First and second phases of biphasic fever: two sequential stages of the sickness syndrome?

ANDREJ A. ROMANOVSKY, VLADIMIR A. KULCHITSKY, NIKOLAI V. AKULICH, STANISLAW V. KOULCHITSKY, CHRISTOPHER T. SIMONS, DANIEL I. SESSLER, AND VALERY N. GOURINE

Thermoregulation Laboratory, Legacy Research, Legacy Portland Hospitals, Portland, Oregon 97227; Institute of Physiology, Belarusian Academy of Sciences, Minsk 220725, Belarus; and Thermoregulation Research Laboratory, Department of Anesthesia, University of California at San Francisco, San Francisco, California 94143

Romanovsky, Andrej A., Vladimir A. Kulchitsky, Nikolai V. Akulich, Stanislaw V. Koulichkis, Christopher T. Simons, Daniel I. Sessler, and Valery N. Gourine. First and second phases of biphasic fever: two sequential stages of the sickness syndrome? Am. J. Physiol. 271 (Regulatory Integrative Comp. Physiol. 40): R244–R253, 1996.—We hypothesized that the systemic inflammatory response undergoes two consecutive stages, each characterized by different nonspecific sickness patterns. To test this hypothesis, we studied thermal, nociceptive, and motor responses to lipopolysaccharide (LPS) in 43 unanesthetized, habituated, and lightly restrained male Wistar rats previously implanted with a catheter in the jugular vein. Escherichia coli LPS was injected intravenously in a dose of 0, 0.1, 1, 10, 100, or 1,000 µg/kg. Colonic temperature (Tc) was measured with a thermocouple. Changes in nociception were assessed by tail flick latency (TFL) to a noxious heat stimulus. Motor activity was evaluated using an observation-based activity score (AS). The two lowest doses were aphyrogenic. The next dose induced a monophasic fever with a maximal Tc rise of 0.9 ± 0.2°C at 108 ± 11 min post-LPS. The next two higher doses caused biphasic fevers with the first and second peaks of 0.7 ± 0.1 and 1.4 ± 0.1°C (10 µg/kg) and 0.7 ± 0.1 and 1.4 ± 0.2°C (100 µg/kg) occurring at 60 ± 6 and 165 ± 17 min and at 45 ± 3 and 141 ± 6 min, respectively. The highest dose of LPS resulted in a Tc fall (nadir, +0.6 ± 0.1°C at 83 ± 6 min). Two different sickness patterns were exhibited. The first (high Tc, low TFL, and high AS) occurred during the monophasic fever and the first (early) phase of the biphasic fevers, and it was termed the early phase syndrome. The second pattern (low or high Tc, high TFL, and low AS) developed during the second (late) phase of the biphasic fevers and LPS-hypothermia (endotoxin shock), and it was termed the late phase syndrome. Occurring at different stages of the systemic inflammatory response and developing through different coping patterns (fight/flight (energy expenditure) vs. depression/withdrawal (energy conservation)), the two syndromes represent two different types of adaptation to infection and have different biological significance. Viewing sickness as a dynamic entity is justified clinically. Such a dynamic approach to the problem resolves several contradictions in the current concept of sickness.

thermoregulation; behavior; pain; adaptation; systemic inflammation; rats

AFTER THE FUNDAMENTAL REVIEW by Hart (6), establishing the adaptive value of the sickness-associated behavior, several attempts (e.g., Refs. 10, 42, 43) have been made to characterize nonspecific behavioral and physiologic responses to infectious challenge and to identify their mechanisms. The revealed sickness syndrome is thought to include fever as its central symptom and be primarily mediated by pyrogenic cytokines: interleukins (ILs)-1 and -6, tumor necrosis factors (TNFs), and interleukins (IFNs). Along with fever, several other symptoms (depression, sleepiness, anorexia, hyperalgesia, etc.) are now recognized as signs of sickness. In his review, Hart (6) mentioned that some symptoms of sickness change as a disease progresses from its onset to outcome. This issue, however, has not been further developed. We hypothesize that, as a disease evolves, the strategy of fighting it changes and that, reflecting this change, the sickness syndrome undergoes two distinct stages characterized by different physiological and behavioral responses.

