

G – GENITAL-URINARY SYSTEM

G01A – Gynaecologic antimicrobial and antiseptic agents

G01AA – Antibiotics

Mepartricine – G01AA09

It is available in Italy since 1983.

We have been unable to locate references on possible human reproductive effects of this agent, or have we found any similar studies on laboratory animals.

Meclocycline – D010AF99 – D06AA49 – G01AA49

It is available in Italy since 1986.

We have been unable to locate references on possible human reproductive effects of this agent, or have we found any similar studies on laboratory animals.

Tyrothricin – G01AA99 – G01BA – R01AX10 – R02AB02

This is a compound of peptic antibiotics available in Italy since 1967.

We have been unable to locate references on possible human reproductive effects of this agent, or have we found any similar studies on laboratory animals.

G01AA Class Conclusions: There is no written evidence of specific studies concerning the use of drugs belonging to this therapeutic class, in human pregnancy. In case of exposure an increase in the population background reproductive risk is not likely, due to its low absorption by vaginal way. Besides, there are no reported anomalies over the long period of commercialization, and their teratogenic action on experimental animals is lacking (records provided by manufacturer for registration, not available in databases).

G01AF – Imidazole derivatives

Imidazole antimycotics used by vaginal way have low systemic absorption of 3-10%. (Ritter 1985).

Clotrimazole – D01AC01 – G01AF02

Patented in 1972.

Case-control studies, nonspecific

- Rosa et al (1987): Analysis of medical discharging records in Michigan hospitals. Cases = 6,564 newborns showing congenital anomalies (suspected), 74 of which were exposed to clotrimazole. Controls = 97,775 showing no congenital anomalies, 1,012 of which were exposed. RR as per exposure by vaginal way over the first period of pregnancy = 1.1 (CI 95%: 0.9-1.4).
- Czeizel et al (1999), Hungarian CCSCA: Cases = 18,515 newborns showing congenital anomalies, controls = 32,804 showing no congenital anomalies. Exposures by vaginal way and/or topically among cases: 7.1% vs. 7.7% among controls (OR = 0.7; CI 95%: 0.6-1.0).

Retrospective cohort studies with internal controls

- Rosa (1993), Michigan MMS: of 2,624 first trimester exposures by vaginal way, 118 had major defects, 112 were expected (RR = 1.1; CI 95%: 0.9-1.3).

- King et al (1998), Michigan MSS: of 1,086 first trimester exposures by vaginal way, 74 had congenital anomalies, 68 expected (RR = 1.1; CI 95%: 0.9-1.4). 112 spontaneous abortions: RR = 1.3 (CI 95%: 1.1-1.7).

Conclusions: ADEC, FASS and WGZ consider clotrimazole a drug of choice in pregnancy. CDCs (1998) recommend the use of clotrimazole by vaginal way in every trimester of pregnancy.

Econazole – D01AC03 – G01AF05

This agent has a high crossing potential, but its absorption is of 1% of the applied dose. As per vaginal way its absorption mean rate is of 6.9%. Patented in 1973.

We have been unable to locate references on possible human reproductive effects of this agent during the first trimester of pregnancy.

Studies on laboratory animals

- Maruoka et al (1978): nonteratogenic in mice (up to 100 mg/kg subcutis); nonteratogenic in rabbits (up to 75 mg/kg subcutis).

Feto-neonatal effects: No adverse outcomes have been noticed in newborns exposed by vaginal way during the late weeks of pregnancy (Goormans et al 1985)

Isoconazole – D01AD05 – G01AF07

It is available in Italy since 1987

We have been unable to locate references on possible human reproductive effects of this agent during the first trimester of pregnancy.

Studies on laboratory animals

- Iida et al (1981): no adverse outcomes on fertility or on post-natal development have been revealed. No teratogenic effects were uncovered in rats (up to 125 mg/kg subcutis) and in rabbits (up to 30 mg/kg subcutis).

Feto-neonatal effects: No adverse outcomes have been uncovered in newborns exposed by vaginal way in the late weeks of pregnancy (Blum and Schweitzer 1989).

Tioconazole – D01AC07 – G01AF08

Its systemic absorption is of 5-16%. It is available in Italy since 1987.

We have been unable to locate references on possible human reproductive effects of this agent.

Studies on laboratory animals

- Noguchi et al (1982): nonteratogenic in rats (up to 100 mg/kg subcutis).

Fenticonazole – D01AC12 - G01AF12

It is available in Italy since 1985.

We have been unable to locate references on possible human reproductive effects of this agent, or have we found any similar studies on laboratory animals.

Azanidazole – G01AF13

It is available in Italy since 1984.

We have been unable to locate references on possible human reproductive effects of this agent.

Studies on laboratory animals

- Tammissa et al (1978): nonteratogenic in rats (300 mg/kg) or in rabbits (110 mg/kg).

G01AF Class conclusions: Clotrimazole has been thoroughly studied and it is considered a drug of choice in pregnancy. A systematic review has highlighted a better therapeutic effectiveness of this drug (7 days of treatment), in comparison with nystatin (OR 0.21; CI 95%: 0.16-0.29). As far as other imidazole derivatives, in case of exposure an increase in the population background reproductive risk is not likely, considering their low absorption by vaginal way and pharmacological class. In fact, they are very similar to the better-studied clotrimazole. Besides, there are no reported anomalies over the long period of commercialization (apart from sertaconazole that has become recently available), and their teratogenic action on experimental animals is lacking (records provided by manufacturer for registration, not available in databases).

G01AX – More antimicrobial and antiseptic drugs

Policresulen – A01AB11 – G01AX03

It is available in Italy since 1989.

We have been unable to locate references on possible human reproductive effects of this agent, or have we found any similar studies on laboratory animals.

Nifuratel – G01AX05

Patented in 1963.

We have been unable to locate references on possible human reproductive effects of this agent.

Studies on laboratory animals

- Scuri (1966): nonteratogenic in mice, rats and rabbits at doses 13, 13 and 7 times the human dose, respectively.

Feto-neonatal effects: No adverse effects have been noticed in newborns exposed orally or by vaginal way just before delivery term (Candela and Romano 1968, Aure and Gjonnaess 1969).

Povidone-iodine – D8AG02 – G01AZ11 – R02AA15 – S01AX18

It is absorbed by vaginal mucosa, with consequent increase of TSH serum concentration (Vorherr 1980; Safran and Braverman 1982; Jacobson et al 1984). Any iodide-based agent rapidly crosses the placenta. It is available in Italy since 1970.

We have been unable to locate references on possible human reproductive effects of this agent.

Studies on laboratory animals

- Claussen and Breuer (1975): nonteratogenic in rabbits when directly injected in the amniotic liquid.
- Siegemund and Weyers (1987): nonteratogenic in rabbits.

Feto-neonatal effects: No teratogenic effects have been detected in newborns exposed to topical vaginal disinfection at obstetric controls and birth. Iodine

levels, along with THS and T4 levels were normal in pregnant women and in the umbilical cord at birth, while T3 level was altered. On day 6 T4 was normal in all neonates; transiently high T3 was noticed in 2 newborns (Sakura et al 1993). Symptoms of hypothyroidism were revealed in newborns exposed for 7 days by vaginal way between 37 and 40 weeks (Gimes and Peter 1997). Dosages administered over a period of 10 days have not caused any risks to the newborn (Herbst and Selenkow 1965, Selenkow and Herbst 1966).

Conclusions: No specific studies are available in literature consistent with the use of povidone-iodine during the first trimester of human pregnancy. In case of exposure an increase in the background reproductive risk is not likely, due to a lack of reported anomalies over the long period of commercialization and considering that teratogenic effects in laboratory animals have not been found (records provided by manufacturer for registration, not available in databases). The use of iodide compounds over an extended period of time can cause hypothyroidism and goiter in the fetus and newborn.

Ciclopirox (Ciclopyroxolamine) – D01AE14 – G01AX12

It is available in Italy since 1984.

We have been unable to locate references on possible reproductive effects of this agent in the first trimester of human pregnancy.

Studies on laboratory animals

- Miyamoto et al (1975): nonteratogenic in rats and mice at 10 mg/kg.

Feto-neonatal effects: No adverse outcomes have been noticed late in pregnancy exposures (Novach et al 1999).

G01AX class conclusions: No specific studies are available in literature consistent with the use of agents in this therapeutic group during the first trimester of human pregnancy. In case of exposure an increase in the background reproductive risk is not likely, due to their low absorption by vaginal way, a lack of reported anomalies over the period of commercialization and considering that teratogenic effects in laboratory animals have not been found.

G02 – More gynecologic agents

G02A – Oxytocics

G02AB – Ergot alkaloids

Prospective cohort studies with internal controls

- Heinonen et al (1977), CPP: of 32 newborns exposed in the early 16 weeks to various ergot-derivatives – 5 of whom to methylergometrine and 18 to ergometrine, 1 had a major defect. ARR for the entire class of ergotamine derivatives = 0.7 (CI 95%: 0.1-4.8).

Methylergometrine (methylergobasine, methylergonovine) – G02AB01

This ergot derivative is mostly used to control postpartum bleeding. It is the most energetic on the uterus, among natural and semi-synthetic alkaloids of ergot. Its activity appears early and has a long duration (6-8 hours). It is available in Italy since 1948.

Case report

- Garcia et al (1990): 1 newborn exposed at 6 weeks of pregnancy to methylergonovine for metrorrhagia, with severe cranial abnormality (closure defect) in absence of part of the cerebellum and cerebral hemispheres, of hypophysis and suprarenal hypoplasia.

Feto-neonatal effects: this drug can cause uterine tetany during labor, and consequent fetal distress (Wong and Paul 1979, Moise and Carpenter 1988). Rhythm anomalies can occasionally occur (Moise and Carpenter 1988, Wong and Paul 1979).

Conclusions: No specific studies are available in literature consistent with the use of this drug in human pregnancy. Its use is not recommended but in postpartum. See also ergotamine (N02CA02).

Ergometrine (ergonovine) –G02AB03

This ergotamine derivative is used to contract the uterus. Patented in 1937.

Case report

- David (1972): 1 newborn that was exposed in the first trimester of pregnancy to massive doses of ergometrine in order to induce abortion had Poland anomaly.

Feto-neonatal effects: intrauterine death due to overdose administered just before birth (Au et al 1985).

Conclusions: There are incomplete studies consistent with the use of methylergometrine and ergometrine in human pregnancy. The action process of this compound inducing uterine contractions and consequent reduction of uterine blood flow, and its analogy with ergotamine (see N02CA02) is associated with congenital anomalies due to hypovascularization recorded in one single report of exposure to ergometrine. The risk cannot be estimated.

G02CA – Sympathomimetic drugs, tocolytic agents

Ritodrine – G02CA01

This is a sympathomimetic beta-2-selective agent used to relax uterine muscles. Umbilical venous drug levels are similar to those in the maternal circulation. Patented in 1964.

