of cotton or other forms of "native" cellulose in the characterization of cellulolytic enzymes from wood-destroying fungi may not be a valid test of the ability to degrade the cellulose in wood.

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Suppressed Multiplication of *Listeria monocytogenes* within Macrophages derived from Thymectomized Mice

THE thymus has been reported to be important in the development of the immunological capacities of several animal species, so that neonatal thymectomy impairs many kinds of immune responses. On the other hand, there have been few reports of the effects of thymectomy on the function of the macrophage, which is known to be involved in immune responses. No differences have been detected between the reticuloendothelial phagocytic activity of neonatally thymectomized and control animals in response to colloidal gold[1] and carbon[2]. On the other hand, Corsi and Giusti[3] reported that although a single injection of colloidal carbon made no difference to either group of rats, after a second injection carbon was removed more quickly in the thymectomized rats. While investigating the infection of thymectomized mice with *Listeria monocytogenes*[4], we observed a phenomenon which may indicate that macrophage function is modified by neonatal thymectomy.

Mice of the *CF*1 strain were used. Thymectomy was performed from 6 to 18 h after birth. Sham-thymectomized mice were used as controls throughout. Mice that developed "wasting" disease and those with remnants of thymus in the thymectomized group were discarded. Five to six weeks after thymectomy the mice were injected intraperitoneally with 0·1 mg of glycogen dissolved in 1 ml. of saline to induce the accumulation of macrophages in the peritoneal cavity. Three or four days after injection, exudate cells were collected from the peritoneal cavity and cultivated in Petri dishes containing four pieces of cover-slips in a moist atmosphere containing 5 per cent CO_2. Hanks BSS containing 0·5 per cent lactalbumin hydrolysate and 25 per cent bovine serum was used as the medium. After 18 h, when the culture consisted of a complete monolayer of macrophages, the medium was discarded and cells of *L. monocytogenes* suspended in Hanks BSS were added. After incubation for 1 h the cultures were washed by a jet stream of Hanks solution to remove uningested bacteria, and were then added to Hanks solution containing 0·5 per cent lactalbumin hydrolysate and 25 per cent bovine serum. At intervals during incubation, cover-slips were fixed with methanol, stained with Giemsa solution, and the bacteria were counted in 50 to 100 macrophages.

We found no significant difference in phagocytic activity towards the bacteria between macrophages derived from neonatally thymectomized and those from control mice (Table 1). After incubation for 8–9 h mean numbers of bacteria per macrophage were consistently greater in the control group (Table 2). The difference in the growth rates in both groups was statistically significant ($P < 0{\cdot}001$) in all experiments listed. This indicates that the multiplication of *Listeria* is suppressed within macrophages derived from the neonatally thymectomized mice. In other words, the multiplication of the bacteria is enhanced in the macrophages derived from the sham-thymectomized control mice. The intracellular growth of *Listeria* was examined using macrophages obtained from mice thymectomized as adults (6 weeks old). The results showed no difference between thymectomized and control mice in phagocytic activity to the bacteria or in multiplication of the bacteria.

Table 1. PHAGOCYTIC ACTIVITY OF MACROPHAGES DERIVED FROM NEONATALLY THYMECTOMIZED AND CONTROL MICE TO *L. monocytogenes*

Experiment No.	% * of macrophages which ingested *Listeria* after 1 h contact	
	Control mice	Thymectomized mice
1	49·3	48·7
2	50·7	43·0
3	44·0	50·7

* Calculated from 300 cells.

Table 2. MULTIPLICATION OF *L. monocytogenes* IN CULTURED MACROPHAGES DERIVED FROM NEONATALLY THYMECTOMIZED AND CONTROL MICE

Experiment No.	Incubation period (h)	Mean numbers of *Listeria* per one macrophage obtained from	
		Control mice	Thymectomized mice
1	0	2·4 ± 0·2*	2·7 ± 0·2
	8	12·5 ± 0·6	9·2 ± 0·6
2	0	2·4 ± 0·1	2·7 ± 0·2
	9	14·2 ± 0·7	9·9 ± 0·5
3	0	1·3 ± 0·1	1·4 ± 0·1
	9	10·9 ± 0·8	7·8 ± 0·4
4	0	2·2 ± 0·1	2·7 ± 0·2
	8	10·3 ± 0·5	8·1 ± 0·4

* Standard error.

These results seem to suggest that neonatal thymus influences the macrophage in enhancing the intracellular growth of micro-organisms such as *L. monocytogenes*. We have also found (unpublished results) prolonged survival of neonatally thymectomized mice after infection by *Rickettsia sennetsu* which is an obligately intracellular parasite within macrophages. Possibly the thymus will affect the growth of *R. sennetsu* within the macrophages in the same way that it affects *L. monocytogenes*.

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Mechanism of Alkylation of Nucleic Acids by Nitrosodimethylamine

SINCE it was discovered[1] that nitrosodimethylamine gives rise to alkylated nucleic acids in the liver when injected into rats, and that an alkylated base, 7-methylguanine, can be isolated from these nucleic acids, there has been much evidence of a correlation between carcinogenicity of *N*-nitrosamines and their transformation *in vivo* into an alkylating agent[2–4]. There was evidence that the initial step in the conversion of the nitrosamine to an alkylating agent was an enzymatic oxidative dealkylation to a hypothetical monoalkylnitrosamine which was then converted to a diazoalkane or, by some other route, to a carbonium ion. No evidence of the production of a diazoalkane in this way has been presented, although this possibility has been widely postulated[5–8].

In our studies of the alkylation of nucleic acids by cyclic *N*-nitrosamines it was found that the very small quantities of alkylated bases isolated from the hydrolysates of nucleic acids of animals treated with the nitrosamines

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