The present study addresses this hypothesis by investigating the biphasic febrile response of rats to intravenous administration of a bacterial lipopolysaccharide (LPS, endotoxin). This model is widely used in research of both fever and stress. The two phases of biphasic fever appear to differ in several respects. The first (early) phase is characterized by precise body temperature (Tb) control (although at a new, elevated level). The precision, however, drastically decreases during the second (late) phase (34, 37, 39) because of the development of threshold dissociation (i.e., separation between the threshold Tb for activation of cold-defense mechanisms and that for triggering heat-defense responses) and possibly a decrease in the effector sensitivity to Tb. The first phase of fever is accompanied by a rise in blood pressure, although normotension (30) or even slight hypotension (unpublished observations) occurs during the second phase. It has been further suggested that the early phase is coupled with wakefulness, whereas the late phase is associated with sleep (28). Furthermore, according to the majority of authors (for references, see 26), the two phases are differently mediated: the first is likely to involve prostaglandins of the E series (PGE), whereas the mediators of the second phase, yet unidentified, are thought to be different from PGE. A different physiological pattern and different biochemical mediation of the early vs. late febrile phase suggest that the two phases are associated with different sickness symptoms. The primary aim of the present study was to reveal the changes in Tb, pain perception, and motor activity during LPS-induced biphasic fever and thus to investigate whether
the early and late phases are characterized by distinct sickness patterns.

Although the most typical thermal response to an intravenous bolus injection of LPS is biphasic fever, lower (just above subpyrogenic) doses of LPS cause monophasic $T_a$ rises, and very high doses induce hypothermia and endotoxin shock (11, 27). The secondary aim of the present study was to investigate whether the sickness patterns revealed during the early and late phases of biphasic fever would also occur in response to different doses of LPS, from subpyrogenic to shock inducing.

METHODS

Animals

Forty-three male Wistar rats weighing ~280 g at the time of surgery were used. The rats were housed in individual cages with food and water available ad libitum. Ambient temperature ($T_a$) was maintained at 21 ± 1°C. All rats were habituated to the experimental conditions and procedures in five 3- to 4-h-long training sessions. During training, the animals were kept in cylindrical plastic restrainers that prevented them from turning around and restricted their back-and-forth movements; the same restrainers were used in subsequent experiments. Each training session also included handling and colonic thermocouple insertion. During both training and experimental sessions, the rat had no access to water or food. Each animal was experimented on only once. After an experiment, the rats were observed for 24 h and then killed with a lethal intraperitoneal injection of pentobarbital sodium.

Surgical Preparation

One-half hour before surgery, each rat received a prophylactic intramuscular injection of gentamicin sulfate (50 mg/kg). The animal was then anesthetized with an intraperitoneal injection of ketamine-xylazine (75 and 5 mg/kg, respectively), and a 1-cm longitudinal incision was made on the ventral surface of the neck, 1 cm to the right of the trachea. The muscles were retracted, and the right jugular vein was exposed and freed from its surrounding connective tissue. A silicone catheter (ID 0.5 mm, OD 0.9 mm) containing heparinized (100 U/ml) pyrogen-free saline (PFS) was passed into the vena cava superior through the jugular vein. The 15-cm-long free end of the catheter was tunneled subcutaneously, exteriorized in the interscapular area, flushed with heparinized PFS, knotted, rolled into a coil, and secured with tape. The wound on the neck was sutured. The following day, the catheter was flushed and its conductance verified. The animal was used in an experiment 2 days after surgery.

Experimental Protocols

To assess thermal, nociceptive, and motor responses to LPS, colonic temperature ($T_a$) was monitored, pain perception was determined in the repeated tail flick (TF) test, and motor activity was evaluated by observing the animal. The tail skin temperature ($T_{sk}$) was also recorded to exclude its potential confounding effect on the results of the TF test (1). Each rat was instrumented with colonic and tail skin thermocouples, placed in its restrainer, and transferred to the experimental chamber. After a 1-h adaptation to the experimental conditions, measurements were started. The parameters under investigation were monitored from 0.5 h before until 5 h after the intravenous injection of LPS or PFS (control). To obviate possible effects of circadian rhythms, all experiments were started around 10:00 A.M. To create optimal conditions for responding to LPS with a fever, $T_a$ in the experimental chamber was maintained at 29–31°C, i.e., at the upper limit or slightly above the rat’s thermoneutrality (verified by tail skin vasodilatation).

