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Long-term preservation of intellectual functioning in sapropterin-treated infants and young children with phenylketonuria: A seven-year analysis

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Abstract

Sapropterin dihydrochloride has been approved for the treatment of hyperphenylalaninemia in infants and young children with phenylketonuria (PKU). Sapropterin can reduce phenylalanine (Phe) levels in tetrahydrobiopterin (BH4)-responsive patients, potentially preventing the intellectual impairment caused by elevated Phe levels. The long-term effect of sapropterin on intellectual functioning was assessed using the Full-Scale Intelligence Quotient (FSIQ) in 62 children who began treatment before the age of 6 years. Over each 2-year interval, the estimate

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Declaration of Competing Interest

BKB has received personal fees from BioMarin outside the submitted work. AF has received payments from BioMarin during the conduct of the study. SEM and NL have received grants and personal fees from BioMarin during the conduct of the study. AS-V is a principal investigator for several BioMarin clinical trials, has received payment from serving in advisory boards funded by BioMarin, and participated in a speakers bureau for BioMarin. SW has received consulting fees from BioMarin during the conduct of the study, and from Homology, Pfizer and Synlogic outside the submitted work. JL, KBW, and RR are employees of BioMarin Pharmaceutical inc. LLK has nothing to declare.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ymgme.2021.01.001.

of mean change in FSIQ was —0.5768 with a lower limit of the 95% confidence interval (CI) of —1.60. At the end of the follow-up period (Year 7), the least squares mean estimate of the change in FSIQ from baseline was 1.14 with a lower limit of the 95% CI of —3.53. These lower limits were both within the clinically expected variation of 5 points. During the whole study period, mean blood Phe levels remained within the American College of Medical Genetics (ACMG) target range of 120–360 μmol/L. In addition, height, weight, and head circumference were maintained within normal ranges throughout follow-up, as defined by growth charts from the World Health Organization (WHO) and Centers for Disease Control and Prevention (CDC) for children below and above the age of 24 months, respectively. All patients (n = 65) enrolled in this study experienced at least one adverse event, as expected from previous studies. In conclusion, long-term use of sapropterin in individuals with PKU helps to control blood Phe, preserve intellectual functioning, and maintain normal growth in BH4-responsive children who initiated treatment between the ages of 0 to 6 years.

Keywords

Phenylketonuria; Sapropterin; Intellectual functioning

1. Introduction

Phenylketonuria (PKU) is a rare autosomal recessive disorder caused by mutation(s) in the gene coding for the hepatic enzyme phenylalanine hydroxylase (PAH). PAH converts the essential amino acid phenylalanine (Phe) to tyrosine, a process that requires the enzyme cofactor tetrahydrobiopterin (BH4) [1]. Untreated PKU is characterized by elevated blood and brain Phe levels resulting in detrimental effects on brain development and function. Current treatment guidelines recommend lifelong blood Phe control with a goal range of 120–360 μmol/L [2–5].

Among children with PKU, it is well established that poor control of blood Phe during the first 12 years of age leads to a decrease in intelligence quotient (IQ), with the strongest inverse association between IQ and blood Phe levels appearing in children under 10 years of age [6]. Based on this knowledge, treatment guidelines consistently recommend initiating PKU management immediately after diagnosis, which is usually soon after birth, since PKU is part of all newborn screening programs in developed countries [4,5]. The mainstay of PKU treatment is a Phe-restricted diet supplemented with medical food. Early dietary treatment can prevent most severe long-term neurological and cognitive complications of PKU and can maintain intellectual functioning within the normal range [6,7]. However, dietary management is challenging and poor adherence to dietary restrictions results in blood Phe levels above the guideline-recommended threshold in approximately 12–28% of children with PKU under the age of 4 years [6,8]. Furthermore, care should be taken to prevent nutritional deficiencies related to dietary management, which may result in suboptimal growth outcomes [9].

Sapropterin dihydrochloride (KUVAN®, BioMarin Pharmaceutical Inc., Novato, CA, USA) is an oral synthetic formulation of the 6R-isomer of BH4 marketed in over 58 countries

(Data on file. BioMarin Pharmaceutical Inc., Novato, CA). Both the Food and Drug Administration (FDA) and the European Medicines Agency (EMA) have approved the use of sapropterin for the treatment of hyperphenylalaninemia (HPA) in BH4-responsive PKU patients of all ages, including patients below the age of 4 years [10,11].

The current study (PKU-015, NCT00838435) evaluated the long-term safety of sapropterin and its effect on the maintenance of blood Phe levels, intellectual functioning, and growth in children with PKU aged 0–6 years at treatment initiation. The aim was to complement existing evidence in older individuals with PKU from completed sapropterin clinical studies, including double-blind placebo-controlled trials that established the safety and efficacy of sapropterin [12–15]. Additionally, this study supplements the evidence in children <4 years of age at enrollment from the European Safety Paediatric efficAcy phaRmacokinetic with Kuvan® (SPARK, NCT01376908) study [16]. The SPARK study consisted of an initial, 26-week long, open-label, randomized phase IIIb trial, which demonstrated that sapropterin in conjunction with diet was well-tolerated and significantly increased Phe tolerance in children <4 years of age with BH4-responsive PKU [16]. All patients from the initial stage of the SPARK study entered in a 3-year extension study in which all patients received sapropterin [17]. The extension study confirmed that the increase in dietary Phe tolerance was maintained in the long-term [17]. A previous interim analysis of the study presented here was the first to demonstrate that also intellectual functioning and growth were maintained over 2 years of follow-up in children who began treatment with sapropterin below the age of 6 years [18]. Herein, we report the final 7-year outcomes of this study with long-term intellectual functioning as the primary endpoint.

2. Materials and methods

2.1. Study design

The PKU-015 study was a multicenter, open-label phase IIIb study consisting of two parts, which have been previously described [18]. In summary, during Part 1 (4 weeks), responsiveness to oral sapropterin (20 mg/kg/day) was assessed. At the end of Part 1, patients who had a ≥30% average reduction in blood Phe and an IQ ≥ 80 continued to Part 2 for long-term safety and efficacy evaluation over 7 years. Exemptions were granted to eight patients whose mean blood Phe reduction was <30% during the first four weeks (Fig. 1). Patients were instructed to maintain a stable diet throughout the study. However, dietary Phe could be increased per investigator discretion if Phe levels dropped below 120 μmol/L (the upper limit of normal physiological blood Phe levels) in both Part 1 and 2 of the study.

