

Perinatal Pharmacotherapy and the Risk of Late Neuro-Immuno-Behavioural Deviations

Olga Benešová

Prague Psychiatric Center, Prague, Czech Republic

Keywords: Perinatal pharmacotherapy; Teratogenetic risk

Abstract: Adverse environmental conditions, both biological and social, during gestation and postnatal development may interfere with gene controlled, time and space determined programme of ontogenic processes and induce irreparable defects. These epigenetic variables also include drugs which are prescribed in ever increasing amounts in late pregnancy, during labour and neonatal care to save the lives of high-risk fetuses/neonates. Drug administration in the perinatal period which is characterized by intensive cytodifferentiation and receptor formation in the brain and immune system, may initiate disorders on a cellular/subcellular level and give rise to functional defects which become apparent during later maturation or even in adulthood as various deviations of neuro-psychoimmunocompetence. This functional teratogenic risk is especially high in drugs with receptor-mediated effects (psychotropics, hormones) since the majority of receptors necessary for their action develop in the perinatal period. The functional teratogenic potential of three perinatally applied drugs (dexamethazone, phenoterol, diazepam) was evaluated using model experiments in rats and compared with the outcome of clinical pilot follow-up studies.

Introduction

Every individual – both human and animal – represents an entity with a special pattern of neuro-immuno-behavioural reactivity which reflects the interaction between the genetic equipment and the modulating impact of environmental variables operating in the course of the life. These epigenetic factors are extremely important at early ontogenic stages (embryonic, fetal, neonatal), in the period of the fastest development and highest plasticity of the organism. Adverse environmental conditions (maternal metabolic disorders, infection, radiation, xenobiotics including drugs) may interfere with the gene controlled, time and space determined programme of ontogenic processes (which are of a “once-and-for-all” nature) and thus induce irreparable defects. Recent success of perinatal medicine in protecting high-risk pregnancies and in saving the lives of very immature and injured fe-

Correspondence to: Prof. MUDr Olga Benešová, DrSc, Prague Psychiatric Center, Ústavní 91, 18103 Prague 8 – Bohnice, Czech Republic

tuses/newborns is connected with a substantial increase of drugs used in obstetrics and neonatology. However, minimal attention has been paid so far to the potential teratogenic risk of these drugs for the developing fetal brain and immune system. After all, why care when all drug information leaflets and even pharmacology textbooks warn of teratogenicity risk only in case of drug administration during the first trimester of the gravidity.

Drug Teratogenicity

The teratogenic effect of a drug – or of a chemical compound in general – depends not only on the quality and quantity of the noxious agent, but primarily on the developmental stage of the organism at the time of the interference. In the embryonic and early fetal period in which organogenesis takes place and organs are differentiated, drug interference results in various gross or minor structural deformities, e.g. focomely, known from the history of thalidomide, cleft palate, heart defects etc. which were studied by the classical structural teratology.

The perinatal period is characterized by intensive histogenesis and cytodifferentiation of already shaped organs. This concerns especially the brain and immune system which have similar receptortransmitter equipment as well as memory function. This similarity and close functional relationship inspired the psychoneuroimmunologists (Ballieux and Heijnen 1987) to the concept of the immune system as the “mobile brain”.

Even fine deviations induced by drugs in the programme of these developmental processes initiate disorders in neural network, cytoarchitectonics and receptortransmitter communication systems. These alterations on the cellular/subcellular level are not evident at birth, but form the basis for various functional defects of the brain, immune and endocrine systems (functional teratology). Disturbances of the brain function appear gradually during further maturation in school age, adolescence or even in adulthood as various neuro-psycho-behavioural deviations such as hyperkinetic syndrome, dyslexia, dyspraxia, mental retardation, social maladaptation or psychic liability which may represent the predisposition to psychoses. Late functional defects may concern also immune system and endocrine regulations which are controlled by hypothalamic centers.

It is thus evident that the risk of teratogenicity is not limited to the first three months of pregnancy. This still prevalent opinion of the medical public arises from the traditional concept of teratogenic defects as structural malformations. On the contrary, the risk of drug teratogenic sequellae exists during the entire gestation and lasts even throughout the postnatal development, only the manifestation of the teratogenic outcome is changing (Benešová 1989, 1995).