Drugs

The Escherichia coli 0111:B4 LPS (List Biological Laboratories, Campbell, CA; lot no. LPS-25E) was used in this study. It was suspended in PFS and stored at 4°C. On the basis of preliminary experiments and our recent study (27), the following doses of LPS were administered (in µg/kg): 0 (PFS only; control), 0.1 (subpyrogenic dose), 1 (monophasic fever-inducing dose), 10 and 100 (biphasic fever-inducing doses), and 1,000 (shock-inducing dose). Injections were made in a volume of 0.1 ml/kg and followed by flushing of the intravenous catheter with 0.1 ml PFS.

Physiological Measurements and Data Analysis

Temperatures. $T_a$, $T_{sk}$, and $T_F$ were measured with copper constantan thermocouples. A colonic thermocouple was inserted 8.5 cm beyond the anus. A skin thermocouple was attached to the ventral surface of the tail, on the border of its proximal and middle thirds. The reference junction of each thermocouple was kept at 0°C. The signal from a thermocouple was fed to an amplifier (model F116/2 or B7-34; KINAP, St. Petersburg, Russia) and sent through an analog-to-digital converter (SETU-10; MIS, Moscow, Russia) to an Apple IIC computer.

TF response. For TF testing, a noxious radiant-heat stimulus was delivered by focusing the beam from a 250-W quartz-halogen light bulb on the dorsal surface of the tail (~75 mm from the base). The heated area was ~4 mm in diameter (as determined in a model experiment by measuring the coagulation zone in egg white after 30-s heating). The TF response was evaluated using its latency (TFL: the time from the onset of the stimulation to tail withdrawal). The stimulus intensity was adjusted so that under normal conditions TFL was ~7 s. This value of TFL corresponded to ~50°C $T_{sk}$ in the focus of the light beam (measured in separate experiments). To prevent burns, the cut-off time was set at 20 s; if the TF did not occur within this time, stimulation was terminated, and the TFL was assigned a value of 20 s.

Motor activity. An animal’s motor activity was evaluated by observing the rat over 5-min periods. For each period, an activity score (AS) was assigned according to the following scale: 0, no movements within the period of observation; 1, sporadic movements; 2, substantial motor activity occupying less than one-half of the observation period; and 3, marked activity during more than one-half of the period; one additional point was added if the movements were accompanied by vocalization. The scored movements included rotation around the longitudinal axis of the body, grooming, rearing, attempts at turning around the sagittal axis of the body, and movements of the head, tail, and paws. Because the spontaneous activity of the habituated and restrained rats was quite low, AS could change only in one direction (i.e., increase). The scoring was done by an observer who was “blind” to the administered dose of LPS. In addition to the scored behaviors, we also noted shivering, chattering, eye closure, relaxed elongation of the body, and hunched posture; these behavioral reactions were not accounted for in AS.

Data processing and analysis. For each recorded variable ($T_a$, $T_{sk}$, TFL, and AS), the null hypothesis was that the responses to all six doses of LPS are the same. For $T_a$, $T_{sk}$, and
AS, 1,000 data points per variable were obtained (40 animals, 25 time points per animal, 6 doses), resulting in 150 means per variable to estimate. For TFL, the data were collected at eight time points, resulting in 48 means to estimate. Statistical evaluations were assessed by repeated measures of a two-way analysis of variance. The recorded variables are presented as their absolute value or deviation ($\Delta$) from the initial (time 0) value.

