several studies ([4–6]) show that specifically mobilized circulating macrophages perform this function.

Until such time as the details of Schwann cell-*M. leprae* interaction *in vivo* are elucidated, it might be prudent to apply the term "ingress" of bacilli rather than "phagocytosis."

What do the experts think?

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Drs. Band and Talwar Reply

TO THE EDITOR:

We thank Dr. Pandya for his comments on our work. May we offer some justification for the use of the term "phagocytosis" for the entry of mycobacteria into Schwann cells.

Several workers in the past have studied the behavior of the Schwann cell toward particulate matter of different kinds *in vivo* as well as *in vitro*. These studies have demonstrated that morphologically identifiable Schwann cells *in vivo* are capable of taking up particles such as myelin debris ([1, 7, 9–11, 20–22]), carbon particles ([15, 17]), and mycobacteria ([8]). A similar behavior of Schwann cells *in vitro* toward myelin debris ([23]), latex particles ([2]), and mycobacteria ([2, 13, 14, 16, 18]) has been known for a long time. Moreover, a similar, though more avid, uptake of nonmycobacterial particles such as carbon particles ([12]) and latex particles ([2]), as well as that of mycobacteria ([2, 12, 13]), has been observed with Schwannoma cells *in vitro*. Nearly all of the workers have used the term "phagocytosis" to describe such phenomena. It is difficult to envision that a mechanism other than phagocytosis accounts for the uptake of inert particles, such as that of carbon and latex. It is, thus, clear that Schwann cells are endowed with phagocytic capabilities. We, therefore, used the term "phagocytosis" for the uptake of mycobacteria by Schwann cells in recognition of their phagocytic nature, and to conform with common usage. An added reason for the use of this term was the relative lack of discrimination between different mycobacteria by Schwann cells ([2, 18]) and the inhibition of this interaction by inhibitors of macrophage phagocytosis ([3, 19]). As used in our work, the term was an operational one and did not imply any passive role of the mycobacteria during their entry into Schwann cells.

The use of the term "phagocytosis" should not by itself prevent investigators from defining the role of mycobacteria in the process of their entry into Schwann cells. Although we observed several parallels between the uptake of latex and mycobac-

teria by Schwann cells ([3, 4]), we have also observed marked differences between the two ([6]). In fact, our recent work ([5]) has shown that certain components of the mycobacterial surface may be important in their interaction with the Schwann cell surface. Treatment of mycobacteria with antimycobacterial antibodies inhibited their uptake by Schwann cells *in vitro*. However, in the absence of any direct evidence to suggest that mycobacteria, especially *Mycobacterium leprae,* actively invade Schwann cells, it seems reasonable to continue to use the term "phagocytosis" (which is rather widely used by investigators in the field) to describe their uptake by these cells. Coining new terms like "ingress" to describe the mycobacteria-Schwann cell interaction may only serve to complicate the nomenclature. In addition, this term may indirectly imply an invasive nature of *M. leprae* for Schwann cells which has not been documented so far.

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Microbe Dependence of *Mycobacterium leprae*: A Possible Intracellular Relationship with Protozoa

TO THE EDITOR:

Kato has postulated that *Mycobacterium leprae* may be dependent on concomitant microbes for its growth ([3, 4]). We offer an extension of this hypothesis by postulating that the original host of this mycobacterium may have been an amoeba or other free-living protozoan. The human (and armadillo) macrophages, which have certain features in common with amoebae, may thus be alternative host cells. This possibility was raised by one of us (TR) who was intrigued by the paradox that *Legionella pneumophila* has very fastidious growth requirements yet lives in water. Investigations showed that this bacterium survived and multiplied within amoebae ([7]). Of particular relevance is the fact that *L. pneumophila* replicates within human macrophages, and parallels have been drawn between infection by this pathogen in immunocompromised patients and lepromatous leprosy ([1]). Further, the more fastidious *Legionella micdadei* (Pittsburgh pneumonia agent) can in tissue be as acid-fast as *M. leprae* ([5]), suggesting a behavioral similarity.

We studied the interaction at 30°C between *M. avium* and an endosymbiont-carrying strain of *Acanthamoeba polyphaga* (Ap-1) which has previously been successfully used to isolate, via amoebal enrichment, *L. pneumonphila* serogroup-1, *L. pneumophila* serogroup-3 and a *Legionella*-like amoebal pathogen (LLAP-3) from human cases of pneumonia ([7]). Endosymbionts are bacteria that lie within the amoebal cytoplasm, replicate together with the host cell, and cannot be cultivated *in vitro,* indicating a close mutual dependence ([6]). Intriguingly, some human sera contain antibody to the small, curved, gram-negative, non-acid-fast endosymbiont.

A PYG broth culture of the amoebae was inoculated with a recent clinical isolate of *M. avium* in a bacilli-to-amoeba ratio of 10:1. Half of the mycobacteria were phagocytosed within 1 hr, and virtually all within 18 hr. After 3 weeks, all amoebae contained clumps of strongly staining, intact, acid-fast bacilli. The amoebae were subcultured every 3 weeks to a total of seven subcultures, and were found to replicate at the same rate as uninfected controls. During this time, all amoebae contained the clumps of acid-fast bacilli. Amoebae that became distended with bacilli appeared to be able to liberate these into the medium.

These studies showed that *M. avium* was readily phagocytosed by the amoebae, and appeared to establish a relationship with the host cell. This leads us to postulate that *M. leprae* may likewise exist, or have evolved, in close association with protozoa, and we suggest three possibilities. First, *M. leprae* may replicate within certain species or strains of amoebae, as previously suggested by Jadin ([2]). Second, the intracellular growth of *M. leprae* may be dependent on the pres-