In this connection, it is necessary to note that in contrast to the traditionally emphasized caution during the first trimester of gravidity when considering drug teratogenicity risk, there exists experimental evidence that some drugs may have a quite opposite trend of teratogenic hazard in the course of gestation (Jelínek et al. 1983). The sensitivity of the organism to the teratogenic effect of drugs with general cytotoxic effects is highest during the first stages of development, in the period of rapid growth and primary differentiation with extremely high rate of cell proliferation. On the contrary, the teratogenic potential of drugs with receptor-mediated

effects (psychotropic drugs, hormones) may be negligible during early stages of gestation when relevant receptors, necessary for their action, are not yet present. However, it increases very quickly with advancing development along with the formation of relevant receptors. This refers especially to the perinatal period in which the majority of receptor systems are developing and, at the same time, formed under the impact of external stimuli. Some authors (Csaba 1986) speak even about the period of “receptor imprinting” analogically to the known phenomenon of “behavioural imprinting” (Broadhurst 1963) described in monkey, goose and other animals. Consequently, drugs with receptor-mediated effects which interfere with these processes have high teratogenic effect (it means functional teratogenic risk) when given during late pregnancy or early neonatal life.

Considering the social relevance of various congenital defects, functional deviations, especially when concerning brain and mental efficiency, may represent sometimes a heavier burden for the individual, the family and the whole society than several congenital organ malformations which can be surgically repaired as e.g. cleft palate and heart defects.

Clinical Studies of Drug Functional Teratogenicity

Clinical research in the field of drug functional teratogenicity meets with great difficulties. The long time-interval (up to decades) which exists between the administration of the drug in perinatal period and the manifestation of functional aberrations in school age, puberty or even in adulthood hampers the identification of causal relations. In addition, the final expression of the functional defect may be modified by the impact of various interfering variables and social environment.

Nevertheless, there exist several clinically controlled studies with long-term follow-up of children whose early development was influenced by drug treatment and who revealed various late neuropsychobehavioural deviations without any somatic defects (Table 1). In all these trials, the treated group was compared with a parallel monitored control group matched for gestational age and weight at birth, sex, family status etc.

Diethylstilboestrol (DES) is a synthetic non-steroid estrogen which evades all placental barriers for physiologically circulating hormones and reaches the same concentration in the fetus as in maternal blood. It was prescribed in the period 1945–1971 to pregnant women beginning from the 3rd month of gravidity as a preventive measure in cases of habitual miscarriages with the total number of 4 million treated women in USA. It was only after 26 years of this treatment that DES was withdrawn from the market due to cervical adenocarcinoma (Herbst et al. 1971) which was diagnosed during puberty in adolescent daughters of DES-treated women (transplacental carcinogenesis). However, there was a much higher proportion of DES-exposed offspring, both males and females, who revealed in adulthood functional deviations (disorders of gender-related behaviour, psychic aberrations, reproduction deficits) without any somatic defect (Ehrhart et al. 1985; Vessey et al. 1983).

The history of DES demonstrates the main hazards of drug functional teratogenicity: long time period (in this case 3 decades) necessary to identify the causal relationship between prenatal drug administration and functional deviations in pu-

Table 1. Survey of drug induced functional teratogenic deviations found in clinical follow-up studies.

DRUG	INDICATION	STUDY	COHORT			DEVIATION
			AGE years	N treated	N control	
DIETHYLSTILBOESTROL	pregnancy disorders	Ehrhart et al. 1985	17 - 30	30	30	bi- or homosexuality depression, anxiety, reproductive defects
		Vessey et al. 1983	24 - 30	259	270	
PROGESTIN	pregnancy disorders	Reinisch 1981	6 - 18	25	20	physical aggressivity social maladjustment
GLUCOCORTICIDS	neonatal respiratory distress syndrome	Dobos et al. 1983	6	135	100	minimal brain dysfunction immunodeficiency
		Gunn et al. 1981	5	7	6	
CLONIDIN	hypertension	Huisjes et al. 1986	3 - 9	22	22	sleep disorders, night terror, somnambulism
ALFA-METHYLDOPA	hypertension	Ounsted et al. 1980	4	12	10	sleep disorders, myoclonic jerks
RITODRINE	preterm uterine contractions tocolytic drug	Hadders-Algra et al. 1986	6	76	154	lower No of „best pupils“

