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Transfer by Bone Marrow Cells of Increased Natural Resistance to *Klebsiella pneumoniae* Induced by Lipopolysaccharide in Genetically Deficient C3H/He Mice

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In a previous study we demonstrated that lipopolysaccharide failed to elicit nonspecific resistance in C3H/He lipopolysaccharide low-responder mice against *Klebsiella* infection in contrast to its activity in a closely related histocompatible high-responder subline, C3HeB/Fe. Complete restoration of lipopolysaccharide-induced protection against 10^5 *Klebsiella* was obtained in the present study by transferring bone marrow from high-responder mice to the highly deficient C3H/He mice. The ability of C3H/He mice to clear and destroy bacteria in 5 h was also transferred by C3HeB/Fe marrow cells. In contrast, when high-responder C3HeB/Fe mice were reconstituted with low-responder bone marrow, the clearance and destruction of *K. pneumoniae* were similar to what is observed in the high-responder strain, but survival was only temporary. Collectively, our data show that the failure of C3H/He mice to respond to lipopolysaccharide with nonspecific immunity is due to a defect in two types of bone-marrow-derived cells—radioresistant and radiosensitive.

Gram-negative lipopolysaccharides (LPS) have been shown to increase nonspecific immunity in vivo against several unrelated infections (14, 23). However, we reported previously that in contrast to what is observed in most murine strains, LPS failed to elicit nonspecific resistance to *Klebsiella pneumoniae* infection in the C3H/He low-responder subline (5). The latter mice have proven to be valuable tools for investigation since they are unable to respond with many of the effects that LPS elicits in other murine strains, including the closely related, histocompatible, high-responder strain C3HeB/Fe (2, 5, 8, 11-13, 15, 18, 19, 22, 25-30). In spite of intensive investigation, the biological events that lead to LPS-enhanced resistance remain unclear. The protection that develops 24 h after endotoxin administration may be related to enhanced reticuloendothelial activity (24) which delays bacterial proliferation and allows time for the establishment of the immune response (6, 20).

Recently, we demonstrated that LPS given shortly after lethal irradiation still was capable of prolonging the survival time (but not protection) of mice challenged with *K. pneumoniae* (9). However, when injected 1 month after adult thymectomy, irradiation, and bone marrow reconstitution, LPS insured a definitive protection against this infectious challenge. We therefore suggested that complete enhancement of resistance by LPS did not require T-lymphocytes, but was mediated by an early nonspecific stimulation of radioresistant cells followed by the activation of radiosensitive bone-marrow-derived cells.

The experiments reported here were designed to determine whether the LPS-enhanced resistance to *Klebsiella* could be established in the low-responder C3H/He subline by transfer of high-responder bone marrow. Bone marrow was reciprocally transferred between high- and low-responder thymectomized, lethally irradiated mice, and the effect of this transfer was studied on clearance and survival after challenge by *K. pneumoniae*. Our results show that LPS-induced nonspecific resistance to *K. pneumoniae* was successfully reconstituted in C3H/He thymectomized and lethally irradiated recipients by C3HeB/Fe bone marrow.

MATERIALS AND METHODS

Mice. Five- to six-week-old C3H/He Orl and C3HeB/Fe Orl male mice originating from Jackson Laboratories (Bar Harbor, Maine) were raised and obtained from the Centre National de la Recherche Scientifique (Orléans, France). These C3H/He and C3HeB/Fe mice are histocompatible ($H\text{-}2^k$) as measured by the absence of a mixed lymphocyte reaction and of skin graft rejection between them. Previous experiments have shown that our C3H/He subline retained their unresponsiveness to the mitogenic activity of LPS (5). They will be referred to as He for

C3H/He Orl and eB for C3HeB/Fe Orl.

Preparations. LPS was extracted by the phenol-water procedure, and the trichloroacetic acid extract was prepared by the Boivin procedure (4) from *Salmonella enteritidis* Danysz. NWSM, a water-soluble mitogen extracted from *Nocardia opaca,* was prepared by Ciorbaru et al. (7). Concanavalin A (ConA) (Miles Laboratories) was used as a T-cell mitogen control. These mitogens were used at optimal concentrations established by previous experiments.

