Drug safety in pregnancy

Studying and communicating teratogenic risks

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RIJKSUNIVERSITEIT GRONINGEN

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Studying and communicating teratogenic risks

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Table of contents

General Introduction

Background

For a long time it was thought the placenta protects the foetus against all possible harmful influences.[1] Despite earlier publications on possible negative effects for the unborn child as radiation and measles,[2,3] it was the thalidomide tragedy at the end of the 1950s and early 1960s that increased the awareness of the fact that the placenta is not a perfect barrier to protect the foetus.[4] Thalidomide, prescribed for anxiety, insomnia, gastritis, tension and as an anti-emetic drug in pregnancy, was marketed as a safe drug for adults as well as for children.[5] But, despite the apparent absence of any toxic effect, it turned out to cause phocomelia and other congenital anomalies in thousands of children exposed in utero.[6-9]

The thalidomide case was the beginning of birth defects research, and the term teratogenicity came into use, meaning the ability to interfere with normal foetal development. Initially, research was focused on obviously noticeable defects. Later, it became clear that the manifestation of teratogenicity also includes infertility, spontaneous abortions, intra-uterine death, premature birth, low birth weight, pre-or postnatal growth delay, (neuro)behavioural disorders and organ function disorders.[4] The latter might be difficult to detect, like in the case of diethylstilbestrol (DES). Daughters of women using DES during pregnancy were more likely to develop clear cell adenocarcinoma in early adulthood.[10] Moreover, recently it was suggested that the sons of these daughters have an increased risk on hypospadias.[11] This illustrates not only the possible long-term effects of intrauterine exposure but it also shows our lack of knowledge of the mechanisms of teratogenic substances like some drugs.

In contrast, also some benefits of intrauterine exposure have been established over the years. The most obvious example is the protective effect of folic acid on neural tube defects and probably also on other anomalies.[12-14]

Studying safety of intrauterine drug exposure

Although it is generally recommended not to use medicines during pregnancy unless it is absolutely necessary, several studies show that up to 80% of women do take at least one type of drug during pregnancy.[15-18] Establishing the safety of drugs used by women of childbearing age is thus of high importance. The two options to establish this safety are animal studies and observational epidemiological studies in humans with data of pregnant women that were already exposed to the drug. The first is very useful in many cases, for example with isotretinoin where animal studies prevented a disaster in humans like thalidomide.[19] Unfortunately, results from animal studies can not simply be translated into

risks in humans. A drug can be teratogenic in one species while it has little or no effect in another, as was the case for thalidomide.[20] Teratogenic effects cannot be detected in human trials before marketing a drug. Not only because of the small numbers in these trials but also because women that might become pregnant are mostly excluded from these studies. This leaves us with the unfortunate situation that possible teratogenic effects are mostly discovered after the release of the drug on the market and also after it has been used by pregnant women.

Studying teratogenicity in humans and some limitations encountered

The diversity in numbers of both drugs and congenital anomalies, and therefore the many possible combinations between them which could all be subjects to study, is an enormous challenge for researchers in this field. To study possible associations between a drug and a congenital anomaly, large datasets with detailed information on drug exposure, the outcome and many other variables that might influence the study are needed. The low exposure rates in pregnancy of some drugs, like the disease modifying anti-rheumatic drugs (DMARDs), increase this challenge.[21]

Fortunately, much effort has been made over the years to collect data necessary to study drug-anomaly associations. In Groningen, the EUROCAT Northern Netherlands registry for congenital anomalies was established in 1981 to collect data on children and foetuses with anomalies in the region. Until 1996, all information about the pregnancy outcome, and the mother's condition, diseases and drug use was collected through the physician or midwife who reported the birth. Since 1997, data collection is extended with a questionnaire to the parents and pharmacy data of the mother which provides researchers with complete and detailed information about drug exposure.[22] Comparable registries are established in many locations all over the world. Still, one registry does not always have the number of cases needed to investigate specific associations. To tackle this problem, several drugs or defects can be joined and investigated as a group. Such groups can be formed based on drugs being from the same class or anomalies categorised according to the embryologic tissue of origin. Although grouping increases the numbers in the analyses, it is known that teratogenic effects cannot be predicted reliable from the class of drugs a specific compound belongs to, nor from the existing knowledge of pharmacology and toxicology. Chemical relationships are not generally predictive of teratogenesis as can best be illustrated by the structural relation between thalidomide and glutethimide: there is only a slight difference in structure but there is no evidence of teratogenicity of the latter. Furthermore, by joining drugs or defects an existing association can be diluted by the group

members and therefore overseen. Another solution for increasing the power of a study is collaboration of different registries. In Europe, over 40 registries participate in the EUROCAT network. Other examples of collaboration are the International Clearinghouse for Birth Defects Surveillance and Research (ICBDSR)[23] and the National Birth Defects Prevention Network (NBDPN)[24] in the US.

Counselling women of reproductive age about (anti)teratogenic effects

The first eight weeks of pregnancy are most critical for developing anomalies.[25] Although most Dutch pregnancies are planned thus reasoned behaviour can be planned as well, all pregnancies are only recognised after a few weeks. Therefore exposure of the foetus to teratogens can easily take place in that period. Furthermore, due to long half lives of certain drugs e.g. chloroquine for malaria prophylaxis (3-5 days), clomiphene to induce ovulation (5-7 days), and leflunomide to treat rheumatoid arthritis (2 weeks),[26] the drug can be still in the mother's body during early pregnancy even if it was used before pregnancy. Informing women of reproductive age about their drugs is therefore important.

Widespread preconception care is not present in the Netherlands. Therefore, most women only consult a health care provider about their pregnancy after actually getting pregnant. But, due to the unique situation that most Dutch citizens are registered in one community pharmacy, which keeps their medication history, pharmacists can use this relationship to provide certain populations with specific information. Besides the obvious role of the pharmacy to check whether a new prescription does not conflict with existing medication, pharmacies can also be pro-active and educate people on health issues that might be of interest to them. They can for example educate women of childbearing age about the risks of drugs they get prescribed. Moreover, since over 70% of the Dutch women take prescribed contraceptives before their first pregnancy,[27] they can reach even those women that do not use other drugs but do visit their pharmacy regularly to collect their contraceptives. Thus, in spite of the lack of organized preconception care, education about teratogenic risks of drugs but also about the protecting effect of folic acid on birth defects can be organized through pharmacies. To be able to do so, pharmacies have to develop strategies to implement this form of patient education in their daily practice.

Objectives and outline of the thesis

From the foregoing it is clear that, for establishing the risks and benefits of intrauterine exposure, epidemiological studies have to be performed. Fortunately, ongoing data

collection provides the possibility to do so. Nevertheless, broadening data collection methods or collaboration might improve the possibilities of these studies. Subsequently, new insights should be communicated with the population that actually needs the information, e.g. the women of reproductive age. Therefore, three objectives are formulated in this thesis:

A. To study risks and benefits of intrauterine exposure in relation to congenital anomalies;
B. To investigate the strengths and limitations of current available datasets with respect to studying associations between intrauterine exposure and congenital anomalies;
C. To investigate if and how pharmacies can counsel women of reproductive age about decreasing risks on congenital anomalies, for example by using folic acid supplements.

This thesis consists of three consecutive parts, each dealing with one of the objectives. The intake of folic acid is a repeating theme throughout the thesis. In the first part of the thesis, part A, periconceptional exposure to drugs and/or folic acid is topic of study. After describing drug use among pregnant and non-pregnant women (chapter 1) three studies are presented with folic acid as communal factor (chapters 2 to 4). The last chapter of this part of the thesis describes the association between preconceptional clomiphene use and the occurrence of hypospadias among male offspring. This study demonstrates the limited methodological possibilities if exposure is rare and numbers are small.

In part B, improvements to overcome certain limitations of current available data are explored. In chapter 6, the potential of the EUROCAT network to study teratogenicity of drugs is described. Chapter 7 describes the actions that were undertaken to gather data on non-malformed children as controls for the births in the EUROCAT Northern Netherlands registration.

The following goal is to reach women of childbearing age and to educate them about teratogenic risks. Studies on the possibilities to do so are clustered in part C of the thesis, all with folic acid education as topic. First, determinants of the use of folic acid were studied to provide health care providers with tools to educate this specific population (chapter 8). Subsequently, a pilot study is presented on the introduction of pro-active patient education in daily pharmacy practice (chapter 9) and the effect of this intervention on the knowledge and use of folic acid among the target population (chapter 10).

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Part A

Drug use in pregnancy; studies on intrauterine exposure and foetal risk

Chapter 1

Drug use by pregnant women and comparable non-pregnant women in the Netherlands with reference to the Australian classification system

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Abstract

Objective: The interpretation of the available studies in pregnancy is often hampered because it is unclear to what extent women changed their drug choice. Here, we describe drug use in pregnancy, and compare drug use of pregnant women with non-pregnant women with respect to possible teratogenicity.

Study design: A cross-sectional study based on pharmacy records from 1997-2001 was performed. Pregnant women and matched non-pregnant women (same physician and age) were identified. Prescriptions were set against the Australian risk classification.

Results: 35% of all prescriptions for non-pregnant women were safe in pregnancy (Australian classification A), of 14% the risk was unknown (B1, B2), 49% were potentially harmful drugs (B3,C,D,X), and for 3% no classification was available. For pregnant women these figures were 86, 3, 10 and 2%. In non-pregnant women the highest percentages of prescriptions for unsafe drugs were for psycholeptics (99% not classified as safe), psychoanaleptics (100%), anti-inflammatory/anti-rheumatic products (100%), antihistamines (94%), antacids/anti-ulcer drugs (81%), antiepileptics (100%), beta-blockers (100%), systemic antimycotics (100%), antiprotozoals (97%), diuretics (100%) and immunosuppressives (100%). In pregnant women this pattern was comparable, except for antihistamines (22%) and antacids/anti-ulcer drugs (3%).

Conclusion: We conclude that many drugs used by non-pregnant women should be avoided in pregnancy, and that pregnant women indeed do so. However, for some drug groups the available safe alternatives are limited.

Introduction

Since the thalidomide disaster, treatment with drugs during pregnancy has been a reason for concern. Potential teratogenic risks of treatment, and potential risks for the mother of no treatment have to be weighed each time drug therapy is considered during pregnancy. Several surveys suggested that more than 80% of the women use at least one type of drug during pregnancy.[1-4] Vitamins, iron preparations, analgesics, anti-emetics and antacids were the most widely used drugs, but also nervous system drugs, respiratory drugs and cardiovascular drugs were sometimes used.[1-9]

Several classification systems place drugs in risk groups regarding their known or suspected adverse effects on the unborn child. Examples of these systems include the FDA (Food and Drug Administration) Classification, Swedish Classification, Australian Classification and TERIS risk ratings. Classifications like these may provide guidance to health care professionals and patients in the decision whether or not to use a certain drug during pregnancy, but may also be used in epidemiological research to study drug choice during pregnancy. A study among Danish women showed that 40.9% of all prescriptions during pregnancy were for drug classified as safe, 26% as potentially harmful, and of 28.7% the risk was unknown.[10] Several studies showed that the proportion of women using potentially harmful drugs during pregnancy is lower than just before pregnancy. An example is a study among Dutch women that showed that more than 75% of the women who used a drug classified as potentially harmful before pregnancy did not continue using that drug during pregnancy.[11]

These examples clearly show that drug choice is limited for pregnant women. In a recent study, 486 drugs approved in the United States by the FDA between 1980 and 2000 were examined with respect to their teratogenic risks using the TERIS risk rating system. It was found that the teratogenic risk was still undetermined for 91.2% of drugs approved in this period.[12] This means that for most of the examined drugs inadequate information is available to determine whether the benefits exceed the teratogenic risks. The interpretation of this high proportion of drugs with inadequate information is somewhat hampered, however, because it is not clear to what extent these drugs are needed by women at the fertile age. To gain more insight in this aspect, we evaluated drug use of pregnant women and non-pregnant women, with reference to the Australian classification system. The specific aims were to describe drug use in pregnancy, and to compare drug use of pregnant women with non-pregnant women with respect to possible teratogenicity.

Methods

Population

In the Netherlands, people commonly register with one pharmacy, and obtain all their medication from that pharmacy, so that a complete medication history of an individual is available in the pharmacy dispensing records. Drugs prescribed by specialist doctors to non-hospitalised patients are supplied by community pharmacies and thus included in the dispensing data. Drugs used during hospital stay (inpatients) and over-the-counter medication are not included. Previous studies have demonstrated that dispensing data from Dutch pharmacies offer a valid survey of the use of prescription drugs.[13] This study was performed with pharmacy dispensing data from the InterAction database, which is part of the collaboration between community pharmacists in the northern part of the Netherlands and the University of Groningen.[14] The InterAction database comprises prescriptions from 1994 to 2002, and covered a population of 300,000 people in 2002. Registration is irrespective of health insurance (including people that are not insured), and is thus considered representative for the general population. For this particular study, the prescriptions of a population of approximately 215,000 people were used. Data from the remaining pharmacies could not be used because the address-number used for the identification of the mothers was not supported by their computer system. To identify mothers, first all children aged 0-5-years in 2001 were selected in the database. Per child, the female person 15-50 year older than the child with the same address number was considered to be the mother, providing there were no other female persons 15-50-year older with the same address number. A validation of this method using a 3-year period showed that 65% of all mothers of newborns in the region could be identified, with a correctness of 99%.[15]

Analyses

All drugs were classified according to the Anatomical Therapeutic Chemical (ATC) classification system of the World Health Organization.[16] Individual drugs were defined as different full ATC-codes (e.g. J01CA04, amoxicillin), drug groups were defined as ATC-codes at 2nd level (e.g. J01, antibiotics for systemic use). Drug use was examined in two groups of women: 1) pregnant women and 2) non-pregnant women randomly 1:1 matched by General Practitioner and age (difference ≤ 5 years). Period of pregnancy was defined as the period from birth minus 270 days till birth[17], and was divided in trimesters of 90 days. Non-pregnant women were defined as women who where not identified as pregnant in any

Table 1: The Australian categorisation of risk of drug use during pregnancy

Category	Description
A	Taken by a large number of women without any proven increased risk on the foetus.
B1	Taken by a limited number of women without any proven increased risk on the foetus. Animal studies haven not shown an increased risk.
B2	Taken by a limited number of women without any proven increased risk on the foetus. Studies in animals are inadequate or may be lacking, but available data show no evidence of an increased risk of foetal damage.
B3	Taken by a limited number of women without any proven increased risk on the foetus but animal studies have shown an increased occurrence of foetal damage the significance of which is considered uncertain in humans.
C	Drugs which may have harmful effects on the human foetus or neonate without causing malformations. These effects may be reversible.
D	Drugs which have caused or are expected cause, an increased risk of human foetal damage. These drugs may also have adverse pharmacological effects.
X	Should not be used during pregnancy or when pregnancy is possible.

part of the corresponding pregnancy period of the corresponding pregnant woman. For each non-pregnant woman the period of analysis was equal to the period of pregnancy of the matched pregnant woman. The extent of drug use was expressed as the prevalence rate, and calculated as the number of women per 100 women to whom at least one prescription of the drug or drug group was dispensed in pregnancy, or in the corresponding study period for the matched non-pregnant women.

To compare the use of drugs between pregnant women and non-pregnant women in relation to teratogenic risks, drugs were classified according to the risk categories of the Australian Drug Evaluation Committee (see table 1).[18] The Australian Classification was chosen because this classification is internationally well-known, and the Australian Drug Evaluation Committee has a current version of the classification available online. For all drugs not included in the Australian classification, for instance drugs that are not on the market in Australia, the risk classification of the Health Base Foundation, the Netherlands, was consulted.[19] Next, the following groups were constructed: safe drugs (class A), drugs with an undetermined risk (B1 and B2), potentially harmful drugs (B3, C, D, and X) and drugs that were not included in the classification. For the 35 drug groups that were most widely used by non-pregnant women, the proportion of prescriptions for safe, undetermined, potentially harmful and unclassified drugs was determined both for non-pregnant women

and pregnant women. We used the 35 drug groups most widely used by non-pregnant women as a starting point because these are the drug groups that women at the fertile age are likely to use if there are no limitations for drug use. Oral contraceptives and ovulation stimulating drugs were excluded from these analyses.

Results

Between 1995 and 2001 7500 pregnant women could be identified in the database. Table 2 shows the prevalence of drug use by these pregnant women: 85.6% had used at least one drug during pregnancy. If folic acid, iron preparations and vitamins were not taken into account this figure was 69.2%. More than half of all women used between 1 and 3 different types of drugs during pregnancy and 16.0% used 5 or more different types.

As can be seen in table 3, drug use increased with trimester: 45.3% of all pregnant women used drugs in the first trimester, 57.1% in the second, and 70.3% in the third. Folic acid was the most widely used drug in the first trimester (8.4%), but was also used in other trimesters. Meclozine/cyclozine (3.6%) was a top-10 drug in the first trimester only. The use of iron preparations (7.7, 30.6 and 44.4%) and antacids (2.2, 9.5, and 18.5%) both strongly increased with duration of pregnancy. Other top-10 drugs include dermatologicals, gynaecological preparations, lactulose, salbutamol and paracetamol; the use of these drugs remained approximately constant during pregnancy.

For 6489 of the 7500 pregnant women it was possible to find a non-pregnant woman in the same age group with the same GP. Table 4 shows drugs used by both non-pregnant women and pregnant women, with reference to the Australian classification system. In the

Table 2: Prevalence of drug use during pregnancy (n=7500 women)

	% of women (number)
All pregnant women in the study	100 (7500)
Pregnant women using drugs[1]	85.6 (6423)
1 type of drug	23.2 (1742)
2 types of drugs	21.2 (1588)
3 types of drugs	14.5 (1091)
4 types of drugs	10.7 (803)
5 or more types of drugs	16.0 (1199)

[1]Type of drug defined as full ATC-codes (e.g. J01CA04, amoxicillin)

Table 3: Ten most widely used drugs per trimester (n=7500 women)

Trimester 1		Trimester 2		Trimester 3	
Drug	% of women (n)	Drug	% of women (n)	Drug	% of women (n)
Folic acid	8.4 (632)	Iron preparations	30.6 (2298)	Iron preparations	44.6 (3343)
Iron preparations	7.7 (579)	Antacids	9.5 (716)	Antacids	18.5 (1391)
Amoxicillin	3.8 (286)	Amoxicillin	6.2 (463)	Amoxicillin	7.9 (595)
Meclozine with cyclozine	3.6 (270)	Folic acid	5.2 (393)	Folic acid	7.9 (590)
Miconazole (gyn)	3.0 (224)	Miconazole (gyn)	4.8 (360)	Miconazole (gyn)	4.9 (368)
Antacids	2.2 (162)	Clotrimazole (gyn)	2.7 (200)	Clotrimazole (gyn)	2.8 (211)
Miconazole (derm)	1.8 (137)	Lactulose	1.9 (146)	Lactulose	2.6 (197)
Triamcinolone (derm)	1.5 (114)	Miconazole (derm)	1.9 (142)	Miconazole (derm)	2.1 (158)
Lactulose	1.5 (113)	Emollients and protectives	1.6 (122)	Lidocaine (antihemorrhoidal)	1.6 (118)
Salbutamol	1.4 (108)	Salbutamol	1.4 (103)	Paracetamol	1.5 (110)
Any drug	45.3 (3395)	Any drug	57.1 (4282)	Any drug	70.3 (5273)

gyn = gynaecological; derm = dermatological

majority of drug groups the percentage of safe drugs was higher among pregnant women than among non-pregnant women. Overall, 35% of all prescriptions for non-pregnant women were classified as safe for use in pregnancy (Australian classification A), 14% were classified as drug with an unknown risk (B1,B2), 49% were potentially harmful drugs (B3,C,D,X), and for 3% the drug was not included in the classification. Among pregnant women these figures were 86, 3, 10 and 2% respectively, or 77, 5, 15 and 3% when folic acid, iron preparations and vitamins were not taken into account. Taking women as unit of analysis instead of prescriptions results in a comparable pattern: 37% of all non-pregnant women received at least one drug classified as safe, 17% risk unknown, 62% potentially harmful, and 5% not classified, compared to 83%, 9%, 21% and 6% of the pregnant women (data not shown).