berty and adulthood and, consequently, high numbers of individuals endangered by the treatment with the drug before its withdrawal from the market. This situation contrasts with the condition of classical morphological birth defects which provide – owing to the early detection after delivery – the chance of easier identification of the causative agent and, consequently, a prompter elimination of the relevant drug from therapeutic use. This fact may be illustrated by the tragedy of neonatal focomely induced by maternal consumption of the hypnotic thalidomid (Contergan). In this case, the drug was withdrawn after 5 years of use (1956–1961) and the number of malformed children was estimated at about 10–15 thousand (Schardein 1985). The history of DES is the example of an inconsiderate prescription of a drug for longterm preventive administration to pregnant women, a drug which finally appeared to be ineffective in sustaining pregnancy. Nevertheless, it caused harm to many individuals.

The administration of another synthetic hormone, progesterin, to pregnant women induced increased physical aggressivity and social maladjustment in their sons and daughters when tested at the age of 6–18 years in comparison to the control offspring of the same mothers (Reinisch 1981).

Glucocorticoids are administered to pregnant women in the condition of imminent preterm labour or to premature newborns for the prevention/therapy of neonatal respiratory distress syndrome because they stimulate the surfactant production in the immature fetal lung. Dobos and coll. (1983) carried out a prospective controlled clinical study with the cohort of 235 newborns with a follow-up until the age of 6 years, and found a significantly higher incidence of minimal brain dysfunction (defects of fine motor coordination, concentration deficit, decreased social adaptation) in children treated neonatally with prednisolon, in comparison to controls matched for gestation age, birth weight and social position of the family. In a rather small cohort of children (N = 13), treated neonatally by another glucocorticoid-hydrocortizone (Gunn et al. 1981), signs of immunological deficit were diagnosed at the age of 5 years (frequent infections, decreased number of T-cells).

Functional teratogenic deviations induced by the treatment of hypertension during pregnancy with alfa-noradrenergic blockators clonidin (Huisjes 1988) and alfa-methyl dopa (Ounsted et al. 1980) concern various sleep disorders (night terror, somnambulism, myoclonic jerks). It is necessary to say that these drugs induced sleep disorders also in animal model experiments using prenatal drug administration and EEG record in adult rats with implanted electrodes (Mirmiran et al. 1983; deBoer et al. 1989).

Ritodrine is a beta-adrenomimetic drug used as tocolytic i.e. for disruption of preterm uterine contractions in cases of imminent parturition. Although this study was performed with a sufficiently large cohort (N = 234) the difference between treated and control children at the age of 6 years was negligible, only the the number of “best pupils” was lower in the ritodrine group (Hadders-Algra et al. 1986).

The above mentioned clinical studies confirm on a human level the existence of drug functional teratogenic risk reported previously only in animal experiments and indicate also the diversity of functional teratogenic deviations. It is worth mentioning that all drugs listed in the table are agents with a receptor-mediated effect.

Experimental Research of Drug Functional Teratogenicity

It is evident that – to ensure the safety of perinatal pharmacotherapy – it is necessary to study drug functional teratogenicity in animal model experiments in which all variables can be controlled, pathogenetic processes studied even on cellular and molecular levels and functional deviations monitored during the whole life. This approach enables a comprehensive, quantitative and qualitative estimate of the risk of a drug or of drug combinations. An additional advantage is the fact that it is possible – due to the short lifespan of an experimental animal – to acquire the necessary information within a reasonable time. These experimental data give then useful cues for the strategy of clinical follow-up studies which are longterm and hampered by many interfering factors.

The validity of the results from animal experiments for the extrapolation on the human level depends on the adequacy and consistency of the animal model with the clinical condition. The common approach to models in teratology is based on the assumption that drug administration to pregnant woman is simulated by drug administration in pregnant female animals. This approach, however, disregards the species specific differences in the timing of birth during ontogeny. The vulnerable phase of brain intensive histogenesis and cytodifferentiation which occurs in the human perinatally (i.e. it begins in the third trimester of gravidity, culminates just before term and continues during the first two years of life) is shifted to the first postnatal decade in the rat and rabbit whose young are born at a lower stage of maturity (Dobbing 1970).