2.2. Patients

Eligibility criteria of the study have been previously described [18]. In summary, children aged 0–6 years with an established diagnosis of PKU with HPA were included if they had documented blood Phe control prior to study enrollment, if applicable (e.g. the patient was old enough for these data to be collected), and able to adhere to a Phe-restricted diet to maintain blood Phe within the recommended ranges. Patients were excluded if they were taking methotrexate or other medications that inhibit folate metabolism, or if they used phosphodiesterase type 5 inhibitors. Patients were divided into four age groups depending

on the age of sapropterin initiation: <1 year, 1 to <2 years, 2 to <4 years, and 4 to <7 years. Informed consent was obtained from the parent or guardian and the study was conducted in accordance with the Declaration of Helsinki.

2.3. Drug administration

A 20 mg/kg/day dose of oral sapropterin tablets was administered at home, at approximately the same time each day with a meal to increase absorption. Tablets were allowed to be dissolved in 4 to 8 oz. (120 to 240 mL) of water or apple juice or crushed and stirred into soft foods, such as applesauce or lemon pudding. A dosing table for patients weighing 10 kg and less was provided. In Part 2, a dose reduction was permitted after week 5 if the patient did not tolerate 20 mg/kg/day, for any reason.

2.4. Assessments

The primary objective of the study was to evaluate the long-term efficacy of sapropterin in preserving intellectual functioning in children with PKU when treatment is initiated between 0 and 6 years of age. Intellectual functioning of patients was assessed every 6 to 24 months (depending on the type of assessment) by age-appropriate scales, i.e. the Bayley-III tool, the Wechsler Preschool and Primary Scale of Intelligence, Third Edition (WPPSI-III), and the Wechsler Intelligence Scale for Children, Fourth Edition (WISC-IV). The Bayley-III tool was used to assess the development in infants and toddlers within the age range of 0 to <30 months, while the WPPSI-III and WISC-IV were used to assess the intelligence of children aged ≥30 months to <7 years and >7 years, respectively [19–23]. The results from the WPPSI-III and the WISC-IV tests are considered equivalent because studies have shown a 0.89 corrected correlation between scores from the two tests, which is within the reliability of each test [22]. Full-Scale Intelligence Quotient (FSIQ) score was measured by the WPPSI-III or WISC-IV tests. All investigators conducting the intellectual testing provided evidence of appropriate education and credentials prior to selection for participation in the trial. Subsequently, these investigators received training regarding the study protocols and test materials. All assessments were re-scored to verify the results, ensure rater reliability, and prevent bias.

The secondary objectives were to evaluate the effect of sapropterin on anthropometric parameters over time and the long-term safety of the drug in the study population. Anthropometric measurements, including height, weight, and head circumference (up to 4 years of age), were assessed at each study visit. In addition, plasma blood Phe and prescribed dietary Phe intake were monitored monthly.

Safety assessments included the patient's medical history and recording of adverse events (AEs), vital signs, physical examinations, clinical laboratory tests (chemistry, hematology, urinalysis, Phe, tyrosine, tryptophan), and electrocardiogram (for at least the first 80 patients). AEs were coded in accordance with version 15.1 of MedDRA.

2.5. Statistical analysis

Descriptive summaries of continuous variables included number of observations, mean, standard deviation, and standard error. Categorical endpoints were summarized by frequency

and percent. All tests were conducted at an alpha level of 0.05 and all confidence intervals (CIs) were given at a 2-sided 95% level, unless stated otherwise. All analyses were performed using statistical analysis system (SAS®) Version 9.4 (SAS Institute, Cary, NC).

The primary endpoint analysis was limited to patients who had at least two post-treatment results from the WPPSI-III and/or WISC-IV tests, conducted at least 2 years apart. A random coefficient model was used to calculate the change (per year) of FSIQ over the entire study period and for each patient in the efficacy population. The treatment was considered successful if the lower 95% confidence limit of the mean change in FSIQ excluded a decline of greater than 5 points over a 2-year window. This benchmark reflects the average margin of error for Wechsler tests [22]. Accounting for the possibility of attrition, the enrollment target for Part 2 was 60 patients, providing 90% power to detect a mean 2.5-point loss in FSIQ per year. Sensitivity analyses were performed to justify the missing at random assumption used in the random coefficient model. The last observation carried forward (LOCF) imputation used the last observed value carried forward for all subsequent missing time points, while the worst-case observation carried forward (WOCF) imputation used the minimum IQ score a patient had experienced. In addition, multiple imputation was performed, using a Markov-chain Monte Carlo sampling method, which assumes data are missing at random, and a multiple imputation sensitivity analysis, using fully conditional specification methodology, that further tests the robustness of the inference from the missing at random assumption.

For the secondary efficacy analysis, the height, weight, and head circumference

z-scores were determined based on the clinical growth charts and the percentiles from the World Health Organization (WHO) growth charts for children < 24 months and Centers for Disease Control and Prevention (CDC) clinical growth charts for children >24 months [24]. *Z*-scores and change in z-score from baseline of these growth variables were summarized for each age group and all ages combined at each scheduled time point.

Changes in total dietary Phe prescription and weight-based dietary Phe prescription were analyzed over time. The mean daily Phe prescription of study patients was compared to age-specific recommendations for daily Phe intake [25].

As an exploratory analysis, the least squares (LS) mean estimate of the change in FSIQ from baseline, defined as the first measurement of WPPSI-III or WISC-IV after determination of sapropterin responsiveness, over the follow-up period of 84 months was calculated using a repeated measures model. In addition, absolute mean changes in blood Phe levels from baseline to each follow-up time point were analyzed. Other exploratory endpoints were long-term FSIQ measurements, scores on the intelligence subtests, blood Phe concentration, relationship between intelligence test score and blood Phe, and prescribed dietary Phe intake. In addition, the indices of dietary control (IDC) were assessed by averaging the half-year Phe level medians over the first 6 months, over the first year, and annually thereafter.