Also shown in table 4 is that in non-pregnant women drug groups with the highest percentages prescriptions for unsafe drugs were psycholeptics (99% of the prescriptions not classified as safe), psychoanaleptics (100%), anti-inflammatory and anti-rheumatic products (100%), systemic antihistamines (94%), drugs for acid related disorders (81%), antiepileptics

Table 4: Drugs used by non-pregnant women (n=6489) and pregnant women (n=7500), with reference to the Australian classification system (contraceptives and ovulation stimulation drugs excluded)

		Non-pregnant women						Pregnant women					
ATC	Drug group	N of women	N of presc	% A	% B1 B2	% B3, C,D, X	% NC	N of women	N of presc	% A	% B1 B2	% B3, C,D, X	% NC
N05	Psycholeptics1	534	2778	1	14	85	0	227	540	2	4	94	0
N06	Psychoanaleptics2	322	1835	0	16	84	1	99	367	0	11	89	0
M01	Anti-inflammatory and anti-rheumatic products (incl NSAIDs)	935	1690	0	1	99	0	197	268	0	0	100	0
J01	Antibacterials for systemic use	1038	1635	37	11	51	0	1545	2237	82	5	12	0
N02	Analgesics	420	1006	52	2	46	0	323	448	89	0	10	0
D07	Dermatological corticosteroids	577	994	85	0	15	1	643	973	93	0	7	0
R03	Anti-asthmatic drugs	269	967	44	7	49	0	291	912	54	3	43	0
R06	Systemic antihistamines	421	839	6	84	10	1	614	1145	78	12	10	0
R01	Nasal preparations	364	686	46	5	48	0	413	658	55	1	43	1
A02	Drugs for acid related disorders	202	572	19	33	48	0	1829	5119	97	2	1	0
D01	Dermatological antifungals	332	509	71	27	0	2	598	762	83	13	0	4
D02	Emollients and protectives	246	483	97	0	0	3	372	807	99	0	0	1
S01	Ophthalmologicals	263	431	70	2	14	15	259	400	70	2	12	17
G01	Gynaecological antiinfectives / antiseptics	293	397	91	6	0	3	1184	1811	97	1	0	2
R05	Cough and cold drugs	289	384	86	13	0	2	324	386	84	16	0	0
A06	Laxatives	145	376	94	0	3	2	473	905	92	0	1	7
A03	Drugs for functional gastrointestinal disorders	199	363	50	48	0	2	114	145	60	40	0	0
N03	Antiepileptics	54	356	0	3	97	0	19	92	0	7	93	0
C07	Beta blocking agents	124	341	0	1	99	0	49	130	0	0	100	0
B03	Antianemic preparations	165	335	100	0	0	0	4303	12561	100	0	0	0
A10	Drugs used in diabetes	38	265	65	0	35	0	47	244	100	0	0	0

Table 4 (continued): Drugs used by non-pregnant women (n=6489) and pregnant women (n=7500), with reference to the Australian classification system

		Non-pregnant women						Pregnant women					
ATC	Drug group	N of women	N of presc	% A	% B1 B2	% B3, C,D, X	% NC	N of women	N of presc	% A	% B1 B2	% B3, C,D, X	% NC
H02	Systemic corticosteroids	126	219	100	0	0	0	74	127	100	0	0	0
H03	Thyroid therapy	40	218	89	0	11	0	83	423	95	0	5	0
D06	Dermatological antibiotics	162	201	32	32	36	0	135	154	38	25	36	0
D10	Anti-acne preparations	96	191	60	5	6	30	72	98	62	4	0	34
A01	Stomatological preparations	139	175	75	0	0	25	129	143	72	0	0	28
J02	Systemic antimycotics	110	170	0	0	100	0	21	21	0	0	100	0
S02	Otologicals	93	158	62	0	0	38	98	127	62	0	0	38
A11	Vitamins	48	139	100	0	0	0	68	113	100	0	0	0
A07	Antidiarrheals and intestinal anti-inflammatory agents	69	128	37	0	61	2	73	162	20	0	65	15
J07	Vaccines	106	125	58	42	0	0	58	64	56	44	0	0
P01	Antiprotozoals	89	114	3	81	17	0	40	49	8	80	12	0
C03	Diuretics	34	107	0	0	100	0	13	16	0	0	100	0
L04	Immunosuppressive agents	9	102	0	0	100	0	5	23	0	0	100	0
D11	Miscellaneous dermatologicals	59	91	10	11	2	77	70	107	14	0	1	85
	Other drug groups	501	1181	33	25	32	10	994	2118	57	10	22	11
	All drug groups	4535	20561	35	14	49	3	6423	34655	86	3	10	2

N means number, presc means prescriptions. Letters denote the Australian risk categorisation (see table 1), NC means no classification available. Due to rouding percentages do not always add up to 100%.

[1] Psycholeptics include antipsychotics, anxiolytics, hypnotics and sedatives

[2] Psychoanaleptics include antidepressants and psychostimulants

(100%), beta blocking agents (100%), systemic antimycotics (100%), antiprotozoals (97%), diuretics (100%) and immunosuppressive agents (100%). In pregnant women this pattern was comparable, except for systemic antihistamines (22%) and drugs for acid related disorders (3%).

Discussion

The main findings of this study are that pregnant women tend to use safer drugs than non-pregnant women (overall 86% and 35% of all prescriptions were classified as safe, respectively), but that for several drug groups nearly all prescriptions for both pregnant and non-pregnant women were unsafe.

The major strengths of this study are the population based coverage, the prospective recording of pharmacy dispensing data, and the completeness of pharmacy data in comparison with other data sources, such as registration of general practitioners. An important limitation of our study design is that our exposure definition is based solely on the day of dispensing the drug from the pharmacy, and we have no information about actual drug use. Other weak points of the study are the absence of over-the-counter and medication used during hospital stay. In addition, it is likely that there has been some misclassification of pregnant women (identified pregnant women were not truly pregnant), and of non-pregnant women (women identified as non-pregnant were pregnant). A validation of our identification method showed that 99% of the pregnant women were correctly identified, suggesting the first type of misclassification will be rather small.[15] However, because our identification method is only able to identify 65% of all pregnant women in a region - thus leaving 35% unidentified - it is likely that there will have been pregnant women classified as non-pregnant. Another type of misclassification will occur because many pregnancies will last 1-2 weeks shorter or longer than the 270 days we assumed; a prescription issued in the first week of pregnancy according to our database, for example, could be issued just before pregnancy in the actual situation. All types of misclassification mentioned above will result in a underestimation of a possible difference between pregnant women and non-pregnant women. Finally, because we used pharmacy data to identify pregnant women, selection towards drug-using women might be expected; the validation of the identification method showed that this selection was limited, however.[15]

The usefulness and reliability of systems for the classification of drugs for teratogenic risk is currently a topic of debate.[20-22] The major classification systems are the Swedish system, the Australian system and the FDA system. These systems use almost the same

codes (A, B, C, D and X), but with different contents and sense; the FDA category B, for example, is to a certain extent comparable with Australian and Swedish category C, which obviously easily leads to confusion. Moreover, in a recent study Addis *et al* showed that only 26% of the drugs common to all three systems were placed in the same risk category.[20] Although risk classification systems were set up to facilitate drug prescribing for pregnant women, the use of different categories of drugs have been proven to be ambiguous and difficult to evaluate, both for professionals and patients.[22] Therefore it is currently recommended that ratings of drugs for use in pregnancy should be replaced by narrative statements that summarise and interpret the available studies, and provide estimates of teratogenic risks.[21,22] These proposed narrative statements are suitable for counselling at individual level, but not for assessing and comparing risks at group level. For describing drug utilisation patterns at an aggregated level, therefore, the risk classification systems are still of great value.

Overall 86 percent of all pregnant women used at least one drug during pregnancy. As demonstrated in table 3 the most widely used drugs during pregnancy were for presumably ordinary conditions like anaemia, nausea and infections. The predominant use of vitamins, iron preparations, analgesics, antiemetics and antacids was also found in previous studies.[1-9] The folic acid use in the first trimester may seem low (8.4%), but it has to be kept in mind that folic acid is also available over-the-counter, and our database includes only prescribed folic acid. Analyses like these are useful to gain insight in drug use during pregnancy. However, by studying pregnant women only, it is not possible to determine to what extent women are hampered in their drug choice by pregnancy. As can be seen in this and other studies, pregnant women tend to avoid potentially harmful drugs, and therefore it is necessary to include non-pregnant women in the study as well.[11] A number of surveys have included the trimester prior to conception in their analysis, but as at least a part of the pregnancies is planned (80% in the Netherlands), also this period is likely to be biased by an altered drug choice behaviour.[6,10,11,23]

The fact that only 35% of the prescriptions used by non-pregnant women are considered safe in pregnancy clearly indicates that many women have to reconsider the choice of their drugs in case of a pregnancy. Indeed 86% of all prescriptions for pregnant women were for safe drugs, indicating that pregnant women choose other drugs. Several problems are connected to this shifting, however. First, not for all situations safer alternatives will be available. On the assumption that pregnant women will choose a safe drug when available, the low percentages of prescriptions for safe drugs in the groups of psychotropic medication, anti-inflammatory and anti-rheumatic drugs, antiepileptics and

several cardiovascular drug groups suggest that these groups are problematic. Second, the alternatives might not always be fully therapeutic equivalent. In the group of systemic antibiotics for example, drugs may not be interchangeable due to different microbiological spectra. Third, pregnant women may be denied relatively new drugs. In the group of acid related disorders, for example, non-pregnant women frequently used proton pump inhibitors such as omeprazole or pantoprazole (both classified as B3, undetermined risk), whereas pregnant women almost exclusively used ordinary salt combinations (all classified as A, safe). These examples illustrate that shifting does not always offer a perfect solution. It is not inconceivable that for some drug groups the problems described above are of such magnitude that women decide not to become pregnant.

It should be noted that the category potentially harmful does not mean that these drugs are definitely not safe; a suspicion of possible adverse effects is enough to classify a drug as potentially harmful. It is evident that observational studies may be of great value to gain more insight in the safety of these drugs. The finding that 14% of prescriptions used by pregnant women were not classified as safe shows that, if we put enough effort in the registration of exposure and outcomes, these drugs are used frequently enough to gain some new safety information. It is promising that this is done increasingly often, for instance in the case of omeprazole, metoclopramide, venlafexine and fluoroquinolones.[24-27]

We conclude that many drugs used by non-pregnant women should be avoided in pregnancy, and that pregnant women indeed do so. Our findings suggest, however, that for a number of drug groups, under which psychotropic medication, anti-inflammatory and anti-rheumatic drugs, antiepileptics and several cardiovascular drug groups, the available safe alternatives are limited.

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Chapter 2

Folic acid reduces the risk on neural tube defects as well as other birth defects - a registry based case-control study in the Netherlands

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Abstract

Background: Besides neural tube defects (NTDs) folic acid (FA) might also protect against heart anomalies, oral clefts, urinary tract anomalies, limb reduction defects, omphalocele and anal atresia. We investigated the association of FA exposure and the occurrence of these FA sensitive defects.

Methods: With data from the EUROCAT Northern Netherlands registry, case-control analyses were performed. Cases were all isolated FA sensitive defects (N=2057) whereas controls were defined as children and foetuses with no FA sensitive defect present (N=4621). Exposure was defined as daily use of at least 0.4 mg of FA during the FA sensitive period of the foetus and two weeks prior to this period. Since not all anomalies origin in the same weeks of foetal development, this sensitive period differed per defect.

Results: This study shows a protective effect of FA for the group of FA-sensitive anomalies as a whole, but this effect is not significant anymore after adjusting for maternal age and year of birth. Furthermore, a significant effect is found for the heart anomalies in particular. The odds ratios for NTDs, urinary tract anomalies and limb reduction defects are, although not significant, indicative for a protective effect of FA.

Discussion: Our results support the positive findings of the effect of FA in other studies. This study also stresses the importance of availability of significant numbers of affected births to perform valid studies in this field.

Background

The protective effect of folic acid (FA) on neural tube defects (NTDs) is known since the early 1990s.[1,2] More recently, many studies were published on the association between FA or multivitamin use and other defects namely heart anomalies, oral clefts, urinary tract anomalies, limb reduction defects, omphalocele, and anal atresia. Some studies report a protective effect of FA or multivitamins, while others do not, as shown in an overview of Botto *et al*.[3] Using data from EUROCAT Northern Netherlands, we have investigated the possible preventive effect of FA on all seven groups of malformation mentioned above.

Methods

Case-control analyses were performed using data of all life births, still births and abortions in the EUROCAT Northern Netherlands registry between 1981 and 2000 (N=7852). The following defects were considered FA sensitive: NTDs, heart anomalies, oral clefts, urinary tract anomalies, limb reduction defects, omphalocele and anal atresia. Within these groups, some subgroups were investigated as well: spina bifida, conotruncal heart anomalies, cleft lip with or without cleft palate and cleft palate without cleft lip. Cases were defined as having an isolated FA sensitive birth defect (N=2057) and controls as all children and foetuses with no FA sensitive anomaly present, including all chromosomal and monogenic disorders (n=4621). Births with a non-isolated FA sensitive defect were excluded from the study, unless the defect is part of a syndrome (than included as control).

It takes about two weeks to reach an optimal FA blood level. We therefore defined exposure as daily use of at least 0.4 mg of FA during the FA sensitive period of the foetus and two weeks prior to this period. Because this sensitive period differs per defect, the exposure windows also differed per birth defect as shown in table 1. Exposure of controls was defined as daily use of at least 0.4 mg FA, from two weeks before conception until eight weeks after. ‘Not exposed’ was defined as reported no use of FA during the entire preconceptional and pregnancy period. Births with other exposure windows or with unknown FA use were excluded from this study (n=1931). Logistic regression was used to determine odds ratios (OR) and 95% confidence intervals (95%CI). An adjusted OR was calculated for all FA sensitive anomalies only, because of the small numbers elsewhere.

Results and discussion

In total we included 1614 cases with an isolated FA-sensitive defect with a known FA exposure. Of these cases, 5.0% were periconceptionally exposed to FA. The proportion

Table 1: Folic acid use and outcome measures in several groups of congenital anomalies.

	Exposure window*	FA / no FA	FA (%)	OR	95% CI
Controls	-2 to 8	221 / 2,912	7.1		
All FA-sensitive anomalies	variable per defect	80 / 1,534	5.0	0.69 0.77[a]	0.53 – 0.89 0.52 – 1.14[a]
NTDs	-2 to 4	16 / 213	7.0	0.75	0.44 – 1.26
Spina bifida	-2 to 4	8 / 123	6.1	0.64	0.31 – 1.33
Heart anomalies	-2 to 8	33 / 735	4.3	0.59	0.41 – 0.86
Conotruncal anomalies[b]	-8 to 8	20 / 581	3.3	0.45	0.29 – 0.72
Clefts	variable per cleft	18 / 301	5.6	1.09	0.66 – 1.80
Cleft lip (+/- cleft palate)	-2 to 7	20 / 243	7.6	0.98	0.61 – 1.57
Cleft palate (- cleft lip)	-2 to 10	5 / 57	8.1	1.60	0.63 – 4.04
Urinary anomalies	3 to 10	7 / 183	3.7	0.59	0.27 – 1.27
Limb reduction defects	-2 to 8	2 / 67	2.9	0.39	0.10 – 1.62
Omphalocele	-2 to 10	1 / 14	6.7	1.30	0.17 – 9.95
Anal atresia	-2 to 7	3 / 21	12.5	1.70	0.50 – 5.73

* expressed in weeks in relation to calculated conception date

[a] adjusted for age mother and year of birth.

[b] conotruncal anomalies: persistent truncus arteriosus, vsd (except perimembraneous), fallot, pulmonary valve anomaly, aortic valve stenosis, hypoplastic left heart syndrome, coarctation aortae, pulmonair artery anomalies and other anomalies of aortae.

exposed varies per birth defect. Of the 3133 included controls, 7.1% were exposed to FA (see table 1).

This study shows a protective effect of FA for the FA-sensitive anomalies as a whole group, but this effect is not significant anymore after adjusting for maternal age and year of birth. Furthermore, a significant effect is found for the heart anomalies in particular and the ORs for NTDs, urinary anomalies and limb reduction defects are, although not significant, indicative for a protective effect of FA. Therefore this study partly supports the positive findings of the effect of FA in other studies. If only chromosomal and monogenic anomalies are taken as controls (N=1402) instead of all non-cases, comparable results were found (not shown in table 1).

The strongest limitation of the study is the small numbers, especially regarding the exposure rates, and therefore the low power. For example, with our 229 cases of NTDs and an exposure of 7% among controls, we can detect a OR < 0.37 (α=0.05 and power of 80%).[4] The MRC trial found a 72% protective effect of FA on the recurrence1 of NTDs and

calculating from the prevalences of Czeizel and Dudas, a reduction of 43% is found for the occurrence of NTDs.[2] Several observational studies found a risk reductions between 40% and 60%.[5-8] Thus, since we only have the power to detect a reduction of >63%, the lack of significant findings on NTDs in our study might be more the result of our small numbers than of absence of a real effect of FA itself. Interpretation of our findings is therefore difficult and should take place with caution. Another limitation of the study is the lack of healthy controls. By using malformed controls recall bias is avoided but FA might have a till so far unknown protective effect on some defects included as controls in this study. This possible non-differential misclassification might bias our findings towards unity.

Although this study finds comparable results with other studies which is indicative for validity of this database, it mainly shows the importance of availability of significant numbers of births in order to execute valid studies in this field, especially if exposure rates are low as well.

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Chapter 3

Folic acid sensitive birth defects in association with intrauterine exposure to folic acid antagonists

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Abstract

Since the protective effect of folic acid (FA) on birth defects is well known, it is reasonable to assume intrauterine exposure to FA antagonists increases the risk on these defects. We have therefore performed case-control analyses to investigate the risk of intrauterine exposure to FA antagonists, using data on births from the EUROCAT Northern Netherlands registry from 1997-2002. Of the 815 cases, 11 were exposed to a FA antagonist compared to 16 of the 1,402 controls. For FA sensitive defects as a group the study showed no effect after exposure to a FA antagonist (OR=1.18, 95% CI: 0.55-2.57). We found no effect after exposure to a dihydrofolate reductase inhibitor (DHFRI) (OR 0.44, 95% CI: 0.12-1.54) but we did find a statistically significant effect after exposure to an antiepileptic drug (OR=3.45, 95% CI:1.04-11.48). This study supports the findings of various other studies on the teratogenicity of antiepileptics. An association between DHFRIs and FA sensitive defects was not found.

Introduction

Experimental as well as observational studies have shown a protective effect of folic acid (FA), alone or in a multivitamin, on neural tube defects (NTDs)[1,2] and other defects like heart anomalies,[3,4] orofacial clefts,[5,6] limb reduction defects,[3,5] urinary tract anomalies,[5,7] omphalocele,[8] and anal atresia.[9] If FA decreases the risk of having a child with one or more of these congenital anomalies, it is reasonable to assume that intrauterine exposure to a FA antagonist increases this risk. Two studies have been published on this association by Hernandez-Diaz *et al*.[10,11] They found an increased risk of NTDs, heart anomalies and orofacial clefts after exposure to FA antagonists. A study by Werler *et al* describes the possible teratogenicity of folic acid antagonists among other drug exposures, in an Australian population.[12] Unfortunately, their numbers were too small to distinguish between different groups of folic acid antagonists.

