Expression Profile and Function of Triggering Receptor Expressed on Myeloid Cells–1 during Melioidosis

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Background. Triggering receptor expressed on myeloid cells–1 (TREM-1) amplifies Toll-like receptor–initiated responses against pathogens. We aimed to characterize TREM-1 expression and function during sepsis caused by Burkholderia pseudomallei (melioidosis).

Methods. TREM-1 expression was determined on leukocytes and plasma from 34 patients with melioidosis and 32 controls and in mice with experimentally induced melioidosis. Responsiveness toward B. pseudomallei of TREM-1+/H11001 and TREM-1-/H11002 leukocytes was tested in vitro. TREM-1 function was inhibited in mice by a synthetic peptide mimicking the ectodomain of this receptor.

Results. Patients demonstrated increased soluble (s) TREM-1 plasma levels and TREM-1 surface expression on monocytes but not granulocytes. Similarly, mice inoculated with B. pseudomallei displayed a gradual rise in sTREM-1 level and an increase in blood monocyte but not granulocyte TREM-1 expression. At the primary infection site, however, granulocyte TREM-1 expression was enhanced, and the rise in sTREM-1 level occurred earlier. Additionally, purified human TREM-1+ granulocytes showed reduced responsiveness to B. pseudomallei relative to TREM-1+ granulocytes, a difference not detected for TREM-1+ and TREM-1+ monocytes. Treatment with a peptide mimicking a conserved domain of sTREM-1 partially protected mice from B. pseudomallei–induced lethality.

Conclusions. During melioidosis, TREM-1 expression is differentially regulated on granulocytes and monocytes; measurement of TREM-1 expression on blood granulocytes may not provide adequate information on granulocyte TREM-1 expression at the infection site. TREM-1 may be a therapeutic target in melioidosis.

Triggering receptor expressed on myeloid cells–1 (TREM-1) is a new mediator in the inflammatory arena that amplifies Toll-like receptor (TLR)–initiated responses against microbial challenges by potentiating the secretion of proinflammatory cytokines [1]. TREM-1, which signals through the adaptor protein DAP12, is strongly and specifically expressed on monocytes and neutrophils from patients with sepsis [1, 2]. Elevated concentrations of soluble (s) TREM-1 in bronchoalveolar lavage fluid (BALF) can indicate ventilator-associated pneumonia in patients receiving mechanical ventilation [3], and high concentrations in plasma can indicate infection in patients with systemic inflammatory response syndrome [4]. Excitingly, blockade of TREM-1 protects mice against lipopolysaccharide (LPS)–induced shock as well as against sepsis caused by Escherichia coli or cecal ligation and puncture (CLP) [1].

In Southeast Asia and northern Australia, the gram-negative bacillus Burkholderia pseudomallei is an important cause of community-acquired sepsis [5, 6]. More than half of these persons with melioidosis (as this severe infection is named) present with pneumonia, which is frequently associated with bacterial dissemination to
distant sites [6, 7]. In the present study, we aimed to characterize the expression and function of TREM-1 during melioidosis. To do this, we examined TREM-1 expression on peripheral blood leukocytes obtained from patients with severe melioidosis and in different compartments of mice inoculated with a lethal dose of *B. pseudomallei*. The functional role played by TREM-1 during melioidosis was studied in vitro by investigating the responsiveness toward *B. pseudomallei* of TREM-1+ and TREM-1− leukocytes as well as in vivo by treating mice infected with *B. pseudomallei* with a synthetic peptide mimicking a short, highly conserved domain of sTREM-1.

