Letter to the Editor

On Bacterial Tactic Response Times and Latencies

In our recent paper on "Chemotactic responses of *Escherichia coli* to small jumps of photoreleased L-aspartate" measured by computer-assisted motion analysis, we found that the responses were initiated without measurable latency upon photolysis of a photolabile (caged) precursor (Jasuja et al. 1999). Measurement of a latent or lag phase before initiation of a biochemical or physiological process provides important clues regarding the number of steps and their rate in a sequential chain of reactions (see Guttfreund, 1995, for examples). Response latencies documented in earlier studies of bacterial phototaxis and chemotaxis measured different parameters. This letter is intended to provide common ground on which future work can build by clarification of these differences.

Sundberg et al. (1986) first used computer-assisted motion analysis to measure bacterial tactic response times. They found a minimum latency period of 0.70 ± 0.14 s for phototactic responses mediated by sensory rhodopsin I (SRI) and 0.40 ± 0.07 s for those mediated by sensory rhodopsin II (SRII) in *Halobacterium salinarum*, where latency was conventionally defined as the "period after stimulation in which the population reversal frequency is unchanged from its pre-stimulus value." These latencies are due to long-lived intermediates in the photocycles of the sensory rhodopsin signaling complexes (McCain et al., 1987; Hoff et al., 1997). Exponential fits to our data make clear that there is no resolvable latency for *E. coli* chemotactic responses given the nominal temporal resolution (33 ms) of the video technique. This and the stimulus strength dependence of response times imply that signal generation by the *E. coli* aspartate chemoreceptor, Tar, is rapid relative to subsequent signal processing. Thus signal generation by *H. salinarum* SRI is an order of magnitude slower than that by *E. coli* Tar. It will be of interest to determine whether this difference is a consequence of the different tactic stimuli or motile behavior. *H. salinarum* swims three to four times slower than *E. coli* and accomplishes 180° changes in direction during a reversal, as opposed to the less acute reorientation undergone by *E. coli* during a tumble. It may require more time for integration of spatial light gradients before committing to a reversal.

Excitation responses of *E. coli* were first measured by Segall et al. (1982), who reported a 0.23 ± 0.07 s response latency to strong aspartate step stimuli applied by iontophoretic stimulation of tethered bacteria. This result, which we took to be at odds with our data, was due to the different definition of the term. Segall et al. (1982) defined the response latency as the mean time for response, which is approximately equal to the time for the half-population response, i.e., our response time. They did not commit themselves on whether this time was due to "a sequence of one or more reactions that signal the motor with a delay" or whether "the motor, having received the signal is not able to initiate a reversal in a shorter time." It is clear, however, from this and later work (figure 2 of Segall et al., 1986), that motor rotation bias begins to differ from the prestimulus value, even for the first 0.1-s poststimulus interval recorded, for weak as well as strong stimuli. Thus their data and ours largely agree.

A twofold discrepancy remains between *E. coli* chemotactic response times observed when aspartate step stimuli are administered by photorelease as opposed to iontophoresis. Using photorelease, we have documented response times as rapid as 0.07 ± 0.03 s to strong attractant step stimuli (Jasuja et al., 1999; Lux et al., 1999). Similarly, Dowd and Matsumura (1997), using a commercially available nitrobenzyl L-aspartate derivative, measured response times of 0.01 ± 0.03 s. It remains to be seen whether the discrepancy is significant. The outer membrane could be a significant permeability barrier limiting access of iontophoretically released effectors to their target chemoreceptors (Delcour, 1997). Spatial assays have shown that wild-type bacteria are not limited for chemotaxis by outer membrane permeability, but chemotactic sensitivity was affected by alterations in the outer membrane porin composition (Ingham et al., 1990). An important advantage of the photorelease approach is that the caged precursors may be equilibrated between cellular compartments and the medium before photolysis.

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