RESULTS

The dependence of the thermal response to LPS on the dose is shown in Fig. 1. The analysis of variance [$F(125,850) = 28.0; P < 10^{-6}$] easily rejected the hypothesis that the thermal responses to various doses of LPS were all alike. At a dose of 1 $\mu$g/kg, LPS caused a monophasic fever with the peak of 0.9 ± 0.2°C reached at 108 ± 11 min postinjection. Administration of two higher doses resulted in biphasic fevers with the first and second peaks of 0.7 ± 0.1 and 1.4 ± 0.1°C (10 $\mu$g/kg) and 0.7 ± 0.1 and 1.4 ± 0.2°C (100 $\mu$g/kg) occurring at 60 ± 6 and 165 ± 17 min and at 45 ± 3 and 141 ± 6 min, respectively. In response to the highest dose of LPS (1,000 $\mu$g/kg), all animals similarly exhibited the initial $T_c$ fall (nadir, −0.6 ± 0.1°C at 63 ± 6 min) and a slow $T_c$ restoration thereafter. However, the individual responses diverged at ~150 min postinjection: in some animals, $T_c$ started falling again; in others, it was maintained for the next few hours at a relatively constant level; and yet, in still others, $T_c$ continued to gradually increase. This diversification resulted in the mean $\Delta T_c$ showing no dynamics for the last 3 h of the experiment and stabilizing at about −0.3°C. In contrast to lower doses of LPS, administration of the 1,000-$\mu$g/kg dose resulted in some mortality: three out of eleven (27%) animals receiving this dose died ~3 h postinjection (results of these three experiments were excluded from the data). Both the subpyrogenic dose of LPS (0.1 $\mu$g/kg; for the sake of figure clarity, not shown in Fig. 1) and PFS induced no changes in $T_c$.

The thermal responses to LPS followed responses of the tail skin blood vessels (Fig. 2). Each rise of $T_c$ (during both mono- and biphasic fevers) was preceded by a decrease in $T_{sk}$, indicating vasoconstriction, and the fall of $T_c$ at the onset of LPS-induced shock was preceded by a $T_{sk}$ rise, indicating vasodilation. The subpyrogenic dose of LPS (not shown) and PFS induced no changes in $T_c$. The analysis of variance for $T_{sk}$ [$F(125,850) = 10.2; P < 10^{-6}$] allowed the rejection of the null hypothesis.

The dependence of the nociceptive response to LPS on the dose is shown in Fig. 3. The analysis of variance for TFL [$F(40,271) = 8.0; 1$ missing data point; $P < 10^{-6}$] rejected the null hypothesis. Animals that received the subpyrogenic dose of LPS (not demonstrated) or PFS exhibited no changes in their pain perception throughout the experiment and consistently responded to the noxious heat stimulus with a TFL of ~7 s. During the monophasic fever, TFL decreased to 5.3 ± 0.6 s at 72 ± 7 min post LPS. The biphasic fever caused by 10 $\mu$g/kg LPS similarly induced a decrease in TFL (5.0 ± 0.3 s at 48 ± 7 min) during the first febrile phase and an increase (15.2 ± 1.8 s at 246 ± 36 min) during the second phase. As the LPS dose increased to 1,000 $\mu$g/kg, the nociceptive response transformed into a monophasic rise in TFL, with the peak of 18.2 ± 1.1 s occurring at 158 ± 26 min postinjection. Note that, at late stages of their response to LPS, some animals did not respond to the tail

Fig. 1. Changes in colonic temperature ($\Delta T_c$; mean ± SE) in response to the intravenous injection of lipopolysaccharide (LPS; doses indicated). For the sake of figure clarity, the response to the apyrogenic dose of LPS (0.1 $\mu$g/kg) is omitted; for other doses, only 19 of 25 time points are shown. Preinjection value of $T_c$ (mean for the whole study) was 38.4 ± 0.1°C.
heating with the TFL before the cut-off time (i.e., within 20 s): one rat at 210 min after the injection of 10 μg/kg, another at 300 min after the injection of 100 μg/kg, and five others at 90 (2), 210 (2), and 300 min (1) after the injection of 1,000 μg/kg. These responses were assigned a TFL of 20 s.

