

Welcome to the “CONNECTING THE WORLD OF PKU” congress highlights editorial!

The “CONNECTING THE WORLD OF PKU” forum congress took place in Hamburg, Germany, from 29 to 30 October 2025. It is an international forum designed to convene all stakeholders actively engaged in the management and care of patients affected by phenylketonuria (PKU).

The scientific programme covered a wide range of topics, providing a unique and valuable opportunity for networking and discussion among colleagues dedicated to the management of PKU. A revised understanding of the pathology of PKU was explored, including the potential identification of novel biomarkers and metabolic control parameters, together with updates on dietary and pharmacological treatments. Furthermore, critical assistance-related issues such as the burden of illness and unmet needs were addressed.

In this congress highlights editorial, all sessions and expert insights/discussions of the 2-day forum are summarised. We hope that this educational editorial proves beneficial to your clinical practice, offering valuable insights into current research and into new ways the PKU community can contribute to collectively address global challenges in PKU.

CONTENTS

- ▶ **Modifying paradigms in PKU**
- ▶ **Long-term outcomes**
- ▶ **Revised PKU guidelines**
- ▶ **Advances in dietary treatment**
- ▶ **Burden of illness and unmet needs of PKU**
- ▶ **Changing landscape: pharmacological options**
- ▶ **Future perspectives**
- ▶ **Industry sponsored sessions: meet the expert**

BIOMARIN[®]

All opinions and thoughts discussed in this editorial are entirely the speakers' own, based on their clinical experiences, and not necessarily represent those of BioMarin. BioMarin does not recommend the use of any product in any matter different from that described in the approved prescribing information. Treatments mentioned in this document may not be approved for use in your country. Please consult local licensing authorities for further information. Sapropterin and pegvaliase indications and prescribing information are listed at the end of this document. Some links in this document are “external links” to websites over which BioMarin has no control and for which BioMarin assumes no responsibility. When visitors choose to follow a link to any external website, they are subject to the cookie, privacy, and legal policies of the external website. Compliance with applicable data protection and accessibility requirements of external websites linked to from this website falls outside the control of BioMarin and is the explicit responsibility of the external website. PKU.expert is organised and funded by BioMarin. For healthcare professionals only. This congress highlights editorial has been developed and funded by BioMarin. ©2025 BioMarin International Ltd. All Rights Reserved. For healthcare professionals only. COM-ET-1003 Date of preparation: December 2025

Palynziq[®]
(pegvaliase) Injection

Modifying paradigms in PKU

Although PKU has been studied for decades, several aspects remain to be fully understood. This section includes expert insights on current approaches to PKU diagnosis and monitoring and explores emerging biomarkers that may improve understanding and management of PKU.

Screening, diagnosis, and monitoring of PKU: what have we learned?

Dr. Pasquali's centre investigated receiver operating characteristic (ROC) analyses (tool to quantify predictive accuracy of diagnostic test) to evaluate phenylalanine (Phe) concentration and Phe/Tyrosine (Tyr) ratio obtained from NBS as potential predictors of classic PKU vs. hyperphenylalaninaemia (HPA). Results indicated that the Phe/Tyr ratio can aid in differentiating classic PKU from HPA, while Phe concentration alone was found to be insufficient.

With regards to monitoring, Phe and Tyr levels can be measured using both dried blood spots (DBS) and plasma samples, although clinical decisions should take into account that Phe levels from DBS can be 15-30% lower than the corresponding plasma levels. Another important consideration is that plasma samples collected from patients receiving pegvaliase could indicate larger reduction in Phe concentration if not handled properly due to residual pegvaliase activity.

Variants and correlations: can we predict phenotype?

Dr. Blau reviewed phenotype prediction rules and tools for PKU. Generally, a patient's genotype determines the residual phenylalanine hydroxylase (PAH) activity, which in turn determines both the metabolic phenotype and tetrahydrobiopterin (BH₄) responsiveness. Milder variants dominate over the severe ones and are phenotype-determining. A severe genotype (2 null-variants) is associated with classic PKU and can exclude BH₄ responsiveness. Overall, the PKU phenotype can be predicted from the genotype with 88% confidence.

Phenotype prediction can be performed using two distinct approaches: pathogenic classification according to ClinGen/ClinVar criteria and genotype-phenotype correlation using the large BIOPKU database. The BIOPKU phenotype data for variants occurring in homozygous or functionally hemizygous state can be used to calculate the allelic phenotype value (APV)¹, which was identified as a globally used powerful tool for phenotype prediction.

Biomarkers in PKU: what can we do more?

Dr. van Spronsen discussed the use of biomarkers to assess clinical outcome measures in PKU. Plasma Phe levels as surrogate marker for brain Phe levels was shown to explain only 62% of clinical outcomes, highlighting the need for other biomarkers. However, many studies investigating e.g., lipidomic, metabolomic or protein biomarkers, failed to report any relation with clinical outcomes, sparking the hypothesis that part of the poor outcomes observed might be consequences of (dietary) treatment itself instead of PKU as disease. This was further illustrated with examples of diet burden leading to hidden outcomes including anxiety, depression and eating disorders. It was emphasised that not only biomarkers can be used to improve understanding of PKU outcomes, but also that there is a need for more in-depth questions and novel tools that address and measure the burden of PKU as a disease, its treatment and care for PKU patients.

Phe target levels: what is normal?

Prof. Burlina addressed the concept of Phe homeostasis and normal Phe levels in PKU. In people without PKU, there is a steady-state between Phe input and runout. This Phe homeostasis was described as a complex quantitative trait involving genetic, metabolic and environmental factors. Disruption of PAH enzyme function impacts homeostasis and leads to HPA and PKU, highlighting the importance of metabolic regulation. In the context of PKU, it was highlighted that 'normal' may have different meanings, and to prevent ambiguities inherent to the term 'normal values', the concept of reference values was introduced. 'Normal' Phe levels in individuals with PKU refer to a therapeutic range minimising neurotoxicity while maintaining quality of life (QoL). Although such target ranges are described in PKU guidelines, it was highlighted that differences in the United States (US)² vs European guidelines³ should be resolved to address the question of what is 'normal' in PKU.