**PATIENTS, MATERIALS, AND METHODS**

**Patients.** Thirty-four patients with melioidosis (mean age, 52 years; age range, 18–86 years; 50% male) were recruited prospectively in 2004 at Sapprasithiprasong Hospital, Ubon Rat Chathani, northeastern Thailand. Sepsis due to melioidosis was defined as culture positivity for *B. pseudomallei* from any clinical sample plus a systemic inflammatory response syndrome (SIRS) [8]. To meet the SIRS criteria, patients had to meet ≥3 of the following 4 criteria: core temperature of ≥38°C or ≤36°C; heart rate of ≥90 beats/min; respiratory rate of ≥20 breaths/min; PaCO2 of ≥32 mm Hg, or use of mechanical ventilation for an acute respiratory process; and white cell count of ≥12 × 10⁹ cells/L or ≤4 × 10⁹ cells/L or differential count showing >10% immature neutrophils [8, 9]. Exclusion criteria were the use of dialysis and/or immunosuppressive therapy, known coagulation disorders, and concomitant infection with HIV. Blood samples were drawn within 36 h after start of appropriate antimicrobial therapy. Thirty-two healthy blood donors (mean age, 41 years; age range, 21–59 years; 71% male) recruited from the Sapprasithiprasong Hospital blood bank served as a control population. The study was approved by both the Ministry of Public Health, Royal Government of Thailand, and the Oxford Tropical Research Ethics Committee, University of Oxford, United Kingdom; written informed consent was obtained from all study subjects.

**Flow cytometry analysis.** All samples were analyzed directly after sample collection by flow cytometry using a FACSCalibur instrument (BD Biosciences). In patients and controls, the cell-surface expression of the molecules of interest was determined on peripheral monocytes and granulocytes by using fluorochrome-conjugated mouse anti-human CD14 (BD Biosciences), TREM-1 (R&D), and CD11b (Bioscience) antibodies in accordance with the manufacturers’ recommendations. Granulocytes were defined according to their scatter pattern and monocytes according to their scatter pattern and CD14 positivity. To correct for nonspecific staining, appropriate isotype control antibodies (BD Biosciences) were used. Data on mean fluorescence intensity (MFI) are presented as the difference between MFI intensities of specifically stained cells and nonspecifically stained cells. Data on the number of positive cells were obtained by setting a quadrant marker for nonspecific staining. In mice, immunostaining for TREM-1 on blood cells, on cells obtained from BALF, and in whole-lung cell suspensions was performed using directly labeled antibodies against GR-1 (GR-1–fluorescein isothiocyanate [FITC]; Pharmingen) and TREM-1 (TREM-1–phycoerythrin [PE; R&D]) and a biotin-labeled antibody against F4/80 (S erotec), in combination with streptavidin allophycocyanine. Antibodies were used at the concentrations recommended by the manufacturer. After staining, cells were fixed in 2% paraformaldehyde. TREM-1 MFI was measured in the Gr-1−high gate (granulocytes) and in the sidescatter-low and F4/80-positive (monocytes) and the sidescatter-high and F4/80-positive (macrophages) gates.

**Evaluation of mRNA levels by quantitative reverse-transcriptase polymerase chain reaction (RT-PCR).** Total RNA was isolated using the RNeasy Mini Kit System (Qiagen), was treated with RQ1 RNase-Free DNase (Promega), and was reverse transcribed using an oligo(dT) primer and Moloney murine leukemia virus RT (Invitrogen Life Technologies), in accordance with the manufacturers’ recommendations. RT-PCRs were performed on cDNA samples that were 4-fold diluted in H₂O using FastStart DNA Master SYBR Green I (Roche), with 2.5 mmol/L MgCl₂ in a LightCycler apparatus (Roche). PCR conditions were as follows: a 5 min at 95°C hot start, followed by 40 cycles of amplification (95°C for 15 s, 60°C for 5 s, and 72°C for 20 s). For quantification, standard curves were constructed by PCR on serial dilutions of concentrated cDNA; data were analyzed using LightCycler software (Roche) as described by the manufacturer. Gene expression is presented as a ratio to the expression of the housekeeping gene β₂-microglobulin [10]. All PCRs generated a single DNA product of the expected length, as evaluated by ethidium bromide–stained 1.2% agarose gel electrophoresis. Primers used for human TREM-1 were 5′-CTGGGTCTCAATTGAGAATTC-3′ and AS717 (5′-CAGGACAGAGACCCGGG-3′). The forward primer for the housekeeping gene was hB2M 5′-TCAATAGGACTGGATGACAGAGAAGACCAGGC-3′, and the reverse primer was hB2M AS231 (5′-GCCCTGTACCTTGATCCCTTG-3′). Oligonucleotides were purchased from Eurogentec.