Irradiation. Mice confined singly in a Lucite container were exposed under identical conditions to 950 R of ^{60}Co delivered at a rate of ca. 100 rads/min (type Lysa III, Micker International Corp.). A total of 950 R represent a 100% lethal dose for the He and eB strains (all noninfected controls die 10 to 20 days after irradiation).

Tx-BM mice. Five- to six-week-old He and eB mice were anesthetized with Hypnorm (UVA) (Innovar, MacNeill) and thymectomized by the method of Miller (16). One week after surgery, mice were lethally irradiated (950 R), and within 3 h after irradiation they were injected intravenously with 10^7 bone marrow cells flushed from the femur of normal donors. They also received a single injection of 500 U of bipenicillin and 500 μg of dihydrostreptomycin, subcutaneously, the following day. These mice, referred to as Tx-BM, were used ca. 1 month later (10, 17). At postmortem, the mediastinum was checked, and any mice with thymic remnants were discarded from the experiment.

Infectious challenge. Mice were injected intravenously with a virulent strain of *Klebsiella pneumoniae* capsular type 2, biotype d (21). Clearance of viable organisms was measured by plating samples of liver, spleen, and blood 5 h after the challenge by procedures already reported (20). Protection was evaluated by survival time.

Statistical analysis. Student's *t* test was used to analyze the variance between experimental groups in clearance experiments, and the chi-square test was used in survival experiments. Data were considered significant at a level of $P < 0.05$.

Blast transformation. Spleen cells (1.5×10^6) were cultured in triplicate in plastic tubes containing 1 ml of RPMI 1640 (Flow Laboratories) supplemented with 10% decomplemented fetal calf serum (Flow Laboratories), 1% glutamine, and antibiotics. The cells were incubated with mitogens for 2 days, and 1 μCi of [^{3}H]thymidine (1 Ci/mmol, Saclay, France) was added 18 h before harvest. Thymidine incorporation was determined by liquid scintillation counting.

RESULTS

Response of spleen cells from LPS high- and low-responder mice to T and B mitogens 4 days after lethal irradiation. The nature and function of residual spleen lymphocytes still present in lethally irradiated mice were evaluated by their ability to respond to mitogens in vitro. Because 24 h after irradiation the total number of viable nucleated cells represents about 10% of the original number, these experiments were performed 4 days after irradiation with spleen cells pooled from eight mice. Cell suspensions were incubated with three mitogens (LPS, trichloroacetic acid extract and ConA).

The results in Table 1, representing a typical experiment, show that whereas the control high-responder eB subline responded well to all three mitogens, the control He subline responded to the T-mitogen and as previously observed to trichloroacetic acid extract (5) but not to the phenol-water-prepared LPS. Moreover, irradiation effectively destroyed both B- and T-lymphocytes because spleen cell cultures from all such mice were not stimulated by mitogens.

Spleen cell response to T- and B-mitogens after reciprocal transfer of high- and low-responder bone marrow cells into thymectomized lethally irradiated eB and He mice. Similar experiments were performed in thymectomized, lethally irradiated, and bone-marrow-reconstituted (Tx-BM) mice. These assays were performed 1 month after irradiation and bone marrow reconstitution, and, in addition to the previous three mitogens, NWSM, which is also a B-mitogen, was tested. As can be seen in Table 2, which represents a typical experiment: (i) Normal eB controls were strongly stimulated by all mitogens. He controls were selectively unresponsive to LPS obtained by the phenol-water procedure because these same mice were stimulated by the trichloroacetic acid extract and NWSM. (ii) Spleen cells of both high- and low-responder recipients reconstituted with He donor bone marrow cells were not stimulated by LPS but were stimulated by trichloroacetic acid extract and by NWSM. (iii) Spleen cells of He recipients reconstituted with eB donor cells became responsive to the three B-mitogens, including LPS extracted by the phenol-water method. (iv) All thymectomized and irradiated mice did not respond to ConA, showing that they had been essentially depleted of T-lymphocytes.