We have been unable to locate references on possible reproductive effects of this agent in the first trimester of human pregnancy.

Studies on laboratory animals

- Imai et al (1984 a,b,c): intrauterine death and developmental delay have been noticed in rabbits at maternal toxic doses (>35 mg/kg e.v. >750 mg/kg per os). At oral doses: digit skeleton defects, DIV and external defects.

Feto-neonatal effects: the following malformations have been reported, due to late in pregnancy exposure. Fetal supraventricular bradycardia and tachycardia (Gramissans et al 1969, Kleinhout and Veth 1975, Lauersen et al 1977, Hermansen and Johnson 1984), neonatal hypoglycemia (Epstein et al 1979, Kazzi et al 1987), neonatal hypocalcemia, paralytic ileus, ketoacidosis, fetal death. Controlled studies have besides revealed the following. The fetus was superior in weight and length vs. controls, no difference as for Apgar score (Thiery et al 1973), no difference vs. controls as for neonatal weight (Seidl et al 1973), no difference vs. controls as for psychomotor development, Denver test, ECG and urine test (Freysz et al 1977). No difference vs. controls as for glycemia, insulinemia, arterial pressure, heart rate, blood volume (Leake et al 1983), no difference vs. controls as for umbilical pH, Apgar score, cranial circumference, neonatal neurological condition (Huisjes and Touwen 1983). No difference vs. controls as for growth, neurological development and psychometric test (Polowczyk et al 1984), no difference vs. controls as for blood gas analysis, hemato-chemical test, platelet counting, bilirubin and oxygen therapy length (Hancock et al 1985). Higher incidence of transitory hypertrophy of the ventricular septum (Nuchpuckdee et al 1986), no difference vs. controls as for height, weight, cranial circumference, neurological and behavioral development, but poor progress at school (Hadders, Algra et al 1986). No difference vs. controls as for Apgar score and weight at birth, but 28.6% revealed ECG anomalies, such as ischemia (Gemelli et al 1990). Lower incidence vs. controls as for respiratory distress, no difference as for transitory tachypnea or other complications (Kerem et al 1977), lower risk of intracranial hemorrhage vs. controls exposed to other tocolytic agents (magnesium sulfate or indomethacin) (Weintraub et al 2001).

Lossisoprine – G02CA49

It is a non-selective beta-2- mimetic with light alfablocker activity. It is used as muscle relaxant and as peripheral vasodilator. Patented in 1956.

Case report

- Sakuma et al (1993): 1 newborn exposed between 16 and 18 weeks showing aplasia of the limb cutis and soft tissue hypoplasia.

Prospective cohort studies with internal controls

- Heinonen et al (1977), CPP: of 54 exposures during the early 16 weeks, 2 newborns with congenital anomalies were detected. ARR for any type of malformation = 0.8 (CI 95%: 0.2-3.2).

Feto-neonatal effects: the following malformations have been reported due to late pregnancy exposure. Fetal tachycardia, hypocalcemia, hypoglycemia, ileum, hypotension and death (Brazy et al 1979, Brazy et al 1981a, 1981b), neonatal respiratory depression (Kero et al 1973), maternal pulmonary edema (Evron et al 1983, Nimrod et al 1984a, 1984b). Cohort studies: higher frequency vs. controls

as for hypocalcemia, ileum, hypoglycemia, hypotension and neonatal mortality (Brazy and Pupkin 1979); lower incidence vs. controls as for respiratory distress (Brazy et al 1981); no difference vs. controls as for Apgar score and birth weight, but 66.7% had ECG anomalies such as ischemia (Gemelli et al 1990).

G02CA class conclusions: There is no written evidence of specific studies concerning the use of ritodrine and isoxsuprine in human pregnancy. In case of exposure during the first trimester an increase in the population background reproductive risk is not likely. This is due to a lack of reported anomalies over the long period of commercialization (first trimester use sometimes specified) as well as to the absence of teratogenic action on laboratory animals (records provided by manufacturer for registration, not available in databases). Newborns exposed late in pregnancy should be kept under strict control, because of possible arise of hypocalcemia, hypoglycemia, hypotension, paralytic ileus and ECG anomalies, spontaneously reversible in few weeks.

G02CB – Prolactin inhibitors

Bromocriptine – G02CB01

This is an ergot derivative with no oxytocic action that has been used in the treatment of hyperprolactinemia and in Parkinsonian disorders. Its use to induce ovulation is no longer approved. Patented in 1968.

Cohort studies without controls

- Griffith et al (1978): of 448 bromocriptine-induced pregnancies (administered up to 3 weeks after conception), 358 healthy newborns (among whom 6 twins), 2 newborns showing major defects, 9 having minor defects, 49 spontaneous abortions, 30 VIP.
- Bergh et al (1978) 17 healthy infants born to mothers whose pregnancy was induced with bromocriptine (administered up to 3 weeks after conception).
- Nencioni et al (1978) of 24 bromocriptine induced pregnancies (administered up to 3 weeks after conception), 7 miscarriages (29%) and 17 healthy newborns (follow-up until 1 year of age).
- Griffith et al (1979): of 137 pregnancies of women suffering from hypophysis tumors and treated with bromocriptine, 116 newborns. No increased risk of spontaneous abortion, multiple pregnancies and congenital anomalies, in comparison with the rest of the population.
- Campagnoli et al (1980) has reported on 15 pregnancies induced with bromocriptine (administered up to 3 weeks after conception and in one single case throughout the first trimester). There were 11 healthy newborns and 2 miscarriages (2 pregnancies in progress at the moment of publishing).
- Audibert and Turkalj (1982), Krupp et al (1986): of 1,410 pregnancies treated with bromocriptine (average duration of the exposure 27-30 days after conception), 12 extrauterine pregnancies (0.9%) 3 mole idatiformi (0.2%), 157 miscarriages (11.1%), 25 VIP, 31 multiple pregnancies (2.2%), 1,236 live births (87.7%), 5 stillbirths (0.4%). 43 out of 1,241 newborns had congenital anomalies (3.5%), 12 of them were considered by the authors "major" (1%) and 31 "minor" (2.5%) malformations. There was no difference as for dosage and duration of exposure between the newborns with congenital anomalies and the healthy newborns.
- Canales et al (1981), Bergh et al (1981), van Roon et al (1981), Yuen et al (1982), Maeda et al (1983), Crosignani et al (1984): of 145 pregnancies exposed to bromocriptine altogether, 133 were live births, 2 multiple malformations, 9 spontaneous abortions, 1 VIP. 5 newborns were showing "minor" congenital anomalies.

- Jasonni et al (1983): of 48 pregnancies treated with bromocriptine (administered up to 3 weeks after conception), 6 miscarriages and 42 healthy newborns.

Retrospective cohort studies with internal controls

- Rosa (1993), Michigan MSS: of 50 first trimester exposures, 3 newborns with major defects, 2 expected (RR = 1.5; CI 95%: 0.3-4.4).

Feto-neonatal effects: regular psychophysical development in exposures throughout pregnancy (Konopka et al 1983, de Wit et al 1984), no adverse outcomes on exposed newborns (Krupp et al 1985, Sas et al 1986).

Conclusions: The available studies – although almost all cohort studies without controls – consistent with the use of bromocriptine in the first trimester of pregnancy, have assessed a large number of exposures in pregnancy (around 2,200) and have not uncovered any increased population background risk.. The use of this agent in other periods of pregnancy has not revealed any adverse outcomes on the newborns.

Cabergoline – G02CB03

It is an ergot derivative, dopaminergic agonist used in hyperprolactinemic states and recently used also in the treatment of hypophysis tumors. It is available in Italy since 1994.

Literature review

- Rains et al (1995): of 199 exposures, 10 newborns had congenital anomalies, not showing a specific malformative pattern.

Cohort studies without controls

- Robert et al (1996), TIS Lione: of 204 exposures, 24 miscarriages, 3 VIP (Down syndrome, hypo-agenesia of limbs, hydrocephalous), 149 live births out of which 2 (1,7%) with congenital anomalies (megauretere and scafocephaly); 5 congenital anomalies therefore out of 183 exposures (2.7%). No developmental abnormalities were uncovered in 61 newborns followed up to the first year of age.
- Ricci et al (2002): of 61 exposures, 6 miscarriages, 5 VIP, 1 mola idatiforme and 49 live births 1 of which with trisomy 18.

Conclusions: The available studies consistent with first trimester exposures (about 500 exposures in cohort studies without controls) have not uncovered any increased background population risk. The use of this agent in other periods of pregnancy has not revealed any adverse outcomes on the newborns.

Metergoline – G02CB05

This is an ergotamine derivative, dopamine agonist with dynamic pharmacological properties, similar to bromocriptine. It is available in Italy since 1995.

We have been unable to locate references on possible human reproductive effects of this agent, or have we found any similar studies on laboratory animals.

Conclusions: There are no studies in literature, consistent with the use of metergoline in human pregnancy. In case of exposure an increased background reproductive risk is not likely, due to a lack of reported anomalies over the long period of commercialization. The pharmacological similarities with bromocriptine have been thoroughly studied and teratogenic effects in laboratory animals have not been found (records provided by manufacturer for registration, not available in databases).

G03 – Sexual hormones and modulators of the reproductive system

Estrogens and Progestins are endogenous hormones active in many physiological ways. These compounds are mainly used as:

- **Oral contraceptives.** The agents currently marketed consist of an association of estrogens and progestins, but there are also contraceptives only made of progestins. Ethynylestradiol is the estrogen used in oral contraceptives at variable dosages ranging between 0.02 and 0.05mg. Levonogestrel (0.05-2.25mg), gestodene (0.05-0.1mg), desogestrel (0.15mg), drospirenone (3mg) and cyproterone (2 mg) are the progestins used in present contraceptives. The “old fashioned” oral contraceptives contained norgestrel and norethisterone, progestins with a possible androgen activity.
- **Hormone pregnancy tests.** In the past they were carried out administering progesterone alone (20 mg/i.m.) by intramuscular way or per os. Sometimes in association with estrogens (10-50mg of ethisterone+0.1-0.5mg of ethynylestradiol; 12.5-50mg of progesterone + 2-3mg of estradiol; 5-10mg of norethisterone/norethindrone + 0.01-0.02 mg of ethynylestradiol; 10mg of dimethisterone + 0.5mg of ethynylestradiol).
- **Regular abortion therapy or to prevent miscarriage.** In this case 50-100 mg of progesterone and 250-375 mg of hydroxyprogesterone has been administered, although their effectiveness in routine use has not been proved (probably except for a sub-group of women with 3 or more abortions treated with progesterone). (World Health Organization 1981, Oates-Whitehead 2003). Various progestins were used in the past: allilestrenol, noretisterone, didrogestrone, medroxyprogesterone and diethylstilbestrol (DES).