3. Results

3.1. Patient disposition and baseline characteristics

Patients were recruited from 20 study centers in the United States and Canada. Of the 95 children enrolled in Part 1, 71 (74.7%) were considered sapropterin responders and eligible for Part 2, of which 65 (91.5%) enrolled (Supplementary Table 1). Overall, 63 (96.9%) patients met the criteria for inclusion in the efficacy analysis, while 62 (95.4%) patients had at least one year of treatment duration and met the inclusion criteria for the primary endpoint analysis. Ultimately, 49 (75.4%) patients completed Part 2 with 27 subjects (41.5%) having FSIQ assessments through 84 months (Fig. 1).

Demographic and baseline characteristics are shown in Table 1. The mean (SD) age at enrollment was 3.1 (2.0) years. The mean (SD) z-scores at baseline were 0.4 (1.0) for height, 0.4 (0.8) for weight, and 0.3 (1.0) for head circumference, which were within the expected growth percentiles. The mean (SD) baseline blood Phe concentration was 333.9 (135.0) μmol/L.

3.2. Sapropterin exposure and adherence

The mean total duration of sapropterin exposure for Part 1 and Part 2 was 6.5 years, with a maximum of 7.1 years. The mean treatment duration of the 62 patients included in the primary endpoint analysis was 6.8 years. After week 5, 25 subjects (38.5%) adjusted dose at least once, totaling 57 events. In Part 2, the mean daily dose of sapropterin was 20.2 ± 1.3 mg/kg/day across all age groups, closely approximating the initial dose of 20 mg/kg/day. Adherence to treatment, based on the prescribed dose taken and on the reconciliation of used and unused study drug, was 99.1 ± 1.6% with the overall minimum level of adherence being above 94%.

3.3. Phenylalanine, tyrosine, and tryptophan control

The mean changes in blood Phe concentrations from baseline over 7 years follow-up are shown in Fig. 2.

Overall, blood Phe declined from a mean (SD) of 332.8 (136.2) μmol/L at baseline to 217.7 (142.0) μmol/L and 251.1 (207.1) μmol/L at 3 and 6 months after initiation of sapropterin treatment, respectively. After this initial drop, there were no clinically relevant differences in mean blood Phe levels from baseline over 7 years follow-up in any age group. However, mean blood Phe levels remained within the American College of Medical Genetics (ACMG) target range of 120–360 μmol/L, with approximately 60% of children maintaining blood Phe levels within this range throughout the study. Furthermore, mean IDC were maintained between 133 μmol/L and 375 μmol/L blood Phe for all age groups at all time points (Fig. 3).

Relative to recommended daily Phe prescription by age [26], 30.5% were above, 64.6% were within, and 5.1% were below guidelines at baseline, indicating that some of the patients had a milder phenotype. After the first year of sapropterin use, the percentage of patients prescribed dietary Phe in excess of guidelines had increased to 45.9%, i.e. a 50% increase from baseline. Overall, mean (SD) prescribed dietary Phe increased from a

baseline of 396.8 (276.8) mg/day to 605.2 (443.3) mg/day at 12 months and 804.2 (518.9) mg/day at 84 months. The largest change was seen in those subjects who were 0 to <1 year of age at study entry, who had mean baseline prescribed dietary Phe intake of 283 (97) mg/day, increasing to 991 (637) mg/day by end of study, i.e. a 250% increase from baseline. The average weight-based dietary Phe prescription initially increased from a mean (SD) of 30.1 (26.2) mg/kg/day at baseline to 38.0 (35.4) mg/kg/day at 12 months, and then slowly decreased for all age groups over the course of the study (Supplementary Fig. 1). Out of the 65 enrolled patients, six patients (9.2%) had at least one tyrosine level below the age-related norms, and four patients (6.2%) had at least one tryptophan level below the age-related norms, but low values were infrequent and most patients only had one abnormally low value for both tyrosine and tryptophan. Conversely, 35 patients (53.8%) had at least one tyrosine level above the age-related norms, while eight patients (12.3%) had at least one tryptophan level above the age-related norms. Baseline values for tyrosine and tryptophan were largely missing for patients <2 years of age at study entry.

3.4. Preservation of intellectual functioning

The mean baseline FSIQ for the efficacy population was 101.1 ± 14.0, which is not significantly different from the population norm of 100 [18]. The long-term change in FSIQ compared to the population norm for all patients combined is shown in Supplementary Fig. 2.

Intellectual functioning was preserved throughout the duration of the study, and overall remained above the population norm. The slope of the change in FSIQ over the entire study period, calculated using the random coefficient model, was estimated at −0.29 per year with a 95% CI of −0.80 (lower limit) to 0.22 (upper limit) (n = 62). This yields a 2-year window estimate of −0.58 with a lower limit of the 95% CI of −1.60, excluding a decline in the FSIQ greater than 5 points. In addition, the 2-year lower limit of the 95% CI was not greater than 5 points based on the LOCF (−1.56), WOCF (−1.99), and multiple imputation (−1.51) sensitivity analyses. An additional sensitivity analysis, multiple imputation by fully conditional specification, was performed to confirm the original multiple imputation result, which used a missing at random (MAR) assumption. The sensitivity analysis assumes not missing at random (NMAR) and results in a 2-year lower limit of −4.71 when a 10% FSIQ decrease is observed (the lower limit is smaller when the decrease is smaller as well). Since 10% is unlikely in a given year, a lower limit greater than −5.0 is maintained under the NMAR assumption and the −1.51 lower limit is acceptable.

An exploratory analysis assessing the stability from baseline in the FSIQ score over the longest follow-up period of 84 months using a repeated measures model supported the results of the primary analysis (Table 2 and Supplementary Fig. 2). At 84 months (n = 27), the LS mean estimate of the change in FSIQ from baseline was 1.14, and the lower limit of the 95% CI was −3.53.

No correlation was found between scores on intelligence tests and blood Phe control or prescribed dietary Phe intake (data not shown). This is likely due to the fact that blood Phe and IDC were maintained within a tight metabolic range throughout the whole study, per protocol.