TABLE 1. *Spleen cell responses of LPS high- and low-responder mice to T- and B-mitogens 4 days after lethal irradiation*

Mouse strain	Treatment	Mitogenic response (cpm)[a]			
		Saline	ConA (3 μmg)	LPS (30 μg)	Trichloroacetic acid extract (30 μg)
eB	Controls	4,220	47,133	15,734	36,771
eB	950 R	186	728	192	242
He	Controls	5,758	46,740	6,521	25,418
He	950 R	45	63	63	62

[a] Arithmetic mean of counts per minute of [^{3}H]-thymidine from triplicate cultures of 1.5×10^6 cells.

TABLE 2. *Spleen cell responses to T- and B-mitogens after reciprocal transfer of high- and low-responder bone marrow cells into thymectomized, lethally irradiated eB and He mice*

Mouse strain	Mitogenic response (cpm)[a]				
	Saline	ConA (1 μg)	LPS (10 μg)	Trichloroacetic acid extract (10 μg)	NWSM (10 μg)
eB[b]	6,100	65,000 (10.7)[c]	21,900 (3.6)	31,200 (5.1)	30,800 (5.1)
eB/eB[d]	6,300	7,500 (1.2)	22,600 (3.6)	49,700 (7.9)	42,100 (6.7)
eB/He	3,800	9,400 (2.4)	3,800 (1.0)	8,200 (2.2)	14,200 (3.7)
He	4,200	61,600 (14.6)	6,900 (1.6)	28,100 (6.7)	44,900 (10.7)
He/He	3,500	4,500 (1.3)	3,500 (1.0)	12,400 (3.5)	15,800 (4.5)
He/eB	5,200	7,900 (1.5)	26,700 (5.1)	33,700 (6.5)	29,700 (5.7)

[a] Arithmetic mean of counts per minute of ^{3}H[thymidine] from triplicate cultures of 1.5×10^6 cells.

[b] Normal non-thymectomized, non-irradiated, non-reconstituted controls.

[c] Numbers in parentheses represent index of stimulation—counts per minute of mitogen-stimulated cells/counts per minute of control cells.

[d] Adult thymectomized, lethally irradiated recipient reconstituted with 10^7 donor bone marrow cells: recipient/donor.

Influence of LPS on clearance of viable *K. pneumoniae* in high- and low-responder mice 4 days after lethal irradiation. In these experiments, mice were sacrificed 5 h after an intravenous injection of 4×10^4 *K. pneumoniae*. Twenty-four hours previously they received by the intravenous route 1 μg of LPS (controls received nonpyrogenic saline only). Several groups were irradiated with 950 R 4 days before the infectious challenge. The total number of viable organisms recovered (Table 3) shows that eB mice were stimulated by LPS pretreatment as was reported previously (5). About 3×10^5 (5.54) microorganisms were recovered in the LPS-treated high-responder as compared to 68×10^5 (6.82) in their nontreated controls. The average numbers of organisms recovered from irradiated eB mice was also of the same order: 133×10^5 (7.12) versus 6×10^5 (5.73) after stimulation by LPS.

As was reported before (5), the total number of organisms found in He nontreated controls was greater, indicating the higher sensitivity of this substrain to the infection. Moreover, He mice were refractory to stimulation by LPS because the number of organisms recovered was of the same order in all groups, and there was no change in their distribution. Thus, it can be concluded that this aspect of LPS refractoriness was not abolished by irradiation.