The three following main issues make it complicated to estimate the teratogenic risk associated with sexual female hormones:

1. the used dosages are often not specified and they vary along with the period and the group of studied women;
2. the evaluation of progestins is difficult since it is often not clear the reason why they have been administered. Under the generic expression “threatened abortion”, in fact, quite different conditions are to be considered, such as: threatened abortion in preceding pregnancies, cervical incompetence, conception difficulty, blood loss due to various causes, stomach pains and, eventually, a real lack of progesterone. It is therefore impossible to figure out the confounding role of the condition that has required the use of the drug;
3. lack of evaluation as for specific molecules.

Finally, one more thing should be considered: many of the studies carried out on female sexual hormones are old and based on no-longer used agents, such as progestins with high androgen activity (noretinodrel, noretisterone or noretindrone, linesterol, norgestrel, allilestrenol etc.). Sometimes the studies are instead based on associations of estrogens and progestins at dosages much higher than those actually marketed.

Table of female sexual hormones

	Possible association	ATC	Therapeutic use
Estrogens	+ progestins	G03AA G03AB	Fixed dose oral contraceptives Sequential oral contraceptives

	_____	G03CA	Postmenopausal substitute hormone therapy – Gonadal dysgenesis – Menstruation pathologies
Progestins	+ estrogens	G03AA G03AB	Fixed dose oral contraceptives Sequential oral contraceptives
	_____	G03AC	Oral contraceptives with progestin as their single component (desogestrel)
	_____	G03AC	Postcoital contraceptives (levonorgestrel)
	+ estrogens	G03FA G03FB	Postmenopausal substitute hormone therapy
	_____	G03DA02	Postmenopausal substitute hormone therapy – Menometrorrhagia – Amenorrhea – Mammary carcinoma
	_____	G03DA03 G03DA04	Sterility from lutein insufficiency – Threatened abortion
	_____	G03DB03 G03DB04	Threatened abortion – Pathologies of menstruation – Infertility
	_____	G03DC02	Pathologies of menstruation – Endometriosis – Mammary carcinoma
	_____	G03DC05	Postmenopausal substitute hormone therapy

GENERAL STUDIES ON FEMALE HORMONES

Systematic review

- Raman-Wilms et al (1995). A systematic review was carried out for the metanalysis of the risk magnitude as for external malformations of genitals (i.e.: hypospadias, cryptorchidism, ambiguous genitals, and clitoral hypertrophy). The above mentioned were then associated with exposure of the various female hormones in the first trimester of pregnancy (except for DES) also used as oral contraceptives, in hormonal pregnancy test, and treatment in case of threatened abortion. The authors have made a research using Medline and other sources, such as reviews, original articles, books and essays, as follows:
 - 186 studies published between 1962 and 1992 (August) have been identified;
 - 23 controlled studies specifically allowing risk assessment have been evaluated. (Cosgrove 1977, Czeizel et al 1979, Janerich et al 1980, Kallen and Winberg 1982, Depue 1984, Resseguie 1985, Calzolari et al 1986, Stoll et al 1990 were not included, due to lack of information or other essential information enabling risk estimate).
 - 14 studies have been used for the metanalysis: 7 case-control studies (Jacobson 1972, Sweet et al 1974, Monteleone et al 1981, Polednak and Janerich 1983, Beard et al 1984, Kallen 1988, Kallen et al 1991, McBride et al 1991) and 7 cohort studies (Harlap et al 1975, Harlap and Heldor 1980, Mau 1981, Katz et al 1985, Harlap et al 1985, Yovich et al 1988);
 - the cumulative odds ratio has been calculated in all studies and sub-groups of studies (see table);
 - the authors have therefore stated that there is no evidence of increased risk as for genital anomalies due to the most common sexual hormones, except for noretindone, that is associated with a risk of clitoral hypertrophy and other symptoms of virilization in female subjects.

Type of study	Exposures	Controls	Cumulative Odds ratio (IC 95%)
Total (14)	5.755	59.812	1.1 (0.9-1.3)
Case – control (7)	1.731	2.365	1.0 (0.3-3.7)
Cohort (7)	4.030	57.447	1.2 (0.2-8.9)
CO exposure only (5)	1.931	42.660	1.0 (0.2-3.9)
Males only (8)	2.259	5.398	1.1 (0.0 – 29.2)
Females only (1) §	59	99	29.9 (3.8-234.2)

Studies on cases with inappropriate controls or without controls

- Nora et al (1978): this is a study the authors consider conclusive and confirming some of the clinical observations previously carried out by the same authors. The authors refer as study 1 case report (the way it has been recorded is not specified) of 30 patients with VACTERL syndrome (at least 3 of the 7 defects of the complete syndrome), interviewed at diagnosis, varying as far as time left prior to delivery. 13 of them (43%) had been exposed in the first trimester to female hormones due to hormone pregnancy test (7 cases) or threatened abortion (5 cases). The authors in this very article refer also another study (with 2 types of controls referred as case-control 2 and 3) concerning 236 congenital cardiopathies of various type. Due to a procedure aiming at identifying the controls, since 21% of cardiopathies were not sufficiently described, it turned out that they had been exposed to various

types of hormones used for hormone pregnancy test or in the treatment for threatened abortion. Elsewhere hormones had been used following the failure of contraceptive pill. Some women had been treated with clomiphene, so that they were subfertiles. The prevalence of exposure among controls given in this study is estimated at 8%. The authors suggest an association between hormones and cardiopathies, particularly in the case of VACTERL syndrome. The methodological quality of the reported observations may be considered inadequate. Nevertheless the hypothesis of a possible association (even if at a scarce risk rate) of VACTERL with specific hormones (and/or specific conditions requiring the treatment) has not been dropped, it has been tested, instead, with an appropriate study (see below).

- Aarskog et al (1979): of 130 subjects showing hypospadias, 11 had been exposed in the first trimester to progestins due to threatened abortion, or else to estroprogestins for a pregnancy test. The study proved to be useful in order to work out the hypothesis, but it does not give any evidence, due to lack of controls.

Case-control studies, nonspecific

- Martinez-Frias et al (1998), ECEMC: case-control studies to evaluate the risk of malformation commonly associated with various categories of female sexual hormones. Cases: 20,388 newborns with congenital anomalies (excluding generic syndromes or syndromes due to well-known teratogens); controls: 19,981 healthy newborns. Exposure evaluated with routine interviews after birth: female sexual hormones of various type and used for various reasons in the first trimester (mostly associations of progestins due to threatened abortion: 440 exposures among cases vs. 381 among controls and oral contraceptives; 123 exposures among cases vs. 89 controls). The analysis has been carried out according to a logistic multi-varied model checking the main confounding factors in case of specific groups of malformations. Result: none of the female hormones revealed a high malformation risk. A high risk of esophageal atresia was spotted in women who had threatened abortion (vaginal blood loss). This study clearly shows the effect of the confounding factor "threatened abortion" in the estimate of the risk following the use of female hormones. The association between threatened abortion and esophageal atresia is noteworthy (see below).

Retrospective cohort studies with internal controls

- Hemminki et al (1999): a study to assess the risk of tumors (and of major or minor anomalies) in exposures to female hormones. It has been worked out checking medical records of newborns filled in during prenatal examinations (85% of all the newborns in Finland), between 1954 and 1963. Exposed cohort: systematic sample of all newborns exposed in the first trimester to estrogens or progestins for various reasons (particularly for threatened abortion); control cohort: systematic sample of all newborns not exposed to hormones during pregnancy. Ascertainment of outcome with record linkage to medical records. Follow up until 34-43 years of age. Results: 76 newborns with congenital anomalies of various type (17 of unknown seriousness) among 1,963 exposed, vs. 40 among 2,108 controls. RR for major defects (not specified) = 1.8 (CI 95%: 1.0-3.0); for female tumors = 0.7 (CI 95%: 0.3-1.5). This study, extremely interesting as far as tumors in the long run, reveals some important lacks in the analysis of congenital anomalies.

Prospective cohort studies with internal controls

- Heinonen et al (1977), CPP: of 1,042 exposures in the early 16 weeks of pregnancy to a therapy with various female hormones, 19 newborns had congenital cardiopathies vs. 385 newborns with cardiopathy out of 49,240 not exposed newborns. ARR = 2.3 (CI 95%: 1.5-3.7). In the subgroup of

exposures to oral contraceptives there were 6 cardiopathies out of 278 with ARR = 2.7 (CI 95%: 1.2-6.0). This study has been later reevaluated by:

- Wiseman and Dodds-Smith (1984): once the characteristic of exposure had been controlled they suggested an association statistically insignificant, and nevertheless not reporting the data. In fact, of the 19 reported cases showing association between cardiopathy and exposures to hormones, 2 turned out not to have been exposed to hormonal compounds. Besides, 5 were newborns exposed over a period following cardiac organogenesis (19-50 days of pregnancy), 2 more had been exposed over a period much before cardiac organogenesis, and 2 had Down syndrome.
- Hook (1994) has reassessed RR for congenital cardiopathies taking notice of the considerations by Wiseman and Dodds-Smith but suggesting an ARR = 2.5 (p<0.05).

Case-control studies, specific

- Ferencz et al (1980): 110 newborns with cardiac cono-truncali malformations, 110 healthy controls, 186 random controls and 20 pathological controls with hypoplastic syndrome of left heart. See Table as for the main use of various hormones. No association with various type of hormones.

Ferencz et al 1980 Employment of sexual hormones in the 1 st trimester	Cases 110	Matched controls 110	Random controls 186	Pathologic controls 20
Oral contraceptives	5.5 %	4.8 %	5.5 %	0
Pregnancy test	3.6 %	5.9 %	4.5 %	0
Ovulation stimulants	1.8 %	1.1 %	0	0
Gestation stimulants	4.5 %	3.2 %	6.4 %	5.5 %
Total	15.5 %	14 %	16.4 %	5.5 %
Oral contraceptives used in the month prior to pregnancy and during the 1 st trimester	13.6 %	12.9 %	8.2 %	0

- Milla et al (1982): 64 newborns showing hypospadias, 19 exposed to progestins in the first trimester, vs. 15 out of 128 controls (OR = 3.2; IC 95%: 1.4-7.3) Not considered by Raman Wilms since it was in Italian.
- Depue et al (1983): 108 cases of testicular tumor in men under 30 years of age and as many healthy controls of the same age. The risk was 8 times higher in exposures to estrogens during pregnancy. Very high memory bias.
- Lammer and Cordero (1986): case-control study "in rotation" (see dictionary) out of 1,091 newborns with 12 severe malformation easy to diagnose (i.e. spina bifida, esophageal atresia, cleft lip, anal atresia and Down syndrome), to assess the associated risk to female hormones. For each group of malformations, newborns with other malformations served as control. Among the 12 studied anomalies only esophageal atresia (36 cases in total) had a high OR = 2.8 (CI 95%: 1.5-5.3) not modified by some examined confounding factors (not including threatened abortion!). The association was limited to the use of hormones used for hormone pregnancy test or progestins used against threatened abortion (the results of the two different exposures were not separated) and not to oral contraceptives (few exposures – 28 in total – and low power of analysis for subgroup). This study has not been considered by Raman-Wilms in his systematic review, since it was reckoned inappropriate to set out the exposure risk magnitude, due to the type of used controls.

