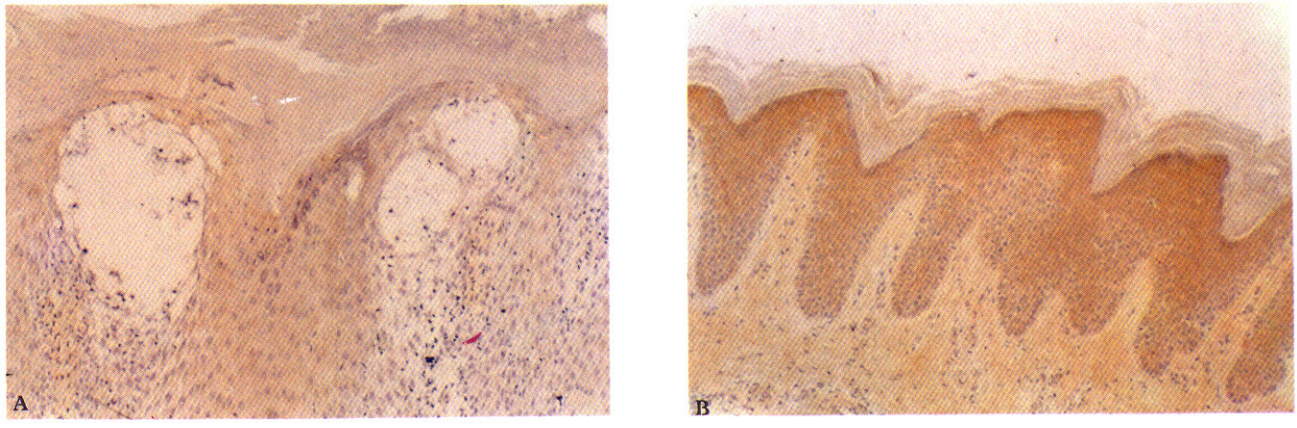

Fig. 2. Weak reactions around spongiosis and reticular degeneration in acute eczematous dermatitis (A) and strong reactions of the upper epidermis in the chronic eczematous dermatitis(B) to GST-Pi(ABC immunoperoxidase staining, × 100).

possibility of GST-pi as a preneoplastic marker in human tissues has become controversial[6,7]. GST-Pi was expressed predominantly in normal epithelial cells of the urinary, digestive and respiratory tracts, suggesting a role for GST-Pi in detoxification and elimination of toxic substances[6] in organs exposed to the outside.

Therefore in this study we tried to compare the distribution patterns of GST-Pi expression with the acute and chronic stages of psoriasis or eczematous dermatitis in order to elucidate the role of defoxifing enzymes against endogenous or exogenous environment in two inflammatory dermatoses.

MATERIALS AND METHODS

Materials

Biopsy specimens of psorasis (n=21) and eczematous dermatitis(n=26, including atopic dermatitis, contact dermatitis and lichen simplex chronicus) were subjected to elliptical biopsy under local anesthesia. A portion of each lesion was processed for routine histologic examination and stanined with hematoxylin and eosin for diagnostic confirmation. Psoriatic specimens were defined by the histologic criteria of Griffin et al[8,9]. The histologic criteria were (1)acute psoriasis:diffuse parakeratosis, Munro microabscess, absent granular layer, scatter neutrophils and perivascular swelling;(2) chronic

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psoriasis:compact hyperkeratosis, spotted or absent parakeratosis, intact granular layer and mild endothelial swelling. Eczematous dermatitis lesions were classified by histologic finding of spongiosis, reticular degenration, acanthosis and exocytosis[10]. We used a control group from normal skin specimen(n=6).

Methods

The formalin fixed-paraffin embeddied specimens were subjected to immunohistochemical analysis with successive treatment of anti-GST-Pi antibody(1:1000 dilution, kindly donated by Sang Chul Park at Seoul National University). For the critical cases, the GST-Pi enzyme isolated from the human placental tissues was added to the reaction mixture to block the primary antibody in order to exclude the non-specific cross-reactive problem. The remaining immunochemicals for the three-step peroxidase- antiperoxidase(PAP) procedure were purchased as a PAP kit from Dako, INc., followed by visualization with diaminobenzidine peroxidation.

