
Short report

An animal model for maternal phenylketonuria

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Persistent maternal hyperphenylalaninaemia during pregnancy is harmful to the fetus. Recently, an international collaborative survey of 69 children born to women with phenylketonuria (PKU) showed that cardiac anomalies, intrauterine growth retardation, microcephaly, and mental retardation are common sequelae in the surviving obligate heterozygous offspring.¹ While the fetal outcome is apparently related to the degree of maternal hyperphenylalaninaemia during pregnancy, the question of whether other still unknown factors are also involved in PKU embryopathy remains open to debate.

As a first step towards addressing this issue, we cultured 22 rat embryos for 48 hours² in sera of 10 patients with either typical PKU or variants on a normal diet (plasma phenylalanine: 0.4-1.6 mmol/l, normal (1 SD) = 0.05 (0.001) mmol/l) and in control human sera. Crown-rump length, head length, yolk sac diameter, and a developmental score taking into account the main parameters of organogenesis were determined. Gross examination was completed by histological serial sections in some specimens.

All embryos but one had malformations and each malformed embryo had several anomalies (figure). Severe neural tube cell necrosis, small telencephalic vesicles, expanded fourth ventricles, and hypoplastic branchial arches were the most frequent features (table). Eye development was also markedly abnormal with severely enlarged optic pedicles, rounded optic vesicles, and absence or extreme underdevelopment of the lens placode. As a consequence, the



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(Above) Rat embryo cultured for 48 hours in PKU human serum. It shows the main malformations described in the table. (Below) Control rat embryo cultured for 48 hours in normal human serum.

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Incidence of anomalies observed in 21 embryos.

	Dead	Developmental score	No of somites	Malformed embryos
Controls (n=26)	4	49.76 (SD 2.32)	32.95 (SD 3.38)	1
PKU (n=22)	1	45.80 (SD 5.04)*	29.95 (SD 4.18)†	21

*p < 0.01, Student's *t* test.†p < 0.05, Student's *t* test.

	No	%
Hypoplastic branchial arches	16	76.2
Small telencephalic vesicles	13	61.9
Fourth ventricle enlarged and swollen	12	57.1
Underdeveloped olfactory pits	12	57.1
Anomalies of body curvature	12	57.1
Short tails without visible segmentation	12	57.1
Anomalies of somites	11	52.4
Large optic vesicles with underdeveloped or absent lens	10	47.6
Hypoplastic limb buds	10	47.6
Hypoplastic maxillae	9	42.9
Neural tube defect	1	4.8

developmental score was significantly reduced ($p < 0.01$).

Our results were compared to those obtained by others. Hamers *et al*³ cultured rat embryos in rat serum containing either phenylalanine or one of the PKU related metabolites (phenylpyruvic acid, phenyllactic acid, 2-OH phenylacetic acid, or phenylacetic acid). The last two metabolites induced dose related embryotoxicity above 0.3 mg/ml.³ Similarly, Denno and Sadler⁴ cultured mouse embryos in rat serum containing either phenylalanine or related metabolites (phenylethylamine, phenylpyruvic acid, phenylacetic acid, 2-OH phenylacetic acid, or phenyllactic acid). They found dose related abnormalities, especially neural tube defects, with all these compounds, phenylethylamine and 2-OH phenylacetic acid being the most toxic. However, it must be emphasised that the concentrations of the metabolites added to rat serum were much higher than those observed in the serum of PKU patients.

It is too early to establish whether a close dose-effect relationship exists between the level of phenylalanine (and/or its metabolites) in human

plasma and the number or severity of malformations in our experimental model, as shown in clinics by the international collaborative study of the MRC:DHSS Phenylketonuria Register.¹ While care of a woman with hyperphenylalaninaemia during her fertile years is important, the present model might contribute to a better understanding and prevention of PKU embryopathy.

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- 1 Drogari E, Smith I, Beasley M, Lloyd JK. Timing of strict diet in relation to fetal damage in maternal phenylketonuria. *Lancet* 1987;ii:927-30.
- 2 New DAT. Whole-embryo culture and the study of mammalian embryos during organogenesis. *Biol Rev* 1978;53:81-122.
- 3 Hamers AEP, Piersma AH, Verhoef A, Jansen EJHM, Peters PWJ. Embryotoxicity of phenylketonuria (PKU)-related metabolites of phenylalanine (phe) in rat whole embryo culture. *Teratology* 1989;40:284.
- 4 Denno KM, Sadler TW. Phenylalanine and its metabolites induce embryopathies in mouse embryos in culture. *Teratology* 1990;42:565-70.