# COMMENTARIES

# Biowaiver Monographs for Immediate-Release Solid Oral Dosage Forms: Primaquine Phosphate

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**ABSTRACT:** Literature data relevant to the decision to allow a waiver of *in vivo* bioequivalence (BE) testing for the approval of immediate-release (IR) solid oral dosage forms containing the antimalarial drug primaquine phosphate as the only active pharmaceutical ingredient (API) are reviewed. On the basis of permeability data and solubility studies, primaquine phosphate was found to be "highly soluble" and "highly permeable" API, thus conforming to Class I of the Biopharmaceutical Classification System (BCS). It has a wide therapeutic index. BCS-conform dissolution studies showed the products to be rapidly dissolving. No data pertaining to BE or bioinequivalence of IR primaquine phosphate products could be located in open literature. On the basis of the available data, a biowaiver-procedure-based approval can be recommended for IR solid oral dosage forms of primaquine phosphate, provided the generic product contains excipients present in products already approved by the International Conference on Harmonisation or associated countries in similar amounts and the test and reference products meet the dissolution criteria for "rapidly dissolving" (>85% drug release in 30 min in standard media at pH 1.2, 4.5, and 6.8; similarity factor ($f_2$) > 50) or "very rapidly dissolving" products (>85% drug release in 15 min in standard media at pH 1.2, 4.5, and 6.8). © 2011 Wiley Periodicals, Inc. and the American Pharmacists Association J Pharm Sci 101:936–945, 2012
**Keywords:** absorption; bioavailability; bioequivalence; Biopharmaceutics Classification System (BCS); permeability; solubility; primaquine phosphate

## INTRODUCTION

The possibility of waiving the need to perform pharmacokinetic bioequivalence (BE) studies for approval of primaquine phosphate oral products is reviewed in this biowaiver monograph. The decision is discussed on the basis of biopharmaceutical and clinical properties of primaquine phosphate obtained from the open scientific literature and additional experimental data. The risks of assessing BE based on surrogate *in vitro* techniques (so called biowaiver procedure) rather than *in vivo* study data for the approval of new immediate-release (IR) solid oral dosage forms, including reformulated and multisource drug products, of primaquine phosphate are then evaluated. Conclusions pertain to drug products containing primaquine phosphate as the only active pharmaceutical ingredient (API) and not to fixed-dose combination (FDC) products.

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The purpose and scope of the biowaiver monograph series have been discussed previously.[1] The prerequisites for biowaiving and approach to the risk–benefit analysis for a given API are based on guidances from the World Health Organization (WHO),[2] US Food and Drug Administration (FDA),[3] and European Medicines Agency (EMA).[4] The risk–benefit analysis includes a consideration of the probability of an incorrect biowaiver decision as well as the consequences of an incorrect decision in terms of public health and individual patient risks. Biowaiver monographs have already been published for various APIs.[1,5–25] These monographs are also available online on the International Pharmaceutical Federation's website (www.fip.org/bcs_monographs).

## GENERAL CHARACTERISTICS

### Name

BANM, rINNM name: Primaquine phosphate[26]

Molecular Formula: $C_{15}H_{21}N_3O,2H_3PO_4$

Chemical Name: 1,4-Pentanediamine,$N^4$-(6-methoxy-8-quinolinyl)-,($\pm$)-,phosphate (1:2)[26];

($\pm$)-8-[(4-Amino-1-methylbutyl)amino]-6-methoxyquinoline phosphate (1:2)[27];

$N$ [4](6-methoxy-8-quinolinyl)-1,4-pentanediamine phosphate (1:2)[28];

(4RS)-$N^4$-(6-methoxyquinolin-8-yl)pentane-1,4-diamine bisphosphate[29]

The structure of primaquine phosphate is shown in Figure 1.