Influence of LPS on clearance of viable *K. pneumoniae* after reciprocal transfer of high- and low-responder bone marrow cells into thymectomized and lethally irradiated mice. In these experiments mice also were sacrificed 5 h after an intravenous injection of 4×10^4 *K. pneumoniae*, and for certain groups the mice were sacrificed 24 h after an intravenous injection of 1 μg of LPS by the same route. The number of viable organisms in the liver, spleen, and blood was evaluated by plating. The eB and He normal controls presented in Table 4 are the same as those in Table 3 because the data were derived from a global experiment. The total number of organisms recovered in eB/eB untreated or LPS-stimulated mice was of the same order as in their respective normal eB controls (6.57 and 5.07). Moreover, reconstitution of eB recipient with He bone marrow did not abolish their capacity to be stimulated by LPS (5.65 instead of 6.65 in their nontreated controls). However, eB/He mice were not stimulated as much as the eB recipients reconstituted with syngeneic bone marrow ($P < 0.01$). In both control and LPS-treated He/He mice the total number of organisms found were elevated and of the same order as their respective He controls and there was no difference in the distribution of these organisms. In contrast, transfer of eB donor cells modified considerably the response of the He recipient low-responder strain to *K. pneumoniae*; eg., (i) in untreated He/eB mice 31×10^5 (6.43) were recovered versus 111×10^5 (7.04) in He/He mice, and (ii) even smaller numbers were recovered after pretreatment by LPS.

Protection by LPS against *K. pneumoniae* challenge after reciprocal transfer of high- and low-reponder bone marrow cells into thymectomized, lethally irradiated mice. In these experiments four normal and eight Tx-BM groups of mice were challenged intravenously with 10^5 *K. pneumoniae* 1 month after bone marrow reconstitution under the same conditions as previously described. Twenty-four hours before the challenge, half of these groups received, by the same route, 1 μg of LPS, whereas the controls received saline only (these groups will be referred to, respectively, as T_1 to T_6 and C_1 to C_6). Figure 1 represents the cumulative results of two assays done under similar conditions. As can be seen, all six groups of unstimulated normal or Tx-BM controls (C_1 to C_6) died within 24 h after challenge. Notwithstanding

TABLE 3. *Influence of LPS on clearance of viable K. pneumoniae in high- and low-responder mice 4 days after lethal irradiation*

Mouse strain	Treatment	LPS[a]	No. of mice	Distribution of living bacteria[b] in:		
				Blood (%)	Liver and spleen	Total no. of living bacteria (log_{10})
eB	Controls	−	10	76.7 (6.70)[c]	23.3 (6.16)	6.82 ± 0.10[d]
		+	9	46.9 (5.22)	53.1 (5.16)	5.54 ± 0.30
eB	950 R	−	4	83.6 (7.03)	16.4 (6.33)	7.12 ± 0.11
		+	4	52.1 (5.44)	47.9 (5.41)	5.73 ± 0.14
He	Controls	−	7	94.1 (7.31)	5.9 (6.07)	7.33 ± 0.08
		+	7	92.6 (7.21)	7.4 (6.07)	7.24 ± 0.11
He	950 R	−	4	87.2 (7.11)	12.8 (5.92)	7.16 ± 0.29
		+	4	83.8 (7.12)	16.2 (6.23)	7.18 ± 0.10

[a] One microgram of *S. enteritidis* (phenol-Westphal) LPS given intravenously 24 h before challenge.
[b] Log_{10} of the number viable organisms recovered in the blood, liver, and spleen ± standard deviation.
[c] Numbers in parentheses represent the log_{10} of viable organisms recovered in the blood, liver, and spleen.
[d] Log_{10} of total number of viable organisms recovered 5 h after intravenous injection of 4×10^4 *K. pneumoniae* (geometric mean ± standard deviation).

