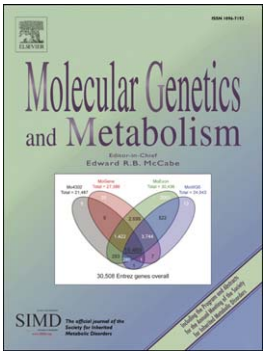

PKU: High plasma phenylalanine concentrations are associated with increased prevalence of mood swings

Karen Anjema, Margreet van Rijn, Paul H. Verkerk, Johannes G.M. Burgerhof, M. Rebecca Heiner-Fokkema, Francjan J. van Spronsen

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Title	**PKU: high plasma phenylalanine concentrations are associated with increased prevalence of mood swings**
Author names and affiliations	Karen Anjema[1], Margreet van Rijn[1], Paul H. Verkerk[2], Johannes G.M. Burgerhof[3], M. Rebecca Heiner-Fokkema[4], Francjan J. van Spronsen[1] [1] Beatrix Children's Hospital, University Medical Centre Groningen, University of Groningen, Section of Metabolic Diseases. [2] TNO Child Health, Leiden. [3] Department of Epidemiology and Statistics, University Medical Center Groningen, University of Groningen. [4] Laboratory of Metabolic Diseases, University Medical Centre Groningen, University of Groningen, The Netherlands
Corresponding author	Francjan J. van Spronsen
Present/permanent address	Francjan J. van Spronsen, MD, PhD Beatrix Children's Hospital, University Medical Centre Groningen, CA80, PO Box 30.001,9700 RB Groningen, The Netherlands. E-mail address: f.j.van.spronsen@bkk.umcg.nl

Abstract

In phenylketonuria, knowledge about the relation between behavior and plasma phenylalanine is scarce. The aim of this study was to determine whether high phenylalanine is associated with disturbed behavior noticed by the patient and or close environment (parents or partners). 48 early treated PKU patients (median age 8.5, range 0-35 years) participated (median phenylalanine concentration in total sample 277 (range 89- 1171) µmol/l; and in patients < 12 years 238 (range 89 -521) µmol/l). After sending blood samples, patients or close environment were interviewed with a standardized questionnaire whether they noticed hyperactivity, annoying behavior, mood swings and introvert or extravert behavior. The interviewer as well as the respondents were blinded with regard to the phenylalanine concentration. Results: Patients reported less deviant behavior compared to close environment. Mood swings were positively associated with phenylalanine concentrations in the total group (P=0.039) and patients <12 years (P=0.042). The relationships between temporary high phenylalanine concentrations and hyperactivity, annoying behavior, introvert and extravert behavior were not statistically significant. Conclusion: there is a positive association between phenylalanine concentrations and mood swings.

Keywords: Phenylketonuria; PKU; Phenylalanine; Behavior; Mood swings; Mood.

Abbreviations: PKU, phenylketonuria; Phe, phenylalanine; IQ, intelligence quotient; UMCG, university medical centre of Groningen; AD(H)D, attention deficit (hyperactivity) disorder.

Introduction

Phenylketonuria (PKU) is an inborn error of metabolism resulting in high levels of phenylalanine (Phe) in blood and brain, which lead to irreversible and reversible damage to the brain. Untreated, this disease causes severe mental retardation, epilepsy, and behavioral problems [1]. Early and continuous treatment has reduced the loss of intelligence enormously. Still, some difference in IQ remains in comparison to the normal population [2-5]. Notwithstanding mild mean deviation in IQ, PKU patients have more school achievement problems [6, 7]. Possibly, these problems in school performance could be attributed to deficits in executive function [8]. Apart from subtle intellectual deficits, behavior problems have been reported frequently in early treated PKU patients. Behavior reported is to be more distractile, impulsive, hyperactive, fluctuating in their work performance, less conscientious and self-reliant, more anxious, unhappy, solitary, and unresponsive [9-11]. Disturbed behavior seems to be more prevalent in those with higher Phe concentrations [9], but research data are scarce. In daily practice patients with PKU and their parents regularly report that high Phe concentrations are affecting behavior. That raises the question if Phe concentrations change behavior. Therefore, we conducted a questionnaire to determine patients' behavior and compared them with the measured Phe concentration. The hypothesis was that high Phe concentrations are related to more disturbed behavior.