Multimodal metabolic and novel metabolic disturbances

Dr. Mailliot discussed the use of metabolomic approaches in PKU. The identification of metabolomic signatures can help to advance the understanding of PKU pathophysiology, as well as to identify new biomarkers that may improve monitoring of PKU patients or to uncover novel

treatment targets. This was illustrated by Dr. Mailliot's research that evaluated human samples using a multiplatform metabolomics approach^{4,5}. Besides disturbances in Phe and Tyr, other metabolites (e.g., cortisol) and pathways (e.g., glycolysis) were found in the metabolic signature of PKU. Furthermore, mice brains were studied, showing similar findings as in humans. Combined with data from other publications⁶, the results led to two more hypotheses for future exploration: based on the presence of indole-3-lactic acid, the gut-brain axis could be involved in PKU, and lastly, data pointing towards DNA methylation pathways in the brain suggest DNA methylation alterations in PKU.

Discussion

Dr. Vockley asked about integrating all available data in search of a biomarker and wondered how to deal with all mentioned approaches in a clinical sense. Dr. Mailliot agreed using multi-omics is needed to study relations with both clinical outcomes and genotype. Furthermore, Dr. Feillet questioned whether individualised medicine should be considered rather than determining what is 'normal' in general. He mentioned some difficulties that were encountered with adopting guidelines based on his own experience with an elderly patient feeling good at Phe levels above the recommended target range. Prof. Burlina added that 'normal' should be viewed in a clinical

sense, meaning healthy, and although he agreed some patients might be feeling well at higher Phe levels, he underlined the majority still benefits from treatment. Dr. Harding agreed, and explained patients might only realise they feel better at lower levels once they are able to reach these levels with pharmacological treatment such as pegvaliase.

Long-term outcomes

There is significant amount of research ongoing on the long-term effects of high Phe levels in adults with PKU. In this section, the impact of PKU and its treatment on long-term neurocognitive outcomes, as well as comorbidities including cardiovascular disease and overweight are discussed.

Neurocognitive outcomes: PKU and the brain

Dr. Nardecchia discussed neurocognitive outcomes in adults with early-treated PKU (ETPKU). Findings from cross-sectional, longitudinal and interventional studies indicate that strict metabolic control and normalisation of Phe levels could improve neurocognitive functions in adult ETPKU patients, pointing to residual brain plasticity and emphasising the importance of Phe-lowering treatment that allow (almost) normal Phe levels. Unexpectedly for adult patients, intellectual functioning (intelligence quotient (IQ), attention and inhibitory control, working memory, memory and learning, cognitive flexibility, and language) also showed a significant improvement after diet resumption and Phe reduction, although the clinical response to low Phe was variable between adults with similar metabolic control, indicating individual vulnerability to Phe. Future research efforts should focus on biomarkers predicting this individual response to Phe values, and on developing more treatments that could lead to Phe normalisation.

Multiple mechanisms of Phe neurotoxicity: could blood Phe levels close to physiological be the solution?

Dr. Gizewska reviewed neuropathological changes in the brain of untreated PKU patients. As there is competition between Phe and other large neutral amino acids (LNAA, e.g., Tyr and tryptophane (Trp)) for the LAT-1 transporter in the blood brain barrier (BBB), elevated blood Phe levels result in reduced transport of LNAA to the brain. This depletion of brain Tyr and Trp leads to reduced protein synthesis, which is necessary for proper cognitive functioning (Tyr/dopamine theory⁷). However, not only neurotransmitter abnormalities are observed in PKU. To explain myelin abnormalities, the myelin/dopamine theory was presented. This theory states that myelin/axonal interactions transduce signals that upregulate the production of Tyr hydroxylase, an enzyme involved in dopamine biosynthesis, which is inhibited by elevated Phe levels. High Phe levels also negatively impact other cerebral processes involved in myelin production. Ultimately, demyelination and hypomyelination result in white matter

abnormalities, which may be associated with suboptimal neurocognitive outcomes. Further studies are needed to investigate how installing physiological Phe levels may improve or reverse brain abnormalities as consequence of Phe toxicity.

Antioxidant status and cardiovascular risk

Dr. Reissman presented cardiovascular (CV) disease as an important comorbidity in PKU patients. This was illustrated with epidemiological insurance claim data⁸ indicating higher rates of ischemic cardiac disease in PKU patients, and with studies⁹ demonstrating changes in cardiac structure in PKU and the important role of high plasma Phe levels in heart aging. It was hypothesised that normal senescence of cardiac tissue is accelerated in PKU patients, especially in case of poor metabolic control. The research in this field focuses on one hand on traditional CV risk factors such as obesity and dyslipidaemia, that are both present in many PKU patients⁸, but also non-traditional CV risk factors are being investigated. Oxidative stress was

demonstrated to be higher in adult PKU patients, while there is no clear correlation discovered yet between proinflammatory state and PKU. It was emphasised that CV risk screening should be incorporated into the routine care of ETPKU patients.

