Temperature Response in Animals Infected with *Bacillus anthracis*

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Rats, rabbits, swine, guinea pigs, and monkeys were infected with anthrax spores, and their temperature responses were recorded. These were characteristic for a species and appeared independent of resistance or susceptibility of the species toward establishment of the disease. The rabbit appeared unique in that it not only failed to demonstrate a dose-response relationship over an 8-log dose range, but acted independently producing erratic body temperatures depending on spore dose. This limits the usefulness of the rabbit in studying anthrax pathogenesis, and poses questions regarding published data with the rabbit as the test animal.

The temperature response of the mammalian species, including man, to *Bacillus anthracis* is extremely variable (1, 4, 9, 13). With the exception of two controlled studies (11, 12), most data have been collected from field cases for which neither the dose of infecting organism nor the state of infection was known.

It is the purpose of this report to demonstrate that the variability of temperature response is characteristic for a species and may be dose-dependent in certain species under controlled laboratory conditions.

**MATERIALS AND METHODS**

*Animals.* Fischer 344 rats weighing 0.25 kg, New Zealand white rabbits weighing between 1.8 and 2.7 kg, rhesus monkeys (*Macaca mulatta*) weighing between 4 and 6 kg, dwarf swine of the Pitman-Moore variety weighing between 10 and 14 kg, and guinea pigs of the Hartley strain weighing 0.25 kg were used in these studies.

*Challenge materials.* Spores of the V1-b strain of *B. anthracis* were used throughout this study. Dose levels of 10⁸ and 10⁹ were administered intraperitoneally (ip) to both rabbits and guinea pigs. The swine received 10⁹ organisms ip, and the monkeys and the rats received 10⁸ organisms intradermally. In addition, rabbits were inoculated subcutaneously with spore doses of 10⁹, 10⁸, 10⁷, 10⁶, and 10⁵ in an effort to determine a mean time-to-death dose relationship.

*Temperature testing techniques.* All animals were housed in temperature-controlled rooms. The average temperature of the housing area was 73 F (22.8 C) with extremes of only 70 and 75 F (21.1 and 23.9 C). Continuous body temperature recordings were made with intraperitoneal thermocouples wired to a multiple-point potentiometer, except that the body temperature of swine was taken by rectal thermometer. Septicemia was determined by serially diluting the blood and plating on tryptose agar plates.

*Analysis of data.* Temperatures were plotted graphically as a function of time after initiation of infection. Relative units of time are plotted on the X-axes of the graphs; 0 is the challenge time and 100 is the time to death. A ratio to determine the relative units to actual times (hr) was calculated by the following formula: 100/TD = X/assay time (hr). TD is the time of death in hours, and X is the relative unit to be determined. Assay time is the time that postchallenge temperatures were read. Each point represents the mean of four to six animals at indicated observation times during the course of infection. Temperature variability is expressed as the highest and lowest temperatures recorded at that observation period. Except for the rat and monkey, control temperatures were obtained from the *Handbook of Biological Data* (10). The upper and lower limits of the normal temperatures are recorded as horizontal lines in the figures.

**RESULTS**

Temperature responses of the guinea pigs and rabbits to anthrax infections are presented in Fig. 1 and 2. Guinea pigs, recognized as susceptible animals to anthrax infection, appeared to give no response initially to either high or low infecting dose, both of which produced 100% mortality (Fig. 1). Temperatures were normal until the start of septicemia, at which time they became hypothermic during the terminal stages. Two types of temperature responses were demonstrated in the
rabbits, depending on the infecting dose (Fig. 2). The high dose \(10^6\) of organisms produced an elevated temperature within the first 10 hr post-challenge that continued until death. The low dose \(10^6\) of organisms produced an elevated temperature that peaked at approximately 40 hr and then declined slowly until death. Both doses of infecting organisms produced 100% mortality in the rabbits. Also, rabbits were found different from other species (R. E. Lincoln et al., Proc. Soc. Exptl. Biol. Med., in press) in that no correlation between time-to-death and dose could be demonstrated (Fig. 3). Monkeys, which are intermediate in susceptibility to anthrax infection (Lincoln et al., in press), showed no febrile reaction of significance, as confirmed with base line data, and progressed to hypothermia terminally (Fig. 4).

The resistant species \(7a\) (rats and swine) to anthrax infection (Lincoln et al., in press), based on dose required to produce death, showed a different temperature response than did the animals more susceptible to establishment of infection. The rats did not demonstrate either a febrile or a hypothermic response to infection when given a lethal dose (100% mortality) of \(10^9\) spores (Fig. 4). Although we were unsuccessful in producing the septicemic disease in swine, the temperature showed a slight elevation at 108 hr post-challenge with a return to normal within 24 hr (Fig. 5).

**Discussion**

These data confirm reports in the literature by Bowen (2) and by Ferguson and Bohl (3) that a rise in temperature appears characteristic of swine in all acute septicemic field cases. The rat, generally regarded as a resistant animal to establishment of B. anthracis infection (6), showed no temperature response to the infecting anthrax organism. Even terminally, a hypothermia was not present. This was in contrast to the finding of Klein et al. (7), who reported a severe hypothermia in rats challenged with a high unitage of sterile in vitro anthrax toxin. This failure of the rats to respond with any change in body temperature after spore infection was most likely the result of the apparent low unitage of toxin necessary to cause death in this animal species (5).

Our studies further indicate that the temperature response was characteristic for a given animal species and did vary with the challenge dose. Also, it was apparent that some animal species
have little, if any, increase in body temperature during the course of infection, and may either succumb to the infection with a normal temperature or become hypothermic. Moreover, in other species the reverse was true.

Nordberg et al. (11), in their work with rabbits, showed a positive correlation between body temperature and increased number of organisms in the blood. Our data differ with their findings and show a definite temperature relationship with the infecting dose. The lack of dose and time-to-death response in the rabbit was the exception among all species challenged in our laboratory (Lincoln et al., in press). This suggests the limited usefulness of the rabbit in studying anthrax pathogenesis in nonimmunized animals and, although not tested by us, most likely in immunized ones as well. This would indicate that results obtained with rabbits may be difficult to relate to observations on other species. However, the failure of the rabbit to respond as do other species may well have opened an intriguing area of investigation.

The clinical use of temperature to indicate the severity of anthrax infection should be dis-
encouraged because of the variability among species as reported in the literature and in our findings. The fact that species vary widely in their temperature response suggests that it is not a primary response to anthrax infection.

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LITERATURE CITED

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