Primaquine phosphate is an orange-red crystalline powder with a molecular weight of 455.3. Its melting point is reported to be 197°C–198°C.[30]

### Therapeutic Indications

Primaquine phosphate, an 8-aminoquinoline antimalarial, is an effective tissue schizonticide against intrahepatic forms of all types of malarial parasite.[26] In the treatment of malaria, primaquine phosphate is prescribed for prophylaxis, presumptive anti-relapse therapy (terminal prophylaxis) against *Plasmodium vivax* and *Plasmodium ovale*, and radical cure after *P. vivax* and *P. ovale* infections.[26,31] In the Model List of Essential Medicines (EML) of the WHO, primaquine phosphate is listed as an antimalarial for the radical cure of malaria in case of *P. vivax* or *P. ovale* infection.[32]

A combination of oral (p.o.) primaquine phosphate along with clindamycin, either intravenous (i.v.) or p.o. depending on the patient's condition, is reported to be effective in treating mild to moderate cases of *Pnemocystis carinii* pneumonia in patients with AIDS and those unresponsive to conventional antipneumocystis agents.[33–34]

### Dosage

Doses of primaquine phosphate are expressed in terms of its free base. Fifteen milligrams of primaquine is equivalent to 26.4 mg of primaquine phosphate. The dose stated in this manuscript refers to the corresponding amount of primaquine base, unless otherwise specified. The maximum recommended therapeutic dose by the FDA is 0.75 mg/kg/day.[35]

#### Prophylaxis

The Center for Disease Control and Prevention (CDC) recommends a prophylactic dose of 30 mg daily, beginning 1 day before exposure and continuing a week after departure from the malaria-infested area. For adults weighing less than 60 kg and children, the CDC recommends a dose of 0.5 mg/kg/day (up to a maximum daily dose of 30 mg).[36] Similarly, for preventing *P. vivax* and *Plasmodium falciparum* infections in adults, the WHO recommends a daily prophylactic dose of 30 mg, taken during exposure and for 1 day after departure from a malarious area.[37]

#### Radical Treatment of P. Vivax and P. Ovale Malaria

Primaquine phosphate is prescribed in conjunction with an effective blood-stage schizonticide for symptomatic patients with *P. vivax* and *P. ovale* in the radical treatment of malaria.

The WHO recommends an adult dose of 0.25 mg/kg or 15 mg/day for 14 days following standard chloroquine therapy or, if glucose-6-phosphate dehydrogenase (G6PD) deficiency is known or suspected, 0.75 mg/kg weekly for 8 weeks. In children over 1 year, 0.25 mg/kg daily for 14 days after standard chloroquine therapy is recommended.[38]

However, several studies performed on patients in Southeast Asia and Western Pacific countries found the standard dose of 15 mg/day to be ineffective against certain strains of *P. vivax*.[39–40] On the basis of these findings, the WHO suggests a higher dose of 0.5 mg/kg for 14 days for anti-relapse treatment of *P. vivax* malaria with primaquine phosphate for Southeast Asia and Western Pacific countries (where the Chesson strain of *P. vivax* occurs). In areas north of the equator, treatment with 0.25 mg/kg for 14 days was found to be effective.[37]
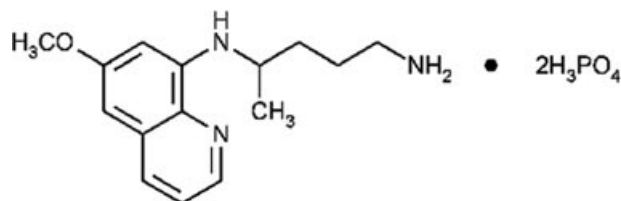


**Figure 1.** Structure of primaquine phosphate.

### Gametocytocidal Therapy

A single dose of 0.5–0.75 mg/kg in adults and children is recommended by the WHO.[38]

### Toxicity and Therapeutic Index

Adverse effects with therapeutic doses of primaquine phosphate are usually minimal, but abdominal pain, epigastric distress, nausea, and vomiting are common if taken on an empty stomach.[41] Clayman et al.[42] noted that the risk of gastrointestinal (GI) upset at any dose essentially disappeared when the drug was administered with food. Higher doses of 60–240 mg/day can lead to moderate to severe abdominal distress with nausea and vomiting, but such doses are not usually administered in clinical situations.