Protein intake and prevalence of overweight

Dr. Rocha presented overweight in PKU as a complex interaction of multiple factors, including nutritional/energy intakes and energy expenditures, PKU severity, and treatment modalities. Overweight in PKU is a relevant issue and is now included in the revised European guidelines, with a novel recommendation stating 'In PKU, preventive life-style strategies should be implemented to avoid overweight and associated comorbidities'.³ Results from a 10-year longitudinal study from Dr. Rocha's centre¹⁰, aiming to identify prevalence of overweight in patients with PKU following a Phe-restricted diet, indicated total and natural protein intake as protective factors against the development of overweight. Furthermore, a non-significant trend towards increased prevalence of overweight was observed, highlighting the importance of annual nutritional status evaluation and continuous follow-up. Additionally, a case report on body composition in infants with PKU illustrated breast feeding may have a positive impact on body fat, although more research is needed to understand the concept of early programming of adipose tissue in PKU.

Discussion

The majority of the discussion was focused on protein intake and overweight. Dr. Ney commented non-PKU studies on obesity also demonstrate that the most satiating nutrient is protein, specifically whey proteins. Dr. Rocha agreed protein type matters, and its effects on metabolism should be further studied. To prevent overweight in PKU, Dr. Rocha mentioned the importance of breast feeding in early life due to its impact on the metabolic programming journey of infants, and emphasised it should be promoted in every centre. Furthermore, he recommended to incorporate normal infant nutrition, as data shows the risk of obesity at 6 years is 50% higher in children treated with high protein formula in their early years. Lastly, he recommended natural food intake in all PKU patients, as long as pharmacological treatment allows it and neurotoxicity can be prevented. Dr. Rocha and Dr. Singh discussed important considerations to treat overweight, which included the quality of weight loss (e.g., loss of muscle mass vs loss of body mass).

Revised PKU guidelines

Guidelines are essential to establish a standard for diagnostics, treatment, and care in PKU, that would lead to optimised clinical and neuropsychological outcomes without overtreatment and unnecessary costs. This section includes insights from the revised US (American College of Medical Genetics and Genomics, ACMG) and European guidelines.

New guidelines: US

Dr. Vockley discussed guidelines in general as a means to evaluate best practices, present evidence on treatment options and identify gaps. He further compared the main PKU guideline recommendations from both the US (ACMG)² and Europe³. A key difference is that the novel ACMG guidelines endorse a single ≤ 360 $\mu\text{mol/L}$ target across life, no longer defining a lower limit, while the revised European recommendations retain the wider 120-600 $\mu\text{mol/L}$ range for individuals aged ≥ 12 (except for pregnancy where they recommend 120-360 $\mu\text{mol/L}$). Threshold to initiate treatment (360 $\mu\text{mol/L}$) is the same, but ACMG frames this as a lifelong target while Europe differentiates by age and physiologic state. Both endorse sapropterin and pegvaliase as pharmacological interventions in eligible patients. In both guidelines, QoL is emphasised and patient education and support is strongly recommended.

New guidelines: Europe

Dr. Van Wegberg discussed the 2025 revision of the European guidelines³. New recommendations cover emerging pharmacological options (e.g., pegvaliase) and a specific neurocognitive test battery. In contrast to the ACMG guidelines, European target Phe levels did not change, as evidence was considered insufficient. Target Phe levels remain a key discussion point globally. This was illustrated by another set of recommendations from German-speaking countries (DACH), where the proposed upper target level for adults is 1200 $\mu\text{mol/L}$. Differences mainly arise from distinct values and preferences amongst healthcare professionals (HCPs), with some prioritising being safe (but maybe overtreat), while others prefer a more lenient approach as the treatment can be burdensome (but maybe undertreat). To align target Phe levels in the future, neurocognitive outcomes in adults need to be captured with better equipped tools, and the impact of Phe levels ≤ 360 $\mu\text{mol/L}$ vs 360-600 $\mu\text{mol/L}$ vs > 600 $\mu\text{mol/L}$ needs to be evaluated further.

Discussion

Differences in guidelines, especially in target Phe levels, sparked a lively discussion. The idea of a global guideline was suggested, but more data is needed to demonstrate improved outcomes of Phe levels of ≤ 360 $\mu\text{mol/L}$ versus < 600 $\mu\text{mol/L}$. Prof. Muntau remarked that currently, achieving target Phe levels remains difficult in both Europe and the US when relying solely on dietary management, and she highlighted the need for pharmacological treatment options (e.g., pegvaliase) that may help meet guideline-recommended goals. Dr. van Wegberg added that such treatments will also allow exploring potential benefits of low Phe levels and help gather the evidence needed to update recommendations. In response to questions regarding higher Phe targets, she stressed that guidelines are meant to inform rather than dictate, and that HCPs can make different choices in their patients' best interest. She reiterated the idea that patients do not know their optimal Phe levels without having experienced lower ones. Lastly, Dr. Singh commented about the need for nutrition-detailed guidelines that consider diet manipulation upon reaching lower Phe levels (including hypoPhe).

Advances in dietary treatment

PKU management primarily consists of medical nutrition therapy, including a low-Phe diet supplemented with specialised medical formulas that provide essential protein, vitamins, and minerals. This section covers data protein source research and dietary interventions for individuals with PKU.

Transitioning of protein substitutes: state of play

Dr. MacDonald reviewed the transition of protein substitutes throughout life. Key aspects to consider in this transition include nutritional requirements, developmental/life stage and product availability, sustainability and reimbursement. The importance of good transition management is evident, as this is associated with easier lifelong administration, better lifelong metabolic control, and reduced burden of treatment. Patient-related factors such as maturity, metabolic control and taste preferences, but also support structures (motivation of caregivers, resilience and community support), impact the transitioning process. This was illustrated by a pilot study¹¹ of Dr. MacDonald investigating a step-by-step transitioning of protein substitutes in children 3-5 years of age. Results indicated that lack of consistency and routine, frequent intercurrent illnesses and low parent resilience may result in a cycle of manipulation, control and defiance. Strategies to improve transitioning included

collaborating with schools, both parents, extended family and directly with the children, as well as using engaging materials and close monitoring.