Methods

Patients

The medical ethical committee of the university medical center of Groningen (UMCG) concluded that this study did not need judgment by them because the study consisted only of short interviews and use of blood Phe concentrations of samples taken by routine. From all 92 treated patients with PKU treated in the UMCG, those who were early (< 21 days) diagnosed and continuously treated (N=81), were sent a written invitation to participate in the study. When they answered positively, informed consent was asked during the later contact by telephone. Eight patients did not participate. Reasons not to participate were language problems (3), pregnancy (3), “always hyperactive and always low Phe concentrations” (1) and without argumentation (1). Of the 73 patients who did participate nine patients could not be reached by telephone frequently enough during the study period and 14 patients did not send sufficient numbers of blood samples during the study period. Data collected during special circumstances, e.g. fever, a cold, birthday party were excluded from analysis. Therefore, for two additional patients we did not obtain the minimum criterion of participation of three questionnaires. In total, data from 48 patients were collected during November 2007 to November 2008.

Procedure

After sending blood samples for measurement of blood Phe concentrations in blood spot, patients, parents or partners were contacted by telephone. Participants were interviewed with a standardized questionnaire. Both participants (patients, parents or partners), and the interviewer did not know the Phe concentration at the time of the questionnaire. The maximum allowed time between taking the blood sample and the questionnaire was set at 5 days. The behavior questionnaire consisted of five questions: 1. Are you more hyperactive then usual? 2. Are you more annoying, aggressive or less willing to listen to other people? 3.

Do you experience more mood swings? 4. Is your behavior more introvert? (more reserved, less outgoing) 5. Is your behavior more extravert (talk a lot, draw attention)? All questions could be answered by no, a little, yes or a lot. Participants were asked to recall the behavior at the time of the blood draw and to compare this behavior to the usual behavior of the patient. Blood phenylalanine concentrations from dried blood specimens were measured by the AccQ Tag method using high-performance liquid chromatography (Waters BV. Breda, The Netherlands).

Statistical analysis

Since all patients participated a different number of times, the median Phe concentration was calculated from the individual medians. Due to the scarcity of the answers ‘a little’ and ‘a lot’, the answers ‘no’ and ‘a little’, and ‘yes’ and ‘a lot’ were combined. The behavior questions versus the Phe concentrations (as natural logarithm) were analyzed with a linear mixed effects model, a model that takes into account the correlation between observations within the same patient. In this analysis maximum likelihood estimation and the covariance structure compound symmetry was used. The natural logarithm of Phe was used to approach normal distribution. A p-value ≤ 0.05 was considered to be statistically significant. Statistics were performed with PASW statistics 18.0; SPSS, Inc., Chicago, IL, USA.

Results

Data on the median age and Phe concentrations of the participants are shown in *Table 1*. The youngest patient was four months old. The age of the other patients varied from 1.4 to 35.0 years of age. Below 12 years of age (N=31) median Phe concentrations were 238 µmol/l (range 89 – 521 µmol/l). In patients 12 years and older Phe concentrations were 558 µmol/l (range 221 – 1171 µmol/l). Regarding the questions of the survey, 84% of the answers were given by parents (for < 12 years 100%, and ≥12 years 39%), so questionnaires of younger patients were more often answered by the patient's close environment than in older patients. In patients ≥12 years, 61% of the answers were self-reports and only 1% was given by a partner. All questionnaires of one patient were answered by the same person in 38 cases, in the ten remaining cases this was done by two different persons. The mean period between blood sampling and the questionnaire was 2.8 days ± 0.8 (min 1 – max 5 days).

Behavior

The frequencies of deviant behavior (questions answered with yes or a lot) are shown in *Table 2*. Patients reported less deviant behavior compared to parents or partners (close environment). Of 432 questionnaires answered by the close environment, deviant behavior was reported 81 times (18.8%), whereas patients reported deviant behavior in seven of 83 questionnaires (8.4%). Deviant behavior was reported in 3 of 34 questionnaires in the four months old patient. Question three (Do you experience more mood swings?) was significantly associated with the natural logarithm of Phe in the total group according to the linear mixed effects model (Estimate 0.28 P=0.039, age 0.05 P=0.000 and intercept 5.52) and in patients < 12 years (Estimate 0.31 P=0.042, intercept 5.74).

Discussion

This paper adds to the scarce information on the relationship between Phe and behavior, given by papers of Smith [9] and Ten Hoedt [12]. We found that some behavioral aspects, especially mood swings, are related to Phe concentrations in PKU.

Since it was not possible to reach participants by telephone after every blood sample, numbers of participation do not reflect the actual adherence of the patients. The relatively small number of participants (N=48) could be explained by the relatively short study period of one year in which we required a minimum of three participations, the delay in the response that patients were willing to participate and not being able to reach often enough. The questionnaire was designed with four possible answers. In reality the answers 'a little' and 'a lot' were hardly used. Therefore, for analysis, answers were combined.