Acute intravascular hemolysis in G6PD-deficient patients is the most serious adverse effect linked to primaquine phosphate. The severity of hemolytic anemia is related to the dose and the variant of the G6PD enzyme. The main subtypes of G6PD-deficient individuals include the African variant A⁻, the Mediterranean variant B⁻, and Asian variants.[31,41] In the A⁻ variant, primaquine phosphate produces mild hemolytic anemia. In G6PD-deficient Asians and B⁻ variant individuals, the reaction is more severe and potentially life-threatening hemolytic episodes may result. Because of high risk of hemolysis in G6PD-deficient individuals, testing of the patient's G6PD status is necessary before prescribing primaquine phosphate for malarial therapy.[31] Similarly, the inability to routinely assess the G6PD status of a fetus *in utero* makes pregnancy a contraindication for primaquine phosphate therapy.[43]

Primaquine-induced methemoglobinemia, although common in clinical doses, is mild, self-limited, and tolerated without signs or symptoms of cyanosis in otherwise healthy individuals.[31] Methemoglobin concentrations of about 25% are well tolerated, but in severe cases appropriate treatment of cyanosis or withdrawal of primaquine must be instituted to reverse the symptoms within 24–72 h.[41,43]

Neuropsychiatric changes seem to be rare. Only one single case of depression and psychosis has been reported after primaquine phosphate use.[44]

High doses (120–240 mg/day) have been associated with neutropenia.[42] However, more recent clinical trials using prolonged dosing that included monitoring of complete blood counts showed no such evidence of toxicity at clinical doses.[45,46]

Primaquine phosphate does not qualify as a narrow therapeutic index (NTI) drug according to the FDA's definition for NTIs.[47] It is reported to have a therapeutic index of 10.[31] A p.o. median lethal dose (LD$_{50}$) of 177 mg/kg was reported in rats.[48]

## PHYSICOCHEMICAL PROPERTIES

### Salt, Stereoisomers, and Polymorphs

Primaquine is commercially available as a diphosphate salt. The oxalate salt, although listed in the literature, has not been reported in commercially available formulations.[49] The European Pharmacopoeia and International Pharmacopoeia list primaquine as primaquine diphosphate, whereas in United States Pharmacopoeia (USP), it is officially listed as primaquine phosphate.[27–29] However, this name implies the diphosphate salt form of primaquine.

Primaquine phosphate possesses a chiral carbon in the diamine side chain and hence shows stereoisomerism. It exists as both R(+) and S(−) enantiomers, but only preparations containing the racemic mixture are commercially available. No polymorphism has been reported for primaquine phosphate.

### Solubility

According to the pharmacopoeial standards, primaquine phosphate is described to be "soluble" in water.[27–29] A solubility of 66.67 mg/mL in water and an intrinsic solubility (−log $S_o$) of 2.77 ± 0.03 at 25°C have been reported.[27,30,49–51] The pH of a 10 mg/mL solution was found to be acidic with a pH range of 2.5–3.5.[28]

Further solubility data of primaquine phosphate were generated by performing solubility studies of the API using the standard shake-flask method in water and compendial buffers of pH 1.0, 1.2, 4.5, 6.8, and 7.5 at 37°C. The experimental studies were carried out according to the Biopharmaceutical Classification System (BCS) guidelines for the determination of API solubility.[2] Around 30 mg of primaquine phosphate was added to 3 mL of the buffer medium. Table 1 shows the concentration of the primaquine phosphate in each buffer along with the corresponding dose–solubility (D/S) ratio. The API showed high solubility

**Table 1.** Concentration and Dose Solubility (D/S) Ratio of Primaquine Phosphate in Buffers at 37°C, Based on 26.3 mg Primaquine Phosphate (Equivalent to 15 mg* Primaquine Base) as the Highest Recommended Dose Strength

| Medium | pH | Concentration (mg/mL) | D/S ratio (mL) |
|---|---|---|---|
| Water | 7.1 | 8.8117 | <2.98 |
| SGF$_{sp}$ | 1.0 | 9.3744 | <2.81 |
| SGF$_{sp}$ | 1.2 | 8.3414 | <3.15 |
| Acetate buffer | 4.5 | 9.8859 | <2.66 |
| SIF$_{sp}$ | 6.8 | 10.8634 | <2.42 |
| SIF$_{sp}$ | 7.5 | 11.5413 | <2.28 |

*Maximum strength of primaquine tablets listed in WHO EML and the maximum single dose recommended in the Summary of Product Characteristics of primaquine phosphate tablets USP (Sanofi-Synthelabo, New York, USA).