- Lammer and Cordero (1986): 34 cases of VACTERL and 1,024 newborns with other malformations, including Down syndrome (as controls), have been studied in order to check the hypothesis suggested by Nora et al of an association between VACTERL syndrome and female hormones. The authors in a previous study had suggested the same hypothesis. OR for VACTERL syndrome = 0.98 (CI 90%: 0.4-2.4). The study had an adequate statistic power to find out 2.8 risks and higher.

STUDIES ON FEMALE HORMONES MAINLY USED AS HORMON PREGNANCY TEST

Literature review

- World Health Organization (1981): Review of studies controlled on exposure to pregnancy hormonal test.

Case-control studies	Defect	Exposed cases n (% defects)	Exposed controls n (% defects)	OR	Comments
Laurence et al 1971	NTD, Anencephaly	293 (20.1)	345 (23.9)	1.3 §	Mothers of the cases had previously delivered infants with NTD
Gal et al 1972	Spina bifida	100 (19)	100 (4)	5.6 **	Possible memory bias, not reported choosing criteria, maternal age differences, incorrect geographical choice of the cases
Oakley et al 1973	All	361 (8.3)	72 (11.1)	1.3 "	Cases collected from registries, controls with chromosomal anomalies, insufficient control of the confounding factors
Greenberg et al 1975	All	836 (11.1)	836 (6.6)	2.1 **	Well done study
Rothman et al 1979	Cardiopathies	390 (3.7)	1.254 (2.4)	1.3 "	Possible memory bias
	APVR	9 (22)	335 (3.6)	11.0 *	

Cohort studies	Defect	Exposed n (% defects)	Not exposed n (% defects)	RR	Comments
Haller 1974	All	617 (2.6)	2.917 (2.1)	1.2 §	No control of the confounding factors
Torfs et al 1981	All	203 (4.4 major), (15.3 minor)	1.021 (3.8 major) 12.4 minor	1.2 § 1.2 §	Low number of exposures, the control of the confounding factors does not modifies the results
	Male GU tract	109 (5.5)	490 2.5	2.3 §	

§ = Not statistically significant * = p<0.05; ** p< 0.01; APVR = anomalous pulmonary venous return

Prospective Cohort studies not considered by WHO review

- Kullander and Kallen (1976): in a cohort of 5,753 newborns there were 128 exposed infants to hormonal pregnancy test (Ethinylestradiol and noretindrone), ascertained with questionnaires filled during pregnancy. Among the exposures there were 4 newborns with "major" malformations: RR = (CI 95%: 0.4-2.5) and 16 "minor" malformations: RR = 1.3 (CI 95%: 0.8-2.1).

G03A – Systemic oral contraceptives

The actual content of estrogens in the current compounds is of 20-50µg, whereas the dose of progestins is varying according to the different power of the used compounds.

G03AA – Fixed estrogen-progestogen combinations

G03AB – Sequential estrogen-progestogen compounds

Levonorgestrel + ethynilestradiol – G0AA07 – G03AB03

Desogestrel + ethynilestradiol – G0AA9 – G03AB05

Gestoden + ethynilestradiol – G0AA10 – G03AB06

Drospirenone + ethynilestradiol – G0AA12

G03AC – Progestins

Levonorgestrel – G03AC03

This is used as a postcoital contraceptive.

Desogestrel – G03AC09

This is a progestin with limited androgenic properties, also used alone in oral contraception. It has a lower androgenic activity compared to 19-nortestosterone and its derivatives. It increases the levels of free testosterone (a consequence of the SHBG reduced levels), but it does not show any androgenic effects when administered in combination with estrogens. Patented in 1974.

STUDIES ON FEMALE HORMONES MAINLY USED AS ORAL CONTRACEPTIVES (G03A)

The studies are based on agents containing progestin-like compounds showing androgenic activity and at dosages about 3 times higher than currently marketed drugs.

Systematic review

- Bracken (1990): Systematic review to be employed in a metanalysis of the risk size of malformations in general, associated only with exposure to oral contraceptives after conception. Only cohort studies “known by the author” have been used (not specifying the method used to systematically find the available studies), excluding those studies analyzing other type of female hormones. 12 studies were taken in consideration (Peterson 1969, Robinson 1971, Haller 1974, Oechsli 1976, Royal College of General Practitioner 1976, Heinonen et al 1977, Rothman and Louik 1978, Vessey et al 1979, Kasan and Andrews 1980, Harlap and Eldor 1980, Linn et al 1983, Harlap et al 1985). The author has estimated the Relative Risk including the studies in total and specific subgroups of studies as follows. (a): studies with well specified control groups in order to tell weather the chosen controls, biased by some confounding factors, might change the estimate of the relative risk; (b): specific subgroups of malformations. Altogether the 12 analyzed groups allowed to evaluate 6,102 exposures. The metanalysis has not revealed any increased risk either in general malformations or in cardiopathies and limb hypo-agenesias (see Table).

Group or subgroup of studies	Cumulative Relative Risk (IC 95%)
Total of available studies (12)	1.0 (0.8-1.2)
Only studies (10) with A control group	1.0 (0.8-1.3)
Only studies (4) with B control group	1.0 (0.7-1.5)

As per congenital cardiopathies (8)	1.1 (0.7-1.6)
As per limb hypo-agenesias (6)	1.0 (0.3-3.6)

Please find in brackets the number of studies used to estimate the cumulative relative risk.

A = use of oral contraceptives before pregnancy

B = use of other contraceptive methods during pregnancy

Literature review

- World Health Organization (1981): Review of studies controlled as for exposure to oral contraceptives. The following have not been considered, due to inappropriate methodology: Robinson 1971, Yasuda and Miller 1975, Nora et al 1976, Nora and Nora 1976. The cohort studies have been analyzed in the systematic review carried out by Bracken (1990). We are hereby listing only case-control studies.

Case-control studies	Defect	Cases	% exposures	Controls	% exposures	OR
Mulvihill et al 1974	Cardiopathies	88	1.1	88	0.0	unknown
Janerich et al 1974	Limb hypo-agenesias	108	5.6	108	0.9	6.2
Janerich et al 1975	Down S.	103	1.9	103	1.9	1.0
Janerich et al 1977	Cardiopathies	104	1.9	104	0.0	n.c
Rothman et al 1979	Cardiopathies	344	2.3	1.126	1.8	1.3
Bracken et al 1978	Anencephaly	81	4.9	2.968	1.8	3.3
	TGV	53	3.8	2.968	1.8	2.3
	Septal defects	200	4.0	2.968	1.8	2.2
	Valvular defects	47	4.3	2.968	1.8	2.7
	Other cardiopathies	85	2.4	2.968	1.8	1.3
	LS+/-PS	38	2.6	2.968	1.8	1.4
	Pyloric stenosis	70	0.0	2.968	1.8	0.0
	Esophageal atresia	48	4.2	2.968	1.8	2.3
	Down S.	52	3.9	2.968	1.8	2.2
OR never turned out statistically significant in single studies						

Case report

- Otten et al (1980): 1 newborn showing hepatoblastoma exposed to oral contraceptives after conception.

Case-control studies, nonspecific

- Janerich et al (1980): 715 newborns showing various congenital anomalies (104 cardiopathies, 103 Down syndromes, 108 limb hypo-agenesias, 201 renal tube defects, 99 hypospadias and 100 various malformations), 715 healthy controls. Exposure to oral contraceptive after conception occurred in 17 cases vs. 8 controls: OR = 2.1 (IC 95%: 0.9-5.7).

Prospective cohort studies with internal controls (not included in the metanalysis)

- Heinonen et al (1977), CPP: of 278 exposures to oral contraceptives during the early 16 weeks, 13 newborns with congenital anomalies. ARR for any type of defect = 1.0 (CI 95%: 0.5-1.6); for cardiovascular defects (6) = 2.4 (CI

95%: 0.9-5.2); for hypospadias (3 out of 134 male babies) = 2.7 (CI 95%: 0.6-7.6).

Nested case-control studies, specific in the prospective cohort of all newborns

- Kallen and Otterblad Olausson (2003), Swedish MBR: 5,015 cases showing cardiovascular defects, 22 of which exposed to oral contraceptives in the first trimester and 577,730 controls among which 1,878 exposures. OR for cardiovascular defects = 1.4 (CI 95% 0.9-2.1).
- Kallen (2003), Swedish MBR: of 1,044 cases born with non-syndromic LPS there were 6 exposed to oral contraceptives in the first trimester, whereas out of a total of 576,873 controls there were 1,878 exposures. AOR = 1.8 (CI 95%: 0.7-4.0).

Case-control studies, specific

- Czeizel et al (1983), Hungarian CCSCA: 247 newborns with limb hypo-agenesias. OR for post-conception exposure = 1.8 (not statistically significant).
- Winship et al (1984): 704 cases born with CNS abnormalities and 764 healthy controls. OR for first trimester oral contraceptive exposures = 2.5 (CI 95%: 0.4-26.3).
 - Krickler et al (1986): this is a study by Mc Credie published in 1983, reproposed by the authors. 155 cases born with limb hypo-agenesias and 274 healthy controls. 18 cases had been exposed to oral contraceptives over a period varying from 2 weeks to 5 months, vs. 1 exposure among controls. OR = 30.2 (CI 95%: 5.3-52.0). This study lay itself open to several criticisms concerning the methodology.
- Werler et al (1992): 76 cases with gastroschisis vs. 2,142 controls with other major malformations. 5 among the cases and 64 among controls were exposed to oral contraceptives. AOR = 1.3 (CI 95%: 0.5-3.5).
 - Li et al (1995): 118 renal congenital anomalies or defects of the urinary tract (renal a-dysgenesis, multicystic kidney, hydronephrosis and ureter anomalies), 369 healthy controls (who had joined the study out of 570 eligible subjects, chosen at random among all newborns). The exposure to oral contraceptives continued after conception has been ascertained by means of direct interview that was worked out after birth (in a not specified period). Neither the interviewer nor the mother knew the hypothesis of the study. No significant difference was pointed out between the characteristics of cases and of controls. The analysis was carried out according to a logistic regression pattern. Results: 9 exposed newborns among cases vs. 8 among controls, AOR = 4.8 (CI 95%: 1.6-14.1). The risk turned out higher (AOR = 8.0, CI 95%: 1.3-48.5) for exposures over the period beyond week 4 (based on 5 exposures among cases) vs. exposures that had lasted less than 4 weeks (AOR = 3.6; CI 0.9-14.3).
 - Olshan et al (1999): the hypothesis was checked concerning the risk of neuroblastoma, raised by Mandel et al 1994, on 504 guys showing the anomaly. There has not been any association with the use of oral contraceptives in the first trimester of pregnancy: OR = 1.4 (CI 95%: 0.9-2.1).