**Murine melioidosis.** The Animal Care and Use of Committee of the University of Amsterdam approved all experiments. Pathogen-free 8–10-week-old C57BL/6 mice were purchased from Harlan Sprague Dawley. For preparation of the inoculum, *B. pseudomallei* strain 1026b (provided by D. Woods, University of Calgary, Canada [11, 12]) was streaked from frozen aliquots into 50 mL of Luria broth (Difco) and incubated overnight at 37°C in 5% CO₂. Thereafter, a 1-mL portion was transferred to fresh Luria broth and grown for ~5 h to midlogarithmic phase. Bacteria were harvested by centrifugation at 1500 g for 15 min, washed, and resuspended in sterile isotonic saline at a concentration of 5 × 10⁷ or 7.5 × 10⁷ cfu/50 µL, as determined by plating serial 10-fold dilutions on blood agar plates. Pneumonia
was induced by intranasal inoculation of a 50-µL (5 × 10^2 or 7.5 × 10^3 cfu) bacterial suspension. This bacterial dose induces 100% mortality within a 5-day period. Twenty-four, 48, and 72 h after infection, mice were killed by bleeding from the inferior vena cava. BAL was performed as described elsewhere [13]. Pulmonary cell suspensions obtained from infected mice were evaluated by fluorescence-activated cell-sorting (FACS) analysis (BD Biosciences), as described elsewhere [14, 15].

**TREM-1 peptide (LP17).** LP17, a synthetic peptide mimicking a short, highly conserved domain of sTREM-1, was chemically synthesized by Pepscan Systems, as described elsewhere [16]. The peptide with sequence order LQVTDSLRYCIVYHPP was obtained in >99% yield and was endotoxin free. A control peptide was similarly synthesized and contained the same amino acids as LP17 but in a different sequence order (TD-SRCVIGLYHPPQVY). Mice were given 100 µg of LP17 or con-
trol peptide via the intranasal route at different time points, as indicated in Results.

**Assays.** Human sTREM-1 levels were measured with an immunoblot technique, as described elsewhere [3]. Mouse sTREM-1 was measured using a commercially available ELISA (R&D Systems). Human tumor necrosis factor (TNF)–α, interleukin (IL)-1β, IL-6, and IL-8 were measured by cytometric bead array (CBA) multiplex assay (BD Biosciences), in accordance with the manufacturer’s recommendations.

**Cell-sorting experiments.** Leukocytes derived from 8 healthy male volunteers (mean age, 30 years; age range, 28–41 years) were sorted into TREM⁺ and TREM⁻ monocytes and TREM⁺ and TREM⁻ granulocytes by FACS. Briefly, unstimulated whole-blood leukocytes were labeled with TREM-1–PE and CD14–FITC. Subsequently, TREM-1⁺ and TREM-1⁻ monocytes and TREM-1⁺ and TREM-1⁻ granulocytes were separated by use of a FACSARia instrument (BD Biosciences). All sorted subsets were >95% pure. Isolated TREM⁺ and TREM⁻ monocytes and TREM⁺ and TREM⁻ granulocytes (1 × 10⁵) were stimulated with LPS (from *E. coli* 0111:B4; Sigma; 10 ng/mL), heat-killed *B. pseudomallei* (clinical isolate strain 1026b; 1 × 10⁵ cfu/mL) [11, 12], or RPMI 1640 medium for 4 h at 37°C, after which supernatants were collected and stored at −20°C until assays were performed.