SGF$_{sp}$, simulated gastric fluid without pepsin; SIF$_{sp}$, simulated intestinal fluid without pancreatin.

at all five pH values tested, thus meeting the criterion for "high solubility."

## Partition Coefficient

Log $p$ (octanol–water) values of 3.2,[30] 2.72,[51] and 2.1[52] have been reported in literature for primaquine. The differences in the log $p$ values observed are likely due to the different methods utilized for the determination. Additionally, the temperature at which the results were obtained was not specified in these reports. Calculations using a ClogP program (version 3.0; Biobyte Corporation, Claremon, California) for Clog$P$ determination and fragmentation methods, based on atomic contributions to lipophilicity, for log $p$ calculation resulted in values of 2.6 and 1.47, respectively.[50]

## p$K$a

Primaquine is a dibasic compound, having p$K$a values of 3.2 and 10.4.[53–54] Bergstrom et al.[51] obtained p$K$a values of 9.99 and 3.74 when determined by a cosolvent method.[51] The temperatures at which the tests were performed were not reported in any of the above-mentioned papers.

## Dosage Form Strengths

The 16th edition of the WHO EML lists primaquine as tablet formulations containing diphosphate salt equivalent to 7.5 and 15 mg of free base.[32] Single API tablets having a marketing authorization (MA) in United States (US), Australia (AU), and India (IN) are available in strengths of 7.5 and 15 mg.[55–57]

## PHARMACOKINETIC PROPERTIES

### Absorption and Bioavailability

Primaquine is readily and completely absorbed from the GI tract after p.o. administration. Peak plasma concentrations are achieved within 2–3 h of administration.[26,58] Absolute bioavailability (BA) determined in five volunteers by simultaneous administration of i.v. [$^{14}$C]–primaquine (7.5 µCi, 1.55 mCi/mmol) and a standard 45 mg p.o. dose showed the extent of drug absorption to be virtually complete with a mean reported absolute BA of $96 \pm 8\%$.[58] In the same study, the area under the curve (AUC) increased linearly when doses of 15, 30, and 45 mg were administered, suggesting first-order kinetics and transcellular drug transport.[31,59] The mean time to reach maximum plasma concentration ($T_{max}$) was $3 \pm 1$ h in healthy volunteers.[58–61]

Pharmacokinetic studies of primaquine phosphate in humans produced a maximum plasma concentration ($C_{max}$) range of 50.7–65 ng/mL for a 15 mg single p.o. dose. The $AUC_{0-\infty}$ for the same dose ranged from 0.5 to 0.547 µg h/mL.[59,62–64] No significant difference was found in plasma primaquine concentration profiles of G6PD-normal and G6PD-deficient Thai individuals.[61,64]

## Permeability

An apparent permeability ($P_{app}$) of $177 \pm 40 \times 10^{-6}$ cm/s using the Caco-2 cell monolayer system was reported in comparison with metoprolol and mannitol, the FDA approved high- and low-permeability markers, which had $P_{app}$ values of $133 \times 10^{-6}$ cm/s and $15 \times 10^{-6}$ cm/s, respectively, in the same set of studies (personal correspondence with author).[51]

## Distribution

Primaquine is extensively distributed into the tissues and has a mean apparent volume of distribution ($V_d$) ranging from 200 to 300 L.[58,59,62] It is concentrated in liver, lungs, heart, and skeletal muscles.[65] In contrast to other aminoquinolines, neither primaquine nor its carboxy metabolite are found to accumulate in blood cells, as studies reporting the ratio of drug in whole blood to plasma found values close to unity.[59,62] Studies with $\alpha_1$-acid glycoprotein (AGP) established that primaquine binds to physiological amounts of glycoproteins in plasma and this can alter the amount of free drug in the plasma.[31,66]