It is very important to reproduce epidemiological studies in databases with different characteristics and compare the findings, especially in this case since trimethoprim, one of the folic acid antagonists, is regarded safe during pregnancy. Therefore, the aim of this study was to investigate whether intrauterine exposure to a FA antagonist in the first weeks of pregnancy is associated with an increased risk of FA sensitive birth defects within the EUROCAT Northern Netherlands database.

Materials and Methods

Study population

In January 2004, we selected live births, stillbirths and abortions from 1997 to 2002 from the EUROCAT Northern Netherlands registry. In this registry, live births, stillbirths and abortion data with congenital anomalies are registered since 1981. Until 1996, information about the pregnancy outcome and the mother's condition, diseases and drug use was collected through the physician or midwife who reported the birth. Since 1997, parents have been sent a questionnaire in order to collect information about their characteristics like age, education, and family history. A request for access to their pharmacy data was included as well. After receiving the pharmacy data, mothers were asked by phone if drugs reported by the pharmacy were actually used and in which time period the drugs were used. Data about Over- the-Counter (OTC) drugs, illness during pregnancy, and life style aspects like smoking and alcohol consumption were provided by the parents as well. Information about the anomaly was provided by physicians, midwives, clinical geneticists and pathologists.

No age limits for discovering an anomaly were applied, which means that besides anomalies leading to abortions and anomalies discovered at birth, anomalies discovered even years after birth are recorded as well.

Of the in total 2,658 births from the selected years, 441 (16.4%) were excluded since data on drug exposure were missing. The mothers of these subjects did not fill out the questionnaire and therefore data of the pharmacy were not collected. The remaining 2,217 births were either case or control. In total 120,214 children were born in the northern Netherlands from 1997 to 2002.[13]

Cases and controls

Cases were defined as having a FA sensitive defect, including NTDs, congenital heart anomalies, orofacial clefts, limb reduction defects, urinary tract anomalies, omphaloceles and anal atresias (N=815). Births with chromosomal or monogenic defect with a FA sensitive defect as well were not defined as cases, unless the FA sensitive anomaly is not part of the chromosomal defect. For example, a Down syndrome with a neural tube defect is a case since a neural tube defect is not part of the syndrome. A Down syndrome with a heart anomaly is not a case. Controls were all births not defined as cases (N=1,402) including 401 chromosomal or monogenic defects.

Exposure assessment

We considered a foetus to have been exposed if the mother reported using a FA antagonist any time during the first 10 weeks after her last menstrual period. FA antagonists can be divided into two groups: the dihydrofolate reductase inhibitors (DHFRIs) and compounds that interact with other enzymes in folate metabolism, mainly antiepileptics. Dihydrofolate reductase is the enzyme that converts the inactive form of FA into active metabolites. Examples of DHFRIs are methotrexate, sulfasalazine, triamterene, pyrimethamine, and trimethoprim. The antiepileptics that influence folate metabolism are carbamazepine, phenobarbital, phenytoin, primidone, valproic acid, and lamotrigine.

If the numbers allowed it, the use of FA was included in the analyses. Exposed to FA was then defined as having used FA during all 10 weeks after the last menstrual period. Other use of FA was defined as not exposed.

Data analysis

P-values were calculated, with chi square tests or t tests where applicable, to compare the cases and controls. Trend analyses were performed for ordinal data. Logistic regression was

used to determine odds ratios (ORs) and 95% confidence intervals (95%CIs). If the numbers allowed, subanalyses were made per group of anomalies and per drug group.

Table 1: Characteristics of the cases and controls

	cases (n=815)		controls (n=1,402)		
mean age mother (sd)	30.28	(4.639)	30.44	(4.511)	p=0.425
Type of birth (n, %)					p<0.001
live birth	632	77.5	1,195	85.2	
died after birth	118	14.5	78	5.6	
miscarriage	6	0.7	16	1.1	
induced abortion	36	4.4	87	6.2	
stillbirth	23	2.8	26	1.9	
Gender (n, %)					p=0.140
male	475	58.3	765	54.6	
female	340	41.7	635	45.3	
could not be determined	0	0	2	0.1	
First pregnancy (n, %)					p=0.001
no	466	63.6	697	54.8	
yes	267	36.4	544	42.8	
Education mother (n, %)					p=0.033
low	220	28.8	314	23.7	trend p=0.067
middle	350	45.8	662	49.9	
high	195	25.5	350	26.4	
Chronic illness mother (n, %)					p=0.628
no	675	86.5	1,186	87.3	
yes	105	13.5	173	12.7	
Smoking (n, %)					p=0.320
never smoked	555	70.4	985	72.6	trend p=0.175
stopped when pregnant	76	9.6	137	10.1	
smoked during pregnancy	157	19.9	235	17.3	
Folic acid 0-10 weeks after lmp (n, %)					p=0.510
yes	187	22.9	339	24.2	
no	628	77.1	1,063	75.8	

Total numbers per column can vary due to missing values. P-values were calculated with chi square tests or t tests, where applicable.
Trend analysis was carried out for ordinal data.

Results

In table 1, the characteristics of 815 cases and 1,402 controls are shown. These groups differ in type of birth, gravidity, and education of the mother. Among the controls, more live births, miscarriages, and induced abortions occurred. For the mothers of the controls, this pregnancy was more often the first one. A low level of educational attainment was slightly more frequent among case mothers and a middle education level more frequent among control mothers although the p value of the trend analysis was not statistically significant (p=0.067). In the case group, more males were born compared to the control group, but this difference was not statistically significant.

Of the 815 cases, 11 (1.3%) were exposed to a FA antagonist in the first 10 weeks after the last menstrual period; three of these (0.4%) were exposed to a DHFRI (all three to trimethoprim) and eight (1.0%) to an antiepileptic drug (two to carbamazepine, four to valproate, one to carbamazepine+valproate and one to carbamazepine+valproate+ primidone). Of the 1,402 controls, 16 (1.1%) were exposed to a FA antagonist of which 12 (0.8%) to a DHFRI (seven to trimethoprim, two to trimethoprim+sulfoxamide and three to sulfasalazine) and four (0.3%) to an antiepileptic drug (one to carbamazepine, two to valproate and one to carbamazepine+valproate). None of the cases or controls were exposed to a DHFRI as well as to an antiepileptic drug.

Table 2 shows the ORs, and 95% CIs of the analyses with all FA antagonists, the DHFRIs, and the antiepileptics in relation to the group with all FA sensitive birth defects and

Table 2: Odds ratios and confidence intervals for folic acid sensitive defects after exposure to a folic acid antagonist in general or a dihydrofolate reductase inhibitor or an antiepileptic drug in specific.

		all folic acid antagonists			DHFRI			antiepileptics		
	non-exp. (n)	exp (n)	OR	(95% CI)	exp (n)	OR	(95% CI)	exp (n)	OR	(95% CI)
controls	1,386	16	ref.		12	ref.		4	ref.	
all fa-defects	804*	11	1.18	(0.55-2.57)	3	0.44	(0.12-1.54)	8	3.45	(1.04-11.48)
heart anomalies	363	5	1.19	(0.43-3.28)	0			5	4.77	(1.28-17.86)
neural-tube defects	75	2	2.31	(0.52-10.23)	0			2	9.24	(1.67-51.25)
clefts	187	3	1.39	(0.40-4.81)	3	1.85	(0.52-6.63)	0		
urinary tract defects	170	1	0.51	(0.07-3.87)	0			1	2.04	(0.23-18.34)
limb reduction defects	48	2	3.61	(0.81-16.14)	0			2	14.4	(2.58-80.75)
anal atresia	33	0			0			0		
omphalocele	15	0			0			0		

* a case can have more than one folic acid sensitive defect; therefore the sum of the cases with the specific defects will be more than 804

to the groups with specific defects. No cases with anal atresia or omphalocele were exposed. In the group with exposure to all FA antagonists, the ORs for NTDs and limb reduction defects were increased but not statistically significantly. Adjustment for the use of FA in the analysis with all FA defects had no effect on the OR (ORadj=1.18; 95%CI 0.55-2.56).

After restriction to the DHFRIs all three exposed cases had an orofacial cleft: one cleft palate and two cleft lips (+/-palate). The OR was 1.85, but not statistically significant (95% CI: 0.52-6.63). None of the three cases or 12 controls exposed to DHFRIs was exposed to FA as well. Therefore, for this subgroup no adjusted OR was calculated. For the analyses of the antiepileptics, we found an overall OR of 3.45 (95% CI: 1.04-11.48). There were no clefts exposed to antiepileptics. For the groups of specific defects, we found statistically significant increased ORs for heart anomalies, NTDs, and limb reduction defects. The OR for urinary tract defects was increased as well, but not statistically significant. Of the 8 cases exposed to an antiepileptic drug, 4 were also exposed to FA compared with 1 of the 4 exposed controls. However, the OR did not change after adjusting for FA use (ORadj=3.50; 95%CI 1.05-11.67).

Discussion

In this study we found no significantly increased risks of having a FA defect affected pregnancy after analysing the FA antagonists as a group. Although the effect was not statistically significant, our results are indicative for an increased risk for clefts after exposure to DHFRIs. Furthermore, we did find significantly increased risks for heart anomalies, neural tube defects, limb reduction defects, and all FA sensitive defects as a group after intrauterine exposure to antiepileptics. These results support the findings of the effect of antiepileptics of many other studies,[14-19] which indicates the validity of our data.

For the analyses with all FA antagonists as a group, we found an OR of 2.31 for NTDs and an OR of 3.61 for limb reduction defects which can be an indication for an increased risk; nevertheless, neither was statistically significant. Possibly, this group as a whole does not increase the risk. On the other hand, there might be an increased risk we could not detect due to the lack of power (small numbers).

The effect of DHFRIs as a group were previously examined by Hernandez-Diaz *et al*.[10,11] Their findings suggest that DHFRIs may increase the risk of NTDs, heart anomalies, and orofacial clefts. Since all cases exposed to a DHFRI were clefts, we could only calculate this OR and the OR for the group as a whole. Our OR for the whole group FA defects does

not support these findings; however, they are based on three exposed clefts only. Our results for the clefts are indicative for an increased risk although the CI does not include one, probably due to small numbers. Difference in time periods of exposure might also explain the dissimilar findings of us compared to Hernandez-Diaz *et al*. In the US, trimethoprim is mostly used for 7-10 days, while in the Netherlands the standard is 3, sometimes 5 days. Probably, length of exposure time is correlated with its influence on the folate levels; this in contrast to the antiepileptics, which are normally used chronically.

Including FA in the analyses did not influence the OR for the antiepileptics. This is in accordance with the findings of Hernandez-Diaz *et al*. Since none of the subjects exposed to a DHFRI was also exposed to FA, we could not confirm nor disclaim their findings that simultaneously use of FA with DHFRI reduces the possible harmful effect of DHFRIs.

The difference in type of birth between cases and controls can be explained by the definition of cases and controls: among the cases are more anomalies with a low survival rate after birth, like NTDs and severe heart defects. Therefore, the percentage live births is lower among cases and the percentage subjects that died after birth is higher among cases, compared with controls. We could not account for the difference in gravidity between the cases and controls, nor for the difference in education of the mother. From the latter we would like to remark that the trend analysis was not statistically significant, thus case mother are not systematically higher educated than mothers of controls or vice versa.

Having malformed births as controls is a strength as well as a limitation of this study. By having only malformed controls, we can not generalise our findings directly to the whole population. Another drawback of using malformed controls might be that FA has an until now unknown protective effect on some of the defects included as control in this study. For example, polymorphisms of folate metabolising enzymes have been linked to Down syndrome[20] although no association between folic acid consumption and Down syndrome is established. If indeed subjects now classified as controls turn out to be cases, misclassification has occurred in our study. In that case, this misclassification will bias the ORs towards unity thus we might underestimate a possible association in this study. On the other hand, by using malformed controls, recall bias is avoided.

A strength of this study is the availability of pharmacy data and the knowledge of actual use of the reported drugs. Therefore, exposure data are as complete as possible. Still, since the numbers in this study were very small, we suggest that further studies are to be conducted in order to be able to confirm these findings. Especially trimethoprim is of interest, since it is considered to be safe during pregnancy and possible harmful effects should be established before causing unnecessary fear among women and their physicians.

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Chapter 4

Can folic acid protect against congenital heart defects in Down syndrome?

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Abstract

Background: Several studies suggested a protective effect of folic acid (FA) on congenital heart anomalies. Down syndrome (DS) infants are known to have a high frequency of heart anomalies. Not all children with DS suffer from heart anomalies, which raises the question whether maternal factors might affect the risk of these anomalies.

Objectives: To investigate whether first trimester FA use protects against heart anomalies among DS children.

Methods: Women with liveborn DS children participating in the Slone Epidemiology Center Birth Defects Study between 1976 and 1997 were included. We performed case-control analyses using DS with heart anomalies as cases and DS without heart anomalies as controls. Subanalyses were performed for defects that have been associated with FA in non-DS populations (conotruncal , ventricular septum (VSD)) and for those that are associated with DS (ostium secundum type atrial septal defects (ASD), and endocardial cushion defects (ECD)). Exposure was defined as the use of any FA-containing product for an average of at least 4 days per week during the first 12 weeks of pregnancy, whereas no exposure was defined as no use of folic acid in these 12 weeks.

Results: Of the 223 cases, 110 (49%) were exposed versus 84 (46%) of the 184 controls. After adjustment for possible confounders, no protective effect of FA was found on heart anomalies overall (OR 0.95, 95%CI: 0.61-1.47) nor separately for conotruncal defects, VSD, ASD and ECD.

Conclusions: Our study does not show a protective effect of FA on heart anomalies among infants with DS.

Introduction

Although clinical features of Down syndrome (DS) are well recognized, it remains unclear how the extra chromosome relates to the high incidence of certain major malformations among DS children compared to the "normal" population. For example, cardiac malformations are commonly seen among DS infants, with incidence rates varying between 40-50%. Shapiro (1983) suggests that besides direct effects of the chromosomal abnormality, it might be possible that maternal risk factors interact with an already susceptible genotype, leading to the development of major anomalies in some individuals but not in others.[1]

Studies on maternal risk factors and major birth defects restricted to infants with DS are few and results are not consistent, but there is some evidence that environmental factors might influence the occurrence of defects among foetuses with an extra chromosome 21. For maternal age, findings differ on the direction of risks: while Källèn *et al* found that DS infants born to teenage mothers had a decreased risk for cardiac defects, which was particularly pronounced for endocardial cushion defects and ventricular septum defects;[2] they also found an increased risk for megacolon for mothers under 25 years of age. Khoury and Erickson also found an inverse association, but with maternal age and oral clefts; additionally, they found an association between maternal race and cardiac defects (40% among blacks vs. 17% in whites).[3] With respect to exogenous factors, Fixler and Threlkeld found no differences in risk of heart defects in relation to maternal illness, medication use, or consumption of caffeinated beverages, cigarettes or alcohol, but others did find an effect of such maternal exogenous exposures.[4] Khoury and Erickson found an association between first trimester fever and duodenal atresia. [3] Although this was not confirmed in a later study by Torfs and Christianson,[5] the latter did find associations between coffee consumption and maternal fever and Hirschsprung's disease as well as between smoking and cardiac defects; alcohol was not associated with any defect. Taken together, these studies suggest maternal factors might play a role in the origin of major congenital anomalies among infants with DS.

No study, however, has evaluated the risks of major birth defects among DS infants in relation to maternal use of folic acid (FA). In the general population, studies suggest that FA use may reduce the risk of cardiac defects. While Werler *et al* [6] found no association between multivitamin use and conotruncal defects or ventricular septal defects, other studies did find evidence of such an association. In a randomized clinical trial, Czeizel found a significant protective effect of FA-containing multivitamins for heart defects overall (OR 0.42, 95%CI 0.19-0.98) and for conotruncal defects (OR 0.29, 95%CI 0.09-.097) in particular.[7] A

number of observational studies were consistent with that finding. For example, Botto *et al* found significant protective effects of multivitamins (which usually contain FA) on outflow tract defects, VSDs, and cardiac defects overall.[8] Furthermore, studies of Hernandez-Diaz *et al* and Meijer *et al* provide indirect evidence of a possible protective effect of FA on cardiac defects.[9,10] Both found an increased risk of cardiac defects after intrauterine exposure to medications that antagonize the effects of FA; this risk diminished if folic acid supplements were taken along with the FA antagonists (Hernandez-Diaz *et al*). Although these studies did not investigate the effect on heart anomalies specifically among DS infants, their results raise the question whether FA might reduce the risk of heart anomalies in this particular population. We therefore sought to evaluate the hypothesis that FA has a protective effect on heart anomalies among infants with DS.

Materials and methods

Since 1976, the Boston University Slone Epidemiology Center Birth Defects Study (BDS) has been interviewing mothers of children with a range of birth defects.[11] Until 1997, mothers from the areas around Boston (since 1976), Philadelphia (since 1977) and Toronto (since 1978) were interviewed in person, within six months of delivery, usually in the subject's home, by a trained study nurse. Because of personnel limitations, not all eligible subjects were approached for interview. Rather those subjects that were approached were selected based on 'priority' diagnoses which reflected changing research interests of the program. For example, from 1983 to 1987 DS was on the priority list but neural tube defects were not. From 1988 to 1992, neural tube defects were on the priority list, but DS was not. Therefore, during the latter period, all subjects with a neural tube defect and only a sample of those with DS were approached for interview. However, selection of subjects for interview was never dependent on exposure to any particular agent. The interview contained questions about demographic, reproductive, and medical factors, as well as details about all medications used, including vitamins. The product name, starting and stopping dates and frequency of use were recorded for each vitamin product taken between two months before through the end of pregnancy.

The present analyses include data on liveborn infants with DS enrolled between 1976, the start of the study, and 1997, the year before food fortification with FA. We excluded infants with gestational age <37 weeks whose only cardiac anomaly was patent foramen ovale, ostium secundum type atrial septal defects (ASD), or patent ductus arteriosus (n=15).

Table 1: Characteristics of cases and controls.

	Controls (n=184)		Heart anomaly (n=223)	
	n	%	n	%
Smoking †				
Never	76	41.3	118	52.9
During (part of) pregnancy	67	36.4	48	21.5
Ex-smoker	41	22.3	57	25.6
Alcohol during pregnancy*				
Yes	33	20.2	49	23.7
Missing N=37				
Maternal race †				
White (vs. non-white)	171	92.9	192	86.1
Maternal age at conception				
<25	32	17.4	37	16.6
25-29	55	29.9	69	30.9
30-34	56	30.4	55	24.7
>34	41	22.3	62	27.8
Maternal years of education				
-12	23	12.5	27	12.1
12	53	28.8	63	28.3
13-15	59	32.1	57	25.6
>15	49	26.6	76	34.1
Planned pregnancy*				
Yes	102	62.6	132	64.7
Missing N=40				
Maternal diabetes* †				
Yes	3	1.6	13	5.9
Missing N=4				
Parity				
Primapara	42	22.8	51	22.9
Sex of baby †				
Male	124	67.4	105	47.1
Center				
Boston	65	35.3	67	30.0
Philadelphia	55	29.9	58	26.0
Toronto	64	34.8	98	43.9

* if missing data: percentages calculated on available data.

† significant difference between cases and controls ($p<0.05$)

Among the remaining 773 DS infants, cases were defined as infants with any heart anomaly. Separate analyses were performed for conotruncal defects, ventricular septal defects (VSD), endocardial cushion defects (ECD) and ASD. The first two defects were included because of the inverse association with FA in the literature and the latter two because they are commonly seen among DS infants. The four groups were not mutually exclusive. Controls were DS infants without a heart anomaly.

Since lunar months two and three are most important regarding development of heart anomalies, exposure to FA was defined as the use of any FA-containing product for at least 48 days during the first 12 weeks of pregnancy (lunar months one to three), which corresponds to an average of 4 days per week during this period. In all exposed study subjects, the exposure occurred on ≥16 days in lunar months two and three. No exposure was defined as no FA use at all in these first 12 weeks of pregnancy.