**Statistical analysis.** Values are expressed as means ± SE. Differences between groups were analyzed by the Mann-Whitney *U* test or Kruskal-Wallis analysis, with the Dunn post-hoc test where appropriate. Correlations were calculated using the Spearman *ρ* test. These analyses were performed using GraphPad Prism (version 4.00; GraphPad Software). *P* < .05 was considered to indicate statistical significance.

**RESULTS**

**Increased TREM-1 expression in patients with severe melioidosis.** To obtain insight into TREM-1 expression during melioidosis, we first measured sTREM-1 in plasma from 34 patients with culture-proven *B. pseudomallei* infection and 32 healthy controls. Fifteen patients (44%) with melioidosis died in the hospital. sTREM-1 was profoundly elevated in the patients with melioidosis, with mean plasma concentrations that were 8-fold higher than those in healthy subjects (*P* < .0001, for

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**Figure 2.** Increased expression of CD11b on monocytes and granulocytes in patients with melioidosis. CD11b cell-surface expression was strongly increased on monocytes (*A*) and granulocytes (*B*) in patients with melioidosis (*n* = 34), compared with that in healthy controls (*n* = 32). No correlation was seen between triggering receptor expressed on myeloid cells–1 (TREM-1) and CD11b cell-surface expression on monocytes (*C*) or granulocytes (*D*). MFI, mean fluorescence intensity; NS, not significant. ***P* < .001.
the difference between the 2 groups (figure 1A). On admission, patients who went on to die had higher sTREM-1 plasma levels than did patients who survived ($P < .01$) (figure 1B). In 8 patients in whom a second blood sample was obtained at the end of a 2-week treatment period, plasma sTREM-1 concentrations had not decreased relative to levels measured on admission to the hospital (data not shown). TREM-1 mRNA levels were significantly higher in peripheral blood leukocytes from patients than in leukocytes from healthy controls ($P < .001$) (figure 1C).

To determine the impact of melioidosis on TREM-1 protein expression at the surface of peripheral blood cells, we compared TREM-1 expression on circulating monocytes and granulocytes from patients with melioidosis and controls by FACS analysis. Relative to controls, patients displayed higher cell-surface levels of TREM-1 on their monocytes, as reflected by higher MFIs ($P < .0001$) (figure 1E), as well as an overall increase in the percentage of monocytes expressing TREM-1 on their cell membranes ($P < .0001$) (figure 1F). The mean intensity of cell-surface TREM-1 expression on peripheral granulocytes, as determined by MFI, was not different between cases and controls ($P = .17$) (figure 1H); however, the percentage of TREM-1–positive granulocytes was lower in the patients than in the controls ($P < .0001$) (figure 1I). Plasma sTREM-1 levels were positively correlated with granulocyte cell-surface TREM-1 expression ($r = 0.40; P < .05$) but not with monocyte TREM-1 expression ($r = 0.30; P = .07$). The levels of neither cell-associated TREM-1 nor TREM-1 mRNA differed between survivors and nonsurvivors (data not shown).

No correlation between cell-associated TREM-1 expression and CD11b expression in patients with melioidosis. To investigate whether TREM-1 expression is associated with the activation state of leukocytes, we analyzed expression of the activation marker CD11b. CD11b cell-surface expression was strongly increased on monocytes ($P < .0001$) (figure 2A) and granulocytes ($P < .0001$) (figure 2B) from patients with severe melioidosis, compared with those from healthy controls. However, no correlation was seen between TREM-1 and CD11b cell-surface expression on monocytes ($r = 0.23; P = .21$) (figure 2C) or granulocytes ($r = 0.08; P = .66$) (figure 2D).