## Metabolism and Excretion

The drug showed high intersubject variability in its disposition in several studies.[60–63,67] The average plasma clearance values of primaquine range between 27 and 37 L/h[58,59,61–64,68,69] and its plasma half-life lies between 3 and 8 h.[26] Primaquine is predominantly cleared by nonrenal elimination, with only about 1%–4% unchanged drug being excreted in the urine over 24 h.[58,6570,71] Elimination of primaquine is mainly by the liver. On the basis of *in vitro* human microsomal studies, CYP1A2 along with CYP2D6 were identified as the main cytochrome P450 isoforms catalyzing the metabolism of primaquine.[72] Primaquine primarily undergoes rapid side-chain metabolism to carboxyprimaquine, the principal plasma metabolite.[65] The carboxylic acid metabolite has a longer half-life in the body than the parent drug and is ultimately subjected to further biotransformation prior to excretion.[58,70] Peak levels in plasma are reached within 3–12 h after dose, and a $C_{max}$ 10-fold higher than that of the parent drug are attained.[58,65] The extent of formation of the metabolite is unaffected by the route of administration. Studies revealed that primaquine is not subject to an extensive first-pass effect.[59]

### Effect of Ethnic Background, Degree of Infection and Gender on Primaquine Pharmacokinetics

Ethnic differences (Caucasians, Thais, and Indians) did not affect primaquine pharmacokinetics in healthy or infected individuals significantly, even

though primaquine showed a wide range of interindividual variation in plasma concentrations within each study group.[60–63,67] However, administration of 15 mg/day dose for 14 days in 30 Korean male *P. vivax* infected patients showed much higher mean $C_{max}$ ($282 \pm 177$ ng/mL) and $AUC_{0-\infty}$ [$1970 \pm 1360$ (ng h)/mL] as compared with other studies.[73]

High mean plasma concentrations of 276.1 ng/mL were also reported after the first dose in acutely ill malarial patients (infected by *P. vivax*) treated with the same standard regimen.[74] A study by Edwards et al.[75] demonstrated that in Thai individuals, the oral clearance of primaquine decreased significantly during infection with *P. falciparum*. The authors attributed the high plasma concentrations to the binding of the drug to AGP, the levels of which are elevated during infection with malaria.[75] However, Bhatia et al.[62] found no such deviations in the disposition following 15 mg/day p.o. dose for 14 days, when studying in seven Indian patients with *P. vivax* malaria.[62] In any case, even though the effect of ethnic background and degree of infection on the pharmacokinetics of primaquine cannot be completely ruled out, the high drug concentration levels observed in several pharmacokinetic studies in patients did not affect the tolerance or safety of the drug.

Cuong et al.[76] found the mean plasma concentration curves between healthy male and female Vietnamese volunteers (10 each) in the fasting state to be similar after administration of 30 mg single p.o. dose. Although not statistically significant, the mean clearance of primaquine in the study tended to be higher in males than females.[76] Another study in the Vietnamese volunteers with a higher degree of power (17 volunteers of each gender) showed that women had a significantly higher $C_{max}$ and $AUC_{0-\infty}$ values for the same dose as compared with their male counterparts after multiple dosing.[69] A similar pattern was observed in a small study performed by Singhasivanon et al.[60] on Thai individuals (four individuals each).[60] By contrast, a study performed on Australian individuals revealed no significant difference in primaquine disposition between male and female volunteers at the same dose.[68] Even though it is difficult to pinpoint what causes gender differences in plasma concentrations, it is clear that these differences are seen only in steady-state levels and not after a single dose. Interestingly, in studies including the Thai and Vietnamese volunteers, women reported lower incidences of drug-associated GI disturbances than men.

## DOSAGE FORM PERFORMANCE

### Dissolution and BE

The USP specifies $Q$ (average amount of dissolved active ingredient) limits of not less than 80% within 60 min in 900 mL of 0.01 N hydrochloric acid at a paddle speed of 50 rpm for dissolution test of primaquine phosphate tablets.[27] For biowaiver approval, BCS-conform dissolution tests need to be performed, and literature research returned no relevant data on this subject. To close this gap, dissolution tests were carried out on pure drug and two commercially available primaquine phosphate tablets USP (Sanofi-Synthelabo, New York, USA) ) and Malirid® (Ipca Laboratories, Mumbai, India) India) at pH 1.2, 4.5, and 6.8 buffer according to the biowaiver guidelines. It was observed that more than 85% of the drug was dissolved in each of the three media from all the tested samples within 15 min. (Fig. 2). The two commercial products passed the WHO, FDA, and EMA criteria for "very rapidly dissolving" products. Thus, with respect to the dissolution studies, these products would qualify for a biowaiver as per WHO, FDA, and EMA guidelines. These very rapid dissolution profiles are consistent with the very small quantities of media needed to dissolve the labeled dose (Table 1).