In addition, Performance IQ remained essentially stable, with the maximum deviation from baseline of +8.3 points at the 48-month time point, and a change of +4.5 points at the final 84-month time point. Verbal IQ likewise remained essentially stable, with a maximum deviation from baseline of +2.6 points at the 72-month time point, and a change of +0.7 points at the final 84-month time point (Supplementary Fig. 3). The Performance/Perceptual Reasoning, Processing Speed, and Working Memory scores remained stable as well during the course of the study for the majority of assessments with exceptions in the positive direction (Supplementary Table 2–4).

3.5. Growth assessments

Height and weight mean z-scores remained within the normal ranges throughout 7 years follow-up in all age groups. In addition, head circumference z-scores were maintained within the expected percentiles in patients between 0 and 4 years of age (Fig. 4).

3.6. Safety assessments

All 65 subjects (100%) reported at least one AE during the study (Table 3). There were no deaths or serious AEs (SAEs) that resulted in permanent discontinuation from sapropterin or from the study. However, two patients (8.7%) in the 2 to <4 year age group discontinued due to non-serious AEs, mild dysphonia in one subject and mild abdominal pain in the other subject, which both resolved. The most common AEs of special interest (>5% of patients) are shown in Supplementary Table 5. Overall, 35 subjects (53.8%), across all age groups, experienced episodic sapropterin-related AEs. The most common AEs assessed as sapropterin-related were upper respiratory tract infections (12 subjects, 18.5%), abdominal pain and vomiting (10 subjects each, 15.4%), and diarrhea (8 subjects, 12.3%). Eleven patients (16.9%) experienced SAEs, affecting two to four patients in each age group. Out of those, two subjects (3.1%) were assessed by the site investigator to experience sapropterin-related SAEs: one subject in the 2 to <4 year age group and one in the 4 to <7 year age group. All SAEs resolved without discontinuation from sapropterin. The overall summary of drug-related AEs is shown in Table 3. Furthermore, laboratory tests and vital signs showed no significant change over time.

4. Discussion

Medical nutritional therapy was initially the only available treatment option for children who were diagnosed with PKU through newborn screening programs. Adoption of a Phe-restricted diet supplemented with medical food within the first days after birth markedly improved the neurological and cognitive outcomes of PKU patients [7,27]. Dietary management should aim to prevent blood Phe above the ACMG target range (120–360 μmol/L) and avoid fluctuations in Phe, especially during childhood, since both are believed to be deleterious for brain development and predictive of late cognitive outcomes [28,29]. However, dietary management poses a significant burden to patients and their caregivers, often limiting adherence [6,8,30,31]. Although studies evaluating long-term growth outcomes in PKU patients on dietary management have shown conflicting results, a recent meta-analysis reported that growth is impaired in children with classical PKU [9]. Nevertheless, children with PKU were recently found to recuperate initially-delayed growth

beyond the age of 12 years [32]. Generally, the inability to achieve optimal growth is related to nutritional deficiencies caused by inadequate dietary management rather than to high blood Phe [9]. This may especially be relevant to patients with classical PKU, in whom growth was found to be significantly reduced as compared with non-classical PKU patients [33].

Sapropterin was the first non-dietary pharmacological therapy approved for the treatment of PKU patients with BH4-responsiveness [10,11]. The results of the PKU-015 study confirm those of previously described observational data, demonstrating the safety and effectiveness of sapropterin to control blood Phe. Safety and efficacy of sapropterin were initially shown in patients above the age of 4 years [12,13]. Shortly afterwards, the safety and positive impact of sapropterin on blood Phe control were demonstrated in small cohorts of children treated between the ages of 0 to 4 years [34,35]. These findings were confirmed by the PKU Demographics, Outcomes, and Safety (PKUDOS) registry, including 97 children in whom sapropterin treatment was initiated below the age of 4 years [36]. In an interim analysis of the PKUDOS registry, sapropterin treatment resulted in a 42% decrease in average blood Phe after 3 years from baseline, when patients were continuously exposed to sapropterin. This drop in blood Phe also allowed an increase in Phe tolerance in the same age group [36]. Additionally, the SPARK study showed a mean increase in Phe tolerance of 30.5 mg/kg/day in children <4 years of age at enrollment who were treated for 26 weeks with sapropterin and a Phe-restricted diet compared with children treated with a Phe-restricted diet alone [16]. As secondary endpoints, the SPARK study attempted to evaluate the impact of sapropterin treatment on intellectual functioning and growth outcomes, demonstrating that anthropometric parameters were stable and neuromotor development was normal in both treatment groups [16]. Although all patients were enrolled in a 3-year open-label extension phase, long-term data on neurocognitive and growth outcomes are not reported, as few patients were evaluated in the SPARK study for these outcomes [17]. Neurological and growth outcomes were also evaluated in the Kuvan® Adult Maternal Paediatric European Registry (KAMPER) study, which is an observational, multi-center drug registry, including patients across all ages with HPA treated with sapropterin for up to 15 years [37]. Across all patients <10 years of age, weight, height, and BMI z-scores were maintained within the normal ranges according to the CDC growth charts. Additionally, 92.8% of all PKU patients (n = 362) between <4 years (n = 3) and 4 to ≤18 years (n = 333) of age were at the appropriate school level. Nevertheless, only 10 patients enrolled in the KAMPER study were below the age of 4 years at enrollment, hampering robust evaluation of early sapropterin treatment on the preservation of intellectual functioning and the maintenance of growth in this age group [37].