Multivariate models were used to calculate adjusted odds ratios (ORs) and 95% confidence intervals (95% CIs). Variables that were related to exposure and/or outcome were included in the model: maternal race (white/non-white), maternal age (<25, 25-29, 30-34, and >34), maternal education (<12, 12, 13-15 and >15 years), maternal diabetes (yes/no), year of birth (<83, 83-87, 88-92, and >92), and geographic center (Boston, Philadelphia, and Toronto).

Results

Of the 773 DS infants in the database, 366 were excluded because the mothers used FA but at frequencies and durations that were inadequate to meet the exposure criteria. Among the 407 who met these criteria, 223 (55%) were cases and 184 (45%) were controls. The cases included 20 births with conotruncal defects, 73 with VSD, 73 with ASD and 85 with ECD. Among the excluded births, a similar distribution of cases and controls was found (58% cases).

The characteristics of the cases and controls are shown in table 1. Among cases, more mothers reported never having smoked and more had diabetes, whereas fewer mothers were white and fewer infants were male (all $p<0.05$). There was little difference between cases and controls for maternal age, education, parity, alcohol drinking, or whether the pregnancy was planned.

Of the 223 cases, 110 (49%) were exposed to FA, versus 84 (46%) of the 184 controls. Logistic regression that adjusted for race, maternal age, maternal education, maternal diabetes, year of birth and center of birth revealed no protective effect of FA for heart

Table 2: Maternal folic acid use in relation to cases with heart anomalies among subjects with Down syndrome..

	Number exposed/non-exp	OR_{adj*}	95% Confidence Interval	
Controls	84/100			
Cases				
Heart anomalies	110/113	0.946	0.608	- 1.471
Conotruncal defects	10/10	0.704	0.213	- 2.323
VSD	40/33	1.456	0.774	- 2.739
Ostium secundum type ASD	40/33	0.943	0.481	- 1.847
Endocardial cushion defects	42/43	0.766	0.419	- 1.397

* adjusted for race, age, education, diabetes, year of birth, center

anomalies overall (OR 0.95, 95%CI: 0.61-1.47), as is shown in table 2. Maternal folic acid use was also not associated with any of the four cardiac subgroups; odds ratios showed some variation, but none of the confidence intervals excluded 1.0.

Discussion

In this study, we examined whether the risk of congenital heart defects among DS infants is decreased by first trimester FA exposure. The literature provides some support for such an effect in the general population, particularly with respect to conotruncal defects and VSDs. However, the present data do not provide evidence for a protective effect of FA on the occurrence of heart anomalies overall among DS infants, nor for the subgroups of conotruncal defects, VSDs, ASDs, or ECDs.

The strength of this study is that we only included children born to mothers who took FA regularly or did not take FA at all. By eliminating occasional FA users, our approach maximized the opportunity to identify a protective effect of FA on heart anomalies among DS infants.

Nevertheless, several limitations of the present study should be considered. Since data about exposure are collected after birth, information bias could occur. We attempted to minimize such bias by using standardized questionnaires and by conducting the interviews relatively soon after the infant's birth. By using other DS infants as controls, we attempted to avoid recall bias, a specific type of information bias

Not all infants with DS encountered in the study hospitals were enrolled in the study because DS was only on the priority list between 1983 and 1987. In the other years, many infants with DS were enrolled not because of the specific diagnosis of DS, but rather because of the presence of other malformations. However, there is no reason to assume that recruitment of subjects was related to use of FA-containing products, making it unlikely that this process introduced selection bias. Another possibility is that defects that may be folic acid-sensitive (such as neural tube or urinary tract defects) were included in the control group. However, the numbers of such defects among controls were small, and the proportions of DS infants with these defects did not differ between cases and controls, suggesting that such bias is unlikely.

Misclassification of cases and controls might have occurred if cardiac defects were not identified or coded. While such misclassification is unlikely to be biased because it is unlikely to be related to FA exposure, non-differential misclassification could tend to obscure a protective effect of FA on heart anomalies.

Finally, residual confounding is still a possible explanation for not finding a protective effect of FA in this population. In our multivariate model, we adjusted for several factors that are associated with either FA use or cardiac anomalies; nevertheless, other variables might differ between the women who take FA in early pregnancy and women who take no FA during that time. If these differences are related to the presence of heart defects, confounding could explain our findings.

The current literature reflects discussion about the effect of FA on the aetiology of DS itself. Polymorphisms of the methylene tetrahydrofolate reductase gene were more prevalent among mothers of children with DS than among control mothers in some studies,[12,13] though other researchers could not confirm this difference.[14] Furthermore, Czeizel and Puho found a decreased risk for DS after periconceptional high-dose FA use in a population-based study.[15] If FA were to protect against DS itself, perhaps the same phenomenon that results in a DS birth despite FA use in pregnancy would also result in a lack of FA protection against heart anomalies. Alternatively, a higher dose might be required to achieve an effect on heart anomalies in DS. Most multivitamin products contain 400 µg of FA; while that dose may be sufficient to reduce cardiac anomaly risks in otherwise normal foetuses, perhaps a larger dose may be needed to protect against development of a heart anomaly when a trisomy 21 is already present.

Findings from previous studies that focused on of the effects of certain exogenous maternal factors on the risks of birth defects in DS have been inconsistent, but maternal age and race might play a role. In our study, we found no differences in maternal age between

cases and controls. Our findings on maternal race are consistent with Khoury and Erickson[3] though not with a later study with more accurate rates of heart anomalies.[16] This is the first study to investigate the effect of FA on heart anomalies among DS children, and it failed to provide evidence of such an effect. Further studies could improve our understanding of this possible relation by taking into consideration the dose and composition of FA-containing products and possible polymorphisms in folate pathway genes.

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Chapter 5

Clomiphene and hypospadias on a detailed level; signal or chance?

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Abstract

Background: Clomiphene, a drug used to induce ovulation, is chemically related to diethylstilbestrol (DES). DES is associated with vaginal cancer and infertility among daughters and with hypospadias among second generation offspring. Since clomiphene has a long half life and metabolites have been found in the faeces up to 6 weeks after administration, foetal exposure is possible.

Methods: Case-control analyses were performed to investigate the association between clomiphene exposure and hypospadias. Cases were all male subjects registered in the EUROCAT Northern Netherlands registry for congenital anomalies with non syndromal hypospadias. Controls were all male births without hypospadias, including chromosomal and monogenic defects. Logistic regression analyses were performed to calculate odds ratios (ORs) and 95% confidence intervals (CIs)

Results: Of the 392 cases, 7 (1.8%) were exposed to clomiphene compared with 64 out of 4538 controls (1.4%). For penoscrotal hypospadias we found a significant increased OR (6.08; 95% CI 1.40-26.33); the ORs for the mild and moderate forms of hypospadias were not increased.

Conclusions: Since penoscrotal hypospadias is rare, the effect is diluted when studying all hypospadias together as a group. Therefore, our study stresses the importance of studying birth defects on a detailed level as possible. Other studies should be conducted to confirm our findings.

Introduction

Hypospadias is a congenital anomaly of the male genital characterized by an abnormal location of the urethral opening, varying from the ventral surface of the penis to the scrotum or even the perineum. The prevalence in Europe is estimated on 2-5 per 1000 male births.[1] The cause of hypospadias is likely to be multifactorial. Sexual differentiation is determined by testosterone and its metabolites. It has been suggested that changes in concentrations of sex hormones during the critical period of the development (up to week 12) may play a part in the development of hypospadias.[2]

Clomiphene, widely used to induce ovulation, has a long half life and its metabolites have been found in blood samples after 22 days and in faeces up to six weeks after administration.[3] Therefore, exposure to the foetus in the first weeks of pregnancy is possible.

Clomiphene is chemically related to diethylstilbestrol (DES), a drug associated with vaginal adenocarcinoma and infertility among women exposed in utero. Klip *et al* reported an increased risk of hypospadias among sons of DES daughters.[4] Furthermore, although the numbers were small, they found more severe cases of hypospadias among these DES grandsons compared with the unexposed group.

Since clomiphene could be present in the mother's body after conception and because of the chemical relation of clomiphene with DES, we aimed to investigate the association between clomiphene use of the mother and hypospadias among male offspring.

Methods

Case-control analyses were performed using all live births, stillbirths and terminated pregnancies from 1981 to 2003 from the EUROCAT Northern Netherlands registry of congenital anomalies. This population based registry monitors about 20,000 births per year. Information about the anomalies is collected through physicians, midwives, clinical geneticists and pathologists. Methods for case ascertainment have not changed over time and are in accordance with EUROCAT Central Registry guidelines (www.eurocat.ulster.ac.uk). Until 1996, information about the mothers' condition was collected through these same health care providers. Pharmacy data have been routinely collected since 1997 and the actual use of the reported drugs is verified with the mother.

Cases were male subjects with hypospadias, either glandular/coronary, penile, penoscrotal or perineal. The hypospadias could be isolated or present in combination with other defects. Subjects with hypospadias recognized as part of a syndrome (n=30) were excluded from this study, as well as subjects with epispadias (n=12). The mildest form,

glandular hypospadias, is only registered if other defects are present, regardless whether these defects are associated with hypospadias. Since the database has no information on non malformed births and no comparable database can be used to validly extract non malformed births, all male subjects without hypospadias were considered controls, including the chromosomal or monogenic defects and other syndromes. This method corresponds with the one used in the MADRE project from the International Clearinghouse for Birth Defects Surveillance and Research.[5]

During the study years 1981-1996, exposure to clomiphene was defined as use of clomiphene by the mother prior to the index pregnancy as recorded by the health care provider who was explicitly asked whether the pregnancy resulted from clomiphene treatment. During the study years 1997-2003, exposure to clomiphene was defined as evidence of any clomiphene prescription dispensed to the mother from 0-3 months prior to the index pregnancy as recorded by the pharmacy. This period was chosen since date of issue of the pharmacy is not necessarily date of use by the mother; she has to adapt her use to her menstrual period. Furthermore, clomiphene can be issued for more than one menstrual cycle. Actual use of clomiphene had to be confirmed by the mother by asking whether the conception was indeed the result of clomiphene use.

Variables that could influence the results, like multiple pregnancy, exposure to DES, periconceptional use of folic acid and maternal age were investigated as well. Folic acid use around conception was asked for in the mother's questionnaire since 1997 and often missing in the years before. The other variables, multiple pregnancy, intrauterine DES exposure of the mother, and maternal age, were collected through the health care providers by asking for these variables specifically (until 1996) and through this questionnaire to the mother since 1997.

Logistic regression was performed to produce odds ratios (ORs) and 95% confidence intervals (CIs).

Results

In total, 392 subjects were identified with hypospadias. Distributions of some characteristics for cases and controls as well as for exposed and not exposed births are shown in table 1. Of all 4,930 male subjects included in the analyses, 71 (1.4%) were exposed to clomiphene. Exposure was 1.8% (n=7) among cases and 1.4% (n=64) among controls. More twins were born in the exposed group. Of the 11 sons of DES-daughters, two had hypospadias (both penile) and none of these 11 DES-grandsons were exposed to clomiphene. Use of folic acid

Table 1: Characteristics by exposure groups and outcome

	not exposed (n=4859)		exposed (n=71)		p*	cases (n=392)		controls (n=4538)		p*
Characteristics	n	(%)	n	(%)		n	(%)	n	(%)	
child part of multiple pregnancy?										
Yes, twin	188	(4.0)	6	(8.5)	0.17	15	(3.9)	179	(4.1)	0.61
Yes, triplet or more	5	(0.1)	0	(0)		1	(0.3)	4	(0.1)	
is mother DES-daughter?										
Yes	11	(0.2)	0	(0)	0.69	2	(0.5)	9	(0.2)	0.21
folic acid use of mother										
yes, periconceptional	600	(15.7)	18	(25.7)	0.015	65	(19.7)	553	(14.6)	0.002
yes, other period	498	(13.0)	13	(18.6)		71	(21.5)	650	(17.2)	
Exposed	-		-			7	(1.8)	64	(1.4)	0.55

Percentages can vary because of missing data

* Two-tailed $\chi 2$ test

differed significant between exposed and not exposed births as well as between cases and controls. A detailed description of the seven exposed cases and their characteristics are shown in table 2. These seven cases were born between 1989 and 2002, all singletons and all resulted from other clomiphene only treatment. Five of these seven cases had isolated hypospadias. The percentages of isolated cases among all cases are presented in table 3.

For hypospadias in general we found an OR of 1.27 (95%CI 0.58-2.79). The results for the different types of hypospadias are subsequently given in table 3. No subject with perineal hypospadias was found. Penoscrotal hypospadias had a significantly increased OR of 6, the milder forms had no increased OR. Of the 38 cases without further information about the severity of the hypospadias, none were exposed to clomiphene. Since the numbers were too small, we could not adjust for possible confounders or effect modifiers.

Table 2: Detailed description of seven exposed cases

case number	year of birth	multiple birth	fertility treatment	periconceptional folic acid use	description of anomalies
1	1989	no	clomiphene	no	isolated coronary hypospadias
2	1991	no	clomiphene	no	isolated penile hypospadias
3	1994	no	clomiphene	no	penoscrotal hypospadias and scaphocephaly craniosy
4	1998	no	clomiphene	advised period	glandular hypospadias and penile chordae
5	1998	no	clomiphene	advised period	isolated penoscrotal hypospadias
6	2000	no	clomiphene	advised period	isolated penile hypospadias
7	2002	no	clomiphene	advised period	isolated coronary hypospadias

Table 3: Total number of cases, number of isolated cases, number exposed cases and odds ratios of hypospadias

Anomaly	N	N isolated cases (%)	exposed	odds ratio	95% confidence interval
Glandular /coronary hypospadias	186	161 (87%)	3	1.15	0.36-3.68
Penile hypospadias	143	131 (92%)	2	0.99	0.24-4.09
penoscrotal hypospadias	25	19 (76%)	2	6.08	1.40-26.33
perineal hypospadias	0				
hypospadias NOS*	38	10 (26%)	0		

* NOS = not otherwise specified

Discussion

Our study shows different ORs for different forms of hypospadias in relation with preconceptional exposure to clomiphene: no association was found for the milder forms but a significantly increased OR was found for penoscrotal hypospadias.

We found no increased risk of hypospadias in general, which corresponds with the findings of Sørensen *et al*.[6] In that study, no discrimination in severity of hypospadias was made. However, in our study, by investigating all hypospadias as a group the results on penoscrotal hypospadias were diluted. This stresses the importance of studying any possible association at the most detailed level possible, which is the strength of this study.

Is the association with penoscrotal hypospadias found on only two exposed cases based on chance or is there a biologically plausible explanation for our findings? Clomiphene has estrogenic and anti-estrogenic characteristics.[3] Kim *et al* studied the effect of oestrogen exposure on hypospadias among mice.[7] Hypospadias, not specified, occurred in about 50% of the male foetuses. It is possible that disturbed concentrations of sex hormones during the critical period of penile and urethral development play a part in the development of hypospadias. Since clomiphene can still circulate in the mother's body after conception due to its long half life, it might influence the first weeks of foetal development, leading to severe cases of hypospadias. In the latter weeks of development, in which the milder hypospadias appears, the time window between exposure and development of the defect might be too large. This may explain the lack of association between clomiphene and the milder forms of hypospadias. Furthermore, there is the chemical similarity to DES and the results published by Klip *et al*.[4] In that study, four of the 250 DES grandsons had hypospadias. Three of the four had penoscrotal hypospadias and for one the urethral opening was located on the distal shaft. Of the 8,729 non DES grandsons eight had hypospadias. Of these only one had penoscrotal hypospadias, three had their urethral opening at the penile shaft, two were described as penile hypospadias and two as (sub)coronal. Because of the chemical relationship between DES and clomiphene, it is interesting to see all four exposed cases had severe forms of hypospadias compared to less severe forms among boys of whom the mother was not intrauterinely exposed to DES.

Although the findings of this study might be biologically plausible which support a possible causal relation, the small numbers are a problem. The OR found on penoscrotal hypospadias, although statistically significant, has a wide confidence interval indicating great uncertainty of the real effect. Although most hypospadias cases are isolated cases (table 3) we did not have the power to limit the study to these isolated cases. Also, adjusting for

relevant variables shown in table 1 was impossible. In spite of the fact information on periconceptional folic acid use is missing for most births before 1997 we did find folic acid use differed between cases and controls and between exposed and non-exposed births. Not being able to adjust for this possibly important confounder is a limitation.

Another problem in this study is the lack of pharmacy data and information from the parents before 1997. On the other hand, use of fertility treatment before 1997 was explicitly asked thus exposure misclassification is likely to be negligible. Also, since clomiphene was not associated with hypospadias in literature, any misclassification would be non-differential and would bias the estimates towards null and underestimate the real effect. Any possible association between clomiphene and anomalies in the control group would also bias the estimates towards the null. Since these associations are uncertain and selecting specific anomalies as controls might introduce selection bias, we felt strongly our study would be better off including all non-cases as control. By using malformed controls, one assumes exposure of clomiphene among the controls is an estimation of exposure in a general population. It is unlikely exposure in our control group is lower than in general population since that would mean clomiphene would decrease the chance of birth defects which is highly unlikely. Also, information about infertility treatment is asked for thoroughly for all malformed births in the registry. If exposure in our control group is higher than in the general population, the differences between exposure among cases and among controls is underestimated in our analysis and the association found might also be an underestimation.

Among exposed births more multiple pregnancies were found compared to the non-exposed, as was to be expected with ovulation induction. Although hypospadias is associated with multiple birth in literature, cases and controls did not differ with respect to this variable (table 1). Probably other defects than hypospadias are associated with multiple pregnancy as well as a result of which our whole population contains more multiple pregnancies than a general population. Again, it is a drawback we could not adjust for this variable.

A further limitation is the possibility that the underlying infertility is accountable for the association found. This is more a methodological problem than a public health issue, which cannot be solved since we cannot distinguish between them because there are no alternative medicines available; for couples with fertility problems that need clomiphene to get pregnant, this difference is of no importance. Moreover, although an OR of 6 is high, penoscrotal hypospadias is a rare condition and therefore the absolute risk remains low. We would not advise against use of clomiphene based on our results. After all, clomiphene

allows many couples to have children who otherwise could not. However, we feel couples that seek for treatment should be informed about possible risks involved.

Despite the statistical limitations of the small numbers, our study stresses the importance of collecting and analyzing data about congenital anomalies at the most detailed level possible. Therefore, we encourage birth defects monitoring registries to pursue their work and to collaborate to enhance the power of future studies. We highly recommend evaluating this relation in other databases to confirm our findings.

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Part B

Improving data collection for epidemiologic studies on safety of drug exposure during pregnancy

Chapter 6

The potential of the European network of congenital anomaly registers (EUROCAT) for drug safety surveillance

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Abstract

Background: EUROCAT (European Surveillance of Congenital Anomalies) is a network of population-based congenital anomaly registries in Europe surveying more than 1 million births per year, or 25% of the births in the European Union. This paper describes the potential of the EUROCAT collaboration for pharmacoepidemiology and drug safety surveillance.

Methods: The 34 full members and six associate members of the EUROCAT network were sent a questionnaire about their data sources on drug exposure and on drug coding. Available data on drug exposure during the first trimester available in the central EUROCAT database for the years 1996-2000 was summarised for 15 out of 25 responding full members.

Results: Of the 40 registries, 29 returned questionnaires (25 full and four associate members). Four of these registries do not collect data on maternal drug use. Of the full members, 15 registries use the EUROCAT drug code, four use the international ATC drug code, three registries use another coding system and seven use a combination of these coding systems. Obstetric records are the most frequently used sources of drug information for the registries, followed by interviews with the mother. Only one registry uses pharmacy data. Percentages of cases with drug exposure (excluding vitamins/minerals) varied from 4.4% to 26.0% among different registries. The categories of drugs recorded varied widely between registries.

