Table I. *5-S-Cysteinyldopa, dopa, and dopamine in tissues and body fluids (ng/g)*

Guinea-pig	Kidney			Urine			Serum			Heart		
	5-S-CD	Dopa	DA	5-S-CD	Dopa	DA	5-S-CD	Dopa	DA	5-S-CD	Dopa	DA
1	230	3.0	2.1	63	4.9	17	7.2	1.6	0.0	0.0	6.2	9.4
2	130	3.0	3.5	120	1.0	71	2.6	0.7	0.5	2.5	7.9	13
3	100	17	3.8	23	2.6	14	2.1	0.6	0.3	2.0	4.6	22
4	140	6.9	6.2	–	–	–	3.9	0.5	0.5	4.1	4.6	17

in this organ. The possibility of some aberrant melanocytes should also be considered.

The levels of dopa and dopamine in heart and spleen are high compared with those in serum and here their localization to special structures that can decarboxylate dopa is the likely explanation for the concentrations observed.

The cervical ganglion analysed from one guinea-pig contained 25 ng dopa/g tissue, but no 5-S-cysteinyldopa or dopamine.

The results obtained illustrate the important role of the kidney in the excretion of 5-S-cysteinyldopa, having tissue levels of the same order as for the urine, and far above the serum levels. The concentrations of the different catechols in the spleen and in the heart indicate the presence of specific catechol-containing structures in these organs. The results obtained show that our method offers new possibilities of investigating dopa metabolism in the body.

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Nuclear DNA-Polymerase Estimation in Human Epidermal Cells

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Abstract. A new technique which detects the presence of DNA-polymerase and primer-template DNA by measuring the incorporation of ^{3}H-thymidine-5'-triphosphate (^{3}H-TTP) showed nuclear labelling of enzymatically liberated epidermal cells of human skin.

Key words: Nuclear DNA-polymerase; Human epidermal cells

Two different types of DNA-polymerase have been demonstrated (7), viz. one cytoplasmic of high molecular weight, 3S. Variations in both the high molecular weight and low molecular weight DNA-molecular weight and low molecular weight DNA polymerase have been shown during the cell cycle, with an increase correlated to DNA synthesis (2, 10). Ooka & Daillie (9) found variations in both the nuclear and the cytoplasmic DNA-polymerase during the cell cycle in synchronized KB cells and suggested that the cytoplasmic 8.3S DNA polymerase may simply be a storage form of the enzyme from which the 3.4S DNA-polymerase can be formed when needed for DNA synthesis. These

Spleen		
5-S-CD	Dopa	DA
19	33	10
7.2	6.7	4.1
16	19	10
11	10	6.0

results are consistent with the notion that DNA-polymerase, particularly the nuclear variety, is needed for DNA replication.

The above-mentioned studies have been based on biochemical analyses. In 1973, Nelson & Schiffer (8) described a technique allowing autoradiographic detection of ^{3}H-thymidine-5′-triphosphate (^{3}H-TTP) incorporated into nuclear DNA of sarcoma-180 mouse ascites tumour cells under direction of their own nuclear DNA-polymerase. This method is a single-cell assay with respect to nuclear DNA-polymerase, provided the cell also contains primer-template DNA (11). After modifying the technique, Lange Wantzin (5) succeeded in demonstrating nuclear labelling of haemopoietic cells (6).

The present work demonstrated ^{3}H-TTP labelling of human epidermal cells dispersed by enzymes.

MATERIALS AND METHODS

Normal mammary skin of five young women was supplied by the Department of Plastic Surgery. Viable cells were dissociated from the epidermis (3). The epidermal cells were smeared immediately on rinsed glass slides, fixed in absolute ethanol : acetone (1 : 1 v/v), and incubation chambers made by attaching a ring on top of the smear. The incubation fluid consisted of 20 mM TRIS buffer, pH=7.8, with 0.135 mM 2′deoxyadenosine-5′-triphosphate (dATP), 2′deoxycytidine-5′,triphosphate (dCTP), and 2′deoxyguanosine-5′-triphosphate (dGTP) with the addition of 5 mmol $MgCl_2$, and finally 10 μCi ^{3}H-TTP were added to each chamber (spec. act. 40 Ci/mmol). Ficoll was added to a final concentration of 40 g/dl to preserve the cells. The slides were incubated for one hour at 37°C. After rinsing in water and phosphate buffer, additional fixation with formaldehyde solution, and rinsing in phosphate buffer, autoradiograms were prepared by dipping, using Kodak NTB-2 liquid emulsion. Exposure time was 2 weeks. Developing took 2 minutes with Kodak D-19b developer. Some 1 000–2 000 cell nuclei were counted to determine the percentage of nuclei labelled with ^{3}H-TTP (^{3}H-TTP labelling index=^{3}H-TTP LI). After background estimations, cells were considered to be labelled when they contained more than 4–6 grains over the nucleus.