Behavior

This study showed that more mood swings are seen in combination with high Phe concentrations. In our study the participating patients reported less disturbing behavior in comparison to the patients' close environment (parents, partners). These results might indicate that the patients' close environment is better able to notice behavioral changes when blood Phe concentrations are high. Another possible explanation could be that patients are less willing to admit disturbing behavior, which results in underreporting. Furthermore it should be taken into account that age could also be a confounder. Unfortunately, data on patients ≥ 12 years could not be analyzed separately since this group was too small. Most data are of children of 2.7 to 12.3 years of age. The data of one child, who started the study at the age of four months, were also included. The parents of this child reported deviant behavior a number of times, showing that the parents were able to observe differences in behavior.

The presence of mood swings during high Phe concentrations has not been described before. Mood swings can be considered as a derivative of irritability, which has been described mainly in untreated PKU [13].

Annoying or aggressive behavior or being less willing to listen has not been reported more in PKU patients [9]. Also in this study there was no association with high Phe concentrations found in patients < 12 years. In patients ≥ 12 years annoying behavior was never reported.

In contrast to Stemerdink et al, we did not find an association between introvert behavior and Phe concentration [14]. Stemerdink et al showed a relationship between introvert behavior and high Phe concentrations, but only in the first two years of life [14].

The present study also showed no evidence that high Phe concentrations lead to hyperactive behavior. This finding did not correlate with most parents' ideas, expecting of a relation between high Phe concentration and hyperactive behavior. Literature with regard to ADHD and PKU showed the same absence of hyperactivity. A study by Antshel et al showed mainly inattentive symptoms associated with ADD [15]. In this study we did not focus on inattentive symptoms.

Recently, ten Hoedt et al also showed that elevated Phe levels are associated with a negative effect on the mood of patients in a double-blind cross-over trial [12]. In the study of ten Hoedt et al, less favorable mood was reported by the patient as well as a friend or relative. It is possible that patients in this trial were willing to admit deviant behavior despite desirable social behavior, because the high Phe concentrations were raised artificially.

A strong point in this research is the blinding of the Phe concentrations for the respondents as well as the interviewer, but there are some possible deficits in this study. The results are based on a questionnaire, giving a risk of over- or under reporting of the behavioral issues. However, by asking these questions by routine, the risk of giving socially accepted responses by patients or their close environment is decreased, so that the answers are considered to

reflect the actual opinion of the respondents. Another inevitable problem of the study was the time gap between blood sampling and the telephone call. In most cases this time gap was three days at maximum, data on a period longer than five days were excluded. This time gap between blood sampling and questioning may have influenced the answers and in consequence may have had an effect on the reliability of the data.,In this study design patients were contacted after the arrival of their blood sample at the laboratory.

Since both the interviewer and the respondents were blinded with regard to the Phe concentration these two forms of bias are likely to lead to non differential misclassification. Non differential misclassification will lead to a bias to the null value of effect estimates. To exclude the possibility that the data on behavior do refer to the day of calling rather than the day of blood sampling, future questionnaires could try to contact at the day of blood sampling.

In conclusion, we found a positive association between Phe concentrations and mood swings. We did not find more annoying behavior, introvert and extravert behavior, and hyperactive behavior with high Phe concentrations. These observations partly confirm our hypothesis that high Phe concentrations are related to more disturbed behavior, and provide us with information to help guide patients and parents in the most optimal way.

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Table 1 Clinical data of the studied PKU patients (N=48)

	Total N	Median	Min-Max
Gender			
- *Female*	26		
- *Male*	22		
Age (years)		8.5	0 - 35
Number of Phe measurements			
(and behavior questionnaires)			
- *Total Group*	515	8.0	3 – 37
- *< 12 years*	380	12.0	4 – 34
- *≥ 12 years*	135	4.0	3 – 37
Phe-level (µmol / l)			
- *Total Group*		277	89 – 1171
- *< 12 years*		238	89 – 521
- *≥ 12 years*		558	221 - 1171
Participant			
- *Parent*	431 (83.7%)		
- *Patient*	83 (16.1%)		
- *Partner*	1 (0.2%)		

Table 2 Numbers of deviant behavior (yes & a lot)

	Whole group (515)	**< 12 years** (380)	**≥ 12 years** (135)	**Environment** (432)	**Patient** (83)
1. Are you more hyperactive then usual?	42	40	2	41	1
2. Are you more annoying, aggressive or less willing to listen to other people?	30	30	0	30	0
3. Do you experience more mood swings?	33	30	3	30	3
4. Is your behavior more introvert?	16	14	2	14	2
5. Is your behavior more extravert?	41	40	1	40	1