Hence it follows, that human perinatal drug treatment should be simulated in the rat – which is used in most functional teratogenic experiments – by relevantly timed postnatal administration of the drug. This approach has also a technical advantage: the drug is injected directly into the target organism with elimination of all interfering variables of maternal body and placenta. In addition, the suckling rat pup may serve as a useful model of the premature neonate as both coincide in the stage of brain development and extrauterine existence.

In our laboratory, the following experimental design is used for testing functional teratogenicity of perinatally applied drugs (Benešová et al. 1984). The tested drug is administered to rats in the first postnatal week as a single or repeated subcutaneous injection. One half of the pups in the litter are injected with the drug, the other half with saline or solvent and serve as controls. The animals are followed-up during development, sexual maturation, adulthood and senescence till the age of 2 years, using tests of behaviour, reproductive ability, immunoreactivity and brain biochemical analysis. The risk of functional teratogenicity was investigated in three drugs with receptor-mediated effects which are used for the treatment of various risk situations in the perinatal period (Table 2). Dexamethazone, synthetic glucocorticoid, is applied for inducing surfactant production in the immature fetal lung and thus in prevention/treatment of the neonatal respiratory distress syndrome. Phenoterol, beta-adrenomimetic agent, is used as a tocolytic drug for disruption of preterm uterine contractions. Diazepam, benzodiazepine anxiolytic, is administered for its tranquilizing and myorelaxing activity to pregnant women in the situation of imminent preterm parturition or in resuscitated neonates when intubated. The comparison of the therapeutic dosage and the effective experimental

Table 2. Evaluation of functional teratogenic effects of dexamethazone, phenoterol and diazepam in model experiments with rats. Clinical indication and therapeutic dose range of tested drugs is indicated in the second column, experimental dosage in neonate rats in the third column. The threshold dose for inducing functional teratogenic deviations is printed in bold letters.

Drug	Clinical use	Experimental dosage in neonatal rat	Functional teratogenic effects
DEXAMETHAZONE	prevention or therapy of the respiratory distress syndrome in preterm neonates 0.6 - 1 mg/kg	0.1 - 0.2 - 0.5 - 1 mg/kg s.c. postnatal day 7	<p><i>Behavioural deviations:</i> Hyperactivity with stereotypy, decreased adaptability, high emotional reactivity which induced lower scores in avoidance learning, deficit of motor coordination <i>Defects of endocrine regulation:</i> Deficits of male and female reproductive ability <i>Neurobiological deviations:</i> Permanent deficit of cerebellum weight Reduced size of cells in cerebellum and hippocampus Decreased concentration of noradrenaline in hypothalamus <i>Deficits of immunocompetence:</i> Depression of humoral and cell-mediated immune response REF: Benešová O., Pavlík A.; <i>Neuropharmacology</i> 28: 89-97, 1989.</p>
PHENOTEROL (Partussisten)	tocolytic drug disruption of preterm uterine contractions 0.5 - 1 mg/kg	1 - 10 mg/kg s.c. postnatal days 5-7	<p>Slight deficit of passive avoidance acquisition joined with increased lipid peroxidation in the cortex of 1-year-old rats REF: Tejkalová H., Benešová O. et al.; <i>Homeostasis</i> 33: 89-93, 1991.</p>
DIAZEPAM	tranquilizer & myorelaxans a) imminent preterm labour b) intubation of resuscitated preterm neonates 0.6 - 1 mg/kg	10 mg/kg s.c. postnatal day 7 5 mg/kg s.c. postnatal days 5-6	<p><i>Behavioural deviations:</i> Increased emotional reactivity, defects in novelty reaction <i>Defects of endocrine regulation:</i> Minor deviations in reproductive functions of females (irregular oestrous cycles) <i>Neurobiological deviations:</i> Decrease of serotonergic transmission in the hypothalamus of 1-year-old rats Slight transitional changes of dopaminergic transmission in the striatum <i>Deficits of immunocompetence:</i> Depression of humoral and cell-mediated immune response REF: Benešová O. et al.; <i>Ann.N.Y.Acad.Sci.</i> 717: 89-101, 1994 Dostál M., Benešová O. et al.; <i>Reproductive Toxicol.</i> 9: 115-121, 1995.</p>

dose inducing functional teratogenic sequellae may serve as a quantitative indicator of the drug teratogenic risk.