Therefore, the PKU-015 study was designed to primarily assess the long-term effect of sapropterin on the intellectual ability and growth outcomes of young children during this critical period of neurological and physiological development. The interim results of this study previously showed that sapropterin treatment of infants and young children aged 0 to 6 years in conjunction with a Phe-restricted diet was well-tolerated, while growth parameters were maintained within the normal range during the first 2 years of follow-up, and intellectual functioning remained stable [18]. These findings are now supported by the longer-term outcomes, which demonstrate that FSIQ and growth rates were maintained over

a follow-up period of 7 years and remained within the normal ranges. Furthermore, Verbal and Performance subset scores remained stable or increased over the duration of the study. The stable intellectual outcomes observed in this study are most likely related to long-term metabolic control, as mean IDC was maintained between 133 μmol/L and 375 μmol/L for all age groups at all time points. These results are in agreement with a previous study demonstrating that sapropterin therapy results in less fluctuating blood Phe levels, with a significant decrease of within-subject variances of mean blood Phe from 417.7 μmol/L (6.9 mg/dL) before to 290.6 μmol/L (4.8 mg/dL) after sapropterin treatment [38]. Additionally, the long-term maintenance of growth among children in this study supports sapropterin use in this population, as growth outcomes in children with PKU treated with diet have been shown to be suboptimal [9]. The increase in dietary Phe observed in this study may be at least partially attributed to increased dietary Phe tolerance, associated with stimulation of residual PAH enzymatic activity by sapropterin. This increased dietary Phe tolerance may allow children to relax some of the dietary restrictions, presumably improving quality of life [39]. The largest changes were observed in the youngest patients enrolled in this study. This is expected due to the rapid early growth expected in this age cohort, an effect that was likely maintained due to the increased Phe tolerance provided by sapropterin. Less change was seen in older subjects, likely due to lower growth rates. In accordance with the 2-year interim results, sapropterin was generally well-tolerated in this young patient population over 7 years. No safety signals other than those listed on the sapropterin package insert were identified [10].

The most significant limitation to this study was the lack of a concurrent control arm (i.e. dietary management only). Nevertheless, results from this study may be apposed to the previously published placebo-controlled SPARK study, demonstrating increased dietary Phe tolerance in sapropterin-treated children below the age of 4 years in comparison with a Phe-restricted diet only [16]. Because sapropterin was already approved by the FDA for treatment of patients of this age prior to the initiation of this study, there was no concern about an increased safety risk in children under the age of 4 years. Another limitation is the challenge in comparing IQ scores over time between different age-appropriate measurement tools. The major restriction was that the assessment of intellectual functioning in children aged 0 to 30 months was measured by the Bayley-III, which yields a development performance score rather than an IQ score. Therefore, Bayley-III baseline data could not be analyzed together with WPPSI-III and WISC-IV scores obtained after later assessments. Additionally, performance assessments are less robust in terms of psychometric properties (validity and reliability) than the FSIQ [40] and are also prone to practice effects, which may be exacerbated in this study by the low patient numbers [21]. The study was also limited to children living in North America, but a similar study (Kuvan®'s Effect on the Cognition of Children With Phenylketonuria [KOGNITO], NCT01965912) is currently ongoing in Europe, with results are expected in 2023.

An important strength of the study is the high adherence rate of the study population to sapropterin. Adherence was approximately 99%, with a mean total duration of exposure of 6.5 years, and a total duration of exposure of up to 7.11 years.

In conclusion, the PKU-015 study allowed evaluation of the long-term safety of sapropterin and the assessment of intellectual functioning and growth parameters in children with PKU who initiated treatment with sapropterin within the age range of 0 to 6 years. Sapropterin was generally well-tolerated in this patient population. In addition, children with PKU treated with sapropterin and a Phe-restricted diet were able to maintain blood Phe concentrations within the target range of 120–360 μmol/L over seven years follow-up, while increasing dietary Phe tolerance. As demonstrated here, long-term maintenance of blood Phe concentration within this target range is associated with the preservation of normal intellectual functioning and growth. This result supports sapropterin response testing in infancy and initiation of sapropterin in those infants and young children who are diagnosed with sapropterin-responsive PKU.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Abbreviations:

ACMG	American College of Medical Genetics
AE	adverse event
BH4	tetrahydrobiopterin
CDC	Centers for Disease Control and Prevention
CI	confidence interval
EMA	European Medicines Agency
FDA	Food and Drug Administration
FSIQ	Full-Scale Intelligence Quotient
HPA	hyperphenylalaninemia
IDC	index of dietary controll
IQ	intelligence quotient
KAMPER	Kuvan® Adult Maternal Paediatric European Registry
LOCF	last observation carried forward

LS	least squares
MAR	missing at random
NMAR	not missing at random
PAH	phenylalanine hydroxylase
Phe	phenylalanine
PKU	phenylketonuria
PKUDOS	PKU Demographics, Outcomes, and Safety
SAE	serious adverse event
SD	standard deviation
SE	standard error
SPARK	Safety Paediatric efficAcy phaRmacokinetic with Kuvan®
WISC-IV	Wechsler Intelligence Scale for Children, Fourth Edition
WHO	World Health Organization
WOCF	worst case observation carried forward;
WPPSI-III	Wechsler Preschool and Primary Scale of Intelligence, Third Edition