TABLE 4. *Influence of LPS on clearance of viable K. pneumoniae after reciprocal transfer of high- and low-responder bone marrow cells into thymectomized, lethally irradiated mice*

Mouse strain	LPS[a]	No. of mice	Distribution of living bacteria[b] in:		
			Blood (%)	Liver and spleen	Total no. of living bacteria (log_{10})
eB[c]	−	10	76.7 (6.70)[d]	23.3 (6.16)	6.82 ± 0.10[e]
eB	+	9	46.9 (5.22)	53.1 (5.16)	5.54 ± 0.30
He	−	7	94.1 (7.31)	5.9 (6.07)	7.33 ± 0.08
He	+	7	92.6 (7.21)	7.4 (6.07)	7.24 ± 0.11
eB/eB[f]	−	6	77.8 (6.46)	22.2 (5.87)	6.57 ± 0.11
eB/eB	+	8	38.3 (4.62)	61.7 (4.85)	5.07 ± 0.21
eB/He	−	4	86.4 (6.57)	13.6 (5.81)	6.65 ± 0.17
eB/He	+	6	48.9 (5.26)	51.1 (5.34)	5.65 ± 0.33
He/He	−	4	93.8 (7.02)	6.2 (5.81)	7.04 ± 0.06
He/He	+	5	95.7 (7.08)	4.3 (5.79)	7.09 ± 0.15
He/eB	−	7	82.2 (6.31)	17.8 (5.63)	6.43 ± 0.30
He/eB	+	8	60.6 (5.80)	39.4 (5.61)	5.99 ± 0.26

[a] One microgram of *S. enteritidis* (phenol-Westphal) LPS given intravenously 24 h before challenge.
[b] Log_{10} of viable organisms recovered in the blood, liver, and spleen ± standard deviation.
[c] Normal non-thymectomized, non-irradiated, non-reconstituted controls.
[d] Numbers in parentheses represent the log_{10} of viable organisms recovered in the blood, liver, and spleen.
[e] Log_{10} of total number of viable organisms recovered 5 h after intravenous injection of 4×10^4 *K. pneumoniae* (geometric mean ± standard deviation).
[f] Adult thymectomized, lethally irradiated mice reconstituted with 10^7 donor bone marrow cells—recipient/donor.

LPS treatment, two other groups (T_2 He normal mice and T_5 He/He) also died very rapidly. As previously reported, LPS protected very significantly both normal eB mice and Tx-BM reconstituted syngeneic eB/eB mice. In contrast, LPS protected only temporarily for 48 h the T_4 group of eB recipients reconstituted with low-responder donor cells because all these mice were dead within 5 days. Therefore, transfer of He bone marrow cells into the eB recipient did not inhibit the early nonspecific stimulation. More surprisingly, LPS protected Tx-BM He low-responder recipients reconstituted with eB bone marrow cells (T_6) as efficiently as normal eB high-responder (T_1) or the reconstituted syngeneic eB/eB mice (T_3).

DISCUSSION

We have reported previously (9) that although LPS stimulated the phagocytic and bactericidal activity of lethally irradiated mice, the protection against death after challenge with *K. pneumoniae* was only temporary. In contrast, a defi-

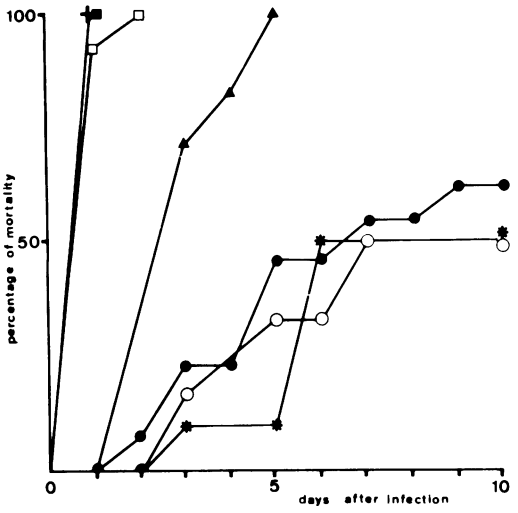

FIG. 1. *Protection by LPS against K. pneumoniae challenge after reciprocal transfer of high- and low-responder bone marrow cells into thymectomized, lethally irradiated mice. Groups of 16 mice were infected with 10^5 K. pneumoniae. Symbols: +, C_1 to C_6, no LPS; ○, T_1 eB; □, T_2 He; ●, T_3 eB/eB; ▲, T_4 eB/He; ■, T_5 He/He; *, T_6 He/eB, all protected by 1 μg of LPS.*