Low protein foods: past, present and future

Dr. Dianin discussed special low protein foods (SLPFs), which are considered essential for patients adhering to a Phe-restricted diet. Although introduced in the 1950's, many challenges with SLPF continue to complicate dietary management. Key issues include the lack of specific maximum limits of Phe content in European legislation, the lack of uniform reimbursement policies, and different global availability. Important future considerations include the need for manufacturers to increase detail on nutritional composition and product labelling. Furthermore, the intake of SLPFs should be carefully monitored given the potential long-term cardiovascular risks associated with high consumption of saturated fats and sugars, and nutritional quality of SLPFs should be improved (e.g., reducing this saturated fat and sugar content). It was also deemed crucial to assess the

overall diet to recommend healthier food choices. Lastly, it was highlighted that patients treated with pharmacological treatments (e.g., pegvaliase) allowing reduced reliance on SLPFs can often transition to a normal, healthy diet.

Natural proteins: fruit and vegetables impact on blood Phe level

Dr. Pinto discussed natural protein intake and its impact on blood Phe levels. Previous studies have shown that consuming fruits and vegetables containing ≤ 75 mg Phe per 100 g does not impact blood Phe levels. Building on this, Dr. Pinto conducted a randomised-control trial¹² to investigate the impact of fruit and vegetables containing 76-100 mg Phe per 100 g on blood Phe levels, and to compare the impact of vegetable vs animal protein on blood Phe levels. Results suggest individual differences in natural protein tolerance exist, and show that animal protein intake is associated with more blood Phe fluctuations, while less variable blood Phe levels were observed with vegetable protein consumption. The study concluded maximum

natural protein tolerance should be assessed more regularly as part of routine PKU care, and indicated that a time frame up to 4 weeks is necessary to establish maximum blood Phe levels when challenging with any additional natural protein.

Glycomacropeptide: a potential nutritional strategy for the treatment of obesity and metabolic syndrome

Dr. Ney discussed glycomacropeptide (GMP) nutritional supplements and its potential for the treatment of obesity. Preclinical studies in mice demonstrated that a GMP diet burned fat and built bone compared with casein, especially in PKU and wild-type female mice. This sparked the GMP Hunger study¹³, a dose-finding pilot study assessing efficacy of GMP supplement to treat obesity in postmenopausal women. Results indicated that GMP increased amylin levels (a satiety hormone co-secreted with insulin), improved glucose homeostasis and altered the faecal microbiome. Furthermore, it was highlighted that GMP may support weight loss and maintenance of lean body mass, while protecting bone health. Given the prevalence of obesity in the adults with PKU, further investigation into the anti-obesity effects of GMP supplementation in this population would be of value.

Actual and future application of use for LNAA in PKU

Dr. Ahring discussed the use of LNAAs in PKU. Although usually considered a single treatment modality, studies demonstrated LNAA treatment can be used in two alternative ways. The first is natural protein intake at recommended daily allowance with LNAA supplementation (for those >18 years of age and/or untreated or late-diagnosed PKU patients, if capable of consuming sufficient natural protein). Alternatively, LNAA can be combined with conventional low protein diet with amino acid supplements (for teenagers with poor compliance). LNAA is considered unsuitable for children and maternal PKU women. It was highlighted that in daily practice, transferring from conventional dietary treatment to LNAA treatment requires frequent monitoring (weekly at start) to ensure optimal treatment (including compliance with intake and timing). While pharmaceutical therapies will likely overtake LNAA treatment, it could still be a valid alternative for adults in countries with no access to treatment advancements, although it is not recommended in the PKU guidelines.

Choline intake and working memory

Dr. Singh presented the impact of total choline intake on working memory performance in adults with PKU. Choline is a precursor for metabolites that impact brain development and function, and research indicates choline metabolic pathways

are perturbed in adults and children with PKU. A cross-sectional study¹⁴ evaluating the association between choline intake and working memory performance in adults with PKU indicated that increased choline intake is associated with improved working memory when good metabolic control is maintained. As the overall total choline intake was found to be suboptimal in PKU patients, it was highlighted that fortified medical foods are essential to ensure adequate consumption of choline and the nutrients supporting the choline metabolism for patients on Phe-restricted diets.

Discussion

Dr. MacDonald stressed the importance of continuous research for redesigning infant formula even in the evolving treatment landscape, as diet remains a powerful tool in regions where pharmacological options may not be accessible. She highlighted that formula for infants with PKU should mimic regular formula as close as possible, with lower amounts of Phe and supplemented with probiotics. Dr. Pinto was asked if he would recommend vegetable or animal protein in clinical practice, and in which amount. He applies an individualised approach, and clarified this would depend on the patient's Phe tolerance. In general, he would advise against using animal protein in classic PKU patients. Dr. Ahring was asked about effects of LNAA treatment on other organs (e.g., heart and bones), indicating further research is needed to explore this.

Burden of illness and unmet needs of PKU

People with PKU, particularly those in special situations (e.g., late-diagnosed individuals) experience a substantial disease burden and ongoing unmet management needs. This section explores a broad range of topics related to unmet needs and other important aspects of PKU care in special PKU patient populations.

Sport activities and catabolic state

Dr. Bélanger-Quintana reviewed catabolism and situations leading to a catabolic state. While catabolism may provide an important source of circulating Phe levels, entering a catabolic state (e.g., due to infections, inflammation, allergies, anxiety or exercise) may result in excessive rise of Phe levels and subsequent poor metabolic control. To improve metabolic control, it is important to find and treat the root cause, and provide nutritional guidance on increased caloric intake and hydration. Generally, reducing protein intake is not necessary during the catabolic state, although in some situations, protein restriction is warranted (e.g., infection and allergies). While physical exercise may cause a catabolic state and higher Phe levels, it should not be restricted in PKU, but diet before, during and after exercise should be carefully considered to avoid dehydration and muscle degradation.