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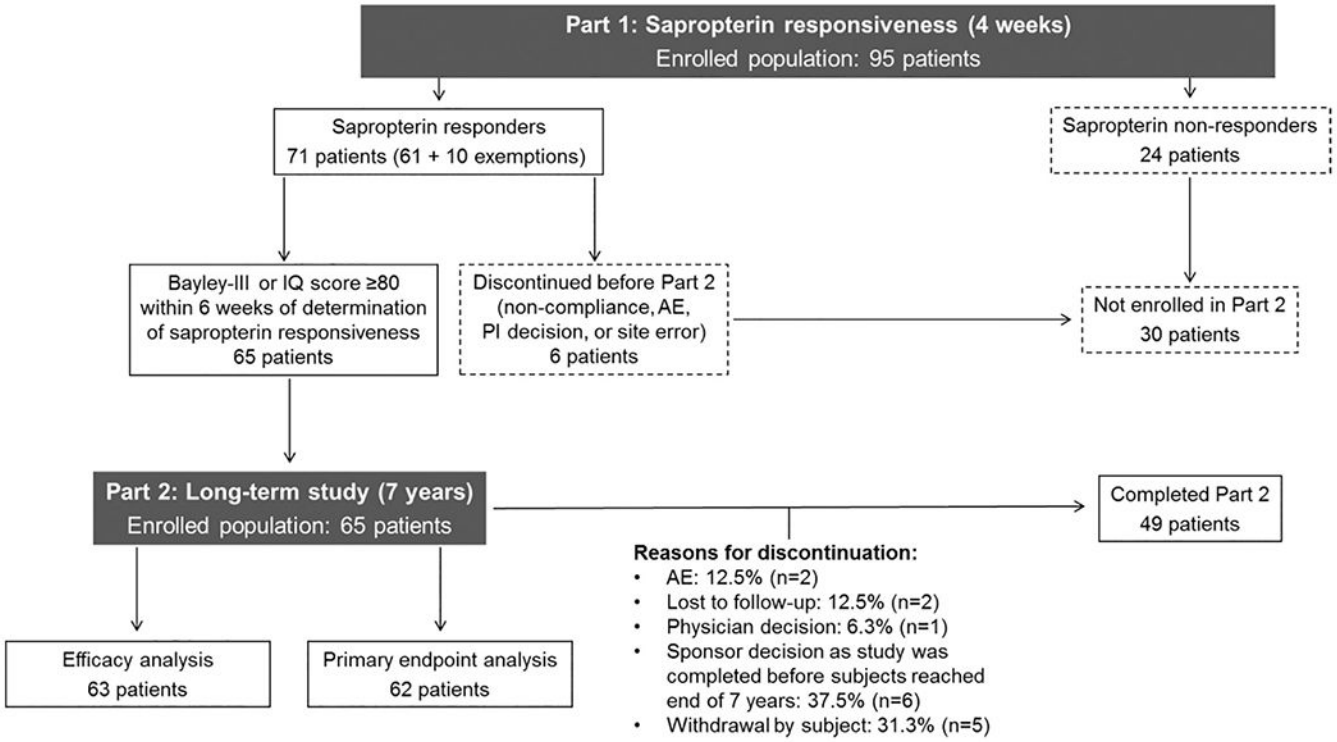

Fig. 1.

Study flow diagram with patient disposition. Sapropterin responders were defined as patients who had ≥30% average reduction in blood Phe concentration during the first 4 weeks. Exemptions were granted to ten patients whose mean blood Phe reduction was <30% during the first four weeks. For eight patients, an exemption was granted to substitute an alternative blood Phe measurement, when a scheduled blood Phe measurement was thought to be abnormally elevated due to dietary Phe overcorrection, illness, or surgery. Two patients with blood Phe reduction close to 30% were also granted an exemption. Exclusion of these ten patients in a sensitivity analysis resulted in no apparent differences in outcome measures (data not shown). AE: adverse event; IQ: intelligence quotient; PI: principal investigator; WISC-IV: Wechsler Intelligence Scale for Children, Fourth Edition; WPPSI-III: Wechsler Preschool and Primary Scale of Intelligence, Third Edition.

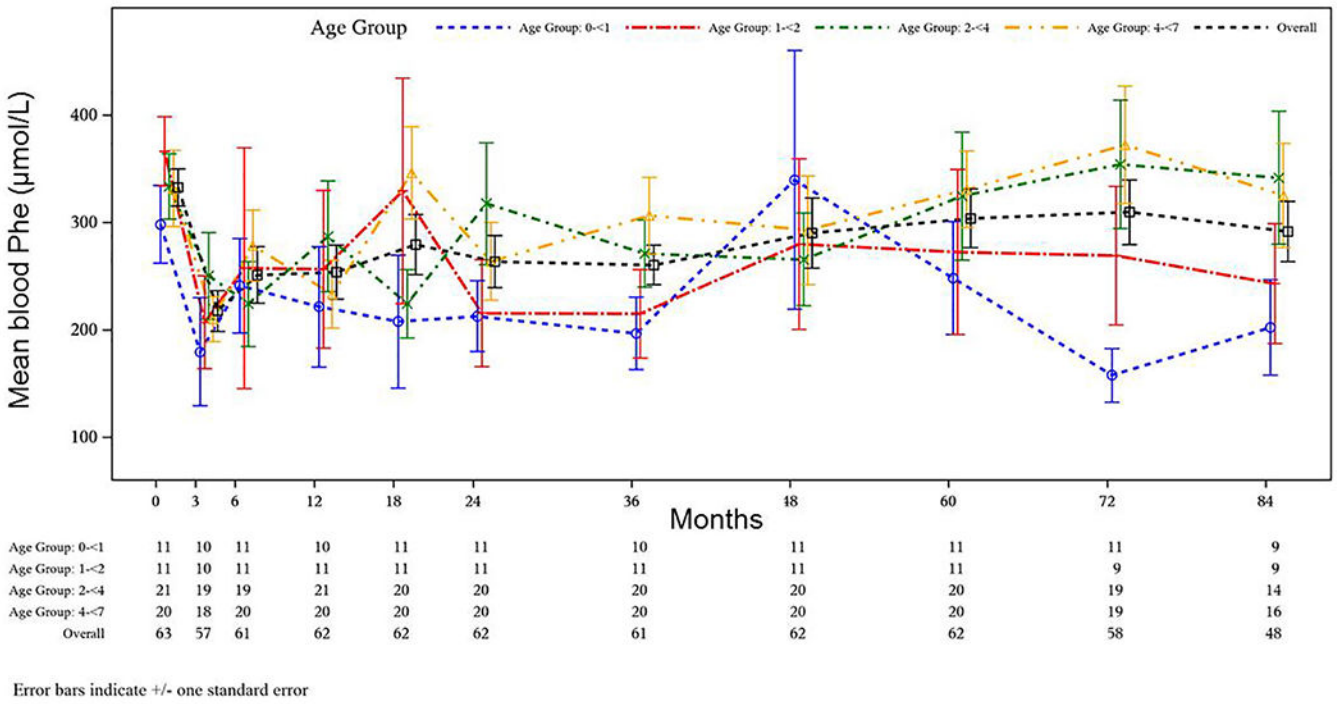

Fig. 2.

Mean blood phenylalanine concentrations from baseline over 7 years follow-up in different age groups and across all age groups (overall). Phe: phenylalanine. Numbers underneath the graph represent the number of patients in each age group.