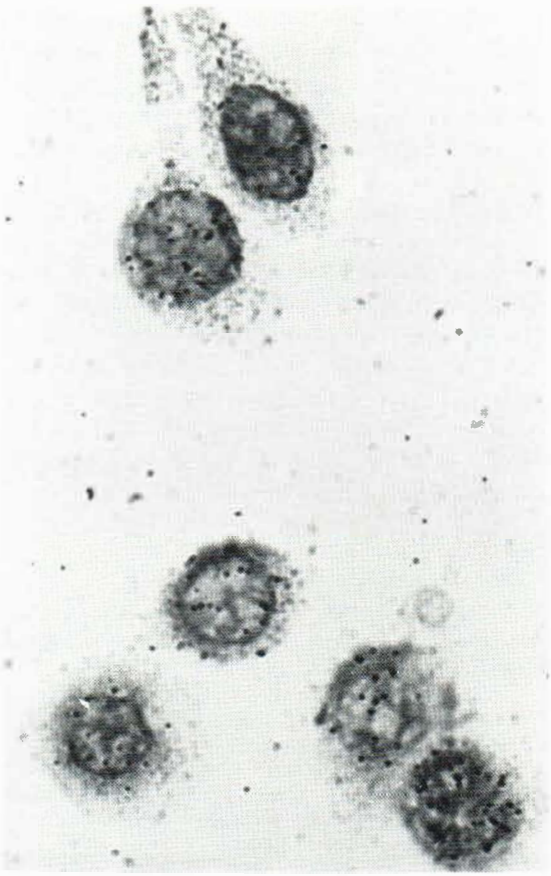

Fig. 1. ^{3}H-TTP-labelled nuclei of epidermal cells.

RESULTS

In all five cases the nuclei were well preserved. Silver grains were mainly found over the nuclei. The grain count of ^{3}H-TTP-labelled nuclei (Fig. 1) was usually between 15 and 20, but a few cells with up to 40 grains were observed. The ^{3}H-TTP LI values were found to vary between 54.2% and 9.2%, with a median value of 66% and an average value of 69%.

DISCUSSION

The incorporation of ^{3}H-TTP under the same experimental conditions as used in this study has been shown to indicate the presence of nuclear DNA-polymerase (8). In 1977, Braunschweiger & Schiffer demonstrated that the nuclear DNA-polymerase is sensitive to *p*-chloromercuribenzoate, which indicates that the enzyme is the replicative DNA-

polymerase (1). The method used involves damage to the cell membrane which allows ^{3}H-TTP and the unlabelled triphosphates to enter the cell body and to be incorporated into DNA after induction of DNA synthesis utilizing nuclear DNA-polymerase and preformed DNA as primer-template.

A cell population may be divided into 'proliferating' and 'non-proliferating' cells. ^{3}H-thymidine labelling indicates the fraction of cells in DNA synthesis (4), whereas the present method indicates the fraction of cells having the potential for replicative DNA synthesis. In experimental tumour systems this method has been shown to be a valid measure of the growth fraction (12). This ^{3}H-TTP single cell assay may provide a new parameter of human epidermal cell kinetics.

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Acute Generalized Pustular Bacterid and Immune Complexes

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Abstract. The case is described of a 39-year-old man with acute generalized pustular bacterid and high $C1_q$-binding activity in the sera during the active stage. Histamine-induced vascular changes studied by immunofluorescence microscopy revealed a perivascular deposition of IgM and C3. These findings support the view that leukocytoclastic vasculitis underlying the subcorneal pustules is mediated by immune complex deposition.

Key words: Pustular bacterid; Leukocytoclastic vasculitis; Immune complex

Recently, Tan (5) reported the case of a 58-year-old woman, who suddenly developed widespread pustules and purpuric lesions after streptococcal upper respiratory infection. The acute clinical course and histopathological observations which revealed a leukocytoclastic vasculitis underlying the pustules, were quite different from those of pustulosis palmaris et plantaris (PPP). The author consequently considered this to be a true case of "acute generalized pustular bacterid".

We have investigated the immune complexes in both sera and tissues of a patient with acute generalized pustular bacterid and found a possible relationship between the immune complex deposition and leukocytoclastic vasculitis.

CASE REPORT

A 39-year-old Japanese man came to our clinic in October 1978, complaining of numerous asymptomatic pustules