nite increase in survivors was observed in lethally irradiated, thymectomized mice reconstituted with bone marrow. Therefore, the complete enhancement of resistance by LPS (i.e., increase in survivors) appears to be independent of T-cells. We have hypothesized that it is initiated by the nonspecific stimulation of radioresistant cells followed by the activation of radiosensitive, bone marrow-derived cells capable of producing specific antibodies. We have also reported that He mice are not protected against a *Klebsiella* challenge by a previous injection of LPS or other immunostimulants (5).

The experiments reported here were designed to determine whether we could modify the immunological status of the He host by passive transfer of high-responder cells. Our results show that complete restoration of nonspecific immunity in a highly deficient mouse strain was produced by transferring such bone marrow cells. Even though the 50% lethal dose of *K. pneumoniae* in He and eB mice is less than 20 organisms administered intravenously, it was possible to protect eB mice against 10^5 *Klebsiella* with 1 μg of LPS, although 10 μg of LPS still did not protect He mice against 20 organisms. For example, when several experiments carried out over a long period of time were analyzed, no He mice out of 224 were protected by 1 or 10 μg of LPS against a *Klebsiella* challenge. Against this background it was surprising that after thymectomy and lethal irradiation, these genetically deficient mice were protected very effectively by 1 μg of LPS against a challenge with 10^5 *K. pneumoniae* 1 month after transfer of 10^7 high-responder bone marrow cells. Their survival was quite comparable to what was observed with high-responder stimulated controls and eB reconstituted with eB cells. In agreement with these results, the viable count and the distribution of *K. pneumoniae* found in He/eB mice were similar to those observed in the high-responder subline (Table 4).

In contrast, high-responder mice reconstituted with low-responder bone marrow retained their capacity to clear and destroy *K. pneumoniae* during the first hours after challenge, but survival was only temporary. Therefore, He bone marrow transfer does not inhibit nonspecific stimulation, but abolishes the sequential establishment of complete recovery which is observed with eB bone marrow transfer. These results confirm our previous report (9) because they show that the first step of the LPS-induced nonspecific resistance against *Klebsiella* is mediated by radioresistant and long-lived cells, and the second step is mediated by radiosensitive bone marrow-derived cells. In addition, the radioresistant cell or cells are also bone marrow-derived cells because lethally irradiated He mice were refractory to the LPS-induced enhanced bacterial activity, but were completely and definitively protected by LPS after reconstitution by eB bone marrow.

All these data suggest that resistance of C3H/He mice to LPS-induced nonspecific immunity is due to a defect in two types of bone marrow-derived cells—one radioresistant and long-lived, and the other radiosensitive.

What is the nature of cells involved in the He immune defect? The correlation between the decrease in the total number of bacteria and the increase in the percentage of bacteria recovered in the liver-spleen compartment after LPS stimulation of high-responder mice, already observed in previous studies (20), suggests a preponderant role of the reticuloendothelial system. Moreover, it has been shown by use of sulfonamide (20) that the equilibrium between multiplication of bacteria and destruction by the host is not related to a bacteriostatic effect in the blood. Due to its life span and radioresistance, the macrophage seems to be the main candidate for effector in the first step.