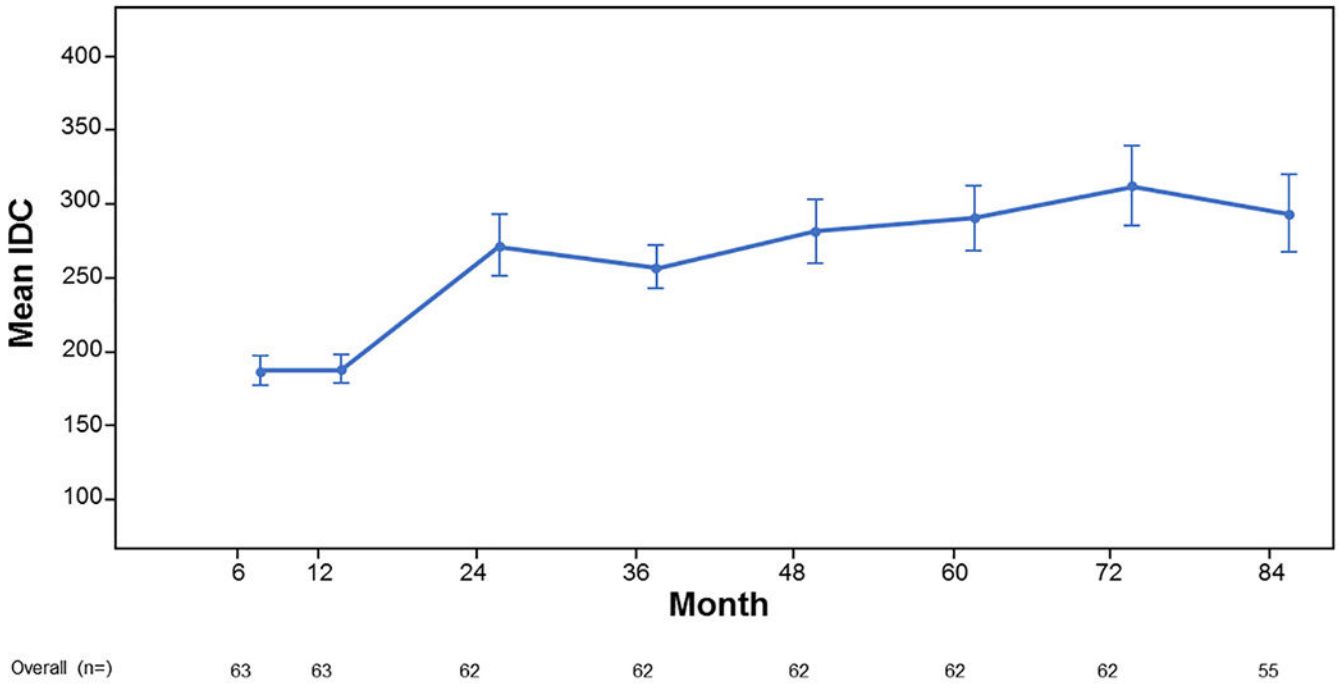

Fig. 3.

Mean index of dietary control (IDC) over time. IDC is measured as half-year Phe level medians averaged over the first 6 months, over the first year, and annually thereafter.

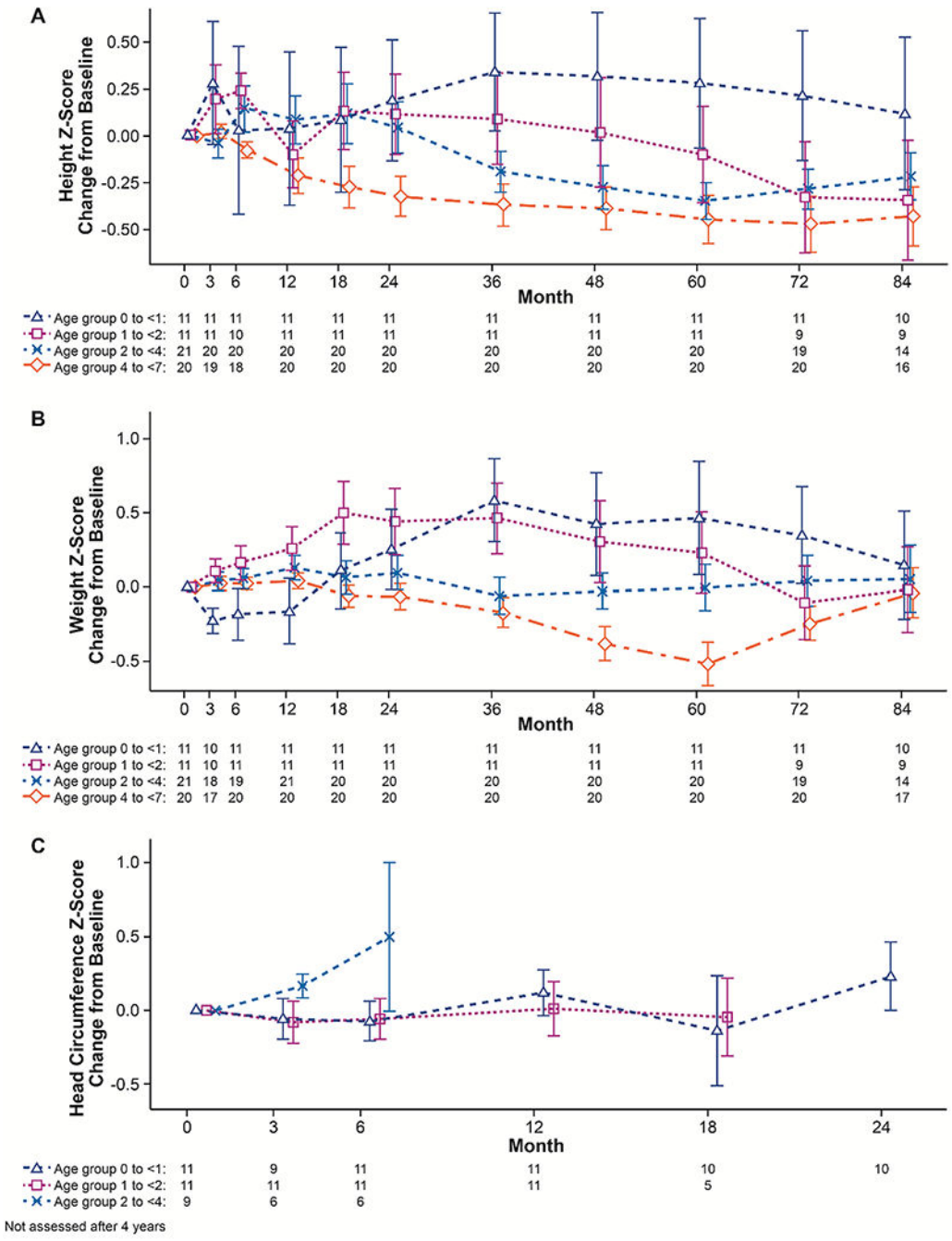

Fig. 4.