Conclusions: Practices vary widely between registries regarding recording drug exposure information. EUROCAT has the potential to be an effective collaborative framework to contribute to post-marketing drug surveillance in relation to teratogenic effects, but work is needed to implement ATC drug coding more widely, and to diversify the sources of information used to determine drug exposure in each registry.

Introduction

Population based registries of congenital anomalies in the European Union collaborate in EUROCAT (European Surveillance of Congenital Anomalies)[1,2] which collects data on more than one million births per year, over 25% of all births in Europe. A sister organisation operating world-wide is the International Clearinghouse for Birth Defects Monitoring Systems (ICBDMS).[3] Many of the congenital anomaly registries were initiated following the thalidomide disaster in the 1960s, resulting in more than 5,000 infants with severe birth defects worldwide.[4] In order to detect new teratogens as early as possible, registries could on the one hand detect trends or clusters in the frequency of congenital anomalies and on the other hand look directly at drug exposure by recording the drugs used during pregnancy.[5-7] One would intuitively expect that collaborative networks such as EUROCAT and ICBDMS to increase the statistical power of any analyses.

While some of the registries participating in ICBDMS (including some EUROCAT registries) collaborate to monitor associations between drugs and malformations,[7] the EUROCAT central database has been less systematically used,[8,9] and awaits full exploitation. Individual EUROCAT registries have investigated drug exposures in relation to birth outcomes.[10-13] The aim of this study was to assess the EUROCAT database as a resource for pharmacovigilance studies, and to assess what changes are required to provide EUROCAT with a stronger role in drug safety surveillance in the future.

Materials and methods

In November 2003, the EUROCAT central database consisted of data from 40 registers, 34 full members contributing individual data to the central database, and six associate members contributing only to the aggregate number of cases. All registries are population based with geographically defined populations. The registries are based on multiple sources of information and include information about live births, stillbirths and terminations of pregnancy. All structural malformations, syndromes and chromosomal anomalies are included in the database. Minor malformations are excluded according to a specified list of exclusions.[14] The methods for case ascertainment of EUROCAT are described elsewhere.[15]

A questionnaire was sent to the registries requesting information on their source of data for drug exposure (mother, medical or pharmacy data), on prospective and/or retrospective collected data, the kind of data available (name of the drug, dose, frequency, etc.), and if there is the possibility to go back to records or parents in case additional information is needed. If information on drug exposure was collected, we asked which system was used

Table 1: EUROCAT codes for drug groups (EUROCAT Guide 1.2 [14])

00	No drugs
01	Atropinics and antispasmodics
02	Anaesthetics, local and general
03	Hypnotics, sedatives and psychotropics
04	Antiepileptics
05	Analgesics, antipyretics and antiinflammatory agents
06	Histamine antagonists
07	Antiasthmatic agents incl. methylxanthines
08	Antiarhythmic and antihypertensive agents
09	Diuretics
10	Tocolytics
11	Antiseptics, antibiotics, -viral, -parasetic, -fungal agents
12	Antiproliferative and immunosuppresive agents
13	Anticoagulant, antithrombotic and thrombolytic drugs
14	Thyroid and antithyroid drugs
15	Oestrogens, progestagens, androgens, incl. contraceptives
16	Adrenocortical steroids
17	Insulin and oral hypoglycemic agents
18	Vaccins
19	Vitamins and minerals
88	Other
98	Drug(s) taken but no information available
99	Not known

for coding the data. The options were (i) the EUROCAT coding system with 23 codes dividing all drugs into groups (table 1), (ii) the ATC coding system (Anatomical Therapeutical Chemical) which is the WHO hierarchical system allowing any level of drug specificity to be coded and/or analysed, or (iii) a unique coding system of the registry itself. We also asked whether data on drug exposure of non-malformed births was also recorded.

The first trimester drug exposure data obtained from the responders, which was available in the central EUROCAT database was summarised for the years 1996-2000. Drug exposure in the first trimester can be completed on a per case basis with space for a maximum of three different EUROCAT drug groups to be recorded (table 1).[14] If all three fields were empty, data was considered to be missing. A case was considered exposed during the first trimester if at least one drug group was reported, including vitamins and

minerals or otherwise mentioned. In addition, there were also three associated text fields that could be used to provide more detailed information, if available. If registries did not work with the EUROCAT drug codes they could use these text fields, but only the 15 registries providing EUROCAT drug codes were analysed.

Results

Of the 40 registries that were invited to participate in this study, 29 (25 full members and 4 associate members) replied to our questionnaire (response 73%). Four of these responders (Styria, Trent, Wessex and Oxford) do not collect data on maternal drugs at all and were therefore excluded from further analysis. Of the remaining 25 useable responses from registries, data sources of drug information and the used coding systems are shown in table 2 and 3. We also examined the EUROCAT central database for the non-responding registries and found 6 out of 11 non-responders had coded drug data.

Seventeen registries (68%) use obstetric or midwife records as source of information on drug exposure. Interview or questionnaire with the mother is the second most used source of information (13, 52%). Northern Netherlands is the only registry that uses pharmacy data. The majority of registries (18, 72%) use more than one source to obtain information about drug use during pregnancy.

Most registries use the EUROCAT drug codes (table 3). ATC-codes are used by Paris, Dublin, North East Italy, Northern Netherlands, Basque Country (until 1998) Finland, Central East France and Norway. Sicily, ECEMC, and North Thames use their own coding system. Emilia Romagna (Italy) and Mainz (Germany) use their own codes in addition to the EUROCAT codes. Drug information on non-malformed births is collected in four registries, Strasbourg, Mainz, ECEMC and Asturias.

Since the central EUROCAT database only contains information on drugs of full members using the EUROCAT coding, 15 responding registries fulfil both criteria and their information about the drugs registered are presented in table 4 and 5. From 1996 to 2000 a total of 2,771 out of 18,576 cases (12.2%) were recorded with at least one drug group exposure, excluding vitamins and minerals (table 4). The percentages of cases with reported drug group exposure during first trimester varied from 4.4% in El Valles to 36.1% in Mainz. The use of vitamins and minerals were highest for the Basque Country and the Asturias registries. Excluding vitamins and minerals, up to 26% (Mainz) of women were reportedly exposed to any medication during the 1st trimester. Most cases were reported to be exposed to only one drug group. The percentages coded as missing or unknown varied from

Table 2: Sources for drug exposure information as stated in the questionnaires (25 registries)

Registry	Recorded before birth (prospectively)				Recorded after birth (retrospectively)	
	Obstetric record	Midwife record	GP record	Pharma-ceutical record	Questionnaire Mother	Interview Mother
Full members						
Antwerp (Belgium)	X	-	X	-	-	-
Zagreb (Croatia)	X	-	X	-	-	-
Odense (Denmark)	X	X	X	-	-	-
Strasbourg (France)	X	X	-	-	-	X
Paris (France)	X	X	-	-	-	-
Mainz (Germany)	X	X	-	-	-	-
Cork & Kerry (Ireland)	X	-	X	-	-	-
Dublin (Ireland)	X	-	-	-	-	-
Emilia Romagna (Italy)	-	-	-	-	X	-
North East Italy	-	-	-	-	-	X
South-East Sicily (Italy)	-	-	-	-	-	X
Tuscany (Italy)	-	-	-	-	-	X
Malta	X	-	-	-	-	Sometimes
Northern Netherlands	-	-	-	X	X	X
Southern Portugal	X	-	-	-	-	X
Asturias (Spain)	X	X	-	-	-	X
Basque Country (Spain)	X	X	-	-	-	-
El Valles (Spain)	X	-	-	-	X	X
Vaud (Switzerland)	X	-	X	-	-	-
Glasgow (UK:Scotland)	X	-	-	-	-	-
North Thames (UK:England)	X	X	-	-	-	-
associate members						
Finland	X	-	X	-	-	-
Central East France	-	-	-	-	X	X
Norway	-	X	-	-	-	X
ECEMC (Spain)	-	-	-	-	-	X

Table 3: Coding systems used by EUROCAT registries participating in this study, as reported in the questionnaires (25 registries)

Registry	Coding system used since 1990			
	EUROCAT			
	Numeric	Text	ATC	Own coding
Full members				
Antwerp (Belgium)	X	X		
Zagreb (Croatia)	X	X		
Odense (Denmark)	X	X		
Strasbourg (France)	X			
Paris (France)			X[1]	X[1]
Mainz (Germany)	X	X		X[2]
Cork & Kerry (Ireland)	X			X[3]
Dublin (Ireland)			X	
Emilia Romagna (Italy)	X			X
North East Italy			X	
South-East Sicily (Italy)				X[4]
Tuscany (Italy)	X			
Malta	X	X		
Northern Netherlands		X	X	
Southern Portugal	X			
Asturias (Spain)	X	X		
Basque Country (Spain)	X[5]	X[5]	X[5]	
El Valles (Spain)	X	X		
Vaud (Switzerland)	X	X		
Glasgow (UK, Scotland))	X			
North Thames (UK, England)				X
associate members				
Finland	X	X	X	X
Central East France			X	
Norway			X	
ECEMC (Spain)				X

1 Own coding until 2000, based on DCI which can be easily translated into ATC-codes; ATC since 2001

2 Own Coding of 36 groups and specific drugs

3 Actual drug is noted

4 Drug description, no code

5 90-98 ATC codes used; 99-present EUROCAT numeric and text codes

0% for Emilia Romagna to over 40% in Antwerp, Odense, Strasbourg, Tuscany, and Basque Country.

Table 5 displays women's exposure to the different EUROCAT drug groups during the first trimester (as a proportion of all exposed cases) for each registry. Large differences existed between the registries, for example, the proportion of cases exposed to any drug where the mother had been exposed to antibiotics varied from 2.8% in Glasgow to approximately 43% in Asturias.

Table 4: Number and Proportion of cases exposed to drugs in first trimester (with and without vitamins/minerals), 15 full member EUROCAT registers 1996-2000.

	Total cases	Missing / not known	Not exposed to drugs	Exposed to drugs	Exposed to drugs excl. vitamins/min
		N (%)	N (%)	N (%)	N (%)
Antwerp (Belgium)	1,909	1,322 (69.3)	447 (23.4)	140 (7.3)	139 (7.3)
Zagreb (Croatia)	483	40 (8.3)	371 (76.8)	72 (14.9)	70 (14.5)
Odense (Denmark)	652	279 (42.8)	318 (48.8)	55 (8.4)	54 (8.3)
Strasbourg (France)	2,003	982 (49.0)	767 (38.3)	254 (12.7)	247 (12.3)
Mainz (Germany)	751	90 (12.0)	390 (51.9)	271 (36.1)	195 (26.0)
Cork & Kerry (Ireland)	807	275 (34.1)	426 (52.8)	106 (13.1)	99 (12.3)
Emilia Romagna (Italy)	2,219	0	1,894 (85.4)	325 (14.6)	325 (14.6)
Tuscany (Italy)	2,632	1,134 (43.1)	1,058 (40.2)	440 (16.7)	397 (15.1)
Malta	798	59 (7.4)	649 (81.3)	90 (11.3)	90 (11.3)
Southern Portugal	1,082	109 (10.1)	871 (80.5)	102 (9.4)	100 (9.2)
Asturias (Spain)	850	183 (21.5)	367 (43.2)	300 (35.3)	150 (17.)6
Basque Country (Spain)*	1,506	890 (59.1)	182 (12.1)	434 (28.8)	85 (5.6)
El Valles (Spain)	298	75 (25.2)	210 (70.5)	13 (4.4)	13 (4.4)
Vaud (Switzerland)	1,380	314 (22.7)	971 (70.4)	95 (6.9)	93 (6.7)
Glasgow (UK, Scotland)	1,206	133 (11.0)	859 (71.2)	214 (17.7)	214 (17.7)
Total	18,576	5,885 (31.7)	9,780 (52.6)	2,911 (15.7)	2,271 (12.2)

* Only data of 1999-2000 since ATC-codes were used before 1999.

Table 5: Proportion (%) of the 2,271 drug exposed cases reportedly exposed to any one of the 20 drug groups (excluding vitamins&minerals) in the first trimester per registry.

Eurocat code groups (code)	Antwerp (Belgium)	Zagreb (Croatia)	Odense (Denmark)	Strasbourg (France)	Mainz (Germany)	Cork & Kerry (Ireland)	Emilia Romagna (Italy)	Tuscany (Italy)	Malta	Southern Portugal	Asturias (Spain)	Basque Country (Spain)	El Valles (Spain)	Vaud (Switzerland)	Glasgow (UK)
N	139	70	54	247	195	99	325	397	90	100	150	85	13	93	214
Atropinics + antispasmodics (01)	1.4	0.0	3.7	23.9	0.5	1.0	0.0	3.5	0.0	1.0	5.3	0.0	0.0	1.1	2.8
Anaesthetics (02)	0.0	0.0	0.0	1.6	0.0	0.0	1.5	0.3	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Hypnotics, sedatives and psychotropics (03)	6.5	7.1	14.8	4.0	0.5	7.1	1.2	6.3	12.2	9.0	6.7	7.1	23.1	6.5	19.6
Antiepileptics (04)	7.9	1.4	7.4	4.9	0.0	4.0	0.0	2.0	4.4	0.0	1.3	12.9	0.0	6.5	4.2
Analgesics, antipyretics and antiinflammatory (05)	12.9	5.7	14.8	11.7	1.0	14.1	8.3	20.7	22.2	13.0	22.0	5.9	0.0	26.9	15.0
Histamine antagonists (06)	1.4	0.0	5.6	0.8	0.0	1.0	0.0	0.8	1.1	3.0	7.3	25.9	0.0	1.1	2.3
Antiasthmatics (07)	4.3	0.0	24.1	1.2	1.0	18.2	0.0	0.8	23.3	0.0	7.3	2.4	0.0	4.3	37.4
Antiarhythmics and antihypertensives (08)	6.5	8.6	1.9	7.7	4.1	2.0	1.2	1.0	2.2	10.0	0.7	9.4	7.7	2.2	4.2
Diuretics (09)	1.4	1.4	5.6	0.4	0.0	2.0	0.0	0.0	0.0	0.0	0.0	1.2	0.0	0.0	0.5
Tocolytics (10)	1.4	37.1	7.4	0.0	0.5	0.0	28.0	24.4	0.0	0.0	2.7	0.0	0.0	0.0	0.0
Antibiotics *et al* (11)	22.3	25.7	9.3	30.4	16.9	15.2	13.2	14.9	5.6	29.0	43.3	7.1	0.0	25.8	2.8
Immunosuppressives (12)	0.7	0.0	0.0	0.0	0.0	0.0	0.0	0.8	1.1	1.0	0.0	1.2	0.0	0.0	0.5
Anticoagulants *et al* (13)	0.7	0.0	0.0	1.6	1.0	0.0	1.2	2.3	1.1	0.0	2.0	1.2	0.0	1.1	0.9
Thyroid and antithyroids (14)	7.9	2.9	9.3	8.5	8.2	7.1	0.0	10.8	4.4	6.0	2.0	9.4	23.1	6.5	4.7
Hormones (15)	15.1	17.1	1.9	14.2	1.0	29.3	18.2	12.1	33.3	6.0	2.7	4.7	0.0	2.2	7.5
Adrenocortical steroids (16)	4.3	1.4	5.6	1.6	3.1	1.0	0.0	1.8	3.3	3.0	2.0	1.2	0.0	1.1	0.0
Antidiabetics (17)	2.9	1.4	9.3	4.5	4.6	6.1	0.0	1.8	6.7	4.0	2.7	7.1	23.1	11.8	5.6
Vaccins (18)	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	1.0	0.7	0.0	0.0	0.0	0.9
Other (88)	28.8	11.4	11.1	10.9	71.8	2.0	5.5	8.6	3.3	10.0	14.7	16.5	23.1	22.6	8.9
Drugs taken but no information (98)	0.0	0.0	0.0	0.0	0.0	2.0	44.3	22.7	0.0	12.0	0.0	0.0	0.0	0.0	1.9

Because a case can be exposed to more than one drug the row percentages can total over 100.

Discussion

Twenty-five of the 29 responding registries participating in EUROCAT collect data on first trimester drug exposure. Of these full members, 15 use the present numeric EUROCAT coding system and six use another coding system, primarily the classification ATC. Three of the four associate members use the ATC system. Among the full members who use the EUROCAT code there was a wide variation in drug exposure during the first trimester, ranging from 4.4 to 26.0 percent, excluding vitamins and minerals.

It is well known that the ascertainment of drug exposure during pregnancy can be difficult, particularly for over-the-counter preparations. In an earlier study, De Vries *et al* compared data on maternal drug use obtained from physicians and midwives from the EUROCAT-registry in the Northern Netherlands (NNL) to prescription data obtained from community pharmacies.[16] The cases were not linked and comparisons were made at population level. Compared to pharmacy data, the estimate of completeness of physician/midwife data varied between 0% for sympathomimetics, dermatological preparations, expectorants and products containing local anaesthetic to 59% for iron preparations. This study has led to changes in the ascertainment of data on maternal drug exposure in EUROCAT-NNL, including pharmacy data and a questionnaire for the mother as sources of information.[17,18]

Our questionnaire showed that the obstetric records are the main sources used for data on drug exposure although other sources, such as an interview with the mother are additionally used. Northern Netherlands was the only registry to directly access pharmaceutical records from community pharmacies.

Compared to the literature, the percentage of cases who were exposed to drugs in the first trimester was low in registry data, probably reflecting the limited sources of information used.[8,19-22] Nevertheless, in the period 1996-2000, 2,271 out of 18,576 cases (12.2%), originating from 15 registries, were recorded in the central database as having been exposed to medication (excluding vitamins and minerals). Although there is no systematic reference information on drug exposure in most registries, the existing data can be used for both hypothesis generation and hypothesis testing. Hypothesis generating analyses look for statistical associations between specific drugs and specific malformations, the method used by the Northern Netherlands register and the international MADRE project.[5-7] For hypothesis testing, the central database can be used to examine whether among the cases exposed to a drug of concern, a specific malformation is in excess relative to other malformations or whether the drug of concern is more frequent among cases of a specific anomaly.[8,9] Where

the EUROCAT codes have been used, it will usually be necessary to use more specific text information on the type of drug, dosage and timing, or re-examine the original records for further information.

The 23 category EUROCAT coding system was specially developed to monitor for any possible drug-related foetal malformation. In order to improve the data coding on drug exposure from 2005, EUROCAT will recommend that registries instead record drug exposure data using the ATC classification[23] to ensure that maximum detail on drug type is coded. With respect to missing data on drug exposure, or cases misclassified as unexposed, individual registries can only solve this problem by using more data sources for drug exposure, for example, utilising pharmacy data. Each type of data source, whether pharmacy data, maternal interview, or obstetric record, can be expected to have its own advantages and disadvantages with respect to knowing which drugs were prescribed, which were actually taken and when, and which over the counter preparations were taken. Linkage with other electronic databases can be particularly powerful, whether pharmacy databases or clinical databases referring to women with a chronic illness such as epilepsy or diabetes or asthma.

A further type of study for which the database can be used is to assess how many cases in the population have been exposed to known teratogens with characteristic malformation patterns, and thus how successful prevention of early pregnancy exposure to known teratogens has been. For example, an assessment of the number of isotretinoin and related drug exposed cases was made in the past by EUROCAT[8] and other registries. The remarketing of thalidomide will require this type of surveillance. For this type of investigation, an additional retrospective drug exposure enquiry should be made for all cases with the characteristic malformation pattern.

Case-control surveillance, where the same exposure information is collected for malformed cases and non-malformed controls would strengthen post-marketing drug surveillance. However, for registries with limited budgets, it is often not possible to allocate resources to collecting information on controls. Moreover, it can be difficult to ensure that the data sources are identical for cases and controls.