Motor responses to LPS are shown in Fig. 4. The analysis of variance for AS [F(125,848) = 4.6; 2 missing data points; P < 10⁻⁶] rejected the null hypothesis. Throughout the experiment, those animals that received the subpyrogenic dose of LPS (not shown) or PFS remained quiet, chattered their teeth from time to time, and made occasional movements (AS at 0 level). Animals that received either a pyrogenic or the hypothermizing dose of LPS all responded to the injection with a monophasic increase of motor activity: AS rapidly changed from 0 to 2, stayed elevated for the next 2 h, and subsequently returned to the preinjection level. In all the animals receiving a pyrogenic dose of LPS, the rise in AS (restlessness) corresponded to a Tc,
Fig. 4. Motor responses of rats to the intravenous injection of LPS (doses indicated). Activity score (AS) is shown as mean ± SE. For clarity, the response to 0.1 μg/kg of LPS is omitted. For other doses, only 12 of 25 time points are presented.

rise and was accompanied by shivering, intensive chattering, and often a hunched posture. The near-zero values of AS at late stages of the response to 10, 100, or 1,000 μg/kg were similar to those during the response to PFS or the subpyrogenic dose of LPS, those at late stages of the monophasic fever, and those before the injection of LPS in higher doses. However, at late stages, responses to the three highest doses were characterized by a different behavior. In addition to elongating their bodies (also expressed during monophasic fever, simultaneously with defervescence), the animals closed their eyes, quit chattering, and often rested their heads on the floor of the restrainer.

DISCUSSION

Physiological Responses to LPS

Thermal response. In this study, rats responded to a low, just above subpyrogenic, dose of LPS (1 μg/kg) with a monophasic fever, to two higher doses (10 and 100 μg/kg) with biphasic fevers, and to the highest dose (1,000 μg/kg) with hypothermia. The latter response, associated with a 27% mortality, represented endotoxin shock. Indeed, a rapid fall in the blood pressure has been demonstrated (27) to accompany hypothermia induced even by a smaller dose of LPS (500 μg/kg). The dose dependence of the rats’ thermal response to LPS is in agreement with previous studies (11, 27).

Biphasic fever, the typical response to moderate LPS doses, is the major focus of the present paper. The first phase of biphasic fever is characterized by equal upward shifts in the threshold \( T_b \) for activation of the heat-defense mechanisms and threshold \( T_b \) for induction of the cold-defense responses (38, 39); as a result, \( T_b \) is precisely regulated during the first phase at a new, elevated level. The second phase of biphasic LPS fever is also often characterized by an elevated level of \( T_b \) (such was the case in the present study), but not always. Under “unfavorable” conditions, such as insufficient nutrition (31, 35), physical restraint (17), or low \( T_s \) (35), the second febrile rise in \( T_b \) readily changes to hypothermia. Moreover, as the stress associated with fever becomes more severe (because of either unfavorable conditions or a greater pyrogen dose), the hypothermia occurs earlier and becomes the predominant change in \( T_b \), as seen in endotoxin shock. The mechanism of this hypothermia is the development of threshold dissociation (38, 39). Within a relatively wide range (between the threshold for activation of heat-defense mechanisms and that for activation of cold defense mechanisms), \( T_b \) becomes the result of passive heat exchange between the body and the environment. The threshold dissociation constitutes, therefore, “the same central thermoregulatory process that leads to biphasic fever in some instances [e.g., when \( T_s \) is high] and results in hypothermia in other cases [e.g., when \( T_s \) is low]” (35). The fact that thermoregulatory manifestations of the response to a pyrogen, especially at its late stages, can be represented by either fever or hypothermia is in agreement with the clinical definition of the systemic inflammatory response syndrome: both fever and hypothermia are listed as key symptoms (19).

Nociception. In the present study, administration of LPS decreased TFL at the onset of the monophasic fever and during the early phase of biphasic fevers, but increased TFL during the late phase of biphasic fevers and endotoxin shock. It should be noted that TFL strongly depends on \( T_{sk} \); an increase in \( T_{sk} \) per se results in a decrease in TFL and vice versa (1). Therefore,
evaluation of nociception by TFL could be a problem if the changes in \( T_d \) are not taken into account. The changes in TFL observed in the present study after LPS administration did not correlate with the changes in \( T_{sk} \). Thus, during the monophasic fever (Fig. 5, left), 5°C changes in \( T_{sk} \) were accompanied by only small changes in TFL, whereas during the endotoxin shock (Fig. 5, right), 200% changes in TFL occurred without changes in \( T_{sk} \). During the biphatic fever (Fig. 5, middle), hyperalgesia developed at the lowest (not highest) values of \( T_{sk} \), and hypoalgesia occurred at the highest (not lowest) values of \( T_{sk} \). This shows that the observed changes in TFL were not due to the changes in \( T_{sk} \), but rather developed despite them. We conclude, therefore, that LPS modulates the pain circuitry per se, inducing hyperalgesia at the onset of monophasic fever and during the early phase of biphatic fever, but causing hypoalgesia during the late phase of biphatic fever and endotoxin shock.