Pregnancy

Dr. Longo presented evolving insights on pregnancy in PKU. It was highlighted that PKU does not impact a women's health during pregnancy, but it does affect the foetus. Maternal PKU is recognised to cause intellectual disability and birth defects (e.g., congenital heart diseases, microcephaly, intrauterine and post-natal growth restriction, facial dysmorphism) resulting from elevated Phe levels and insufficient protein/nutrient intake¹⁵. Outcomes of prospective clinical studies, including the maternal PKU collaborative study (MPKUCS), emphasised the importance of establishing Phe levels between 120 and 360 $\mu\text{mol/L}$ before conception, and maintaining them throughout pregnancy, to reduce risks of PKU pregnancies to those of the general population. Lastly, limited data demonstrates that pharmacological therapies (sapropterin and pegvaliase) allow for improved Phe control during pregnancy and good pregnancy outcomes.

Late-diagnosed and untreated PKU

Dr. Sivri discussed untreated (no treatment by age 7) and late-diagnosed (after >1 month of age) PKU patients. These cases still exist due to absence of NBS in some regions, false negatives, technical or medical errors, migration from non-NBS countries, family non-adherence or therapy discontinuation, and mild HPA previously considered benign. Clinical features of this population include cognitive deficits (e.g., intellectual disability), developmental delays (e.g., language impairment), psychiatric and behavioural manifestations (e.g., aggression, autism spectrum disorder/attention deficit hyperactivity disorder (ASD/ADHD)), neurological features (e.g., microcephaly, magnetic resonance imaging (MRI) abnormalities), and somatic features (e.g., skin and hair changes). This introduces a high burden on caregivers and reduces QoL for patients and their families. However, data demonstrated that initiating Phe-reducing treatment later in life may still lead to meaningful improvements in functioning. It was highlighted treatment options and goals should be individualised based on the

patient's status, patient and caregiver preferences, available resources and social circumstances, and that caregiver support is essential for adequate PKU management.

What is the burden of dietary treatment in PKU patients (and how can we help to decrease this)

Dr. Withaar discussed that although both patients and caregivers are generally highly motivated to maintain dietary treatment in the beginning, lifelong adherence to diet imposes psychological challenges and may conflict with normal social emotional development throughout life. Qualitative interviews (n=20) performed by Dr. Withaar identified restricted social participation and subsequent feelings of exclusion, influences on self-perception and identity, and disrupted eating patterns as important hurdles, but interestingly, the interviews also revealed positive outcomes such as adaptation and resilience in PKU patients and their caregivers. Other qualitative studies demonstrated that family status and parental stress, perinatal psycho trauma and comorbid feeding and eating problems in infancy and early childhood further contribute to the diet burden. Proposed strategies to decrease diet burden include 'asking the right questions' (patient reported outcomes & experience measures or PROEM), regular psychological/pedagogical check-ups, education and support of caregivers, and facilitating peer support to share experiences.

Discussion

Dr. MacDonald asked Dr. Withaar's opinion about the European guideline recommendation to include social services in case of uncontrolled Phe levels (>75% of blood Phe levels outside target range over a period of 6 months and other signs of failure of adherence). Dr. Withaar recommended to include psychological support routinely from the start rather than making referrals when problems start. She also highlighted the importance of normalising psychological support in PKU by making caregivers aware problems are likely to occur. When asked whether she would recommend screening for parental stress in the next guideline updates, and what evidence would support such recommendation, Dr. Withaar commented that all new diagnosed PKU patients and their caregivers should receive a standard psychological consult to assess parental stress and trauma from the start, as evidence from studies performed early in life, even beyond PKU, demonstrated this can prevent problems later in life.

Changing landscape: pharmacological options

Beyond dietary treatment, pharmacological options for treatment of PKU have emerged in the last decades. This section explores the mode of action, efficacy, safety, and latest data of current pharmacological therapies in PKU.

Sapropterin

Dr. Ficicioglu discussed sapropterin treatment in PKU. Sapropterin (6R-BH₄) is a synthetic form of BH₄, the natural cofactor for PAH. It can increase PAH affinity for Phe and enhance residual PAH activity in patients whose responsiveness is proven through a BH₄ loading dose test.¹⁶ Multiple clinical trials and long-term registry data demonstrated that sapropterin increases dietary Phe tolerance while maintaining reduced blood Phe levels in those responsive. Furthermore, results indicated improvements in executive functioning and ADHD symptoms in sapropterin-treated patients vs placebo. Data shows sapropterin is well-tolerated in the long-term, including during childhood, pregnancy and across diverse patient populations.¹⁵ Lastly, real-world registries (Phenylketonuria Demographics, Outcomes and Safety Registry (PKUDOS¹⁷) in the US and Kuvan Adult Maternal Paediatric European Registry (KAMPER¹⁸) in Europe) have shown that sapropterin may be effective, demonstrated improvements in QoL and guided best practices for individualised PKU care.

Sepiapterin

Dr. Longo presented sepiapterin, a natural precursor of BH₄ functioning as intermediate in both the synthesis and recycling of BH₄. Sepiapterin has a dual mechanism of action, leading to improved PAH functioning by increasing the cofactor BH₄, as well as an independent pharmacological chaperone effect on PAH. Phase 1 clinical trial data¹⁹ in healthy volunteers indicates that sepiapterin is more stable and crosses cell membranes including the BBB more efficiently than sapropterin, resulting in substantial and rapid increases in intracellular BH₄. Furthermore, data from phase 2 and 3 clinical trials^{20, 21} suggests that sepiapterin causes a greater decrease of Phe levels and is effective in a larger proportion of individuals with PKU (including some with classic PKU) vs sapropterin. Results from the phase 3 APHENITY trial²¹ indicate responsive individuals may reach Phe levels below 360 µmol/L (84% (n=37/44) in the treatment group vs 9% (4/43) in the placebo group), allowing a gradual increase of natural protein intake (although the diet could generally not be fully normalised). Phase 3 data indicated

sepiapterin was associated with mild increased incidence of headaches and gastrointestinal adverse events (AEs).