From this point of view, dexamethazone reveals the highest risk of functional teratogenicity: single administration of a 5 times lower dose than clinical dosage induced serious functional teratogenic defects in behaviour, motor performance, reproduction and immunocompetence joined with neurobiological deviations in the brain. These experimental findings are in agreement with the picture of glucocorticoid induced deviations reported in clinical studies (see Table 1). The teratogenic potential of phenoterol is significantly lower since only 10 times higher dose than clinical dosage had some minor negative impact. Phenoterol belongs to the same group of drugs as ritodrine which was found to have minimal late consequences in clinical follow-up (see Table 1). Consequently, it seems that beta-adrenomimetic drugs are not as dangerous as glucocorticoids. The degree of functional teratogenic risk of diazepam lies somewhere between the previous drugs since it induced functional teratogenic sequellae following experimental doses relevant to maximal therapeutic dosage used in perinatal treatment. However, the finding of significantly depressed immunoreactivity which persist for the entire life has to be regarded as a serious risk when using this drug.

These results have shown the possibility to evaluate and compare the functional teratogenic risk of various drugs in animal model experiments with sufficient validity of data for extrapolation on the human level.

Discussion and Conclusions

The period of early human development is the phase of highest plasticity and sensitivity of the organism to environmental impact, concerning both social and biological factors. The presented study is concentrated on the problems of biological modulation of developmental processes with special regard to the potential hazard of drugs used in pregnancy. In most studies in this field, attention was paid especially to addictive compounds taken without medical control by pregnant women. In contrast to this point of view, we were interested in drugs prescribed by doctors to treat or prevent various risk situations during late pregnancy, labour and neonatal care. This therapeutic drug administration has an ever increasing tendency reflecting the efforts of perinatal medicine to save the lives of high-risk fetuses/neonates. The carefree behaviour of obstetricians and neonatologists in the use and choice of drugs and their combinations for various treatment or prevention strategies in the perinatal period is, however, based on the false traditional opinion of the medical public that the risk of drug teratogenicity is limited only to the first trimester of gravidity. This has been in line with the late conception of classical structural teratology which specified teratogenic defects as structural malformations. The recent approach to teratogenicity is wider including also defects of function. From this point of view the drug teratogenic risk lasts during the entire gestation and even throughout the postnatal development, only the teratogenic defect is manifested as various functional deviations (brain, immune and endocrine system). Consequently, present medical therapeutic practice with unlimited drug prescription during the perinatal period entails the risk of iatrogenic harm.

This alarming situation initiated our experimental research of drug functional teratogenicity in animal model studies using drug administration in neonate rats and lifelong follow-up of behaviour, learning, reproduction, immunoreactivity and brain biochemical analysis. The results indicate that functional teratogenic risk of drugs with receptor-mediated effects (psychotropics, hormones) is highest in the perinatal period in which the majority of receptors necessary for their action are developing and formed. Experimental findings with tested drugs (glucocorticoids, tocolytics) in rats were in agreement with the outcome of long-term clinical studies. This fact supports the adequacy of the animal model used in our studies concerning the validity of acquired data for the extrapolation to the therapeutic practice. Since some medication is necessary for the treatment of risk pregnancies and risk newborns, it seems very urgent to evaluate the functional teratogenic potential in all drugs which are or will be used in the perinatal period and select such compounds for clinical use which combine sufficient therapeutic potency with minimal functional teratogenic risk.

Acknowledgement. The author wishes to thank to all members of the research team: RNDr H. Tejkalová, Ing Z. Křištofiková, RNDr J. Klaschka (Psychiatric Center Prague), RNDr V. Panajotovová CSc (Research Institute for Pharmacy & Biochemistry), Ing P. Hušek CSc (Institute of Endocrinology), MUDr M. Dostál CSc (Institute of Experimental Medicine ČAV). The research was supported by the grant IGA MZČR No 1811-3.