In several behavioral studies, LPS (18, 40–42), as well as IL-1\( \alpha \) (5), IL-1\( \beta \) (4, 5, 21, 41), IL-6 (4), and TNF-\( \alpha \) (4), have been shown to facilitate pain, but, in other studies (2, 9, 20), LPS, IL-1\( \alpha \), TNF, and IFN-\( \alpha \) all inhibited nociception. To explain this contradiction, Wiertlak and coauthors (41) have proposed that LPS possesses both hyper- and hypoalgesic activities and that the resultant effect on pain perception depends on their interactions. While the present study was in progress, Yirmiya et al. (43) demonstrated that the hyperalgesia occurring at the beginning of LPS fever later changes to hypoalgesia. Our results support the hypothesis of Wiertlak et al. (41) and concur with the observation by Yirmiya and colleagues (43). Thus the contradiction between the reports of pain facilitation by LPS and several pyrogenic cytokines and the studies showing analgesic activities of the same pyrogens has been resolved. If the nociceptive response is measured shortly after pyrogen administration and/or if relatively low doses of the pyrogen are used, there is a high probability of finding hyperalgesia. If, however, nociception is evaluated at later stages of the response and/or the pyrogen dose is high, the likelihood of observing hypoalgesia increases. Similar to LPS and cytokines, a further mediator of fever, PGE\( _2 \), has been shown to induce hyperalgesia at low doses and hypoalgesia at high doses (8, 22).

**Motor activity.** In the present study, pyrogenic and hypoalgesic activities of LPS induced a rapid increase in AS. This rise is in agreement with the observation by Székely et al. (36) that rats sometimes become restless shortly after a pyrogen injection. Similarly, guinea pigs respond to a pyrogenic dose of LPS with a 1.5-h-long increase of locomotor activity (14), and rabbits respond to a pyrogenic dose of muramyl dipeptide with a 1-h-long reduction of sleep (32). Such facilitation of motor activity has been well recognized as “the first sign of stress” occurring during exposure to various stressors (e.g., extreme heat or cold, toxic concentrations of heavy metals or pesticides) in a wide range of animal species (23). In our study, after the initial period of motor hyperexcitability, the activity returned to scores of 0–1 and stayed at this level until the end of the observation period. Because of the design of the study (low spontaneous motor activity of restrained and habituated rats), the return of AS to low values does not allow, in itself, for distinction between the restoration of motor activity just back to its normal level and its actual depression. However, despite the similarly low AS, animal behavior at late stages of the response to LPS was different from that before the LPS injection. For all pyrogenic doses of LPS, the rats exhibited the extension posture with hindlimbs folded outward and toward the rear (heat-loss behavior) during defervescence. In response to the shock-inducing and two highest pyrogenic doses, other behaviors were also observed: the animals rested their heads on the floor of the restrainer, closed their eyes, and quit chattering. In association with this behavioral pattern, the low AS at late stages of the response to pyrogenic and hypothermic doses of LPS is likely to indicate not simply a restoration of the activity back to the normal state, but rather a profound motor inhibition.

![Fig. 5. Relationship between the TFL (mean ± SE) and \( T_{sk} \) (mean ± SE) of rats during 0–300 min after the intravenous injection of LPS (dose indicated). Direction of time is shown with arrows.](image)
Inhibition of motor activity by LPS (12, 15, 43) and pyrogenic cytokines [e.g., IL-1β (3) and IFN-α (29)] have been repeatedly reported to occur in different animal species. This inhibition together with the related sleepiness, lethargy, and reduction of specific behavioral acts (grooming, rearing, head-pokes into a food tray, etc.) is recognized as a sickness symptom (6, 10). It is worth noting that this motor depression could explain why the initial hyperactivity was not reported in some of the papers cited. Indeed, sustained motor inhibition could have masked the preceding short-lasting hyperactivity, especially if activity was averaged over relatively long time periods (as in the majority of studies) or if the motor activity measurement was initiated several hours after pyrogen administration, when the agitation had already ceased (15, 43). The present results suggest that, whereas motor hyperexcitability is an early component of the systemic response to LPS, motor depression is a late component. The same behavioral sequence occurs in various animal species (insects, fishes, poikilothermic mammals, laboratory rodents) in response to several thermal and nonthermal stressors (23, 24).