Compartmentalized TREM-1 expression in a murine model of pneumonia-derived melioidosis. A large proportion of patients with severe melioidosis present with pneumonia with bac-
influx of mainly granulocytes into both the lungs and BALF was seen (figure 3C and 3D). Lung histology showed abscess formation analogous to that found in patients (figure 3E–3G). Plasma sTREM-1 levels remained constant during the first 2 days of infection, followed by a steep increase at 72 h (P < .01) (figure 4A). In BALF, sTREM-1 showed a much earlier rise: at 24 h after infection, BALF sTREM-1 concentrations were already markedly elevated, and these levels remained relatively stable throughout the 72-h period after inoculation (P < .01) (figure 4B). Cell-associated TREM-1 expression in blood, BALF, and whole-lung cell suspensions was evaluated by FACS analysis. Consistent with the data obtained from patients with melioidosis, infected mice showed an up-regulation of TREM-1 expression on blood monocytes (P < .001) (figure 5A). Interestingly, the increase in TREM-1 expression on monocytes occurred only at 72 h after infection, thereby coinciding with the rise in plasma sTREM-1 concentrations. Again consistent with the patient data, granulocyte TREM-1 expression did not increase in the blood of mice with melioidosis (figure 5B). In the pulmonary compartment, cell-associated TREM-1 expression was regulated differently. The most remarkable difference was that granulocytes recovered from BALF and lungs demonstrated an up-regulation of TREM-1 (figure 5F–5J). Macrophages in BALF and lungs, as in blood, increased their TREM-1 expression during the course of the infection. Macrophages in BALF showed a strong constitutive TREM-1 expression, which was reduced after infection with B. pseudomallei (figure 5E), whereas in lungs macrophage TREM-1 expression became up-regulated during melioidosis (figure 5I).

**Diminished proinflammatory response of TREM-1− granulocytes compared with that in TREM-1+ granulocytes.** To investigate the function of the observed TREM-1 up-regulation in patients and mice with melioidosis, we separated TREM-1+ and TREM-1− monocytes and TREM-1+ and TREM-1− granulocytes derived from healthy volunteers by means of a cell sorter (figure 6A). Thereafter, freshly isolated TREM-1+ and TREM-1− cell populations were incubated with medium, LPS, or heat-killed B. pseudomallei. Consistent with the hypothesis that TREM-1 is an amplifier of the inflammatory response, TREM-1− granulocytes released less IL-8 than did TREM-1+ granulocytes after incubation with medium, LPS (P < .05), or B. pseudomallei (P < .01) (figure 6B). The concentrations of other cytokines in the supernatants of purified granulocytes were either below the limit of detection (TNF-α) or very low (IL-1β and IL-6) (data not shown). Purified monocytes released significant quantities of TNF-α, IL-1β, IL-6, and IL-8; however, the supernatants of TREM-1+ and TREM-1− monocytes contained equal concentrations of all cytokines under all incubation conditions (shown for IL-8 in figure 6C).

**Partial protection of mice from B. pseudomallei–induced lethality conferred by TREM-1 peptide LP17.** To further investigate the in vivo role played by TREM-1 in melioidosis, we...
treated mice with a single dose of LP17 a hour before a lethal dose of *B. pseudomallei* was given and found that LP17 pretreatment partially protected mice from *B. pseudomallei*–induced lethality (*P* < .01) (figure 7A). Of interest, the delayed treatment with LP17 at 1, 24, and 72 h after inoculation with *B. pseudomallei* caused a decrease in mortality (*P* < .02) (figure 7A). No difference in organ pathology or cytokine levels could be detected (data not shown). However, consistent with the protective effect, the LP17-treated mice showed less bacterial outgrowth in their spleens, although this could not be detected in the pulmonary compartment (figure 7B and 7C).

**DISCUSSION**

Melioidosis is a debilitating septic disease, with mortality of up to 50% in areas where it is endemic. As with other forms of sepsis, multiple organ failure and death are thought to result from an uncontrolled inflammatory reaction after infection with *B. pseudomallei* [6]. Severe pneumonia with bacterial dissemination to distant body sites is a common presentation of melioidosis [6, 7]. In light of the proinflammatory properties attributed to TREM-1 in pneumonia-derived sepsis, we studied the expression pattern of membrane-bound TREM-1, TREM-1 gene expression, and expression of its soluble form in both humans and mice infected with *B. pseudomallei* in different cell types and in different body compartments. Our results demonstrate that, during severe melioidosis, expression of TREM-1 is differentially regulated on granulocytes and monocytes and in particular that measurements of TREM-1 expression on granulocytes in the circulation may not provide adequate information on granulocyte TREM-1 expression at the site of infection.