Pegvaliase

Dr. Sacharow discussed pegvaliase, a subcutaneous pegylated PAH enzyme substitution therapy that converts Phe to ammonia and trans-cinnamic acid.²² Phase 3 clinical trial data²³ demonstrated pegvaliase leads to sustained blood Phe reductions, for many to within the physiological range, and while often consuming a completely unrestricted diet. Beyond blood Phe lowering, data also suggests improved neurological outcomes for e.g., ADHD, cognitive performance and white matter lesions (MRI).²⁴ Pegvaliase has an acceptable safety profile, with hypersensitivity AEs (e.g., injection site reactions) being most common in early treatment phases and declining over time. Interestingly, results from a risk-benefit analysis indicate most patients are willing to accept the risk of hypersensitivity reactions to achieve recommended blood Phe levels²⁴. Pegvaliase may also be successful in

special patient populations, including those with comorbidities, poor social support, executive function deficits and cognitive impairment, and late-treated patients, and preliminary data of pegvaliase use during pregnancy and breastfeeding, and in adolescents, hint towards possibly improved outcomes in these populations.

Discussion

Regarding weight-based dosing of sapropterin in overweight patients. Dr. Ficicioglu commented he uses the actual weight rather than the ideal weight to calculate the dose, while Dr. Longo and Dr. Sacharow use the highest possible dose. Prof. Muntau and Dr. Singh recommended to dose based on adjusted body weight (using lean mass). In light of the phase 1 results of sapropterin (20 mg/kg) vs sepiapterin (20 mg/kg), an audience member noted that recent results with higher doses of sapropterin (40 mg/kg) showed a tendency towards higher efficacy. They asked how higher doses of sapropterin would compare to sepiapterin. Dr. Ficicioglu explained that in his experience, higher doses of sapropterin do not show higher efficacy. Additionally, Dr. Longo noted that the efficacy increase with higher doses of sapropterin is not worthwhile, and the rise in BH4 levels with sepiapterin is more outspoken compared to higher dose sapropterin. Upon discussing pegvaliase's potential to normalise diet and reduce comorbidities, Dr. Longo highlighted the importance of dietary

counselling in these patients to install healthy food habits and avoid extreme weight changes. Regarding restarting patients on pegvaliase after anaphylaxis, Dr. Longo and Dr. Sacharow highlighted that the original titration/induction protocol may be accelerated when rechallenging a patient, and Dr. Longo clarified patients that stopped 2 years restarted successfully. Dr. Harding highlighted the restart approach depends on the timing of the event. In his experience, patients having an anaphylaxis episode after they are fully desensitised to pegvaliase can generally restart at their last dose, while patients experiencing anaphylaxis early in treatment should be rechallenged more carefully. He advised to use premedication (e.g., H1/H2 receptor antagonists and antipyretics) in all cases. Upon discussing the need to install normal Phe levels, Dr. Sacharow highlighted that most of her patients get an epiphany when reaching normal Phe levels as their brain fog disappears and they can consume a normal diet. Dr. Vockley further highlighted the need to convince HCPs, patients, caregivers and families to at least try reaching lower Phe levels, as the benefits may often only be experience upon reaching normal Phe levels.

Future perspectives

This section explores novel treatments in development for PKU and potential other inborn errors of metabolism (IEM), with data highlighting approaches that pave the way for personalised treatments.

Long non-coding RNA (*HULC*) and PKU

Dr. Feillet discussed the role of *HULC*, a long non-coding RNA (lncRNA) in PKU. It was highlighted that PKU is not a monogenic disease, which partly explains the lack of complete phenotype/genotype correlation. A study in mice²⁶ demonstrated lncRNA *HULC* is involved in PAH functioning, by showing human *HULC* mimics restored PAH enzymatic activity. Furthermore, results indicated that the interaction between PAH-*HULC* could explain the severity of some PAH variants (e.g., R408W). Despite promising results, there are no studies describing *HULC* mutations as the sole cause of human PKU to date. Besides one poster at ICIEM 2025, the last publications about PKU and *HULC* date from 2021-2022, highlighting the need for more studies to understand the influence of *HULC* on the phenotype of PKU patients. Treatments with lncRNA mimics has not yet been developed because of very high costs and the presence of current available treatments.

Gene therapy and gene editing

Dr. Harding presented the potential of gene therapy and gene editing in PKU. Gene therapy uses nucleic acids (e.g., cDNA or mRNA) to replace or add a functional copy of a malfunctioning gene, or repair variants that cause disease. The use of adeno-associated vectors (AAV) for gene addition is a commonly used gene therapy strategy, but only one of five initiated trials is currently continuing (NGGT-002 trial²⁷). This is in part due to challenges associated with AAV gene addition, including the unknown long-term durability of gene expression, inconsistent response, severe inflammatory responses and genotoxicity risk. In gene editing, specific intracellular tools (e.g., prime editing, base editing or CRISPR/Cas9) are used to introduce precise changes to the DNA. This strategy is currently not investigated in PKU, but preliminary data and observations from a phase 1/2 first-in-human clinical trial (OTC-HOPE trial²⁸) in newborn males with OTC (Ornithine Transcarbamylase) deficiency suggest a complete clinical response. More research will be needed to demonstrate the potential of these strategies in PKU.