**Early and the Late Phase Syndromes**

**Definitions.** Our results show that the early phase of a typically biphasic endotoxin fever is characterized by an elevated level of Tb, hyperalgesia, and motor hyperexcitability. These three symptoms form a relatively stable syndrome, which we term here the early phase syndrome. The early phase syndrome also includes precise Tb regulation (39) and, possibly, arterial hypertension (30) and an increase in vigilance (28, 32).

As discussed above, the response to a pyrogen can be manifested at its late stages with either fever or hypothermia. The present data show that, in addition to fever or hyperthermia, the late phase is accompanied by low motor activity and hypoalgesia. It is also known to be associated with low or normal arterial blood pressure (Ref. 30 and our unpublished data) and seems to be coupled with sleepiness (28). Together with sleepiness, the thermoregulatory manifestations of the second febrile phase (threshold dissociation and resultant increased lability of Tb, increased dependence of Tb on Tb, and increased probability of hypothermia) have been termed the late phase syndrome (25). Here, we redefine this syndrome by extending its list of symptoms to include hypoalgesia, motor depression, and possibly hypotension.

**Phenomenology.** Figure 6 shows sickness patterns of animal responses to various doses of LPS. PFS and the subpyrogenic dose of LPS induced no symptoms of either the early or the late phase syndrome. The low pyrogenic dose of LPS induced a monophasic fever: the early phase syndrome (fever, hyperalgesia, and motor agitation) occurred, but the late phase syndrome did not develop. Higher doses of LPS caused biphasic fever: the early phase syndrome was followed by the late phase syndrome (fever, hypoalgesia, low motor activity). The response to the highest shock-inducing dose was more complex. At the very beginning, the two syndromes overlapped, and the motor hyperactivity (a remnant of the early phase symptom) was accompanied by hypothermia and hypoalgesia (rudiments of the late phase symptoms). Later in the dynamics of the response, hyperactivity disappeared, and the late phase syndrome (hypothermia, hypoalgesia, low motor activity) flourished. Figure 6 clearly demonstrates that, as the dose of LPS increased, both the early and late phase syndromes developed earlier. The first febrile peak occurred at 108 ± 11 min in response to 1 μg/kg, 60 ± 6 min in response to 10 μg/kg, 45 ± 3 min in response to 100 μg/kg, and did not occur at all in response to 1,000 μg/kg. Similarly, in response to LPS doses of 10, 100, and 1,000 μg/kg, the maximal change in Tb associated with the late phase occurred at 165 ± 17, 141 ± 6, and 83 ± 6 min post-LPS, respectively. Threshold dissociation, the major thermoregulatory mechanism of the late febrile phase, has been directly shown to develop already at the very early stages of the response to shock-inducing doses of LPS (27). "Stress hormones," such as corticotropin, also shift the time of the development of threshold dissociation during fever toward its onset (38).

**Biological significance.** We propose that the two febrile phases represent two different sequential stages...
of the sickness syndrome. Occurring at the onset of infection, the early phase syndrome constitutes a response of the healthy organism to the forthcoming disease. Its biological significance is the signaling of the pathogenic challenge (hyperalgesia), recruiting active defense mechanisms (fever), and securing the means (wakefulness, hypertension, generalized motor agitation) for the active search for the optimal environment (conditions for behavioral thermoregulation, sufficient water supply, protection from predators, etc.) for fighting the beginning malady. This type of adaptation to infection develops through the active [fight/flight (energetic) activities (see, e.g., Refs. 19), 35). Moreover, the single thermoregulatory mechanism (threshold dissociation), allowing both a rise and fall of $T_{b}$, has been shown to develop in response to pyrogens in experimental animals (39) and to occur in patients in response to various insults such as trauma and burns (33). Apparently, in some areas of research (e.g., thermoregulation and pain studies), the symptoms of only the early phase