The variation between the registries in the proportion of exposure to the different drug groups may be due to variation in drug prescription policies among the European countries[8] as well as to variation in data sources used, variation in coding, differences in populations and population sizes, and the level of interest and resources of the registry for pharmacovigilance. Since the prevalence of anomalies can also vary between registries, careful control for registry is needed when pooling data and looking for drug-malformation

associations. Conversely, diversity in drug prescription policy can be an advantage when aiming to dissociate the effects of the drug for use in pharmacovigilance studies.[24]

Congenital anomaly register-based studies are only one of a variety of approaches needed for effective post-marketing drug surveillance, each with its own strengths and limitations. Other approaches include the yellow card system of cases reports of adverse exposure despite its problem of incomplete and biased reporting and lack of control information, and databases such as the General Practice Research Database (UK) where certain drug data may be well and prospectively recorded[25] but congenital anomaly data would be less well recorded and restricted mainly to surviving live births. In the future, especially with improvements in coding and sources of information used for drug exposure, EUROCAT can be an effective collaborative framework to contribute to the post-marketing drug surveillance to monitor and identify teratogens.

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Chapter 7

Collecting data on non-malformed births as controls for the EUROCAT Northern Netherlands database

Abstract

The possibilities to collect data on non-malformed births as controls for the EUROCAT NNL cases are explored. Data collection through pharmacies was examined first. Two possible methods led to selection and were therefore not suitable. As an alternative, data collection through a midwife practice was explored. From this exploration it became clear that this method is feasible in a midwife practice which gives reasons to be optimistic about the possibilities to include non-malformed births in the standard data collection in the future.

Introduction

In Groningen, the EUROCAT Northern Netherlands (NNL) registry for congenital anomalies was established in 1981 to collect data on children and foetuses with anomalies. Besides description of the anomalies present, data on all kinds of exposures around conception and during pregnancy are collected as well.

Since specific congenital anomalies are rare, the case control design is mostly used to study the association between an exposure and a congenital malformation. Choosing appropriate controls is the biggest challenge with this design. Often there is no so-called 'golden standard' to compare with. Theoretically one can choose either controls with other anomalies than anomalies under study (malformed controls) or controls with no congenital anomaly (non-malformed controls). Mothers of an infant with congenital anomalies recall more events and details about exposure than mothers with healthy infants.[1] By using malformed controls, the effect of recall bias can be decreased.[2] But if the selected malformed controls also have a relation with the exposure, selection bias can occur.[3] This selection bias might lead to an underestimation of the actual risk of the exposure under investigation.

One solution to address both selection bias and recall bias is to use both non-malformed and malformed controls. Currently, EUROCAT NNL does not collect data on non-malformed births leaving the use of malformed controls as only option. In this chapter, possible strategies to expand data collection with non-malformed controls are investigated. Firstly, a description of the EUROCAT NNL data collection is given. After all, controls should be selected from the same population as the cases and data collection methods should be comparable with data collection from EUROCAT NNL to avoid information bias. Secondly, a short overview of possible data collection strategies is given which is followed by a description of the explorative studies we have performed regarding these strategies. Finally the currently explored method is described, the first results are shown and limitations and recommendations for future data collection are discussed.

EUROCAT Northern Netherlands

In the EUROCAT NNL registry, data on live births, stillbirths and abortion with congenital anomalies are registered since 1981. The mother of the child should be a resident of one of the three northern provinces of the Netherlands (Groningen, Friesland or Drenthe). In this area, about 20,000 births per year occur.[4]

Between 1981 and 1996, information about the pregnancy outcome as well as the mother's condition, diseases and drug use was collected through the physician or midwife who reported the birth. Since 1991, this reporting needed the informed consent of the parents. In 1997, data collection methods were extended enormously.[5,6] Information about the anomaly is still provided by physicians, midwives, clinical geneticists and pathologists after informed consent is given by the parents. But for exposure data, parents receive a questionnaire in order to collect information about their characteristics like age, education, and family history. Data about illness during pregnancy, and life style aspects like smoking and alcohol consumption are asked for in this questionnaire as well. A request for the mother's pharmacy data is included with the questionnaire and sent to her pharmacy if she gives her consent. Data from one year before delivery are asked for. After receiving the questionnaire and the pharmacy data, mothers are interviewed by phone to clarify possible uncertainties in the questionnaires and to verify whether drugs reported by the pharmacy were actually used and in which time period the drugs were used. During this interview, information about Over-the-Counter (OTC) drugs is collected through a structured list of possible complaints during the pregnancy or in the three months before pregnancy.

No age limits for discovering an anomaly were applied until 2003, which means that besides anomalies leading to abortions and anomalies discovered at birth, anomalies discovered even years after birth are recorded as well. Since 2003, an age limit of 16 at the moment of enrolment is applied.

Data collection routes and preceding explorations

Possible strategies for continuous data collection on non-malformed births are infant welfare centres (consultatiebureau's), midwives and gynaecologists, and pharmacies. Since about 97% of all children visit an infant welfare centre, normally starting at the age of four months old, this would be an excellent source. However, previous research on drug use during lactation showed co-operation from these centres is not high, among other things because their high workload.[7] Midwives and gynaecologists could help recruiting newborns as well. To select a representative sample of newborns and their mothers, both sources should be combined. After all, women giving birth under supervision of a gynaecologist differ in medical background from women supervised by a midwife. Therefore their drug use might differ as well. Finally, pharmacies could be used as a source. Newborns are registered in pharmacies if they receive a drug prescription, if the insurance company informs the pharmacy about the birth or if the parents inform the pharmacy themselves.

Recruitment of non-malformed births via pharmacies

Because of existing contacts, data collection through pharmacists was explored first. After birth, all children receive one mg vitamin K1 to prevent Vitamin K Deficiency Bleeding (VKDB). Two weeks after birth, 59% of Dutch women still breast-feed her baby.[8] These breast-fed children also receive 0.025mg vitamin K1 supplements from day eight after birth up to three months of age.[8-10] Their vitamin K prescriptions can be used to extract newborns form the pharmacy database. This recruitment was tested in one pharmacy where mothers of children born in 2002 to whom vitamin K was prescribed, received a questionnaire. The response was 61%. Through contacts with the only midwife practice in the same small town, we could verify information from the questionnaire (retrospectively collected) with the information of these same women in the midwife database (prospectively collected). Using two sources, no differences were found for characteristics like age, parity, gravidity, gestational age, birth weight of the responders. But, research in the InterAction database[11] showed differences in drug use during pregnancy among mothers of children receiving vitamin K and not receiving vitamin K: mothers in the group with no vitamin K, indicating they do not breastfeed their baby, use more drugs in general expressed in number prescriptions received.[12] Furthermore, the percentages users of drugs related to rheumatoid arthritis, hormones, antidepressants, antiepileptics, antibiotics, corticosteroids, and antihypertensives were higher among non-breast-feeding women compared to breast feeding women.[12]

Another option of recruitment via pharmacies is through insurance data. This was possible for sick fund (ziekenfonds) patients since these parents have to register their newborn babies with the insurance company shortly after birth and the company notifies the pharmacy. Than it is up to the pharmacy to actually add the child into their database which takes between two and 12 months.[13] But privately insured children (particulier verzekerd) are mostly not registered at the pharmacy until their first prescription. Since insurance differs based on income, thus on social economic status, this data source provides us a selected population.

The conclusion of these two pilot studies was that both methods of selecting newborns via pharmacies lead to a selected population instead of a representative sample of the population of which EUROCAT NNL collects data on malformations. Furthermore, vitamin K is now an OTC drug thus recruiting through a prescription is not possible anymore and the Dutch insurance system has changed since January 2006 which makes that route more complicated as well. Both options above are even more limited now and another data collection strategy than through pharmacies has to be used. Therefore, data collection through a midwife practice was explored.

Recruitment of non-malformed births via midwife practice

Population and setting

In Veendam, a town with just over 28,000 inhabitants, the absolute number of live births varies from over 300 in the end nineties to about 260 in 2005.[4] There is one midwifery to serve pregnant women in and around Veendam. These midwives also provide postnatal care to women that gave birth in a hospital and returned home immediately after. Therefore they have contacts with almost all parents of newborns in the region somewhere between conception and the first week postpartum.

Method of data collection

The method of data collection is based on the method used by EUROCAT NNL, with the exception of the timing we ask for consent: for the non-malformed births we ask for consent before birth instead of after birth as with the EUROCAT NNL children. Figure 1 shows the flowchart of the data collection on non-malformed births. In the Veendam midwifery, all women attending for their first or second consult are provided with both written and oral information about the study by the midwife. If the women decides to participate in the study she is asked to fill out a consent form in which she agrees to receive a questionnaire after delivery, that her pharmacy data can be collected and that the researchers are allowed to call her to verify the collected information. On the consent form, besides personal information like address and date of birth, a question about the expected date of delivery is asked for as well.

Once every month, a list of all women that gave consent and of whom the expected date of delivery has passed, is sent to the midwife practice. They check for each woman whether she indeed gave birth and if the baby appeared to be healthy. After adding the dates of birth to the list, they send it back to the university and the mothers of the non-malformed babies receive a questionnaire that is based on the EUROCAT NNL questionnaire. At the same time, the pharmacy is asked to provide data of the year before the date of birth of the child. If the questionnaire is returned and pharmacy data are collected, the mother is called to verify use of drugs on the pharmacy list and use of possible OTC drugs. For the latter, the same structured list of complaints is used as with EUROCAT NNL. If the questionnaire or pharmacy data are not returned in four weeks, a reminder is

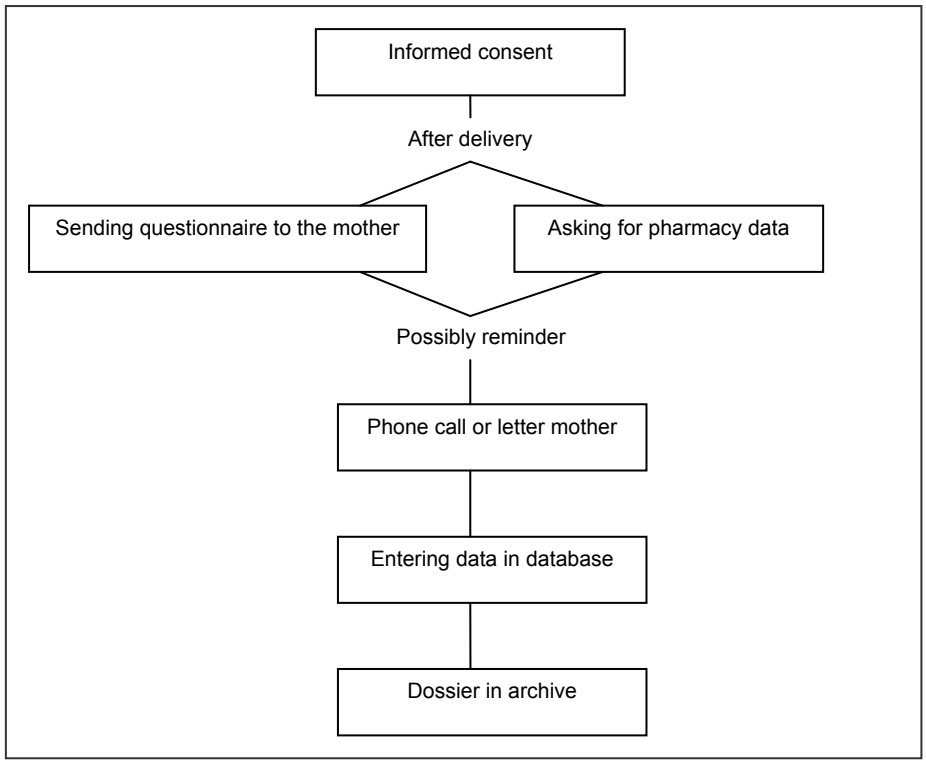

Figure 1: flowchart of data collection

sent. If the questionnaire is still not returned after a reminder, we are phoning the mother and go through the questionnaire together. If the mother cannot be reached by phone after trying multiple times a letter is sent with the remaining questions and the list of complaints, accompanied with a return envelop to encourage responding.

If the newborn has a congenital malformation, the mother is contacted and asked if her name and address can be passed on to EUROCAT NNL.

Comparison first findings with population Veendam practice and regional data

To validate whether the non-malformed births of which we collect data are a representative sample of the midwife practice of Veendam, characteristics of the included births are compared with data of the whole practice population. We could not compare our data with only those mothers whose data we did not have since we did not had their consent to use their data. Therefore we used the practice data all together without distinguishing between

participants and non-participants. Data on births of the first year of our data collection were included (December 2004 until November 2005) and the following characteristics were compared: maternal age, country of birth, employment status, parity and gravidity, gestational age and sex of the child. P values were calculated with t-tests and chi-square tests, where appropriate.

To gather some insight in the representativeness of the social economic status of the participants, we compared the educational level of our participants with the female population aged 15-64 in the three northern provinces of the Netherlands.

Attitudes of midwives and participants

Since this data collection is still in the pilot phase, we also tried to get insight in factors that influence participation rates. We therefore contacted and met with the midwives regularly to evaluate the process and discuss some limitations they encountered. We also interviewed 25 randomly chosen participants. These interviews were executed during the already scheduled phone calls to verify collected data. Open questions were asked about attitude towards the study and study material and reasons to participate in the study.

Results:

Participation rates

Between December 2004 and November 2005, 238 women in the Veendam midwife practice gave birth. Of these, 152 women (64%) consented to collect their data. Of these 152, three newborns had a malformation recognized at birth and all three mothers gave permission to send their information to EUROCAT NNL. The remaining 149 were sent a questionnaire and their pharmacy data were asked for.

Of the 149 eligible births, pharmacy data were collected for all, 112 questionnaires were returned without having to send a reminder, and another 31 after the reminder. Of the remaining six births, one the questionnaire was filled in during the interview by telephone, and one woman refused to cooperate because the questions were too personal. The remaining four could not be reached by phone so far. In total, 144 (97%) questionnaires are completed, and 120 (81%) are also reached by phone thus 120 files are complete. Some of the women (exact number unknown) were not phoned because of some logistic problems in the first months of data collection. After a few months it was decided that the time span between returning the questionnaire and the possibility to phone was too large and we felt we could not bother these women anymore.

Logistics and encountered limitations for midwives

The project, and especially the logistic part of handing out the leaflets, were discussed with the midwives at several meetings. Because they only have to hand out the leaflets and consent forms with a short remark, they pointed out the project is not troubling their daily work at all. Handing out the leaflet to the women they only visit postpartum is not daily routine yet and they indicated they forget to do that most of the times. Another issue that came up is that they sometimes decide not to hand out the leaflet if they think this mother is not capable of filling out a questionnaire.

Results of first year data

The characteristics of the 149 births with informed consent are given in table 1. Maternal age and gestational age could be calculated from the consent form and from the list returned by the midwife and are therefore available for all 149 births. The other characteristics were only available if the questionnaire was filled in (n=144). In table 1, characteristics of all births in the midwife practice are given as well and no differences between the women in the study and the whole practice population are observed (all p>0.05).

The educational levels of the responders were high for 14%, middle for 64% and low for 22%l. These percentages were 19%, 43% and 37% respectively for women aged 15-64 in the northern Netherlands.4

Table 1: Characteristics of mothers and children in the non-malformed database and in the midwife practice.

	Non-malformed births: responders Veendam (n=149)	*Whole population midwife practice (n=237)*
Mean age mother (sd)	30.0 (4.6)	30.1 (4.7)
% mothers born in NL[a]	95.1	92.8
% mothers with a paid job[a]	79.9	79.3
Mean parity[a] (sd)	0.53 (0.59)	0.61 (0.67)
Mean gravidity[a] (sd)	1.9 (1.00)	1.9 (0.97)
Gestational age (%)[b]		
< 259 days	25.5	29.1
259-269 days	43.0	41.4
270-276 days	20.1	21.5
>276 days	11.4	8.0
Birth weight in grams (sd)[a]	3493 (561)	3422 (564)

[a] data available for those with filled in questionnaire only (n=144); [b] percentages do not add up to 100% due to missing data

Interview with participants

Of the women that were phoned as part of the standard procedure, 25 were asked some extra questions about their participation. None of them refused to co-operate with this short interview. Of the 25 women, 20 participated because they think pregnancy research is important and the other five indicated it was just little trouble. None of the responders felt obligated to participate in the study or pressed to do so by the midwife.

The timing of receiving the questionnaire was fine and the questions themselves were clear. Nevertheless, details like specific dates were difficult to remember, especially since some of the questions were about the periconceptional period which is almost a year prior to filling in the questionnaire. Ten women remarked they could have made notes if they knew beforehand such details were asked for after delivery. Also, one participant mentioned, not all women getting pregnant are well informed before pregnancy and information they get during pregnancy can make them uncertain about their own behaviour during the first weeks of pregnancy.

The 25 interviewed participants had no problem being called to verify some details and go through the OTC drugs. Two of them even felt good about the attention paid to them and being able to explain some answers more in detail.

Discussion and conclusion

The availability of non-malformed controls would increase possibilities for research with the EUROCAT NNL data. Therefore a strategy to recruit a representative sample of non-malformed births in the region is needed. From the preceding explorative studies it was clear that recruitment via pharmacies would lead to selection and another data strategy was needed. Therefore, the feasibility of recruiting non-malformed controls via a midwife practice was explored in Veendam. After the first year of implementing the collection method, characteristics of the participants were compared with characteristics of all births in that practice in that year. Encountered barriers on the part of the midwives, the participants and the researchers were summarised.

Form the meetings with the midwives it became clear handing out the leaflet and consent form during the first or second consult of a pregnant woman was implemented easily in their daily practice. However, the few women they only counselled after birth did not always receive the leaflet and consent form. Moreover, the midwives sometimes decided not to ask for participation at all if they thought the woman is not capable. Therefore they introduced selection based on their judgement. Nevertheless, of all women giving birth in

their practice during the first year of data collecting, over 60% gave informed consent to contact them after delivery. From the rough comparison of the characteristics of the participants with all women in the database, these participants seem to be representative for the practice. We therefore can carefully conclude that collecting data via a midwife practice is feasible in contrast with pharmacies, but more effort should be put in to increase participation rates and to support the midwives in handing out the information to all women they counsel.

From the interviews with a sample of women that gave their consent, it is clear the responders are enthusiastic about participating in such a study, and the process and questionnaire are clear. Of course these were the women that already consented to participate instead of a sample of all women in the midwifery. From the comments it was clear some women feel the need to receive more information before pregnancy. Although Dutch health care providers consider preconception care to be important,[14,15] there is no structural preconception care available in the Netherlands at the moment. Furthermore, suggestions were made to make recall more easy. Methods to do so, like providing logbooks or collecting data prospectively, are available but data should be collected the same way as the EUROCAT NNL data to avoid other types of bias.

Although the data collection method seems to be feasible as described in this study, there are some limitations we should address. First, we only explored possibilities within this one practice and adjusted the logistics of the recruitment to the requirements of this practice. If the method is extended, this logistic should be investigated again for each new setting. Second, data collected should be tested more extensively on external validity. We have little information on births in Veendam outside this practice and about the not-participating women within the practice. The participants seem to be a good sample of the whole practice, but we only compared some of all possible characteristics and only from one year. We already know the midwives introduce some selection by not asking all women to participate. Moreover, we can imagine we miss complicated pregnancy where mothers have certain conditions that require intensive auditing of a specialist. Furthermore, we have not investigated the representativeness of the Veendam mothers and babies for the whole region of the northern Netherlands. Third, our current definition of non-malformed controls is restricted. We define a birth as non-malformed when no malformation is feasible at birth and therefore unknown by the midwife. Malformations discovered later after birth are thus included as non-malformed. Since the numbers that are misclassified are small it would not influence future studies. Nevertheless, misclassification should be minimised and this problem should be dealt with in future.

Issues presented in this chapter are only from an explorative study. Future plans should include at least the following three issues: 1) More data should be collected to be able to investigate validity issues more thorough. 2) To address more to other problems and limitations summarised above, we suggest data collection will be extended to another midwife practice with another population, for example to the city of Groningen. 3) To include also the more complicated pregnancies from mothers with health issues requiring a pregnancy and delivery being monitored by a gynaecologist. Therefore, extending recruitment via a gynaecologist is advisable.