SLC6A19 inhibition facilitates urinary neutral amino acid excretion and lowers plasma phenylalanine

Prof. Muntau discussed inhibition of the SLC6A19 transporter, that reabsorbs Phe, as a potential strategy to lower toxic plasma Phe levels in PKU. JNT-517 is designed as an orally bioavailable inhibitor of human SLC6A19, facilitating urinary Phe excretion. Phase 1/2 data demonstrated rapid, dose-dependent reductions in plasma Phe levels (>50% compared to baseline) in patients receiving JNT-517, including patients with high baseline blood levels, failed previous treatments and those consuming diets with substantial amounts of Phe. The 150 mg twice daily (BID) dose resulted in greater blood Phe reduction vs baseline, with 9/11 patients achieving levels <600 µmol/L, 5/11 patients <360 µmol/L and 2/11 patients <120 µmol/L. JNT-517 administered to PKU individuals seemed to be well-tolerated, and 42.9 – 72.7% of participants experienced any treatment emergent AEs. Despite pronounced aminoaciduria, no clinically significant changes in plasma amino acid levels were observed. A phase 3 clinical trial

(JNT-517-301²⁹) is currently ongoing. Beyond PKU, JNT-517 has the potential to treat other amino acid metabolism disorders, including maple syrup urine disease and urea cycle disorder.

Discussion

Prof. Muntau explained the differences between SLC6A19 inhibitors in development by different companies (i.e., JNT-517 vs MZE782). Although they have the same mechanism of action, they differ chemically. This may explain MZE782 appearing more potent than JNT-517, although more data will be needed to allow accurate comparisons. Regarding a question about modifying SLC6A19 inhibitors to specifically reabsorb Phe, Prof. Muntau clarified current data indicate other amino acids blood levels seem not to be affected, but long-term data is required to confirm this hypothesis. Dr. Harding was asked how he envisions the gene editing field to advance into clinical practice when there are effective oral treatments. He explained that, once safety issues are overcome, gene editing uptake will be country depended, hypothesising that non-US countries with more limited treatment options may benefit from a one-time treatment that could potentially cure PKU. Dr. Cirak commented gene therapy may be particularly interesting for neonates, but Dr. Harding highlighted important issues such as safety and the rapid hepatocyte turnover leading

to loss of vector genomes should be addressed first. Looking towards the future, Dr. Vockley proposed the idea to assess multidrug efficacy for PKU, as distinct drugs have complementary mode of actions. Prof. Muntau added the ideal treatment would have compounds combined into one, orally available drug, showing full efficacy and an acceptable safety profile, with the potential to achieve physiological Phe levels and allowing to consume a normal diet. Nevertheless, the overall recommendation of the chairs was installing personalised pharmacological treatment for every patient.

Industry sponsored sessions: meet the expert

Industry-sponsored symposia and panel discussions brought together experts to explore the evolving landscape of pharmacological PKU treatment. These sessions addressed a wide range of topics, from unmet needs with dietary treatment, to clinical experience with emerging therapeutic options.

Challenging the norm: addressing unmet needs in PKU (Organised by PTC Therapeutics)

Dr. Bélanger-Quintana discussed the results from the phase 3 global multicentre double-blind randomised placebo-controlled APHENITY trial¹⁹ and ongoing open-label extension study. Results indicated significant reductions in blood Phe levels in sepiapterin-treated patients vs placebo, including in BH4 non-responsive patients. Interim data from the open-label extension study show that long-term treatment with sepiapterin allows for increased dietary Phe intake and reduced protein substitute use, while maintaining blood Phe levels <360 µmol/L. Furthermore, interim data from the ongoing phase 3 AMPLIPHY trial suggested sepiapterin allows more participants to reach blood target levels (89% <600 µmol/L and 69% <360 µmol/L) compared to sapropterin (51% <600 µmol/L and 39% <360 µmol/L). Sepiapterin was well-tolerated with no serious treatment emergent AEs in both the APHENITY and AMPLIPHY trials. Most frequently reported AEs included diarrhoea, nausea, headache and upper respiratory tract infections.

Redefining targets in PKU: achieving and sustaining normal Phe and diet (Organised by BioMarin)

Dr. Harding discussed the importance of lowering Phe levels with the aim of achieving and sustaining normal Phe levels and diet. Guideline recommendations currently define target Phe levels below 600 or 360 µmol/L, but data suggests that even at Phe levels >240 µmol/L, subtle physical, neuropsychiatric and behavioural symptoms may persist. Given the numerous challenges with the Phe-restricted diet (e.g., access, social pressure), maintaining dietary Phe restriction without pharmacological treatment is near impossible in the long-term for adults. Emerging evidence suggests long-term AEs of dietary therapy (e.g., micronutrient deficiency) exists, while the long-term consequences of even moderately elevated Phe levels are unknown. Therefore, it was hypothesised that Phe-lowering therapy maintaining blood Phe levels within the physiological range while allowing unrestricted consumption of intact dietary protein will lead to improved outcomes. Currently, pegvaliase is

the only approved therapy with the potential to achieve this goal for most eligible individuals with PKU. It was furthermore highlighted that blood Phe is a suboptimal PKU biomarker. The sustained Phe response (SPR)²³ was introduced as a potential solution to overcome the innate volatility of blood Phe, and to enable better comparisons of Phe levels reductions and protein intake across patients over time.

