leprae is determined is the mouse footpad model (Baohong, 1987). This was standardized in the 1950s (Shepard, 1960) and is determined by dosing mice with varying amounts of dapsone in their food and observing whether or not strains of *M. leprae* isolated from patients and inoculated into the footpad are inhibited. Strains that are inhibited by concentrations up to 0.1 mg of dapsone per 100 g of food are said to be susceptible, and strains that are inhibited only by higher doses are said to be resistant. Inhibition by concentrations greater than 0.1 mg/100 g up to 1 mg/100 g of food is labeled low-level resistance, inhibition by concentrations greater than 1 mg/100 g up to 10 mg/100 g of food is intermediate resistance, and growth in the presence of 10 mg or more per 100 g of food is considered total resistance. Administration of 10 mg of dapsone per 100 g of food to mice yields a plasma level of about 1 mg/l, similar to that in adult humans given 100 mg of dapsone daily (WHO, 1982).

An *in vitro* method of determining susceptibility using a system of cultivated macrophages has also proven to be able to determine antimicrobial susceptibility by measuring the incorporation of radiolabeled thymidine in the presence of varying concentrations of the drug of interest; however, it has not been widely introduced into routine *M. leprae* testing (Mittal *et al.,* 1983). Synergy of dapsone with the dihydrofolate reductase (DHFR) inhibitors brodimoprim and K-130 has been demonstrated in mice (Dhople *et al.,* 1990; Dhople, 1999).

OTHER MYCOBACTERIA

The susceptibility of other mycobacterial species to dapsone has been investigated by use of a disc elution method confirmed by a broth dilution method using the Bactec 460 radiometric culture system. *Mycobacterium avium, Mycobacterium intracellulare,* and *Mycobacterium kansasii* were found to have MIC_{90} values of 8 μg/ml. *Mycobacterium tuberculosis* and *Mycobacterium fortuitum* exhibited MIC_{90} values of > 32 μg/ml (Gonzalez *et al.,* 1989). When dapsone is combined with the triazole DHFR inhibitor WR99210, *in vitro* activity against *M. avium* complex organisms is enhanced (Shah *et al.,* 1996). In contrast, dapsone had no activity against *M. avium* in a mouse model of disseminated infection, nor did it add to the effect of clarithromycin. It had only a slight prophylactic effect when used alone, but when used as prophylaxis in combination with clarithromycin, demonstrated no additive advantage (Bermudez *et al.,* 1994).

Mycobacterium ulcerans has been found to be susceptible to dapsone *in vitro* (Pattyn and van Ermengem, 1968); more recent work also confirmed this and demonstrated synergy with the DHFR inhibitor epiroprim (Dhople, 2001).

PLASMODIUM SPECIES

Plasmodium falciparum is, in general, only moderately susceptible to dapsone. The other *Plasmodium* species are inherently less susceptible, and a high rate of clinical failures with high doses of dapsone was reported in the earliest trials. However, the addition of a DHFR inhibitor such as pyrimethamine or cycloproguanil markedly potentiates its action against *P. falciparum,* but less so with the other *Plasmodium* species (Nzila, 2006).

PNEUMOCYSTIS JIROVECI

Dapsone was originally found to be effective in the mouse model of *P. carinii (jiroveci)* infection (Hughes and Smith, 1984). This was confirmed *in vitro* in human lung–derived tissue culture cell lines that showed that growth of rat-derived *P. jiroveci* was > 75% inhibited by dapsone at 0.1, 1.0, and 10 μg/ml (Cushion *et al.,* 1985). Synergy with pyrimethamine and macrolides was demonstrated in a similar *in vitro* cell line system. In this comparison of various combinations, dapsone plus pyrimethamine and trimethoprim combined with sulfamethoxazole were found to be the equally strongest combinations, achieving maximal effect at concentrations readily achievable in plasma (Cirioni *et al.,* 1997).

TOXOPLASMA GONDII

T. gondii DHPS was shown to be highly inhibited by dapsone *in vitro* (Allegra *et al.,* 1990). Dapsone is also active against *T. gondii* in cell culture, but less effective than pyrimethamine. It was not effective in immunosuppressed mice when used alone, but in combination with pyrimethamine it was highly effective (Derouin *et al.,* 1991). In a murine model of disseminated toxoplasmosis, dapsone alone administered at 100 or 200 mg/kg/day protected about 80% of mice from death (Araujo and Remington, 1992). In an immunosuppressed rat model of dual infection with *P. jiroveci* and *T. gondii,* dapsone combined with roxithromycin was effective in preventing infection with both organisms (Brun-Pascaud *et al.,* 1998). Epiroprim in combination with dapsone in a mouse model of acute *T. gondii* infection was found to be more effective than either drug alone and similar in efficacy to the combination of sulfadiazine and pyrimethamine (Chang *et al.,* 1994).

CRYPTOSPORIDIUM PARVUM

Dapsone alone or in combination with macrolides has minimal activity against *C. parvum in vitro* (Giacometti *et al.,* 1996).

2b. Emerging resistance and cross-resistance

MYCOBACTERIUM LEPRAE

Dapsone resistance was suspected clinically soon after widespread use of the drug began in the 1950s for leprosy and was subsequently confirmed in 1964 when *in vivo* testing of patient isolates in the mouse footpad system became routine (Ji, 1985). The prevalence and incidence of secondary resistance to dapsone in a leprosarium in Malaysia were demonstrated to have risen to 7.5% and 0.8%, respectively, through the 1960s (Pearson *et al.,* 1975). By the mid-1970s, dapsone resistance rates of up to 40% were being reported in some areas (WHO, 1977). Dapsone resistance is divided into low