Use of oral contraceptives before conception: Several studies have considered the possible association between the use of oral contraceptives prior to conception and congenital anomalies, but the risk increase has not been proved. The following malformations were taken in consideration:

- Congenital anomalies in general (Satterwaite and Gamble 1962, Peterson 1969, Robinson 1971, Haller 1975, Royal College of General Practitioners 1976, Harlap and Davies 1978, Janerich and Piper 1978, Bracken et al 1978,

Doring and Fresenius 1979, Ortiz-Perez et al 1979, Harlap and Eldor 1980, Janerich et al 1980, Kasan and Andrews 1980, Savolainen et al 1981, Harlap et al 1982, Linn et al 1983) and transposition of the great vessels (Yasuda and Miller 1975)

- Hypo-agenesias of limbs (Yasuda and Miller 1975, Smith et al 1977)
- Down syndrome (Janerich et al 1976, Harlap and Davies 1978, Ericson et al 1983)
- NTD (Cuckle and Wald 1982)
- Isolated cardiac defects of various type (Shaw et al 1992)
- Chromosomal anomalies (Dhadi et al 1971, Bouè 1973, Lauritsen et al 1975, Alberman et al 1976, Klinger et al 1976, Shiono et al 1979, Harlap et al 1982).

G03CA – Natural and semi-synthetic estrogens, non associated

Used as substitutive hormonal therapy in post-menopausal women, in amenorrhea and – at lower dosages, also in association with progestins for oral contraception.

See also classification, therapeutic advice and reported studies in the section dedicated to sexual hormones and genital system (G03).

Ethinylestradiol – G03CA01

This estrogen is an estradiol derivative. It is used in oral contraception, at low dosages and in association with progestins (see above). It is recommended at higher dosages in the pathology of menstruation (amenorrhea, hypo-menorrhea, and oligomenorrhea), menopause and to prevent the beginning of lactogenesis. Synthesized in 1983.

Prospective cohort studies with internal controls

- Heinonen et al (1977), CPP: of 89 exposures in the early 16 weeks due to unspecified reasons, 8 were born with congenital anomalies (ARR = 2.0; CI 1.0-3.9).

Estradiol – G03CA03

This is an estrogen naturally produced by the ovary. It is also a metabolite of testosterone. In the past it was used in some oral contraceptives. It is currently recommended for menopause. Patented in 1976.

Prospective cohort studies with internal controls

- Heinonen et al (1977), CPP: of 48 exposures in the early 16 weeks due to unspecified reasons, 1 was born with congenital anomaly (ARR = 0.5; CI 0.1-3.2).
- Rosa (1933), Michigan MSS: of 29 first trimester exposures due to unspecified reasons, 4 had major defects and 1 expected (RR = 4.0; 1.1- 10.2).

Feto-neonatal effects: no adverse outcomes were reported in case of administration during labor (Luther 1980).

Estriol – G03CA04

It is a natural estrogen, final metabolite of estradiol. It is produced by the feto-placental unity in pregnancy. It is recommended to treat menopause disturbances, as well as post-menopause, dysmenorrhea and sterility. Patented in 1949.

We have been unable to locate references on possible human reproductive effects of this agent, or have we found any similar studies on laboratory animals.

Conjugated estrogens – G03CA57

These estrogens are water-soluble. They mainly contain natural or synthetic estrone sulfate and equilin sulfate. It is recommended to treat menopause and post-menopause. Patented in 1951.

Prospective cohort studies with internal controls

- Heinonen et al (1977), CPP: the compound has been studied along with other estrogens in a total of 13 exposures in the early 16 weeks due to unspecified reasons (11 of them were exposures to conjugated estrogens). 1 newborn had a single congenital anomaly. RR for every type of congenital anomaly in the entire studied group = 1.7 (CI 95%: 0.3-11.2).

G03D – Progestins

Naturally occurring or synthetically obtained, these hormones are administered separately or in combination with an estrogen in contraceptive therapies. They are used to treat threatened abortion and preterm labor.

See also their classification, therapeutic advice and studies reported in the section dedicated to sexual hormones and modulators of reproductive system (G03).

Literature review

- World Health Organization (1981): Review of studies containing controls on exposure to progestins used in the treatment of infertility, habitual and threatened abortion.

Case-control studies	Defect	Cases		Controls		OR *	Notes
		n	% Exp.	n	% Exp.		
Nelson e Forfar 1971	Various	458	4.1	911	2.9	1.4	a
Levy et al 1973	TGV	76	13.2	76	0.0	n.c.	
Janerich et al 1974	Limb hypo-agenesias	108	8.3	108	2.8	3.0	b
Yasuda e Miller 1975	TGV	58	1.7	93	2.2	0.8	c
Hellstrom et al 1976	Limb hypo-agenesias	32	12.5	30	3.3	3.8	
Janerich et al 1977	Cardiac	104	5.8	104	1.0	5.8	b
Rothman et al 1979	Cardiac	390	3.7	1.254	2.4	1.5	b
Cohort studies	Defect	Exposed		Non-exposed		RR *	
		n	% defects	n	% defects		
Harlap et al 1975	Various	432	10.9	11.036	7.8	1.4	
Kullander e Kallen 1976	Various	112	12.5	4.904	12.9	1.0	
Goujard e Romeau-Roquette 1977	Various	830	1.8	10.157	1.6	1.1	

None of the results was statistically significant.

(a) including cortical-steroids and thyroidal hormones; (b) most probable memory bias; (c) controls and cases enlisted in different periods.

Retrospective cohort studies with internal controls

- Matsunaga and Shiota (1979): a very peculiar and interesting study carried out on 667 embryos not impaired after therapeutic abortion due to threatened abortion (vaginal blood loss). 130 of the embryos had been exposed to

progestins, whereas the other 537 had not. The rate of malformed embryos among exposed and non-exposed revealed to be 18.5% and 8.8% respectively, therefore suggesting an influence exerted by the drugs administered to the mothers. Hormones had all been given in the critical period for embryogenesis to develop malformations and the detected anomalies suggest the existence of chromosomal anomalies in a large number of cases. For instance, out of 667 investigated embryos, there were 27 holoprosencephalies, that is 4% of the total. This study clearly suggests that threatened abortion, requiring hormonal treatment, is per se a teratogenic risk factor. More exactly, it is the epiphenomenon of a pregnancy affected by congenital anomaly.

- Resseguie et al (1985): a study concerning clinical records of prenatal examinations at Mayo Clinic between 1936 and 1974. 988 cohort exposed to various progestins (mostly progesterone and 17-OH-progesterone) in the first trimester (middle 2nd month, and 742 at the beginning of the third trimester) mainly for the treatment of threatened abortion. 1,976 controls not exposed to progestins, matched as per sex, age and maternal equality. RR for congenital anomalies in general = 1.2 (CI 95%: 0.9-1.7). No statistically significant combination was noticed in the various groups of considered anomalies.

Follow-up studies on cases

- Money and Mathews (1982): follow-up of 12 women (16-27 years of age, already published case-report) with virilization symptoms at birth, due to exposure during pregnancy to progestins with androgen activity. They were within the norm as far as puberty, menarche and sex relationships.

G03DA – Pregnene derivatives

Medroxyprogesterone – G03DA02

This derivative steroid progesterone is available in Italy since 1979.

Literature review

- Schardein (2000): he collected a total of 2,808 exposures to medroxyprogesterone during pregnancy from various cohort studies. The number of congenital anomalies did not exceed the expectations. (Rawlings 1962, Burstein and Wasserman 1964, Goisis and Cavalli 1962, Lipp 1963, Tronconi 1963, Destro and Paolina 1964, D'Incerti-Bonini 1965, Cubesi 1969, Powell and Seymour 1971, Schwallie and Assenzo 1973, Nash 1975, Heinonen et al 1977, Dahlberg et al 1982, Resseguie et al 1985, Yovich et al 1988, Partdthaisong and Gary 1991).

Medical reports

- Eichner (1963): one female infant exposed in the first trimester with symptoms of virilization.
- Pruett (1965): 1 newborn exposed at 9-10 weeks showing oculo-vertebral syndrome.
- Eller and Morton (1970): one first trimester exposure to medroxyprogesterone and other drugs showing rachis-schisis and clubfoot.
- Lorbert et al (1979): 1 newborn exposed in the first trimester bearing mental retardation, facial dysmorphism and hypospadias.
- Dayan and Rosa (1981), FDA: 14 cases of ambiguous genitals exposed at various stages of pregnancy.

Cohort studies without controls

- Burstein and Wasserman (1964): of 174 exposures to 5-50 mg/day before week 12 of pregnancy, 1 single case of a female newborn with transitory clitoral hypertrophy.

Retrospective cohort studies with internal controls

- Rosa (1993), Michigan MSS: of 407 first trimester exposures, 15 newborns with major defects, 13 expected (RR = 1.1; CI 95%: 0.6-1.9). 7 newborns with cardiovascular abnormalities, 4 expected (RR = 1.8; CI 95%: 0.7-3.6).

Prospective cohort studies with internal controls

- Heinonen et al (1977), CPP: of 130 exposed during the early 16 weeks, 8 newborns had congenital anomalies (ARR = 1.4; CI 95%: 0.7-2.7).
- Yovich et al (1988): 366 newborns exposed from 5th - 7th week to the end of 18th week; 464 controls. 15 exposures with congenital anomalies, vs. 15 among controls (ARR = 1.3; CI 95%: 0.6-2.6).

Hydroxyprogesterone – G03DA03

This is a time-lag progestin with low androgen activity, used in women at risk for miscarriage. It is synthesized by the ovary, adrenal gland and placenta. It is available in Italy since 1984.

Medical reports

- Foley and Wilson (1958): 1 anencephaly exposed in the first trimester.
- Wilkins et al (1958), Wilkins (1960), Leibow and Gardner (1960), Shaffer (1960), Lojodice et al (1964), Cope and Emelife (1965): 7 cases of masculinization of external female genitals in first trimester exposures.
- Burstein and Wasserman (1964): 1 newborn with hypospadias exposed in the first trimester.
- Dillon (1970 and 1976): 6 newborns exposed at different stages of pregnancy showing CNS and cardiovascular congenital abnormalities, and eye defects.
- Yalom et al (1973): 1 newborn with limb hypoplasia exposed in the first trimester also to diazepam.
- Roberts and West (1977): 1 newborn with multiple genital-urinary malformations, exposed in uterus to didrogesterone and 17-OH-progesterone from 8th to 20th week.
- Evans et al (1980): one female infant exposed in the first trimester showing clitoral hypertrophy, hydronephrosis and vesical dilatation due to urethral stenosis. Another female infant exposed in the first trimester had ambiguous male genitalia.