Our data extend the findings of previous studies investigating TREM-1 expression in human endotoxemia and sepsis. Our lab-
oratory previously showed that TREM-1 is up-regulated on blood monocytes of human volunteers injected with LPS intravenously, together with an increase in plasma sTREM-1 levels [17]. In these volunteers, LPS induced a down-regulation of TREM-1 on circulating neutrophils, which is consistent with our current observations in patients with melioidosis. Furthermore, plasma sTREM-1 levels were elevated in patients with sepsis, compared with those in patients with SIRS but without infection [4], and monocyte TREM-1 expression, but not granulocyte TREM-1 expression, was higher in patients with septic shock than in healthy controls [2]. Notably, in our patient population with severe melioidosis, high plasma sTREM-1 concentrations on admission were associated with death.

To investigate whether TREM-1 expression correlates with the activation state of leukocytes, we simultaneously analyzed the cell-associated expression of the established activation marker CD11b in patients with melioidosis [18–20]. Although CD11b expression was strongly enhanced on both monocytes and granulocytes in these patients, no association between CD11b and TREM-1 expression could be found, suggesting that TREM-1 expression is not strictly linked to cellular activation. Hence, TREM-1 likely is constitutively expressed on monocytes and granulocytes and may be further up-regulated in the presence of microbes, such as bacteria or fungi [21].

In patients, it was feasible to study TREM-1 expression in cells drawn from peripheral blood only. Because we were interested in the kinetics of TREM-1 expression at the primary infection site, we made use of a mouse model of melioidosis in which mice are intranasally infected with a lethal dose of *Burkholderia pseudomallei*. In view of the fact that pneumonia with bacterial dissemination to distant body sites is a common presentation of human melioidosis [7], we used a model in which *B. pseudomallei* was administered via the airways. We reproduced the major clinical characteristics of melioidosis, with spread of bacteria to distant organs, multiple organ failure, and abscess formation. By doing so, we were able to precisely report the kinetics of membrane-bound TREM-1 expression on monocytes and granulocytes together with its soluble form in both the lungs and blood compartment. Notably, the differential regulation of TREM-1 expression on blood monocytes and granulocytes (with up-regulation on the former cells and down-modulation on the latter cells), as well as the increase in plasma sTREM-1 concentrations, was repro-

Figure 6. Contribution of membrane-bound triggering receptor expressed on myeloid cells–1 (TREM-1) to the cellular responsiveness to *Burkholderia pseudomallei* in vitro in granulocytes but not in monocytes. TREM-1⁺ and TREM-1⁻ monocytes and TREM-1⁺ and TREM-1⁻ granulocytes derived from healthy human volunteers were separated by means of a flow cytometry–based cell sorter (A). Freshly isolated TREM-1⁺ and TREM-1⁻ granulocytes (B) and TREM-1⁺ and TREM-1⁻ monocytes (C) (1 × 10⁶ cells for each) were stimulated with lipopolysaccharide (LPS) or heat-killed *B. pseudomallei* and incubated for 4 h before measurement of interleukin (IL)–8 in the supernatant. Data are mean ± SE values from 8 subjects per group. FITC, fluorescein isothiocyanate; FSC, forward scatter; NS, not significant; PE, phycoerythrin; SSC, side scatter. *P < .05 and **P < .01.
duced in mice with melioidosis. Of considerable interest, however, granulocytes displayed enhanced TREM-1 expression at the site of the infection, particularly in BALF. In mice, monocytes showed a similar up-regulation of TREM-1 in BALF and lungs, as in blood. Macrophages in BALF, however, constitutively displayed strong TREM-1 expression that diminished after infection, whereas in lungs a relatively low initial TREM-1 expression on macrophages became up-regulated during the course of infection. In addition, whereas sTREM-1 rose relatively late in plasma (after 72 h), in BALF, sTREM-1 concentrations were already markedly elevated at 24 h after infection. Moreover, although the absolute concentrations of sTREM-1 were higher in plasma at a late phase of the infection, sTREM-1 levels likely were much higher in BALF, in light of the dilution factor introduced by the lavage procedure. Together, these data point strongly to local release of TREM-1, which is in line with a recent report on TREM-1 expression patterns in a murine model of septic shock induced by CLP, in which increased TREM-1 expression was seen on the phagocytic cells of the peritoneal compartment together with increased sTREM-1 levels in peritoneal lavage fluid [22].