No BE studies of Malirid® (Ipca Laboratories) with the US comparator product have been published. Considering that primaquine phosphate is a very old API, it is unclear whether the MA for this product was granted based on either *in vivo* BE studies or available toxicity data. Additionally, literature research returned no studies in which BE of primaquine phosphate FDC products has been investigated.

### Effect of Food and Excipients

Cuong et al.[76] found that presence of food (containing ~28 g fat) did not delay the rate of absorption of primaquine, but it significantly increased $C_{max}$ by 26% [127–160 ng/mL, 95% confidence interval (CI) 12–40, $p < 0.001$] and $AUC_{0-\infty}$ by 14% [1222–1396 ng.h/mL, 95% CI 3–27, $p = 0.013$]. However, this rise in concentrations was deemed unlikely to cause any increase in the incidence or severity of adverse events.[76]

There are several IR formulations of primaquine phosphate having a MA in US, IN, and AU. Two products, namely, primaquine phosphate tablets USP (Sanofi-Synthelabo) and Malirid® (Ipca Laboratories), having MA in US and IN, respectively, were tested for dissolution characteristics. The Sanofi-Synthelabo product is a reference listed drug in the FDA Orange book for approved drug products with therapeutic equivalence evaluations. As the dissolution profiles of Malirid® (Ipca Laboratories) did not vary significantly from either the US innovator product or the pure drug, one can conclude that the excipients used had little or no effect on the dissolution characteristics of these products (Fig. 1).

**Table 2.** Excipients* Present in Primaquine Diphosphate IR Solid Oral Drug Products with a Marketing Authorization (MA) in Canada (CA) and the United States (US)**, and the Minimal and Maximal Amount of That Excipient Present Pro Dosage Unit in Solid Oral Drug Products with an MA in the US***

| Excipient | Drug Products Containing Excipient with an MA Granted by the Named Country | Range Present in Solid Oral Dosage Forms $^a$ with an MA in the US (mg) |
|---|---|---|
| Carnauba wax | US (1,2) | 0.1–58 |
| Cellulose, microcrystalline | CA (3), US (1,2) | 4.6–1385 |
| Hypromellose | CA (3), US (1,2) | 0.8–537 |
| Lactose | CA (3), US (1,2) | 23–1020 |
| Macrogols | CA (3), US (1,2) | 0.12–961 |
| Magnesium stearate | CA (3), US (1,2) | 0.15–401 |
| Polysorbates | CA (3), US (1,2) | 0.4–418 |
| Starch, pregelatinised | CA (3), US (1,2) | 5.0–600 |
| Talc | CA (3), US (1,2) | 0.10–220 |