Cohort studies without controls

- Reifenshtein (1958): of 62 newborns exposed between 6th and 25th week, only 1 born with strabismus and 1 with early closure of fontanel.
- McBride (1963): 120 exposures, no cases of masculinization of external genitals in female newborns.
- Serment and Ruf (1968): 1,500 exposures, 2 female infants with clitoral hypertrophy.

Prospective cohort studies with internal controls

- Heinonen et al (1977), CPP: of 162 exposed in the early 16 weeks, 6 newborns had congenital anomalies (ARR = 0.8; CI 95%: 0.4-1.8).
- Varma and Morsman (1982), London 1974-1980: 150 exposures from 6th-8th until 16th-18th week of gestation, 150 controls matched as per age and original pathology that in some cases had required hormonal treatment. One newborn with malformations in the treated group and 3 in the non-treated group. The overall incidence of malformations in the hospital where the study has been carried out was of 2.14%.

Progesterone – G03DA04

This is a natural progestin, available in Italy since 1981.

Medical reports

- 28 cases of masculinization of female external genitals in female infants exposed in uterus have been reported in literature.

Cohort studies without controls

- Rock et al (1985): of 75 newborns exposed in periconception period, 2 newborns (2.6%) had nonspecific congenital anomalies.
- Check et al (1986): 382 exposed pregnancies in the period of implantation, until at least week 14 (in some cases until week 34) of pregnancy to progesterone or 17hydroxy-progesterone. 133 pregnancies had also been exposed to clomiphene in follicular phase and 93 to human menopausal gonadotropins. 5 newborns with congenital anomalies (clubfoot, PS – also exposed to DES); DIV, pulmonary stenosis, onphalocele and Sprengle deformity (also exposed to clomiphene). Incidence of congenital anomalies: 1.3%.
- Michaelis et al (1983): 186 exposures to progesterone. No increase of congenital anomalies.

Case-control studies, nonspecific

- Greenberg et al (1977): 836 newborns with congenital defects, 836 controls. 19 exposures among cases, vs. no exposures among controls.

Case-control studies, specific

- Levy et al (1973): 76 newborns with transposition of the great vessels, 76 controls. 6 exposures in the early 6 weeks to antiabortifacient progestin therapy and 1 exposed to pregnancy hormonal test among cases, vs. no exposures among controls.

Prospective studies with internal controls

- Heinonen et al (1977), CPP: of 253 exposures to progesterone in the early 16 weeks, 9 newborns with congenital anomalies. ARR for any kind of defects = 0.7 (CI 95%: 0.3-1.4).

G03DB – Pregnadiene derivatives

Didrogestone – G03DB01

It is a progesterone derivative steroid, lacking any androgen and estrogen activity. It is available in Italy since 1978.

We have been unable to locate references on possible human reproductive effects of this agent.

Case reports

- Dillon (1976): 1 newborn exposed at 8-12 week of gestation, showing arthrogyposis.

Medrogestone – G03DB03

This progesterone derivative steroid is available in Italy since 1969.

We have been unable to locate references on possible human reproductive effects of this agent, or have we found any similar studies on laboratory animals.

Nomegestrol – G03DB04

This is a synthetic progestin lacking any androgen and estrogen activity, available in Italy since 1993.

We have been unable to locate references on possible human reproductive effects of this agent, or have we found any similar studies on laboratory animals.

G03DC – Estrene derivative

Norethisterone (Norethindrone) – G03DC02

It is a synthetic progestin, 19-nor-ethinil-testosterone derivative. It may exert masculinizing effects. Norethisterone is used in combination with ethinylestradiol for oral contraception. It is available in Italy since 1968.

Medical reports review

- Shardein (2000): review of 81 reported cases of masculinization of external female genitals in female infants exposed in uterus.

Medical reports

- Gardner et al (1970, 1971): 1 newborn exposed in the first trimester to a combination of mestranol and norethindrone revealed multiple defects possibly due to camptomelic syndrome. He also mentions 9 cases collected from the literature or personally reported.
- Pap and Gardo (1971): 1 exposure with camptomelic syndrome exposed to a mestranol-norethindrone combination in the early 3 weeks of pregnancy.

Cohort studies without controls

- Abramson (1958): 90 women were treated throughout pregnancy with 5-20 mg/day of norethisterone and their offspring did not reveal any masculinization outcome in external genitalia.
- McBride (1963): No genital masculinization in 25 female infants exposed at various periods of pregnancy

Prospective cohort studies with internal controls

- Heinonen et al (1977), CPP: of 132 exposures in the early 16 weeks, 11 newborns with congenital anomalies (ARR = 1.8; CI 95%: 1.0-3.2). All of the 25 exposures to mestranol-norethisterone were healthy newborns.

Tibolone – G03DC05

It is available in Italy since 1991.

We have been unable to locate references on possible human reproductive effects of this agent, or have we found any similar studies on laboratory animals.

G03AA-AB-AC-CA and G03D class conclusions: Possible malformation risks following exposure to female hormones have been fully investigated and apparently no risk has been found. Nevertheless, virilizing effects have been noticed (clitoral hypertrophy with/without labioscrotal fusion) in female infants exposed to high doses of progesterone and other synthetic progestins, in particular estrene derivatives (i.e. norethisterone and norethindrone) mostly used in gynecologic pathologies such as endometriosis. The risk to cause these defects is relatively low, in fact lower than 1%, except for norethisterone, revealing a risk

rate as high as 18.3% (Jacobson 1962). Masculinizing effects of progestins is dose-dependent and the low quantity of the drug in oral contraceptives, in case of failure and consequent fetal exposure, does not cause virilization. The prenatal most susceptible period is prior to 10th week, exactly around 8th week of gestation. Hypotheses concerning an overall increased risk of malformations – in particular for hypospadias, congenital cardiopathies, limb hypo-agenesias, VACTERL syndrome and camptomelic syndrome, mostly figured out in the 1960s and 1970s – have not been corroborated by further studies (not even by deeper analysis of existing studies). We have been unable to find further reports to confirm the hypothesis of risk increase relevant, for instance, to camptomelic syndrome. Reported malformations have been in some cases more wisely interpreted as outcomes of the confounding factor that was associated with what has been generally called “threatened abortion”. Nonetheless the risk, although very low, cannot be excluded, in particular considering female hormones administered at high doses and/or over long periods, as it occurred in pregnancy tests (no longer used) or to prevent miscarriages. In short: a) the intake of oral contraceptives prior to conception or exposure to oral contraceptive in the early weeks of pregnancy does not increase the reproductive risk; b) regular intake of female hormones to prevent miscarriages is not recommended due to their therapeutic inefficacy and because of the possible, though minimal risk of malformations; C) progestins with androgens effects are associated to a specific dose-dependant and quite low risk of virilization to the female fetus.

G03B – Androgens

We know little about the effects of androgens on human pregnancies. Only a limited number of reports on exposures to testosterone and its derivatives, most of which no longer marketed.

Review of clinical reports

- Schardein (2000): 59 cases of masculinization of external female genitalia in female infants exposed to various androgens (danazol, metandriol, methyltestosterone, testosterone and normetandrone).

G03BA – 3-oxoandrostene (4) derivatives

Testosterone – G03BA03

It is the main androgen synthesized from testicle, ovary and adrenal gland, with a conclusive role in the virilization of the male external genitalia. It is available in Italy since 1959.

Case report (besides those reported by Schardein in the year 2000)

- Derwhurst and Gordon (1984): 1 female infant exposed to 12 mg/day of methyltestosterone between 5th and 12th week of pregnancy showing virilization of external genitalia.
- Reschini et al (1985): 1 female infant exposed up to 16 weeks of pregnancy to testosterone showing virilization of external genitalia, but uterus, tubes and ovary were normal as well as her secondary sexual development.

G03BB – 5-androstanone (3) derivatives

Mesterolone – G03BB01

G03BA and G03BB class conclusions: Prenatal exposure has been associated with masculinizing androgens in female fetuses, on the ground of about sixty medical reports (Schardein 2000), also when the administered dose was not causative of side effects for the mother (Van Wyk and Grumbach 1968). Female

pseudo-hermaphroditism has been reported. In some severe cases of ambiguous genitals the female infant may appear as a male with hypospadias and cryptorchidism. In most cases clitoral hypertrophy may be more or less evident showing or not showing labioscrotal fusion. Vagina, uterus and ovary appear to be normal. No further symptoms of virilization have been observed, as they are growing, nor precocious puberty and excessive body growth. Masculinization level depends on the administered dose and the period of exposure in pregnancy. Labioscrotal fusion only occurs in cases treated prior to week 13 of gestation (Grumbach et al 1959, Grumbach and Ducharme 1960). Clitoral hypertrophy may occur following exposure to androgens. In this case the genetic sex of the fetus should be known since there are no risks if it is a male. In case of a female the echography, even if well done, is to be considered not reliable for a diagnosis of possible masculinizing process.

G03G – Gonadotropins and other ovulation-inductors

Literature review

- Rosa (1990): this is the widest review of cohort studies without controls (it includes comments already made in other reviews, like for instance Asch and Greenblatt 1976 and Cornel et al 1989) to assess the incidence of NTD in a total of 5,637 pregnancies induced with drugs of various type. Studies not specifying the type of congenital defect and those having less than 25 outcomes have not been included. The review points out a NTD risk of 1.8 /thousand, that the author believes to be similar to the incidence found out among general population, there were surveys have been carried out.

Author	Newborns	NTD
Merrell 1966	2,082	3
Karow-Payne 1968	183	0
Greenblatt 1969	156	0
Baron 1975	92	0
Rutz-Valasco 1976	103	0
Bettendorf-Lehman 1976	64	0
Sas 1977	349	3
Scheumpflug	128	0
Giustini 1977	91	1
Hack et al 1979	240	0
Barrot 1979	59	0
12 more cohort studies	448	0
Goldfarb et al 1968	166	0
Hack et al 1972	104	0
Harlap et al 1976	225	0
Ahlgren et al 1976	148	1
Adaghi et al 1979	64	0
Kurachi et al 1983	935	2
Total	5,637	10
Rate per 1,000		1.8

- Shoham et al (1991): review of cohort studies without controls concerning 11,836 (see Table) induced pregnancies with:
 - **Clomiphene** (Mac Gregor et al 1968, Karow and Payne 1968, Goldfarb et al 1968, Ahlgren et al 1976, Harlap 1976, Hak et al 1971, Adashi et al 1979, Hak and Lunenfeld 1979, Kurachi et al 1983) for a total of 3,751 newborns, 40 of which with major malformations (1.1%) and 82 with

minor malformations (2.2%). No difference was noticed when compared with general population.

- **Gonadotropins** (Hak et al 1970, Spadoni et al 1974, Harlap 1976, Caspi et al 1976, Schwartz et al 1980, Kurachi et al 1983, Lunefeld et al 1986) for a total of 1,160 newborns, 25 of which with major (2.2%) and 38 with minor malformations (3.3%). No difference was noticed when compared with general population.
- **Clomiphene and/or gonadotropins also** subjected to **IVF-ET or GIFT** (Wood et al 1982, Trounson and Wood 1984, Seppala 1985, Andrew et al 1986, Frydman et al 1986; Cohen et al 1988, NPSU 1988).

