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Brief Note

The Effect of DNP Specific Factor on Induction of Contact Sensitivity to DNCB

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In our previous studies,[1] we were able to show that 2, 4-dinitrophenyl groups were distributed on the cells (DNP cells) in the lymph nodes of naive guinea pigs draining the site of painting with 2, 4-dinitrochlorobenzene (DNCB). The DNP cells detected by immunofluorescence using anti-DNP antibody could be increased by treating the animals with cyclophosphamide (CY) before painting with DNCB. Furthermore, culture supernatant from draining lymph node cells of the naive animals was found to possess factor which specifically reacted with DNP groups (DNP specific factor). It is suggested that the factor can effectively block the anti-DNP antibody reacting with DNP cells and possively cause reduction in the number of detectable DNP cells in the draining lymph node of normal animals. The pretreatment with CY is considered to prevent the production of the factor. The object of the experiment reported here is to investigate the effect of the DNP specific factor on inducing contact sensitivity to DNCB.

Animals used were male inbred JY1 guinea pigs weighing 350 to 450 g. A group of them were injected intraperitoneally with 250 mg/kg CY 3 days before painting with DNCB. An application of totally 0.2 ml of 5% DNCB-ethanol solution was given to both sides of inguinal skin of normal and CY treated animals. The inguinal lymph nodes were taken 24 hours after painting with DNCB and cell suspensions were prepared by teasing in Eagle's minimal essential medium (MEM). The cells from naive animals painted with DNCB were cultured in Eagle's MEM, 10% fetal calf serum, penicillin, streptomycin and glutamine for 48 hours at 37°C as described by Zembala and Asherson[2]. The supernatants were collected by centrifugation at 4200g for 30 minutes. The cells from CY treated animals painted with DNCB were incubated in the supernatants for one hour at 37°C (10^7 cells/ml). The cells from normal or CY treated animals, or cells from CY treated animals which were incubated in culture supernatants, were injected subcutaneously to ears of normal recipients. Sensitivity was tested by contact with 0.2, 0.09, 0.05, and 0.01% DNCB in ethanol on the flank 14 days later. The intensities of skin reactions were assessed 24 hours after patch testing as described previously.[3]

The injection of lymph node cells taken at 14 hours post painting with DNCB from normal animals was able to induce a contact sensitivity in the recipient animals as shown by a positive patch test reactions (group 1 and 2 in Table). Injection of 3×10^7 cells from CY treated animals (group 3) was also capable of producing sensitivity, whereas 1×10^7 cells from the animals

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(group 4) were less effective. Induction of the cells in the culture supernatants clearly increased an ability to induce contact sensitivity (group 5). These findings suggest that DNP specific factor play a role in induction of contact sensitivity. Further investigation are necessary to clarify the character of the factor and its participation in the development of contact sensitivity.

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TABLE

The Induction of Contact Sensitivity with the Draining Lymph Node Cells Taken from Guinea Pigs 24 Hours after Skin Painting with DNCB

Animal group	Doner cells			Patch test	
	Treatment of animals		Cell number	Mean intensity	Positive
1		DNCB	3×10^7	2.0	8/8
2		〃	1×10^7	0.4	4/6
3	CY	〃	3×10^7	2.1	6/7
4	〃	〃	1×10^7	0.1	2/8
5	〃	〃	1×10^7(F)	0.8	7/8

F, Doner cells were incubated in DNP specific factor.

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