Our present findings that complete restoration can be obtained by bone marrow transfer argues in favor of the role of radiosensitive B-lymphocytes, but does not exclude other bone marrow-derived cells; among those cells could be mac-

rophages, polymorphonuclear leukocytes, or the marrow-dependent (M) cells described by Bennett and Baker (3) which function in the host's resistance during the early stages of *Listeria monocytogenes* infection. Watson and Riblet (31) postulated that in C3H/HeJ mice, cells other than lymphocytes may have a structurally altered receptor for LPS. Glode et al. (11) showed that macrophages from C3H/HeJ mice are resistant to the in vitro cytotoxic effect of endotoxin. Likewise, LPS-induced colony stimulating factor, as well as the proliferative response of macrophage/granulocyte progenitors in the spleen, is largely reduced in C3H/HeJ mice (1, 2, 18). In addition, because removal of the thymus in He/He mice did not enable them to destroy and clear organisms more efficiently, the presence in an LPS low-responder of a T-suppressor cell inhibiting the expression of nonspecific immune mechanisms is improbable. The exact nature of the cellular defect in LPS-induced responses in the C3H/He strain of mice is still unknown; however, our data indicate that several types of cells could be incriminated. Irrespective of the nature of the defect, this present report demonstrates that complete restoration of LPS-enhanced resistance to *Klebsiella* infection can be obtained by transferring 10^7 normal bone marrow cells to a genetically highly deficient host. Glode et al. (12) have reported that they were able to partially transfer resistance and sensitivity to the lethal in vivo effects of endotoxin with spleen cells; however, with the dose of 450 R of irradiation it is possible that radiosensitive cells may have recovered in the host, whereas in our experiments it was improbable.

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LITERATURE CITED

1. **Apte, R. N., and D. H. Pluznik.** 1976. Genetic control of lipopolysaccharide induced generation of serum colony stimulating factor and proliferation of splenic granulocyte/macrophage precursor cells. J. Cell. Physiol. **89:** 313-324.
2. **Apte, R. N., and D. H. Pluznik.** 1976. Control mechanisms of endotoxin and particulate material stimulation of hemopoietic colony forming cell differentiation. Exp. Hematol. **4:**10-18.
3. **Bennett, M., and E. E. Baker.** 1977. Marrow-dependent cell function in early stages of infection with *Listeria monocytogenes*. Cell. Immunol. **33:**203-210.
4. **Boivin, A., and L. Mesrobeanu.** 1935. Recherches sur les antigènes somatiques et sur les endotoxines de bactéries. Rev. Immunol. **1:**553-569.
5. **Chedid, L., M. Parant, C. Damais, F. Parant, D. Juy, and A. Galelli.** 1976. Failure of endotoxin to increase nonspecific resistance to infection of lipopolysaccharide low-responder mice. Infect. Immun. **13:**722-727.
6. **Chedid, L., M. Parant, F. Parant, and F. Boyer.** 1968. A proposed mechanism for natural immunity to enterobacterial pathogens. J. Immunol. **100:**292-301.
7. **Ciorbaru, R., A. Adam, J.-F. Petit, E. Lederer, C. Bona, and L. Chedid.** 1975. Isolation of mitogenic and adjuvant active fractions from various species of nocardiae. Infect. Immun. **11:**257-264.
8. **Coutinho, A., E. Gronowicz, and B. M. Sultzer.** 1975. Genetic control of B-cell responses. I. Selective unresponsiveness to lipopolysaccharide. Scand. J. Immunol. **4:**139-143.
9. **Galelli, A., M. Parant, and L. Chedid.** 1977. Role of radiosensitive and radioresistant cells in non-specific resistance to infection of LPS-treated mice. RES J. Reticuloendothel. Soc. **21:**109-118.
10. **Gery, I., J. Krüger, and S. Z. Spiesel.** 1972. Stimulation of B-lymphocytes by endotoxin. J. Immunol. **108:** 1088-1091.
11. **Glode, L. M., A. Jacques, S. E. Mergenhagen, and D. L. Rosenstreich.** 1977. Resistance of macrophages from C3H/HeJ mice to the *in vitro* cytotoxic effects of endotoxin. J. Immunol. **119:**162-166.
12. **Glode, L. M., S. E. Mergenhagen, and D. L. Rosenstreich.** 1976. Significant contribution of spleen cells in mediating the lethal effects of endotoxin in vivo. Infect. Immun. **14:**626-630.
13. **Koenig, S., M. K. Hoffmann, and L. Thomas.** 1977. Induction of phenotypic lymphocyte differentiation in LPS unresponsive mice by an LPS-induced serum factor and by lipid A associated protein. J. Immunol. **118:** 1910-1911.
14. **Landy, M.** 1956. Increase in resistance following administration of bacterial lipopolysaccharide. Ann. N.Y. Acad. Sci. **66:**292-303.
15. **McAdam, K. P. W. J., and J. D. Sipe.** 1976. Murine model for human secondary amyloidosis: genetic variability of the acute phase serum protein SAA response to endotoxins and casein. J. Exp. Med. **144:**1121-1127.
16. **Miller, J. F. A. P.** 1960. Studies on mouse leukemia. The role of the thymus in leukemogenesis by cell-free leukemic filtrates. Br. J. Cancer **14:**93-98.
17. **Miller, J. F. A. P., and G. F. Mitchell.** 1969. Thymus and antigen-reactive cells. Transplant. Rev. **1:**3-42.
18. **Montchilo, R., and J. D. Lutton.** 1977. Decreased *in vivo* and *in vitro* colony stimulating activity responses to bacterial lipopolysaccharide in C3H/HeJ mice. J. Cell. Physiol. **92:**303-308.
19. **Parant, M., F. Parant, and L. Chedid.** 1977. Inheritance of lipopolysaccharide-enhanced nonspecific resistance to infection and of susceptibility to endotoxic shock in lipopolysaccharide low-responder mice. Infect. Immun. **16:**432-438.
20. **Parant, M., F. Parant, L. Chedid, and F. Boyer.** 1967. On the nature of some non-specific host response in endotoxin-induced resistance to infection, p. 275-284. *In* N. R. DiLuzio and R. Paoletti (ed.), The reticuloendothelial system and atherosclerosis. Plenum Publishing Corp., New York.
21. **Richard, C.** 1973. Etude antigénique et biochimique de 500 souches de Klebsiella. Ann. Biol. Clin. **31:**295-303.
22. **Rosenstreich, D. L., L. M. Glode, L. M. Wahl, A. L. Sandberg, and S. E. Mergenhagen.** 1977. Analysis of the cellular defects of endotoxin-unresponsive C3H/HeJ mice, p. 314-320. *In* D. Schlessinger (ed.), Microbiology—1977. American Society for Microbiology, Washington, D.C.
23. **Rowley, D.** 1955. Stimulation of natural immunity to *Escherichia coli* infections. Lancet **i:**232-234.
24. **Rowley, D.** 1962. Phagocytosis. Adv. Immunol. **2:** 241-264.
25. **Skidmore, B., J. Chiller, D. Morrison, and W. Weigle.**