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Part C

Implementing knowledge: folic acid education in pharmacy daily practice

Chapter 8

Use and predictors of use of folic acid among women with actual short term child wish

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Submitted

Abstract

Background: Although many studies have been performed on folic acid use among pregnant women, data on the preconception period is scarce.

Methods: Questionnaires were sent to oral contraceptive (OC) using women aged 25 to 35 from 23 pharmacies. Responders that stated they wish to become pregnant within two years, which was asked for explicitly, were selected for this study (n=528). Proportions of responders with specific knowledge about folic acid and self reported folic acid use were shown stratified on time span to pregnancy wish. Logistic regression was used to calculate odds ratios (ORs) and 95% confidence intervals (CIs) for various determinants in relation to folic acid use.

Results: Of the women that wish to become pregnant within six months, 69% knows about the protective effect of folic acid and 81% knows to start before conception and 64% knows both. These percentages drop with increasing term to wish to become pregnant. Among women planning their pregnancy within six months, 59% reports current use of folic acid compared with 6% among those wishing to conceive in 12-24 months. In the multivariate model, attitude, knowledge and source of information are significantly associated with use of folic acid.

Conclusion: Of women planning their pregnancy within six months, 59% reports current folic acid use. Since folic acid use is associated with knowledge, a positive attitude and receiving oral information from a health care provider, interventions to increase folic acid use should be aiming on these variables.

Introduction

Besides neural tube defects, folic acid probably protects against other birth defects as well.[1] In the Netherlands, folic acid fortified foods are not mandatory and women trying to conceive are recommended to use supplements at least from 4 weeks before conception until 8 weeks after. Since the rate of planned pregnancies is high, about 85-90%,[2] using supplements before conception can be planned as well by most women.

To increase use of folic acid among women planning a pregnancy, it is important to get insight in the determinants that predict this behaviour. According to the theory of planned behaviour, intentional behaviour, a good predictor for actual behaviour, is determined by the persons own attitude towards the subject and the attitude of people in their surroundings that are important in relation to the subject (social norm).[3] Thus, before using folic acid, women planning their pregnancy should have a positive attitude towards folic acid, which is partly formed by having correct knowledge.

Although many studies about periconceptional folic acid use have been published, these yielded only scarce valid data about the use in the preconception period. Ray *et al* published a systematic overview of 52 studies that evaluated the rate of periconceptional folic acid or multivitamin tablet supplement use.[4] Among the 34 studies that surveyed about preconceptional use of folic acid, rates of use ranged from 0.9% to 49% of the surveyed women. Periconceptional use, assessed in 49 studies, ranged from 0.5% to 52%. Half of all studies (n=26) were conducted among pregnant women and about a quarter postpartum (n=12, 23%). Information collected during or after pregnancy might give a biased estimate of preconception use. Pregnant women might have received information about folic acid during their prenatal consult which increased their knowledge compared to their preconception knowledge. Also, their answers could show an overestimation of use based on social desirable answering. Five studies (9.6%) were conducted among a general population of non-pregnant women. These non-pregnant women might not be interested in getting pregnant so gaining information from them might be too general. Finally, Ray *et al* found five studies (9.6%) where women were asked about folic acid at the time of family planning. However, these were selected populations, e.g. women visiting a reproduction clinic or married couples receiving preconceptional education. In the Netherlands, no widespread preconception care is present.

In this study we aim to evaluate actual use and intentional use among women with a short term pregnancy wish, and the variables associated with this use in order to refine current knowledge about determinants of pregnancy related folic acid use.

Methods

Design

This study is an observational study for which we used baseline data from a larger study with a pre-post intervention-reference design. The aim of this larger study is to measure the effect of implementing folic acid education in daily practice of Dutch pharmacies. This folic acid education is described in detail elsewhere.[5] Briefly, women visiting their pharmacy to collect oral contraceptives (OC) received oral and/or written information about the benefits of periconceptional folic acid use. Questionnaires were sent before and after the implementation to women aged 25 to 35 from 13 intervention pharmacies and 10 reference pharmacies, who visited their pharmacy seven to twelve months earlier to collect their OC. For the baseline measurement, 2821 questionnaires were sent of which 1965 (70%) were returned and 1838 (65%) were eligible for analyzing. Response per pharmacy varied from 44% to 91%.

For the current study we selected all responders to the baseline questionnaires that stated they wish to become pregnant within two years. In the questionnaire this was asked for explicitly, divided in three mutually excluding questions: “do you intend to become pregnant within 6 months”, “do you intend to become pregnant within 1 year” and “do you intend to become pregnant within 2 years”. If they answered “yes” to one of these questions they were considered having ‘actual short term pregnancy wish’ (n=528 of the 1838).

Outcome measure: Use of folic acid

These 528 women that stated they wish to become pregnant within 6, 12 or 24 months were asked if they were using folic acid. They could choose between the answers “yes, because….”, “no, but I intend to use it because….” and “no, because…..”. They were asked to fill out the reason why they did or did not use folic acid.

For the second part of the study in which we investigate variables associated with folic acid use, we combined the current users of folic acid and the intentional users into one group and compared them with the non-users, to avoid small numbers. The theory of planned behaviour says intentional behaviour is a good predictor of actual behaviour, which partly justifies this conjunction.

Determinants for behaviour

Knowledge of folic acid is explored by two open answer questions to measure the knowledge about folic acid: “What have you heard or read about folic acid?” and “What do

you think is the best period to use folic acid if a women wants to become pregnant?". In the analyses, women were considered to have correct knowledge about the reason to take folic acid if they were able to mention something about prevention of birth defects and/or refer to neural tube defects in the answer of the first question. They were considered to have correct knowledge about timing of use folic acid if they could mention the advised period of use or at least stated one should start using folic acid before conception or directly when ending OC use. For the binary variable "correct knowledge", the responder was considered having correct knowledge about folic acid if both knowledge factors were present.

Attitude was measured by asking whether the responder would advice other women trying to conceive to take folic acid (yes or no/don't know).

In the questionnaire, responders could score one or more sources of information from a list of alternatives. Although we did not directly ask about the opinion of people in their surroundings, the sources they mention can be seen as operationalization of social norm in the behavioural model. We feel the health care professional has a strong role in this social norm, which is supported by several findings. For example, women of childbearing age in the polls of the March of Dimes stated that recommendations of a physician would positively influence their decision to take folic acid.[6] Also, de Jong-van den Berg *et al* showed consulting a health care provider before becoming pregnant was a strong predictor of folic acid use.[7] In the present study we made three groups of source of information on folic acid: oral information from a health care professional (with or without additional information on writing), a leaflet or other written information from a professional and no health care professional as source (other source or no source of information given).

Co-variables

We included the following variables of interest we could derive form the questionnaire: age of women (≤30 and >30), educational level (low is less than high school, middle is high school, and high is college or university), have been pregnant before (yes/no), ethnicity (born in the Netherlands, yes or no).

Data analysis

Data were analyzed with SPSS (version 12). Of all the 528 responders with actual child wish, we had data on knowledge of folic acid. We had missing data on use for 9 responders and another 9 responders had missing data on one or more of the other variables. Knowledge of folic acid was described for women that wish to become pregnant within 6, 12 and 24 months separately as well as for all responders to the questionnaire with no short

term pregnancy wish. Use of folic acid was described for the three groups of women with actual pregnancy wish.

Since two years is a long period to plan behaviour, we chose a smaller population for the second part of the study in which we aim to investigate which variables influence folic acid use. For 289 responders that wish to become pregnant within 6 or within 12 months and of whom we have full data, logistic regression was used to calculate univariate and multivariate odds ratios (ORs) and 95% confidence intervals (CIs) for various risk factors in relation to use.

Results

Of the 1838 questionnaires available for analysis from the baseline measurement, 528 were from women who wish to become pregnant within 24 months: 178 within 6 months, 120 in 6-12 months and 230 in 12-24 months. Of these three groups and of the remaining responders (n=1310) the percentages with correct knowledge on why and when to use folic acid are shown in figure 1. Of the women that wish to become pregnant within six months, 69% knows about the protective effect of folic acid and 81% knows to start before conception (24% mentioned the recommended period) and 64% knows the correct answer to both questions. These percentages drop with increasing term to wish to become pregnant. For responders with no wish to conceive in two years, the percentages are the lowest (43%, 58% and 37% respectively). If we consider all responders together, regardless of pregnancy wish, 47% knows about the protective effect of folic acid, 61% knows women should start taking folic acid before conception and 41% knows both (not shown in figure). Moreover, differences per educational level were observed: among women that wish to become pregnant within six months, 74% of the higher educated women knew about the protective effect of folic acid against 55% of lower educated women. For knowledge on timing of folic acid use, comparable differences in educational level were seen.

A positive attitude towards folic acid is associated with knowledge about the protective effect of folic acid. Of the women that wish to become pregnant within two years and that knows folic acid protects against birth defects, 84% would advise other women to use folic acid compared to 38% among those that did not mention this protective effect of folic acid ($\chi 2$, $p<0.001$). This statistically significant association between knowledge of the protective effect of folic acid and having a positive attitude was also seen in the three separate subgroups of women with actual pregnancy wish (within 0-6, 6-12 or 12-24 months) and among women with no short term pregnancy wish.

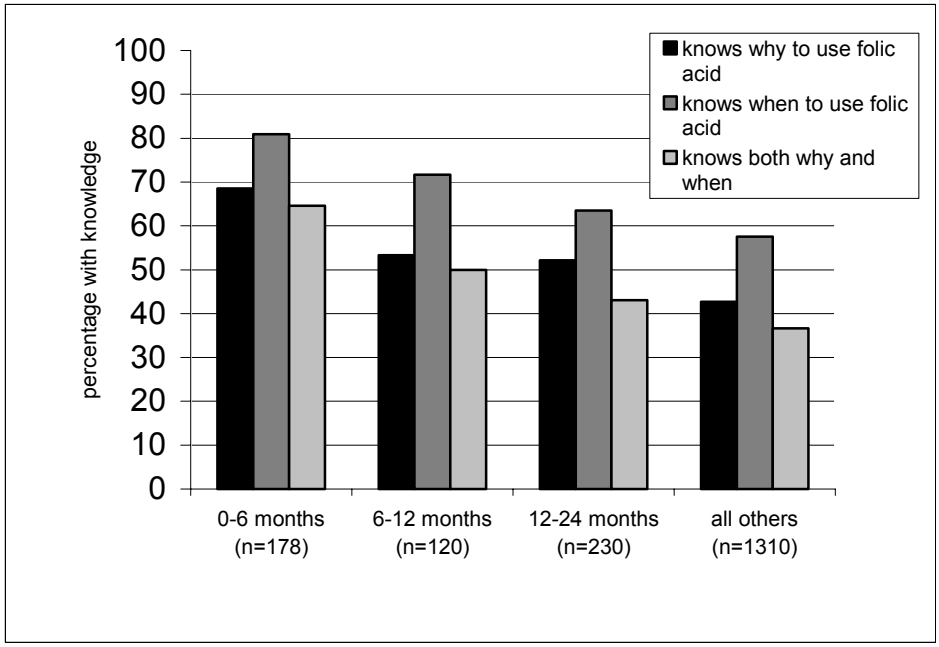

Figure 1. Percentage responders with correct knowledge on folic acid by term women whish to conceive (n=1838).

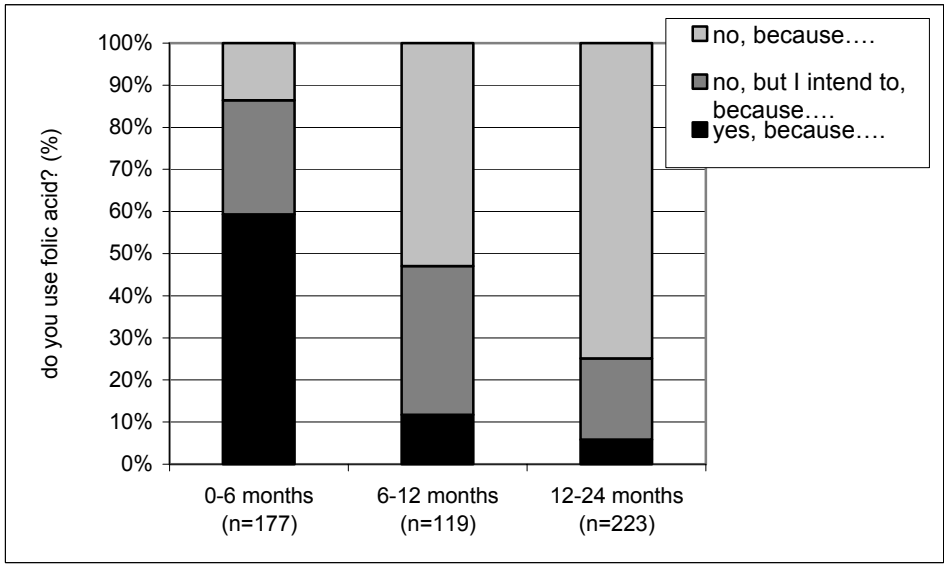

Figure 2. Use of folic acid by term within the women wishes to conceive (n=519).

Table 1: Use of folic acid or intention to use folic acid in relation to possible determinants among women wishing to become pregnant within one year (n=289)

		use or intention to use folic acid					
Factor	total (n)	crude OR	95% CI		adj. OR	95% CI	
correct knowledge on folic acid							
knows why and when	173	7.95	4.02 -	15.71	3.47	1.37 -	8.78
knows why or when	63	4.86	2.21 -	10.70	2.51	0.94 -	6.70
none	53	1					
would advice other women to use folic acid							
yes	209	4.89	3.27 -	7.31	6.10	3.02 -	12.29
no/don't know	80	1			1		
source of information							
oral information professional	61	3.72	1.68 -	8.25	3.32	1.36 -	8.10
leaflet professional	25	1.78	0.68 -	4.65	1.32	0.44 -	3.97
other source/no source	203	1			1		
born in the Netherlands							
yes	268	0.92	0.35 -	2.47	0.32	0.09 -	1.08
no or not known	21	1			1		
education of mother							
high	115	1.80	0.91 -	3.59	0.93	0.38 -	2.28
middle	120	1.49	0.76 -	2.91	0.88	0.38 -	2.04
low	54	1			1		
have you ever been pregnant?							
yes	125	1.19	0.72 -	1.99	0.63	0.32 -	1.25
no	164	1			1		
age of mother							
≤ 30	148	1			1		
> 30	141	1.34	0.81 -	2.22	0.75	0.40 -	1.41

Among the responders with concrete pregnancy wish, use of folic acid or intention to use folic acid differs per term to wish to become pregnant (figure 2). Among women planning their pregnancy within six months almost 60% reports current use of folic acid compared with 6% among those wishing to conceive in 12-24 months. Reasons for not using folic acid but having the intention to start using it were mostly that they did not try to conceive at this

moment. Reasons for not using folic acid and also not have the intention to start using it were that they did not think taking supplements is necessary or they did not know about folic acid.

Table 1 shows the results for the analyses of variables that might influence the use of folic acid. In the multivariate model where each association with (intentional) use is adjusted for all other variables, three variables are significantly associated with use: knowledge (knowing both why and when to use folic acid), attitude (advising other women), and source of information (oral information from professional). Attitude is strongest correlated with use or intention to use (OR=6.10). The remaining variables have no association with use or intentional use, neither in the univariate model nor in the multivariate model.

Discussion

Majority (59%) of women planning to become pregnant within six months report actual use of folic acid and another 27% has intention to use. With decreasing time span to pregnancy planning, percentage of use increases. The same pattern is seen with respect to knowledge of folic acid, which is the highest among those planning to become pregnant within six months. Correct knowledge, attitude and oral information obtained from a professional are associated with folic acid use among women planning their pregnancy within 12 months, in the univariate as well as in the multivariate analysis. Having a positive attitude towards folic acid, one of the determinants for planned behaviour, has the strongest association with reported use of folic acid. As the results show, knowledge is significantly correlated with attitude. Therefore knowledge is a strong predictor of use, both direct as indirect.

The strength of this study is, in our opinion, the fact our population consists of women that wish to become pregnant and of whom we could gather information about folic acid use and determinants of this use prospectively, before they actually are pregnant. Since preconceptional care is not standard in the Netherlands, reaching this population is difficult.

The women were recruited based on a previous OC prescription, which could be a limitation of our study. Although we have no evidence that characteristics of OC users differ from non-users, we can imagine that OC users plan their pregnancies more often. If this is true, the rate of planned behaviour towards folic acid might also be high, which decreases the external validity of our study results. De Walle found indeed different percentages of folic acid use among women that have used OC before pregnancy and those that have not.[8]

Another limitation of our study is that we have no information about our non-responders. There might be more women with actual pregnancy wish among the responders compared

to non responders. But, since that is our population of interest, we expect this does not introduce bias considering our research question. However, we expect responders compared to non-responders have more affection with the topic of our study which might bias our results in a way that our responders have a more positive attitude towards folic acid and a higher use of folic acid. Also, our responders have a relatively high educational level, as is often seen in questionnaire research. On the other hand, educational level did not seem to influence our multivariate model. Furthermore, we have low percentage responders not born in the Netherlands. Therefore our results cannot be generalized to women born outside the Netherlands.

Rate of folic acid use is much higher than in the studies summarized by Ray *et al*.[4] Even of the five studies that investigated folic acid use in the time of family planning, four had a rate under 40% and only one found a rate of almost 50%. This study was performed in a reproductive clinic. But, if we compare our findings with other findings in the Netherlands, we do find similar results. Among pregnant women in the northern Netherlands in 2005, 51% had used folic acid in the entire advised period of four weeks before until eight weeks after conception and another 21% started using the supplement after conception.[8] Our findings show that 59% of the women planning their pregnancy within six months report current use of folic acid and another 27% of those have intention to start using the vitamin. These comparable results might indicate that part of the women with intention to use folic acid start their actual use after conception.

The difference in educational level on knowledge we observed among those planning to become pregnant in six months were comparable with findings among pregnant women in the northern Netherlands in 2000.[9] Although the educational level itself is not associated with use, knowledge is.

Overall, the model of Ajzen[3] seems to apply in this setting. Attitude towards folic acid, correct knowledge and professional source of information are associated with use or intentional use of folic acid in our model after adjusting for all other variables. Also, with decreasing time span in which a pregnancy is wished an increasing rate of folic acid users is observed indicating a strong external validity for the instrument used. Moreover, we show that knowledge about folic acid is higher among responders that wish to become pregnant within six months compared with those that wish to get pregnant in 6 to12 months. This suggests women closer to pregnancy become more open to information on folic acid. This is in accordance with the model of ‘stages of change’ which distinguishes several stages in which people can be within behavioural change: no interest, contemplation, prepare behaviour, actual behaviour, and maintain new behavior.[10] This means interventions aiming

to change behaviour should take the different stages into account since education is only effective if the receiver is 'open' to the given information, if they are interested in the subject. Based on this model and our findings we recommend women should be educated or re-educated about folic acid in the year before they try to conceive. We therefore recommend effort be made to reach this population that changes constantly over time.

We conclude that current folic acid use among Dutch previous OC users that wish to become pregnant within six months is 59%. Since this folic acid use is associated with knowledge, a positive attitude and receiving oral information from a health care provider, interventions to increase folic acid use should be aiming on these variables. We recommend comparable studies should be performed among other populations (e.g. non-OC users, women of non-Dutch origin) in order to provide health care providers with more tools for efficient patient education.

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Chapter 9

Pharmacists' role in awareness about folic acid: the process of introducing an intervention in pharmacy practice

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Abstract

Objective: To determine whether a multiple intervention program is feasible in pharmacy practice; which adjustments in organisation and materials will improve the feasibility; how the target group will experience the intervention.