Although several studies have indicated that TREM-1 can amplify cytokine release induced by TLR ligands [1, 21, 23], to our knowledge, the cytokine-production capacity of TREM-1+/H11001 and TREM-1−/H11002 cells has not been directly compared before. Our data reveal a functional difference between the observed TREM-1 up-regulation on monocytes and granulocytes. In accordance with the hypothesis that TREM-1 is a pivotal amplifier of acute inflammation, TREM-1−/H11002 granulocytes displayed a diminished proinflammatory response after LPS and B. pseudomallei stimulation. Interestingly, however, there was no difference in LPS-induced cytokine release between TREM-1+ and TREM-1− monocytes, suggesting the existence of additional amplifiers of the monocyte TLR cascade. Further research is warranted to ad-

Figure 7. Partial protection of mice from *Burkholderia pseudomallei*–induced lethality conferred by LP17. Mice (12 per group) were randomly grouped and treated with normal saline, the control peptide (100 μg intranasally [inl]), LP17 (100 μg inl) at *t* = 1 h before inoculation, or LP17 (100 μg inl) at *t* = 1, 24, and 72 h after inoculation (A). All mice were inoculated with 7.5 × 10^2 cfu of *B. pseudomallei* inl at *t* = 0 h. No effect of the control peptide was seen compared with the saline group. Both pretreatment (*t* = -1 h) and delayed treatment (*t* = 1, 24 and 72 h) with LP17 partially protected mice from *B. pseudomallei*–induced lethality. Treatment with 100 μg of LP17 inl at *t* = -1 (gray bars) or at *t* = 24 h (black bars) with *B. pseudomallei* had no effect on pulmonary bacterial counts, compared with those in the saline-treated control group (white bars), at 48 h after inl infection with *B. pseudomallei* (B); however, LP17 treatment did cause a decrease in bacterial counts in the spleen in this model (C).
dress this issue. The functional role of TREM-1 during melioidosisis was further investigated in vivo by making use of a TREM-1 peptide called LP17. Monocytes and macrophages produce a soluble form of TREM-1, and the administration of LP17, which mimics this soluble receptor, has been shown to reduce inflammatory hyperresponsiveness and mortality during both endotoxic shock in mice and Pseudomonas aeruginosapneumonia in rats [16, 24]. By thus modulating the TREM-1 signaling, we found that treatment with LP17 partially protects mice from B. pseudomallei–induced lethality. This finding further underscores the importance of TREM-1 signaling in melioidosis and highlights TREM-1 as a potential treatment target in melioidosis.

In conclusion, we have demonstrated that human melioidosis is associated with increased expression of TREM-1 in monocytes but not granulocytes, accompanied by elevated circulating levels of sTREM-1. These findings were replicated in a mouse model of melioidosis, which further demonstrated that cell-associated TREM-1 expression at the primary site of infection was different from that in circulating blood cells. These results provide new information on TREM-1 regulation during severe infection and give further support for the potential usefulness of TREM-1 as a diagnostic and therapeutic target.

Acknowledgments

We are grateful to Marieke ten Brink and Jennie Pater for expert technical assistance.

References