1975. Immunologic properties of bacterial lipopolysaccharide (LPS): correlation between the mitogenic, adjuvant and immunogenetic activities. J. Immunol. **114:** 770–775.

26. **Sultzer, B. M.** 1968. Genetic control of leukocyte responses to endotoxin. Nature (London) **219:**1253–1254.
27. **Sultzer, B. M., and B. S. Nielson.** 1972. PPD tuberculin in a B-cell mitogen. Nature (London) New Biol. **240:** 199–202.
28. **Urbaschek, R., S. E. Mergenhagen, and B. Urbaschek.** 1977. Failure of endotoxin to protect C3H/HeJ mice against lethal X-irradiation. Infect. Immun. **18:** 860–862.
29. **Vas, S. I., R. S. Roy, and H. G. Robson.** 1973. Endotoxin sensitivity of inbred mouse strains. Can. J. Microbiol. **19:**768–769.
30. **Watson, J., and R. Riblet.** 1974. Genetic control of response to bacterial lipopolysaccharide in mice. Evidence for a single gene that influences mitogenic and immunogenic responses to lipopolysaccharides. J. Exp. Med. **140:**1147–1161.
31. **Watson, J., and R. Riblet.** 1975. Genetic control of responses to bacterial lipopolysaccharides in mice. J. Immunol. **114:**1462–1468.