RESULTS

Acute psoriatic lesions are weakly stained in lower epidermis but upper epidermis with spongiform pustulosis and parakeratosis (Fig. 1A). Chronic psoriatic lesions with acanthosis were noted with moderate reactions in the lower epidemis and strong reactions with diffuse distribution in the upper epidermis(Fig. 1B). Acute eczematous lesions were weakly stained around spongiosis and reticular degeneration sites in the epidermis (Fig. 2A). But chronic eczematous lesions showed strong reactions with diffuse distribution in the upper epidermis(Fig. 2B),(Table 1).

Table 1. Immunohistochemical reaction of glutathione S-transferase - Pi in psoriasis and eczematous dermatitis

	Normal	Psoriasis		Eczema	
		Acute	Chronic	Acute	Chrnoic
Horny layer	–	–	+	–	+
Upper epidermis	+~–	–	+++	+	+++
Lower epidermis	+	+	++	+	++
Basal layer	+	+	+	+	+

-,negative; +,weak;++,moderate;+++,strong reactions

DISCUSSION

Glutathione S-transferase(GST) is a family of isoenzymes that have overlapping substrate specificities and play an important role in the protection of cells from cytotoxic and carcinogenic agents[6,11]. GST can conjugate electrophilic substrates with the tripeptide glutathione resulting in less toxic and more readily excreted metabolites. In addition GST can bind and sequester intracellular toxins and certain GST isoenzymes can prevent oxidative damage by an intrinsic organic peroxidase activity that converts toxic peroxides to inactive alcohols[12].

GST isoenzymes can be characterized on the basis of substrate specificity, sensitivity to inhibitors, isoelectric points, immunologic cross reactivity and amino acid composition and sequence[3]. Species independent classificaiton has been proposed establishing three classes of cytosolic GSTs named alpha, mu, and pi[5].

New expression of GST-P form in the murine preneoplastic lesion of hepatocarcinogenic model[13] had been proposed its possibility as a preneoplastic marker in human tissues[5]. However in human study the expression of GST-Pi in the normal liver tissues raised the question of its usefulness as a marker for tumorigenic changes[14].

Multidrug-resistant human breast cancer cells selected for resisitance to adriamycin were noted to have a marked increase in the activity of GST-Pi[15,16], suggesting it to be a marker of drug resistance[6].

Knowledge of the normal tissue distribution of GST-Pi should help to define its significance as it relates to the exposure of tissues to xenobiotic challenge. In normal human tissues GST-Pi levels were higher in epithelia, although strong staining was found in mesenchymal tissues[6,17].

Generally typical psotiatic and eczematous lesions may not be difficult to differentiate but sometimes confusions of diagnosis happens in the course of progression of the diseases. In a study of moleclar mechanism for the defensive roles of skin tissues we thought that GST-Pi may be related to the essential events or a triggering factor in dif-

ferentiation of keratinocytes.

In the present study we tried to compare the expression of GST-Pi in the acute and chronic stage of psoriasis or eczematous dermatitis. As summarized in table 1, in both acute inflammtory lesions the increase of GST-Pi expression was observed in the lower epidermis compared with normal epidermis. But the chronic stage of thicken epidermis shows the strongly stained reactions to GST-Pi antibody rather than acute stages in psoriasis and eczematous dermatitis.

According to Park et al[7], activities to the aged cultured hepatocytes resulted in the expression of GST-Pi. Because tumor tissues are more resistant to a variety of toxic chemicals and the aged cultured hepatocytes might be in the state of stronger endurance to environmental insults, the expression of GST-Pi might be the results of cellular adaptation for longer survival[7]. In the acute stage of inflammatory dermatoses, although the upper epidermis shows a decrease of GST-Pi, pustular formating areas of inflammatory cell infiltration were more strongly stained. It may be related to its role of decreasing the detoxyfing activities against the toxic environment.

Therefore, it is suggested that the higher GST-Pi expression in the chronic stage of inflammatory skin dermtoses might be either an adaptive response or a reparative process and related to its role as the detoxifying system against the endogenous or exogenous toxic environment.

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