*Colorants are not included.
**Sources of data: CA, www.hc-sc.gc.ca (accessed July 21, 2011); US, www.dailymed.nlm.nih.gov. (accessed July 21, 2011).
***US: FDA's Inactive Ingredient Database, http://www.fda.gov/Drugs/InformationOnDrugs/ucm113978.htm (version date July 19, 2011).
$^a$ Refers to all FDA approved products and not just those containing primaquine.
(1), Primaquine phosphate tablet (PD-Rx Pharmaceuticals, Inc., Oklahoma City, Oklahoma); (2) Primaquine phosphate tablet, film coated (Sanofi-Aventis U.S. LLC, Bridgewater, New Jersey); (3) $^{Pr}$Primaquine®, primaquine phosphate tablets USP (Sanofi-Synthelabo). All products contain 26.3 mg primaquine diphosphate corresponding to 15 mg primaquine base
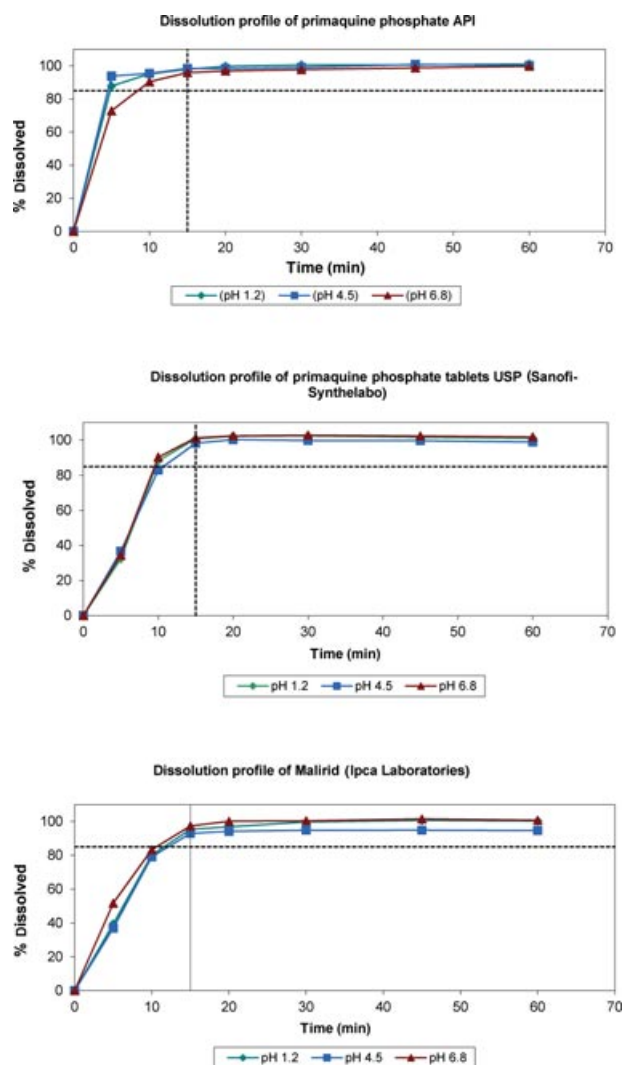


**Figure 2.** Dissolution profiles of primaquine phosphate products and the API in pH 1.2, 4.5, and 6.8 as per BCS guidelines.

## DISCUSSION

### Solubility

pH-dependent solubility data, required to establish the solubility class as per BCS guidelines, were not available in the open literature. Additional tests performed under consideration of the maximum available dosage strength (26.3 mg of primaquine phosphate) of the API revealed that the drug is "highly soluble" over the pH range of 1–7.5. According to the WHO definition of high solubility, the $D/S$ ratio for the API should be 250 mL or less over the pH range 1.2–6.8 at 37°C. The solubility studies on primaquine demonstrated a $D/S$ ratio far below this limit (Table 1). For $D/S$ ratio calculations, the EMA defines dose as the highest single dose of the API administered as IR formulation(s).[4] On the basis of the EMA definition, the API would still undoubtedly pass the $D/S$ criteria even if a higher dose of 45 mg, the dose administered at times in some Western Pacific countries for radical cure of malaria, is considered. Thus, primaquine phosphate qualifies as a highly soluble drug according to the WHO, FDA, and EMA standards.

### Permeability

On the basis of the BCS guidelines by WHO and EMA, an API is "highly permeable" when it is absorbed to an extent of 85% or more. On the contrary, FDA specifies a more stringent criterion of at least 90% absorption.[2–4]

Primaquine is completely absorbed from the GI tract.[26] Mihaly et al.[59] reported an absolute BA well over 90% in humans, indicating the API to be "highly permeable."[59] The *in vivo* data are supported by the Caco-2 study results, in which primaquine phosphate showed higher $P_{app}$ values in comparison with the FDA-approved highly permeable reference marker,

metoprolol.[51] Thus, primaquine phosphate fits WHO, EMA, and FDA criteria for "highly permeable" API.