References

- Ballieux RE, Haijnen J (1987) Brain and immune system: a one-way conversation or a genuine dialogue? In: de Kloet ER, Wiegant VM, de Wied D (eds) *Progress in brain research*. (72, p 71–77)
- Benešová O (1989) Perinatal pharmacotherapy and brain development. *Int J Prenatal and Perinatal Studies* 1:417–424
- Benešová O (1995) Drug treatment in the perinatal period and the risk of functional teratogenicity. In: Seiler JP, Křofťová O, Eybl V (eds) *Toxicology – From Cells to Man*. Springer, Berlin Heidelberg New York (S 89–96)
- Benešová O, Pavlík A (1989) Perinatal treatment with glucocorticoids and the risk of maldevelopment of the brain. *Neuropharmacology* 28:89–97
- Benešová O, Pavlík A, Pětová J (1984) *Brain Maldevelopment and Drugs*. Avicenum, Prague
- Benešová O, Tejkalová H, Křištofiková Z, Klaschka J, Dostál M (1994) Early postnatal diazepam treatment of rats and neuroimmunocompetence in adulthood and senescence. *Ann. N.Y. Acad. Sci.* 717:89–101
- Broadhurst PL (1963) *The Science of Animal Behaviour*. Cox & Wyman Ltd., London
- Csaba G (1986) Receptor ontogeny and hormonal imprinting. *Experientia* 42:450–759
- DeBoer S, Mirmiran M, van Haaren F, Louwerse, A and van de Poll NE (1989) Neurobehavioural teratogenic effects of clo mipramine and alpha-methyl dopa. *Neurotoxicol. Teratol.* 11: 77–84
- Dobbing J (1970) Undernutrition and the developing brain. In: Himwich WA (ed) *Developmental Neurobiology*. Thomas, Springfield (p 241–261)
- Dobos A, Mészáros, J, Szekács, J and Gáti, I (1983) Long-term effect of glucocorticoid administered in the perinatal period. In: Endröczy E et al. (eds) *Neuropeptides and Psychosomatic Processes*. Hung. Acad. Sci., Budapest (p 39–45)

- Dostál M, Benešová O, Tejkalová H, Soukupová D (1995) Immune response of adult rats is altered by administration of diazepam in the first postnatal week. *Reproductive Toxicol.* 9: 115–121
- Ehrhardt AA, Meyer-Bahlburg HFL, Rosen LR, Feldman JF, Veridiano NP, Zimmerman I, McEwen BS (1985) Sexual orientation after prenatal exposure to exogenous estrogen. *Arch. sex. Behav.* 14:57–75
- Gunn T, Reece ER, Metrakos K, Colle E, (1981) Depressed T-cells following neonatal steroid treatment. *Pediatrics* 67:61–67
- Hadders-Algra M, Touwen BCL, Huisjes HJ (1986) Long-term follow-up of children prenatally exposed to ritodrine. *Brit. J. Obstet. Gynecol.* 93:156–161
- Herbst AL, Ulfender H, and Poskanzer DC (1971) Association of maternal stilboestrol therapy with tumor appearance in young women. *New Engl. J. Med.* 284:878–881
- Huisjes HJ, Hadders-Algra M, Touwen BCL (1986) Is clonidine a behavioural teratogen in the human? *Early Human Developm.* 13:1–6
- Jelínek R, Pavlík A, Peterka M (1983) Glucocorticoid receptor mediated teratogenesis in the chick embryo. *Teratogenesis, Carcinogenesis and Mutagenesis* 3:1–7
- Mirmiran M, Scholtens J, van de Poll NE, Uylings HBM, van der Gugten J, Boer GJ (1983) Effects of experimental suppression of active (REM) sleep during early development upon adult brain and behavior. *Dev. Brain Res.* 7:277–286
- Ounsted MK, Moar VA, Good FJ, Redman CWG (1980) Hypertension during pregnancy with and without specific treatment: the development of the children at the age of four years. *Brit. J. Obstet. Gynecol.* 87:19–24
- Reinisch JM (1981) Prenatal exposure to synthetic progestin increases potential for aggression in humans. *Science* 211: 1171–1173
- Schardein CV (1985) *Chemically Induced Birth Defects.* Marcel Dekker, New York
- Tejkalová H, Benešová, Křištofiková Z, Binková B, Dlohožková N, Burešová M (1991) The assessment of the behavioural teratogenic risk of the tocolytic drug phenoterol. *Homeostasis* 33:89–93
- Vessey MP, Fairweather DVI, Norman-Smith B, Buckley J (1983) A randomized double-blind controlled trial of the value of stilboestrol therapy in pregnancy: long-term follow-up of mothers and their offsprings. *Brit. J. Obstet. Gynecol.* 90:1007–1017