Pregnancies	Total newborns	Major defects	Minor defects
Clomiphene induced	3,751	40 (1.1%)	82 (2.2%)
Gonadotropins induced	1,160	25 (2.2 %)	38 (3.3 %)
Subjected to IVF-ET or GIFT	6,925	129 (1.9%)	Usually not reported

The analysis in terms of specific malformations does not reveal any excess

- Van Loon et al (1992): review of data recorded by five registers of assisted fertilization (IVF-ET), to study the association of ovulation inductors with NTD. The review concludes: "Ovulation induction appears not to cause NTD risk in newborns".

NTD after IVF-ET in 5 registries	IVF-ET	NTD, n e x		IC 95%
		1.000		
England (1978-87)	1.586	6	3.8	1.65-8.08
France (1986-90)	4.890	11	2.2	1.09-3.90
Scandinavia (1981-87)	433	0	0	0.00-7.59
U.S.A. (1987)	7.869	2	1.1	0.19-3.58
(1988)	3.583	4	1.1	0.38-2.68
(1989)	4.736	6	1.3	0.55-2.71
(1990)	5.336	7	1.3	0.62-2.58
Australia (1979-88)	4.970	16	3.2	1.93-5.11
Total	27.40	52	1.9	1.39-2.44
	3			
Total - not including encephalocele			1.7	

Case report

- Melamed (1982): 1 infant born after ovulation induction with clomiphene and gonadotropins showed hepatoblastoma.
- Mandel et al (1994): 4 infants born after ovulation induction (clomiphene, gonadotropins and progestins) showed neuroblastoma.

Retrospective cohort studies with external controls

- Lancaster (1987), 1979-1986: 16 centers for in vitro fertilization recorded 1,696 newborns (live and still births) and 3 VIP for congenital anomalies. 37 fetuses/newborns with congenital anomalies: OR = 2.2 (CI 95 %: 1.5-2.9). 6 had spina bifida vs. 1.2 expected (RR = 5.0; CI 95%: 1.8-10.8). 4 had transposition of the great vessels vs. 0.6 expected (ARR = 6.7; CI 95% 1.8-17.7). 2 newborns had trisomy 21 and sirenomelia, respectively.

Prospective cohort studies with internal controls

- Kallen et al (2002): cohort study on all pregnancies following ovulation induction (mostly by means of clomiphene), not subjected to IVF and ascertained among all Swedish population between 1955 and 1999. Overall number of exposed newborns = 4,307; control cohort (not induced pregnancies) = 438,582. OR as per malformations (187 cases) = 1.2 (VI 95%: 1.1-1.40). The period of involuntary subfertility and the consequent multifarious analysis that considered this confounding factor was reported, along with the year of birth, maternal age and parity. The study revealed an OR of 1.1 (CI 95% 0.9-1.3). Without this analysis (that only partially corrects the weight of the confounding factor of fertility) the outcome would be positive. It therefore shows how important confounding factors are in such studies. There were 41 cardiopathies (9.5 per thousand) and 10 hypospadias (2.3 per thousand) among the observed malformations, but no NTD. The most evident result emerging in this study is the exceeding number of twin birth (OR = 4.3; CI 95%: 3.8-4.8), absolute risk = 5.9% to be attributed to agents used for ovarian induction rather than to confounding factors. Twin birth is higher, but not absolute, as for dizygotic twins.

Case-control studies, specific

- Mills et al (1990): cases = 571 mothers of NTD offspring; controls = (a) 546 mothers of offspring showing other malformations than NTD (to minimize memory bias) and (b) 573 mothers of healthy offspring. Exposures to ovulation inductors prior to conception (clomiphene 79%, gonadotropins 17% and bromocriptine 4%) have been evaluated by means of interviews carried out mostly three months after diagnosis (prenatal or neonatal) by "blind" interviewers. The study recorded 8 newborns with NTD out of 571 infants exposed to ovulation inductors shortly before pregnancy; 6 newborns out of 546 showed other congenital anomalies (OR = 1.3; CI 95%: 0.3-2.3) and 10 out of 573 were healthy newborns (OR = 0.8; CI 95%: 0.3-2.3). Exposures to ovulation inductors at any stage prior to pregnancy: 24 out of 571 with NTD vs. 17 out of 546 newborns with other congenital anomalies (OR = 1.4; CI 95%: 0.7-2.7) and vs. 23 out of 573 healthy newborns (OR = 1.1; CI 95%: 0.6-2.0).

G03GA – Gonadotropins

Chorionic gonadotropin – G03GA01

Human menopause gonadotropin (Menotropin) – G03GA02

This is a compound made of lyophilized human menopausal gonadotropin (hMG), standardized with human chorionic gonadotropin (hCG). It is used to treat sterility as ovulation inductor. It is available in Italy since 1998.

Urofollitropin –G03GA04

This is a hypophyseal hypothalamus hormone, a glycoprotein follicle-stimulating hormone obtained from the urine of menopausal women.

Follitropin alpha – G03GA05

Follitropin beta – G03GA06

It is available in Italy since 1987

Lutropin alpha – G03GA07

Case reports

- Caspi and Weinraub (1972): 1 child with sacrococcygeus teratoma, born to pregnancy induced by chorionic gonadotropin, menotropin and progestins.
- Bishai et al (1999): persistent primary hyperplasia of hyaline has been diagnosed to a girl aged 5. She had been exposed to clomiphene in the early weeks of gestation. Causal association is hypothesized by the authors in consideration of side effects of clomiphene on the ocular apparatus, as well as on adult sight (up to 10% of the cases) and on the appearing cataract in mice and rats. No side effects were reported on monkeys exposed to clomiphene in uterus.

Cohort studies without controls (not considered in the literature review reported above)

- Tyler (1968): 236 births of pregnancies induced by menotropin, 1 newborn with esadactyly.
- Alberman (1978): 150 births of pregnancies induced by menotropin are quoted in his book, from a study not previously published. 6 of the newborns had all different malformations, therefore suggesting a lack of risk.

Retrospective cohort studies without internal controls

- Kurachi et al (1983) (also used in metanalysis of clomiphene, see): the study was carried out in 9 Japanese hospitals where 3 major defects (2 clubfoot and 1LCA) were detected out of 213 exposures. The incidence being 1.4%, not different from the incidence observed in 30,033 infants born after non induced pregnancies (1.1%). No difference was noticed in the 935 births of pregnancies induced by clomiphene (1.0%). A bias is likely of under-ascertainment of congenital anomalies. It is worth mentioning that unlike other investigations, the present study has not revealed an increase in miscarriages.

Prospective cohort studies with internal controls

- Heinonen et al (1977), CPP: 2 exposures to gonadotropins in the early 16 weeks, both with no congenital anomalies.

Case-control studies, specific

- Werler et al (1994) (also used in metanalysis of clomiphene, see): 1.034 cases of NTD among which 11 exposed between 4-6 months before the date of the latest menstruation and 1 month afterwards. 32 exposures out of 4,081 controls (with other than cerebral malformations) matched as per mother age and year of birth. OR for cardiovascular defects = 1.8 (CI 95%: 1.0-3.1). The authors suggest that the result may be attributed to the confounding effect of sub-fertility.

G03GA class conclusions: The quality and number of available studies suggest a lack of association between gonadotropins and increased congenital anomalies. Ovarian stimulation with gonadotropins increases the number of multiple pregnancies (11-42% of twin births and 5-6% of triplet births) and it depends on the dosage as well as on the agent used (Scialli 1986). Controlled studies do not suggest an increase in miscarriages and the increase of ectopic pregnancies is not sure.

G03GB – Synthetic stimulants of ovulation

Cyclophenyl – G03GB01

Synthetic non-steroid anti-estrogen, that stimulates pituitary gonadotropin incretion. It is available in Italy since 1974.

We have been unable to locate references on possible human reproductive effects of this agent.

Studies on laboratory animals

- Eiher-Jensen (1968), Suzuki (1970): it is nonteratogenic in rats, but fetal reabsorption is higher.

Clomiphene citrate – G03GB02

It is a synthetic non-steroid anti-estrogen. It acts on hypothalamus and hypophysis inhibiting the negative feedback of estrogens on the release of hypothalamic GnRH, consequently increasing gonadotropin secretion. Its action length appears to be as long as up to 6 weeks in human body (Cunha et al 1987). It is available in Italy since 1966.

Systematic review

- Greenland and Ackerman (1955): Systematic review of cohort studies with internal controls and case-control to estimate the risk magnitude possibly due to clomiphene exposure. Eleven studies out of 16 have been collected after accurate research, back to march 1955 and in some cases the authors have been asked for further information. One study was not included (Karaback et al 1989), due to a difficult reconstruction of the data needed for the final metanalysis. The study by Cornel et al (1989) has been included despite the doubts on the real existence of internal controls, since it does not affect the final risk assessment. The analysis of 10 studies (Kurachi et al 1983, Cornel et al 1989, Czeizel 1989, Czeizel & Racz 1990, Cuckle & Wald 1982 and 1989, Milunsky et al 1990, Mills et al 1990 and 1991, Milli et al 1995, Werler et al 1994, Shaw et al 1995 and 1995, and Lammer & Cordero 1986) has given an overall evaluation of OR = 1.1 (CI 85%: 0.8-1.5), that is not heterogeneous among the various studies (p=0.16). OR evaluation is the same when related to both case-control and cohort patterns, or when exposures have been gathered by means of interview or medical records. Publication bias has been excluded. There is a major difference, instead, between the studies carried out in Europe - 3 studies with OR = 2.9 (CI 95%: 1.7-7.1) and those carried out in Japan and the US - 7 studies with OR = 0.9 (CI 95%: 0.61.3). The hypothesis of a different definition of the exposure period (periconception in Europe, while preconception exposure was considered in Japan and the US) is not convincing. In fact: (a) no differences have been noticed in one study that has compared preconception vs. periconception (Werler et al 1994), (b) a study on animal pattern has found an association only with preconception exposures and (c) clomiphene effects on the body can be persistent. In conclusion, this systematic review does not support the hypothesis of association between NTD and clomiphene use, and it excludes a risk rate exceeding the basic one of 50%.

Medical reports

- Dyson and Kohler (1973), Sandler (1973), Barrett and Hakim (1973), Field & Kerr (1973), Biale et al (1978): 7 newborns of clomiphene induced pregnancies showing anencephaly and one birth following 5 stimulating cycles (two of which after conception). These reports have suggested an association between clomiphene stimulation and anencephaly. See conclusions.
- Ylikorkala (1975): 1 newborn with esophageal atresia and tracheal fistula, cardiopathy, hypospadias and left renal agenesis, exposed in uterus to clomiphene and methyl dopa in the first trimester of pregnancy.
- Halal et al (1980): 1 newborn with multiple malformations (mega-urethra, hypospadias, imperforate anus, clubfoot, right renal agenesis and left renal hypoplasia with hydroureter, and persistent cloaca with rectum-vesical

fistula), exposed in uterus during the third week of pregnancy and for 5 days to 50 mg/day of clomiphene.