Growth assessments over time in different age groups. (A) Mean (SE) changes from baseline in height z-scores over 84 months (7 years) follow-up. (B) Mean (SE) changes from baseline in weight z-scores over 84 months (7 years) follow-up. (C) Mean (SE) changes from baseline in head circumference z-scores up over 6 months for the 2 to <4 years age group, 18 months for the 1 to <2 years age group, and 24 months (2 years) for the 0 to <1 year age group. SE: standard error; z-scores were calculated based on the World Health Organization (WHO) growth charts for children < 24 months and Centers for Disease Control and

Prevention (CDC) clinical growth charts for children > 24 months. Values within 1 SD of the population mean are within the expected growth percentiles. Numbers underneath the graph represent the number of patients in each age group.

Table 1

Baseline characteristics of subjects enrolled in Part 2.

	Age groups				Total (n = 65)
Characteristic	**<1 year (n = 11)**	**1 to < 2 years (n = 11)**	**2 to < 4 years (n = 23)**	**4 to < 7 years (n = 20)**	
Age (years) at enrollment, mean (SD)	0.44 (0.28)	1.37 (0.33)	3.03 (0.67)	5.62 (0.95)	3.11 (2.04)
Sex, N (%)					
Female	6 (54.5%)	5 (45.5%)	15 (65.2%)	14 (70.0%)	40 (61.5%)
Male	5 (45.5%)	6 (54.5%)	8 (34.8%)	6 (30.0%)	25 (38.5%)
Race, N (%)					
White	9 (81.8%)	9 (81.8%)	18 (78.3%)	18 (90.0%)	54 (83.1%)
Asian	1 (9.1%)	1 (9.1%)	0	1 (5.0%)	3 (4.6%)
Black or African American	0	0	1 (4.3%)	0	1 (1.5%)
Other	1 (9.1%) 1	1 (9.1%)	4 (17.4%)	1 (5.0%)	7 (10.8%)
Ethnicity					
Not Hispanic or Latino	11 (100.0%)	11 (100.0%)	20 (87.0%)	20 (100.0%)	62 (95.4%)
Hispanic or Latino	0	0	3 (13.0%)	0	3 (4.6%)
Weight (z-score), mean (SD)	0.25 (0.94)	0.00 (0.86)	0.48 (0.75)	0.69 (0.77)	0.42 (0.83)
Height (z-score), mean (SD)	0.21 (1.15)	0.47 (0.72)	0.41 (1.05)	0.47 (0.91)	0.41 (0.96)
Head circumference (z-score), mean (SD)	−0.01 (0.66)	0.38 (1.01)	0.48 (1.25)	NA (NA)	0.28 (0.99)
Blood phenylalanine concentration (µmol/L), mean (SD)	298.41 (119.67)	366.50 (107.17)	337.02 (135.47)	332.02 (158.81)	333.93 (135.03)

N: number of patients with data available for each variable; NA: not applicable; SD: standard deviation;

z-score was calculated based on the World Health Organization (WHO) growth charts for children < 24 months and Centers for Disease Control and Prevention (CDC) clinical growth charts for children > 24 months. Values within 1 SD of the population mean are within the expected growth percentiles.

Table 2

The least squares (LS) mean estimates of the long-term change in FSIQ score from baseline using a repeated measures model.

Analysis visit	N	LS mean change (SE)	95% CI	
			Lower	Upper
Month 12	59	1.24 (2.155)	−3.05	5.52
Month 24	58	1.44 (2.174)	−2.89	5.76
Month 36	55	1.40 (2.183)	−2.94	5.74
Month 48	48	1.41 (2.238)	−3.04	5.85
Month 60	38	−1.62 (2.289)	−6.16	2.92
Month 72	23	0.21 (2.439)	−4.62	5.04
Month 84	27	1.14 (2.358)	−3.53	5.82

Baseline is defined as the first available measurement after determination of sapropterin responsiveness.

CI: confidence interval; FSIQ: full-scale intelligence quotient; LS: least squares; N: number of patients with data available; SE: standard error.

Table 3

Summary of drug-related adverse events.

Drug-related AEs occurring in > 10% of patients, N (%)	**Age group**				**Total (n = 65)**
	<1 year (n = 11)	**1 to < 2 years (n = 11)**	**2 to < 4 years (n= 23)**	**4 to < 7 years (n = 20)**	
Upper respiratory tract infection	3 (27.3%)	2 (18.2%)	3 (13.0%)	4 (20.0%)	12 (18.5%)
Abdominal pain	2 (18.2%)	0	6 (26.1%)	2 (10.0%)	10 (15.4%)
Vomiting	2 (18.2%)	3 (27.3%)	4 (17.4%)	1 (5.0%)	10 (15.4%)
Diarrhea	1 (9.1%)	1 (9.1%)	4 (17.4%)	2 (10.0%)	8 (12.3%)
Drug-related SAEs, N (%)	**Age group**				**Total (n = 65)**
	<1 year (n = 11)	**1 to < 2 years(n= 11)**	**2 to < 4 years (n = 23)**	**4 to < 7 years (n = 20)**	
Colitis ulcerative	0	0	0	1 (5.0%)	1 (1.5%)
Diarrhea	0	0	0	1 (5.0%)	1 (1.5%)
Convulsion	0	0	1 (4.3%)	0	1 (1.5%)

AE: adverse event; N: number of patients; SAE: serious adverse events.