

## SHORT REPORT

### Tetrahydrobiopterin responsiveness in a large series of phenylketonuria patients

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**Summary:** In a group of 87 consecutive patients with hyperphenylalaninaemia born since 1990, only 3 patients showed a (temporary) decrease of serum phenylalanine levels after tetrahydrobiopterin (BH<sub>4</sub>) loading in usual doses (20 mg/kg body weight).

Kure and colleagues (1999) reported the first identification of a tetrahydrobiopterin (BH<sub>4</sub>)-responsive phenylalanine hydroxylase (PAH) deficiency in four patients with a positive PKU screening result (phenylalanine >120 µmol/L). Most recently, Lindner and colleagues (2001) described three patients with PAH deficiency and the same genotypes but different responses to standardized BH<sub>4</sub> loading. They concluded that BH<sub>4</sub> responsiveness in PAH deficiency is at least partly independent of PAH genotype. Trefz and colleagues (2001) reported a successful treatment with BH<sub>4</sub> (10 mg/kg body weight) in one patient with mild PKU and concluded that BH<sub>4</sub> supplementation instead of a phenylalanine-restricted diet might be possible in at least some patients with classical or mild PKU. The prevalence of BH<sub>4</sub> responsiveness in PAH deficiency is still unclear. We therefore retrospectively evaluated results of the BH<sub>4</sub> loading test in the newborn period of our 87 patients born between 1990 and 2001.

The loading test was routinely performed in all patients with a positive newborn screening (phenylalanine >120 µmol/L) with 20 mg/kg body weight BH<sub>4</sub> (Dr Schircks Laboratories, Jona, Switzerland), continuing a normal protein diet. Serum phenylalanine levels were measured by HPLC before and 4 h and 8 h after BH<sub>4</sub> intake. A BH<sub>4</sub> coenzyme deficiency was excluded in all patients (normal values for neopterin and biopterin in urine, normal dihydropterin reductase activity in red blood cells, measured by N. Blau, Zürich, Switzerland). A 50% decrease of the serum phenylalanine level after BH<sub>4</sub> intake was defined as a significant decrease as in all reported BH<sub>4</sub>-responsive patients after BH<sub>4</sub> supplementation. Pre-loading serum phenylalanine (Phe) levels were within a wide range (132–3036 µmol/L). Three of the 87 patients (3.5%) showed a significant decrease of serum phenylalanine concentrations 8 h after BH<sub>4</sub> intake (84 patients, 96.5% did not). These three patients will be described in more detail.

*Patient 1:* A boy, born 11/99, normal pregnancy and birth. Phe levels: 678  $\mu\text{mol/L}$  in the screening test; 864  $\mu\text{mol/L}$  before, 684  $\mu\text{mol/L}$  4 h and 252  $\mu\text{mol/L}$  8 h after  $\text{BH}_4$  loading. On free nutrition supplemented with 10 mg/kg body weight  $\text{BH}_4$  per day Phe levels increased within 6 weeks to 972  $\mu\text{mol/L}$ .  $\text{BH}_4$  treatment was stopped and a Phe-restricted diet was started. Genotype: IVS3–22G>A/Y414C.

*Patient 2:* A boy, born 11/00, normal pregnancy and birth. Phe levels: 360  $\mu\text{mol/L}$  in the screening test; 396  $\mu\text{mol/L}$  before, 192  $\mu\text{mol/L}$  4 h, and 90  $\mu\text{mol/L}$  8 h after  $\text{BH}_4$  loading. On free nutrition (without  $\text{BH}_4$  Phe levels increased to 642  $\mu\text{mol/L}$ . On free nutrition supplemented with 10 mg/kg body weight  $\text{BH}_4$  per day Phe levels were stable between 540 and 660  $\mu\text{mol/L}$ .  $\text{BH}_4$  treatment was stopped and a protein-restricted diet was started. DNA for genotyping not available.

*Patient 3:* A girl, born 12/00, normal pregnancy and birth. Phe levels: 978  $\mu\text{mol/L}$  in the screening test; 1032  $\mu\text{mol/L}$  before, 702  $\mu\text{mol/L}$  4 h and 432  $\mu\text{mol/L}$  8 h after  $\text{BH}_4$  loading. On free nutrition supplemented with 10 mg/kg body weight  $\text{BH}_4$  per day Phe levels persisted around 600  $\mu\text{mol/L}$ . Continuing  $\text{BH}_4$  treatment, the initiation of a protein-restricted diet (450 mg Phe/day) was necessary to achieve satisfactory metabolic control. Under this treatment Phe levels were measured in a range of 300–420  $\mu\text{mol/L}$ . A protein-restricted diet without  $\text{BH}_4$  will be tested. Genotype: Y414C/A104D.

Recently, an alternative treatment of at least a subgroup of patients with a PAH deficiency with  $\text{BH}_4$  supplementation has been discussed (Kure et al 1999; Trefz et al 2001; Lindner et al 2001). In our group of 87 consecutive patients born since 1990, only 3 patients showed a significant decrease of Phe levels after  $\text{BH}_4$  loading. In none of the patients did  $\text{BH}_4$  treatment prove to be an effective therapy.

In conclusion,  $\text{BH}_4$  supplementation in usual doses might be an alternative treatment in rare cases of PAH deficiency. However, even a positive  $\text{BH}_4$  loading test in the newborn period does not suggest  $\text{BH}_4$ -responsive phenylketonuria in the long term. It is still unclear why phenylalanine levels were rising in our initial  $\text{BH}_4$ -responsive patients in spite of ongoing  $\text{BH}_4$  treatment.

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#### REFERENCES

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