- Sceusa and Klein (1990), Haring et al (1992), Martinez-Roman et al (1995): 3 cases of arcadius acephalus exposed to clomiphene. They are evidently associated with the problem of twin birth induced by the agent.
- White et al (1990): 5 cases of infant tumors (3 neuroblastomas, 1 medulloblastomas; 1 neuroblastoma; 1 neuroectoderm tumor) in newborns of clomiphene-induced pregnancies.

Case studies, without controls

- Kobayashi et al (1991), Japan Children’s Registry (1985-1989): 6,236 cases of children suffering from tumors. A large number of tumors in newborns of induced pregnancies (the unspecified stimulant was presumably clomiphene). It is not clear how the prevalence of exposure has been assessed.

Type of tumor	Studied cases	Ovulation induction
Neuroblastoma	887	4 (p=0.001)
Cerebral tumors	556	1
Malignant Lymphoma	517	2
Reticuloendothelial tumors	174	2
Leukemia	2,301	0
Others	1,801	0
Total	6,236	9

Case-control studies, specific

- Reefhuis et al (2003): cases – 99 newborns with non-genetic craniosynostosis, controls – 777 healthy newborns. OR for craniosynostosis in offspring of clomiphene-induced pregnancies = 3.8 (CI 95%: 1.1-12.3), following artificial insemination = 4.2 (CI 95%: 0.8-9.4) and following assisted fertilization = 4.2 (CI 0.5-27.3). The possible confounding factor of twin births has not been evaluated (craniosynostosis is more frequent in twin births) and the exposure period is not clear.

Nested case-control studies, specific in the prospective cohort of all newborns

- Kallen and Otterblad Olausson (2003), Swedish MBR: Cases – 5,015 newborns with cardiovascular defects, among which 14 exposures to clomiphene in the first trimester, controls – 577,730 newborns, among which 874 exposed. OR for cardiovascular defects = 1.9 (CI 95%: 1.1-3.2). The authors reckon that the result may be attributed to the confounding factor of sub-fertility.

Conclusions: Clomiphene has been thoroughly studied. Reports of anencephalies in 1973 (all letters to Lancet) represent the first concerns regarding an association between ovulation stimulants and malformations, followed by a large number of debates, studies and reviews. The first evaluations (cohort studies without controls) had not uncovered an increased number of congenital anomalies and a couple of reviews suggested that there were no additional risks (Scialli 1986, Lunenfeld et al 1986), basing themselves on around 4,000 studied pregnancies. The majority of the authors also suggested that anencephaly and other NTD might be directly related to woman’s subfertility, rather than to clomiphene treatment (Dyson and Kohler 1973, Sandler 1973, Field and Ker 1974, James 1973,1974,1977, Ahlgren et al 1976, Schardein 1980). Paul Lancaster, who in 1987 (letter to Lancet) reported an excess of NTD (6 vs. 1.2 expected) among

1,697 pregnancies following IVF analyzed in Australia and New Zealand, has re-proposed the problem. Cornel et al (1989) and Czeizel (1989) made some comments (Letters to Lancet) soon after. The analysis of registries for birth defects in Northern Netherlands and Hungary pointed out an additional risk of NTD in clomiphene treated pregnancies. These reports have been followed by further studies and specific reviews on NTD and on birth defects in general. The following has been made clear once and for all. A) Additional malformations due to ovulation drug stimulants are not well grounded. B) If any NTD risk exists, this is very little. C) Studies showing positive results should be interpreted in the light of the confounding factor of subfertility. On the other hand, this is a varying factor since, for instance, two different type of patients are identified depending on whether they have just been treated with the stimulant or an assisted fertilization has followed. Two questions have not yet been answered: a) what if clomiphene exposure occurs after conception and b) is there any (minimum?) risk of infant tumors. Both these problems do not find their solution with the available information. The incidence of multiple births (and their related problems) following the use of clomiphene is of 8-12% (Goldfarb et al 1968). Most of them are multi-zygotic births, but also monozygotic pregnancies are increasing.

G03HA – Antiandrogens

Cyproterone acetate – G03HA01

Cyproterone acetate + ethynilestradiol – G03HB01

This is an antiandrogen that has anti-gonadotropin and progestin activity. Showing a competitive mechanism it prevents androgens from linking to cell receptors of the aimed organs. It also halts stimulating effects of androgens on various androgen-dependent organs. It is the most powerful available antiandrogen and it also has glucocorticoid and progestational activity. Its half-life is of 33-43 hours. Depot formulation remains over a period of 10-14 days. It is also available in combination with ethynilestradiol as oral contraceptive. It is available in Italy since 1975.

Case reports

- Statham et al (1985): one healthy newborn exposed to the drug (2mg/day) contained in an estroprogestin compound during the early 5-6 weeks of gestation. Authors report that Schering, the manufacturer, has recorded 6 cases of male healthy newborns exposed to 50 mg/day up to the 20th week following conception.
- Berg and Bakos (1987): 1 fetus (IVG) with no macroscopic congenital anomalies exposed to cyproterone (100 mg/day) during the early 17 weeks of pregnancy. Authors report that Schering, the manufacturer, has recorded 16 cases of healthy newborns exposed to low doses of cyproterone during pregnancy.

Cohort prospective studies without controls

- Vaudre-Williams et al (1993), TIS Paris: 139 exposures in the first trimester to cyproterone. 75 of them were exposed to 2 mg in combination with a progestin and 19 to 50 mg. None of the 33 exposed male newborns was showing feminization of genitalia. 3 newborns with congenital anomalies were reported exposed to the drug in combination: 1 IVG female fetus with gonad agenesis, 1 newborn with distorted prepuce, 1 newborn with malformation of the auditory canal.
- Jahn (1996): 44 male healthy fetuses inadvertently exposed in pregnancy to doses ranging from 2 to 100 mg/day.

Conclusions: The commonly used dosage of cyproterone in combination with estroprogestins is extremely low and far from the hypothesis of adverse outcomes on the development of external male genitalia. The use of cyproterone at high dosages in the treatment of hirsutism requires a much closer evaluation of exposure timing. If exposure occurs in the critical period (after the 7th week) for the differentiation of external genitalia the opportunity of a prenatal diagnosis to find out the fetus sex should be considered.

G03XA – Anti-gonadotropins and similar drugs

Danazol – G03XA01

This ethisterone derivative is a modified synthetic androgen. It has a low antiandrogen activity and it inhibits pituitary release of gonadotropins, consequently diminishing estrogen secretion. Patented in 1963.

Case review

- Rosa (1984) has collected 46 exposures to danazol occurred between 8 and 20 weeks of gestation (Duch and Katayama 1981, Schwartz 1982, Castro Magana et al 1981, Hamond & Talbert 1981, Peress et al 1982, Shaw and Farquhar 1984). 29 unknown outcomes: 7 miscarriages, 7 male newborns (1 with multiple unspecified congenital anomalies), 5 female newborns showing no virilization, 10 female newborns showing virilization (1 also with ovarian hernia). No cases of virilization have been reported among exposures prior to week 8 of pregnancy. All of them had been treated with 800 mg/day of danazol.
- Brunskill (1992), manufacturer Sterling-Winthrop: 129 exposures to danazol over pregnancy from the United Kingdom reported to Australian Drug Reactions Advisory Committee (ADRAC 1986, 1989, Kingsbury 1985: 4 cases) and to FDA (Rosa 1984) plus the case reported by Quagliarello & Alba Greco (1985). The outcome of 95 pregnancies is reported: 12 miscarriages, 23 VIP, 37 male healthy newborns, 34 female newborns with no evidence of virilization and 23 female newborns with evidence of virilization. No evidence of virilization has been noticed in female infants exposed prior to week 8 of pregnancy. Despite virilization has mostly occurred at high dosages of danazol, one single case had been exposed to 200 mg/day, only.

Medical reports (not included in 1992 Brunskill review)

- Wentz (1982): 1 female newborn exposed to 200 mg/day of danazol between 1st and the 6th week of pregnancy did not show any genitalia changes.
- Within a project of Monitoring the Italian Health Department reports one single case of pseudo-hermaphroditism in the female newborn to a woman with endometrial cyst exposed to 400 mg/day of danazol during 87 days following the date of the non-menstruation.
- Insiripong et al (1996): 1 healthy female infant exposed to danazol in the second half of pregnancy, due to maternal trombocytopenia.

Conclusions: All available clinical reports and danazol pharmacological properties suggest masculinization effects in female fetuses. Nonetheless, the risk rate evaluation is based on the exposure timing (8-13 week of pregnancy being the most critical) and on the dosage (outcomes reported only for 200 mg/day and over). Clitoral hypertrophy is frequent after week 14. Female fetus virilization may occur in about 50% of danazol exposures when the drug is administered over a longer period and between week 8 and 13 of gestation (Brunskill 1992). The available data do not suggest an association with nongenital malformations.

Gestrinone – G03XA02

This is a synthetic steroid hormone, progesterone antagonist. It is used in the treatment of endometriosis associated or non-associated with infertility. It is available in Italy since 1992.

We have been unable to locate references on possible human reproductive effects of this agent, or have we found any similar studies on laboratory animals.

Conclusions: There are no available studies in literature on the use of gestrinone in human pregnancies. In case of exposure the sole reliable point is the lack of an evident teratogenic action on laboratory animals (records provided by manufacturer for registration, not available in databases).

G04 – Urologic drugs

G04BD – Urinary antispasmodics

It is a group of synthetic anticholinergic agents and tertiary amines. They have relaxing, nonspecific effects on smooth muscles.

Flavoxate – G04BD02

It is available in Italy since 1988.

Cohort studies without controls

- Capra and Paggi (1972): 39 healthy newborns exposed in the first trimester to the drug used to prevent spontaneous abortion.

Oxybutynin – G04BD04

It is available in Italy since 1984.

We have been unable to locate references on possible human reproductive effects of this agent.

Studies on laboratory animals

- Edwards et al (1986): nonteratogenic in neither rats nor rabbits when administered a dose 10-15 times the therapeutic human dose and 1-120 times, respectively. It shows DIV in rats at toxic maternal doses (250 times the human dose).

Tolterodine – G04BD07

This is a competitive antagonist of cholinergic receptors. It is particularly used for urinary bladder. It is available in Italy since 1999.

We have been unable to locate references on possible human reproductive effects of this agent, or have we found any similar studies on laboratory animals.

G04BD class conclusions: There is no written evidence of specific studies concerning the use of these drugs in human pregnancy. In case of exposure the following points should be noticed: flavoxate has been little studied and there is a lack of teratogenic action on laboratory animals (records provided by manufacturer for registration, not available in databases).