Just as the sickness pattern changed with the increase of the LPS dose in the present study, clinical symptoms of sickness change as the severity of the disease increases. A short-lasting, slight infectious challenge (e.g., common intoxication with bacterial exotoxins via food) is usually accompanied by the early phase syndrome only, thus resembling a monophasic fever. In the course of a more serious but still relatively mild infection (e.g., a typical case of influenza), the initial sickness symptoms that correspond to the early phase syndrome are gradually changed to at least some of the symptoms of the late phase syndrome. As the severity of disease further increases, the late phase symptoms become more and more pronounced, thus resembling biphasic fever. With still a further increase in the severity and increasing threat of energy deficiency, the late phase syndrome becomes predominant and endures for a longer time until, finally, it completely overwhelms the early phase syndrome. The latter situation is characteristic of some clinical cases of septic shock and resembles experimental endotoxin shock. As a general rule, the early phase syndrome develops in a previously healthy organism at the onset of its response to an infection. Conversely, the late phase syndrome occurs when the organism is already exhausted (damaged) by the preceding early phase syndrome, weakened by a preexisting pathology, or exposed to an extremely severe, immediately damaging homeostatic challenge.

Perspectives

There are several inconsistencies in the current concept of sickness. Thus both hyperalgesia (40-43) and sleep (6, 13) have been recognized as sickness symptoms, although their simultaneous occurrence contradicts common sense. Hart (6) points out another contradiction: in animals facing a problem of survival with no apparent solution (such as infant separation from mothers), depression is usually accompanied by hypometabolism and hypothermia, but a similar depression in infection is traditionally thought to be associated with fever. Furthermore, the current concept completely ignores hypothermia as a sickness symptom despite the accumulated body of clinical and experimental evidence. Thus clinicians view both fever and hypothermia as symptoms of the systemic inflammatory response syndrome (19), and basic scientists have repeatedly demonstrated that pyrogens possess both hypertherming (pyrogenic) and hypotherming (cryogenic) activities (see, e.g., Refs. 11, 27, 35). Moreover, the single thermoregulatory mechanism (threshold dissociation), allowing both a rise and fall of $T_{b}$, has been shown to develop in response to pyrogens in experimen-
syndrome (hyperalgesia, precoice control of Tβ fever) have been viewed as symptoms of sickness, whereas, in other areas (behavior and sleep/wakefulness regulation), the research has been focused on the late phase symptoms (sleepiness, motor depression, anorexia). We concur with Hart (6) that, in infection with "microorganisms associated with a deadly disease, taking a pattern of sick behavior is the most extreme and only strategy an animal in the wild has for surviving." We further suggest that this unique strategy undergoes two consecutive stages, each characterized by a distinct complex of sickness symptoms and representing a different type of adaptation to infection. Viewing the sickness syndrome from this dynamic perspective thus explains the contradictions listed above.

The authors thank Dr. L. D. Huner for statistical consulting and Drs. Y. Benikova, L. I. Crawshaw, L. A. Kiesow, R. C. Kramis, and M. Székely for reviewing the first draft of the manuscript. A. A. Romanovsky also thanks Dr. M. Székely for the discussions that brought to life some of the ideas developed in the present paper. The editorial assistance of J. Emerson-Cobb and R. S. Hunter is greatly appreciated.

This research was made possible by an Anesthesia Research Foundation grant-in-aid to A. A. Romanovsky and V. A. Kulchitsky, intramural support by Legacy Portland Hospitals, Portland, OR, and internal funds of the Institute of Physiology, Minsk, Belarus. Gifts of gentamicin sulfate, ketamine-xylazine, and pyrogen-free saline by Dr. C. M. Blatteis are gratefully acknowledged.

Preliminary results of this study are briefly reported elsewhere (13a).

Address for reprint requests: A. A. Romanovsky, Director, Thermo-regulation Laboratory, Legacy Research, Legacy Portland Hospitals, 2901 N. Gaston Ave., Portland, OR 97237.

Received 11 October 1995; accepted in final form 14 February 1996.

REFERENCES