ABBREVIATIONS

AAV	adeno-associated virus	DBS	dried blood spot	OTC	Ornithine Transcarbamylase
ACMG	American College of Medical Genetics and Genomics	ETPKU	early-treated PKU	PAH	phenylalanine hydroxylase
ADHD	attention deficit/hyperactivity disorder	GMP	glycomacropeptide	Phe	phenylalanine
AE	adverse events	HCP	healthcare professional	PKU	phenylketonuria
APV	allelic phenotype value	HPA	hyperphenylalaninemia	PKUDOS	Phenylketonuria Demographics, Outcomes, and Safety Registry
ASD	Autism spectrum disorder	IEM	inborn error of metabolism	QoL	quality of life
BBB	blood brain barrier	IQ	intelligence quotient	ROC	receiver operating characteristics
BH4	tetrahydrobiopterin	KAMPER	Kuvan Adult Maternal Paediatric European Registry	SE	standard error
BID	twice daily	LAT-1	L-type amino acid transporter 1	SLPFs	special low protein foods
Cas9	CRISPR-associated protein 9	LNAA	large neutral amino acid	Tyr	tyrosine
CRISPR	clustered regularly interspaced short palindromic repeats	lncRNA	long non-coding RNA	Trp	tryptophan
CV	cardiovascular	MPKUCS	maternal PKU collaborative study	US	United States
DACH	Germany, Austria and Switzerland	MRI	magnetic resonance imaging		
		NBS	newborn screening		

REFERENCES

1.

Hillert A et al. Am J Hum Genet 2020;107:234-50

2.

Smith WE et al. Genet Med 2025;27:101289

3.

Van Wegberg AMJ et al. Mol Genet Metab 2025;145:109125

4.

Blasco H et al. JMID Rep 2016;32:69-79

5.

Dos Santos Y et al. JMID Rep 2025;66:e70010

6.

Dobrowolski SF et al. Mol Genet Metab 2016;119:1-7

7.

de Groot MJ et al. Mol Genet Metab 2010;99:S86-9

8.

Trefz KF et al. Orphanet J Rare Dis 2019;14:181

9.

Tanacli R et al. J Am Heart Assoc 2021;10:e020351

10.

Rodrigues C et al. Mol Genet Metab 2025;144:109068

11.

Yilmaz Nas O et al. Front Nutr 2025;31:1507464

12.

Pinto A et al. Nutrients 2022;14:4268

13.

Hansen KE et al. J Nutr 2023;153:1915-29

14.

Schoen MS et al. Orphanet J Rare Dis 2023;18:222

15.

Longo N et al. Genet Med 2019;21:1851-67

16.

Trefz FK et al. J Pediatr 2009;154:700-7

17.

Longo N et al. Mol Genet Metab 2015;114:557-63

18.

Trefz FK et al. JIMD Rep 2015;23:35-4

19.

Smith N et al. Mol Genet Metab 2019;126:406-12

20.

Bratkovic D et al. Metabolism 2022;128:155116

21.

Muntau AC et al. Lancet 2024;404:1333-45

22.

Sarkissian CN et al. Mol Genet Metab 2005;86:S22-6

23.

Harding CO et al. Mol Genet Metab 2024;39:101084

24.

Burlina AP et al. Eur J Neurol 2024;31:e16508

25.

Bhashyam SS et al. Mol Genet Metab 2019;21:100507

26.

Li Y et al. Science 2021;373:662-73

27.

Clinicaltrials.gov: NCT06332807

28.

Clinicaltrials.gov: NCT06255782

29.

Clinicaltrials.gov: NCT06971731

INDICATIONS AND PRESCRIBING INFORMATION PER REGION

Sapropterin

Europe: KUVAN® (sapropterin dihydrochloride) indicated for the treatment of HPA in adults and paediatric patients of all ages with PKU who have been shown to be responsive to such treatment. KUVAN® is also indicated for the treatment of HPA in adults and paediatric patients of all ages with BH4 deficiency who have been shown to be responsive to such treatment. The latest SmPC can be found on the PKU.expert website or click [here](#) for the SmPC.

Canada: Kuvan® (sapropterin dihydrochloride tablets and powder for oral solution) is indicated in conjunction with a Phe-restricted diet for the reduction of blood Phe levels in patients with HPA due to BH4-responsive PKU. The product monograph can be found [here](#). **Japan:** Biopten granules is indicated for the reduction of serum phenylalanine levels in hyperphenylalaninemia (atypical hyperphenylalaninemia) caused by dihydrobiopterin synthase deficiency or dihydropteridine reductase deficiency, and for reduction of serum phenylalanine levels in hyperphenylalaninemia (tetrahydrobiopterin-responsive hyperphenylalaninemia)

caused by tetrahydrobiopterin-responsive phenylalanine hydroxylase deficiency. The prescribing information can be found [here](#).

Brazil: Kuvan is indicated for the treatment of hyperphenylalaninemia (HFA) in adult and pediatric patients from one month of age with phenylketonuria (PKU), who were responsive to this treatment. Kuvan is also indicated for the treatment of hyperphenylalaninemia (HFA) in adult patients, and pediatrics from 4 years of age with tetrahydrobiopterin (BH4) deficiency, which is showed responsiveness to this treatment. The SmPC can be found [here](#).

Pegvaliase

Europe: PALYNZIQ® (pegvaliase) is indicated for the treatment of patients with phenylketonuria (PKU) aged 16 years and older who have inadequate blood phenylalanine control (blood phenylalanine levels greater than 600 µmol/l) despite prior management with available treatment options. The latest SmPC can be found on the PKU.expert website or click [here](#) for the SmPC. **Canada:** Palynziq (pegvaliase injection) is indicated to reduce blood Phe concentrations in

patients with PKU aged 16 years and older who have inadequate blood Phe control (blood Phe levels greater than 600 µmol/L) despite dietary management. The product monograph can be found [here](#). **Japan:** PALYNZIQ (pegvaliase) is indicated for the treatment of adult patients with PKU. The prescribing information can be found [here](#). **Brazil:** PALYNZIQ is indicated for the treatment of patients with phenylketonuria (PKU) from 16 years of age with inadequate control of phenylalanine in the blood [phenylalanine levels in the blood greater than 600 micromol/L (10.0 mg/dL)] with existing treatment.