### BCS Classification

On the basis of the permeability data and solubility studies, primaquine phosphate, a "highly soluble" and "highly permeable" API, conforms to BCS class I according to all current guidances. Several literature articles are available that agree with this classification.[2,50–51,77–78]

Pharmacokinetic study reveals that primaquine is subjected to extensive metabolism in the body, with only 1%–4% of the intact drug being excreted with the urine. Consequently, according to the Biopharmaceutics Drug Disposition Classification System, primaquine phosphate would also be categorized as a class I API.[78]

### Risk of Bioinequivalence with Respect to Excipient and/or Manufacturing Variations

No studies addressing the effect of excipients on BA, interaction of primaquine phosphate with excipients, or bioinequivalence of primaquine products could be found in the open literature. Dissolution tests performed on the two commercially available tablet formulations showed that they were "very rapidly dissolving" products. The dissolution profiles of these products were comparable to that of the pure API. Therefore, it can be inferred that the excipients used in these products did not affect the release of the API from the products.

Along similar lines, excipient or manufacturing process that could affect the release of the drug from the formulations would be evident during the surrogate BE testing, that is, BCS-conform dissolution tests. Although comparative dissolution testing is incapable of elucidating the effect of excipient and manufacturing variables on absorption of primaquine, the high permeability of primaquine indicates that the risk of excipients affecting permeability to an extent that would impact BE is low. By using excipients listed in Table 2, the probability of an excipient effect on primaquine permeability is further reduced.

### Patient's Risk Associated with Bioinequivalence

Primaquine phosphate, a drug with a wide therapeutic index, is the only antimalarial recommended for the radical cure of *P. vivax* and *P. ovale* infection. A bioinequivalent product could, on the one hand, produce subtherapeutic concentrations, which could lead to inadequate and ineffective therapy. On the other hand, it could produce supratherapeutic concentrations, which could precipitate adverse effects in patients.

Several studies have substantiated that high doses of primaquine phosphate are well tolerated and showed good safety in nonpregnant G6PD-normal patients.[31,42,43,45] Baird et al.[46] found that a 30 mg adult primaquine phosphate dose was well tolerated and safe in a randomized parallel placebo controlled trial, as no serious adverse events occurred and no volunteer was removed from the study because of intolerance or evidence of toxicity. The authors also reported that the volunteers taking primaquine phosphate registered complaints of adverse events less often than the volunteers in a parallel placebo group.[46] Similarly, in a trial conducted on Thai patients to study high-dose primaquine phosphate regimens against relapse of *P. vivax* malaria, the group ($n = 66$) that was administered the dose of 30 mg twice a day (60 mg per day) over 7 days tolerated the drug without serious adverse events.[79] Thus, these reports further substantiate that high-dose primaquine phosphate regimens are safe and well tolerated in G6DP-normal patients and that the risk of toxic effects at supratherapeutic concentrations due to bioinequivalence is low.

Although patients with low G6DP levels are likely to have higher concentrations of primaquine, this is not a parameter that will be affected by formulations differences, that is, it is a BA issue and not a BE issue.

In summary, the risk of a bioinequivalent product being formulated, passing all the BE acceptance criteria on the basis of *in vitro* data and yet giving rise to subtherapeutic or supratherapeutic plasma levels seems low and is further reduced by restricting excipient choices to those listed in Table 2. Furthermore, the consequent risk to the patient should such a situation arise lies mostly on the side of suboptimal therapy rather than an acute, life-threatening toxic effect.

### CONCLUSION

A biowaiver-procedure-based approval for IR solid oral dosage forms of primaquine phosphate can be justified, provided the new multisource or reformulated product contains excipients in amounts equivalent to those present in the products approved by the International Conference on Harmonisation or associated countries. Additionally, both test and comparator products need to comply with the BCS-conform dissolution test criteria for "rapidly dissolving" (>85% drug release in 30 min in standard media at pH 1.2, 4.5, and 6.8, and the dissolution profiles should be compared by a similarity factor ($f_2$) > 50 or equivalent statistical criterion) or "very rapidly dissolving" (>85% drug release in 15 min in standard media at pH 1.2, 4.5, and 6.8) products; in the absence of which, BE should be investigated using an *in vivo* pharmacokinetic study.

### ACKNOWLEDGMENTS

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