Method: In each of the 4 pharmacies a core team (1 pharmacist and 1-2 technicians) was formed that was responsible for the organisation and implementation of the intervention. The intervention existed minimally of adding a label about folic acid (FA) on the box of oral contraceptives (OCs) and handing out a leaflet about FA at least once during the intervention period. The intervention was planned or modified during core teams meetings every six weeks. Adjustments were made based on experiences of the pharmacy team, on responses in the pharmacy and on the results of a questionnaire sent to women one week after they visited the pharmacy with a OC-prescription. This cycle of planning, action, observation and reflection was repeated twice.

Setting: four pharmacies in eastern region of The Netherlands.

Key findings: The minimal intervention was carried out by all 4 pharmacies. Other activities differed per pharmacy: two introduced an age limit of 40 years for handing out the leaflet, two installed an electronic display, three worked with posters and display windows, and in two pharmacies the pharmacy technicians wore buttons and a portfolio was placed in the public area. Of the target group, 44% was positive about the received sticker, 49% was neutral and 4% was negative. Besides, 56% of the target group stated that they appreciate the preventive information given through the pharmacy.

Conclusion: Working with core teams seems to be a successful strategy. By editing the intervention per research cycle during the core team meetings an optimal intervention can be reached that fits in the existing organisation and possible barriers can be overcome. The target group was positive about the received information, which motivated the core teams enormously.

Introduction

Since the protective effect of folic acid on having a child with neural tube defects is known,[1,2] many efforts have been made to get women of childbearing age to take enough folic acid. In 1993, the Dutch Inspectorate of Public Health recommended that all women planning a pregnancy should consume 0,5 mg folic acid tablets daily in the periconceptional period from 4 weeks before conception until 8 weeks after conception.[3] This recommendation lead to a mass media campaign about the periconceptional use of folic acid in September 1995.

In 2000, De Walle *et al* asked pregnant women at one of their antenatal visits about their knowledge about and use of folic acid.[4] Although 82% of the respondents had a planned pregnancy, only 36% used folic acid in the advised period and 25% stated not to have used folic acid at all. Among these non-users 14% reported this was a conscious choice and the remaining responders stated they never had heard about folic acid or heard about it too late.

Also in 2000, the Health Council of the Netherlands advised against fortification with folic acid of staple food. Fortification should be limited to specific foods that are aimed at the target group.[5] However, even this fortification is not allowed at the moment; thus consumption of FA supplements is the only option. Therefore the problem of reaching the target group (women planning their pregnancy) and inform them about the benefits of folic acid exists. In the Netherlands no structural preconception consultation exists and because folic acid should be taken before conception, regular antenatal visits of pregnant women to their physician or midwife, even in early pregnancy, are no option for advice in time. However, over 80% of the Dutch women plan their pregnancy and over 70% take oral contraceptives (OCs) somewhere before their first pregnancy, at least for a while. This means that young women visit their pharmacy regularly and thus folic acid education through pharmacies seems a good opportunity to increase periconceptional folic acid use. Generally spoken, it is the responsibility of health care providers to give information about negative as well as positive effects of drug exposure during pregnancy. Traditionally pharmacists give information about dispensed drugs. However, in pro-active patient education clients are exposed to the intervention without an existing latent or manifest actual need or request of the client to be exposed. This role in preventive health care as well as the pro-active approach in patient education is new to pharmacists. Thus new questions and problems will rise. Yet, comparable cases in which daily routines and practices are subject of intended quality improvement in general practice care, including pro-active patient education, are known. Evaluation and controlled studies conducted in the area of health

care are therefore very informative for our case. These studies came forward with several applicable conclusions. First, the chances on a successful voluntary introduction and implementation of changed or new methods of working increase if alleged actors of change are deliberately led through the stages of orientation, interest, evaluation, intention to change and planning to change. Second, in the stage of introduction of the change and implementation, action research can help to identify barriers and create solutions that are fully supported by the health care workers themselves.[6] Furthermore, feedback of outcome measures can add significantly to the magnitude of the improvement achieved.[7] Finally, multifaceted intervention programmes can induce relevant improvements in health care practice with more chance than non-multifacet programms.[8,9] To be able to execute the multiple intervention a set of procedures and facilities was required, that was not yet part of the normal workflow. This implied that the pharmacy had to initiate procedural and behavioural changes in daily practice actively. The main objectives of this study were therefore whether a multiple information intervention program can be carried out in daily practice of a pharmacy; which adjustments in organisation and materials will lead to a better feasible intervention; and how the target group will experience the intervention.

Methods

After introducing the research proposal to a group of pharmacists, several pharmacies wanted to participate in this project. The study was carried out in four pharmacies (A, B, C and D) of which C and D have a partnership. These four were randomly chosen from the volunteers. In each pharmacy a core team of one pharmacist and one or two pharmacy technicians was formed. These teams were responsible for the organisation and implementation of the intervention in their pharmacy. The intervention itself was carried out by the whole pharmacy team. These teams consist of 1-3 pharmacists and 8-12 qualified technicians, depending on the number of patients served.

Intervention

The intervention should minimally consist of adding a sticker about folic acid on the box of oral contraceptives and handing out a leaflet about folic acid at least once during the intervention period. There was one ready-made leaflet available that could be used, but pharmacies were free to make one themselves. No sticker was available. Other means to

reach the women of childbearing age like posters and display windows were up to each core team to decide on.

Research cycles

The introduction and implementation if the intervention was set up over three cycles. Each cycle took six to eight weeks and consisted of four stages: planning, action, observation and reflection. During core team meetings with the researchers the intervention was planned (initial plan on first meeting, modified plans on following meetings). Each core team planned the intervention in a way they thought it was feasible in their pharmacy organisation at that moment. By this approach differences in intervention programs were possible between the four pharmacies. After each meeting whatever actions were thought necessary were undertaken in order to carry out the intervention. In all cases at least (modified) intervention plans were communicated with the whole team and then the team tried to carry out the (modified) intervention for six to eight weeks. During this phase of the cycle, it was actively observed how the intervention was carried out. Observations were done by the core team and by the researchers (see below). At the end of each cycle reflection of the former weeks took place, based on the experiences of the pharmacy team, and the answers on a questionnaire among the target group (see below). All results were discussed under supervision of an experienced conductor (DdS). Based on this reflection the next cycle was planned. At the end of the third cycle the optimal intervention per pharmacy should be reached.

Observations

To collect information about reactions at the counter, a small form was made on which the pharmacy technician could register questions or remarks (positive or negative) from women and their response. At each core team meeting these forms should be collected by the research team.

A questionnaire to measure the attitude of the target group contained among others closed questions about the sticker on the pillbox, the leaflet, opinions about these items and socio-demographic variables (table 1). Per cycle, 30 questionnaires per pharmacy were sent, spread over three weeks (a total of 360). The questionnaire was sent about one week after the women visited her pharmacy with a pill-prescription. Although the questionnaire was pre-tested and agreed on by a senior investigator, some small change in questions and lay-out were made after the first cycle based on the scored answers. Reminders were sent to all subjects of the second and third cycle, about 2 weeks after they received the questionnaire.

Table 1: questions and answers (frequencies) of questionnaires and characteristics of responders

	Frequency (%)
Send questionnaires	360 [1]
Response	164 (45.6)
Characteristics of responders (N=164)	
Mean age (range)	29.4 (18-49)
Born in the Netherlands	154 (94.5)
Mother born in the Netherlands	145 (89.0)
Father born in the Netherlands	149 (91.4)
What do you think about the label? [2] (N=144)	
Good that the pharmacy informs me like this	64 (44.4)
Fine, it doesn't trouble me	5 (3.5) only asked for in first cycle
Doesn't apply to me but it doesn't disturb me	66 (45.8)
Annoying / Very annoying	3 (2.1)
Else, like…	3 (2.1)
Missing	3 (2.1)
How would you like it if from now on you would always find a label on your pillbox? [3] (N=152)	
Good that the pharmacy informs me like this	66 (43.4)
Fine, it doesn't trouble me	11 (7.2) only asked for in first cycle
Doesn't apply to me but it doesn't disturb me	61 (40.1)
Confusing	3 (2.0)
Annoying / Very annoying	3 (2.0)
Else, like…	2 (1.3)
Less often, say once a year, would be enough	6 (3.9)
What do you think about this attention for folic acid by the pharmacy? (N=163)	
Very good that pharmacy gives preventive information	91 (55.8)
Leave it to the GP	0
Don't trouble me with it	1 (0.6)
Very exaggerated, I've no intention getting pregnant before long	4 (2.5)
I've noticed nothing about it	16 (9.8)
Else, like…	6 (3.7)
No opinion	5 (3.1)
No answer	40 (24.5)

[1] Per pharmacy 3*10 =30 per cycle, so in total 3*4*30=360 questionnaires were send. Of these, 164 returned, one was unfit to use the answers of so 163 useable questionnaires. [2] Percentages are based on 144 responders who stated to have seen the label.

[3] The percentages are based on 152 responders: 144 who stated they've seen the label and 8 in the 2nd and 3rd cycle who stated not have seen the label but who were referred to this question anyway. In the 1rst cycle this question was skipped after not have seen the label.

Results

First cycle

At the first core team meeting the core teams together formulated the text of the sticker they all used: “Child wish? Ask for information about folic acid in your pharmacy.” With regards to the leaflets, the pharmacies had different approaches. Pharmacies C and D placed leaflets in the public area of the pharmacy while in pharmacies A and B a leaflet would be handed out to every woman who collected her OC. They all decided to use the available leafl*et al*though they did not like the layout (only text, no pictures). In pharmacy B personnel added a letter about the project with the leaflet. Core team A explicitly mentioned in their plan to give the women oral information. An overview of all interventions executed is given in table 2.

Table 2: intervention per cycle per pharmacy

	Pharmacy A			Pharmacy B			Pharmacy C			Pharmacy D		
Interventions	1	2	3	1	2	3	1	2	3	1	2	3
Label												
on all OC	X	X		X	X	X	X	X	X	X	X	X
age limits (19-42)			X									
Leaflet												
with all OC	X	X		X	X			X			X	
age limit 19-42			X									
age limit <42						X						
only in public area of pharmacy							X		X	X		X
Letter with leaflet		X	X	X	X	X		X			X	
Notation in computer about given information	X	X	X	X	X	X		X			X	
Information by other means												
electronic information display						X		X				
poster(s)		X							X[1]			X[1]
display window /showcase		X									X	
button(s)												
information portfolio in public area									X[1]			X[1]
									X[1]			X[1]

[1] for one week during this cycle.

From the reflection of this first cycle it came forward that putting the labels on the boxes was fully integrated in the daily routine of all pharmacy team members and that leaflets were handed out as planned. For informing their colleagues about the intervention, each team used their usual methods (post boxes, general notice board and/or regular team meetings).

The forms for the patient comments at the counter were not used. It was considered too much work and no direct relevance was felt by the team members. A few negative reactions at the counter were brought up anyway and it was felt that those were the main dilemma during the project within the pharmacy teams. The discussion was about handling negative or painful reactions as well as the fact that team members (especially technicians) felt uncomfortable giving information about folic acid to women above childbearing age. In this discussion it was brought up that giving proper (oral) information at the desk can prevent negative reactions and maybe the letter used by pharmacy B might help as well. For example, a woman over 40 can be explained that the information given might not be applicable for her but she could pass it through to younger women in her surroundings. Besides, one pharmacist stated: “sometimes giving information is more important than the few negative reactions at the counter and the latter should not lead to stop giving this information.” It was agreed on that if a woman states, even after explanation, that she wishes not to receive this information again, a notice could be made in the computer so she would not receive the sticker and leaflet anymore.

Second cycle

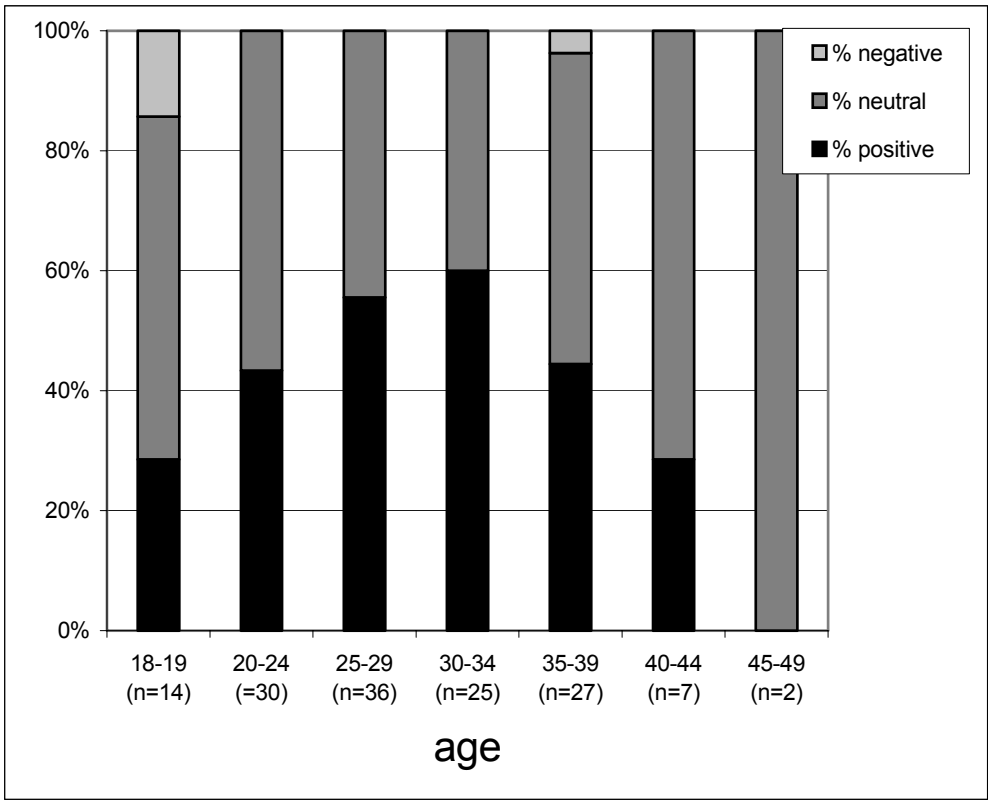
After evaluating the first cycle, all core teams planned the second cycle. As table 2 shows, no changes in the intervention were made by team B. The other pharmacies took over the letter with the leaflet and used posters, a showcase and an electronic display. In pharmacies C and D the leaflet was handed out actively with the OCs.

During the third meeting the second cycle was reflected upon. Again the negative reactions and the age of women receiving the intervention were the main discussion subjects. The counter forms were hardly used again and therefore the counter teams were asked to sit down with their colleagues once or twice in the third cycle to collect the reaction everyone could remember.

Third cycle

To avoid some of the negative reactions and feelings core team A decided to use age limits (19-42) for both label and leaflet while in pharmacy B only an age limit (<42) was introduced for the leaflet. These ideas were enthusiastically accepted by their colleagues, to whom it

Figure 1: percentage responders with a positive, neutral or negative attitude towards sticker on pillbox.



was a kind of relieve. In pharmacy B was decided that women who already had received a leaflet and entered the pharmacy again with an OC prescription, would not receive a leaflet again. Although pharmacies C and D stopped the active distribution of the leaflet to the individual OC users they did organise a theme week. An information portfolio about folic acid and other pregnancy topics was placed in the public area. Buttons and posters were used to encourage people to ask questions. The core team stated: “No concrete questions were asked, but at the end of each day the portfolio was obviously used and had to be tidied up”. No core team had found the time to sit down with their colleagues and collect the counter reactions.

Questionnaires

Out of 360 questionnaires, 164 were returned (response 47%) and 163 were eligible for further analysis. Because the pharmacies delivered us the ages of the non-responders we could compare ages of responders with non-responders. No difference was found (T-test, p=0.92). Although the response rate differed per pharmacy and per cycle, these differences were not statistically significant (Chi-square p=0.06 for pharmacies and p=0.34 for cycles). Compared to the overall Dutch population aged 15 to 64 years, our responders have a higher education and compared to the female working population of this area our population has relatively more women with a paid job (64% vs. 45%).[10]

Of the 163 responders, 144 (88%) claimed to have received the label on the box. The percentages that did not receive the label differed per cycle: 22% in the first cycle and 3% and 11% in the second and third cycle. Differences per pharmacy were due to differences in the first cycle. Of the total of 144 who stated having received the sticker, 18% had not seen it until filling in the questionnaire.

The attitude toward the label was mainly positive ('good that the pharmacy informs me like this') or neutral ('fine, it doesn't trouble me' and 'doesn't apply to me but it doesn't disturb me'). The same kinds of results were seen for the question "how would you like it if you would always find a label on your pill-box from now on?". Six (4%) negative reactions ('confusing', 'annoying' and 'very annoying') occurred. Figures 1 shows that among the age groups 25-29 and 30-24 a higher percentage was positive towards the label than in the youngest and older age groups. The same pattern was found for the question about always receiving the label on the pill (figure not shown).

Over half of the responders (57%) claimed to have received the leaflet of whom 53% read this leaflet. Keeping the leaflet was not dependent of having read the leaflet (41% in both 'read it' and 'not read it').

After three cycles

At the last core team meeting the whole intervention period was evaluated. The core teams experienced the meetings as very helpful, especially the contacts with other core teams. It was encouraging to hear from others about same kind of problems and finding useful solutions together. Discussing the results, mainly of the questionnaire, was experienced very positive. It was motivating to see direct effects of the efforts made. Making a total plan for each cycle was stated to be a bit laboriously, due to the fact the intervention was implemented in a good way from the beginning thus only small changes were made in the second and third cycle. They all would recommend this intervention to their colleagues.

Implementation of this intervention is relatively simple, but it is very important to have the support of the whole pharmacy team.

All four pharmacies made plans to continue their actions on giving information about folic acid to women who visit the pharmacy with an OC prescription.

Discussion

Working with core teams and meetings around research cycles seems to be a successful strategy for two reasons: the tailor-made intervention and reflection during the meetings. The core teams formulated plans as they thought to be feasible in their daily practice and therefore they were able to introduce the intervention by different models without losing the fixed conditions regarding label and leaflet. Since all core teams intended to continue with the intervention after the third cycle we have showed chances of success are high with this approach. Because of time and workload in pharmacies nowadays, it would be impossible to reach this kind of intensive co-operation if you work with the whole pharmacy team. The second reason for success is the opportunity to discuss between core teams during the meetings. Especially the pharmacy technicians felt more certain to confront their colleagues with their plans. Although the core teams stated that redefining the plan was not needed, to our opinion they underestimate the effect of a structural reflection during the meetings and discussions about negative items as age limits. Without core team meetings the intervention would have more chance of failing.

The label was a success. All teams felt satisfied about the text; it should trigger women planning a pregnancy and not make women afraid about certain anomalies that can occur. The leaflet used was unpleasant (to much text, not clear what it is about at first sight) and suggestions were given for a better one. We hope this can be achieved in the future since currently this is the only available ready-made leaflet. Non-attractiveness of a leaflet might be a reason for counter assistants not to hand it out.

All core teams stated introducing the sticker was fully integrated in their daily routine since cycle one. The results of the questionnaire show that the core teams overestimated the success of the implementation of their intervention, especially in the first cycle. Furthermore, 18% of the responders stated to have received the label but not have seen it until they filled in the questionnaire. We assume these responders did not receive oral information while it is known from other research that written information combined with oral information is most effective. We therefore plead for consistent oral information for example once a year with the leaflet. This would also decrease the number of remarks that women

find it strangely to receive information about possible pregnancy with a pill that should them protect of getting pregnant and it could probably avoid the few negative reactions which might be a threat for the intervention as a whole.

The target group reacted mainly positive about the information received. But the few negative reactions were threatening the intervention because they discourage the pharmacy technician at the counter to give the information. The negative reactions in the questionnaires were found in the age groups 18-19 and 35-39. Among the age groups where most children are born (maternal age 25-29 and 30-34), more positive reactions occurred. The results of the questionnaire were very helpful in the reflection phase after each cycle because the results gave an indication how the target women experienced the intervention. Positive feelings encouraged the technicians to continue to educate about folic acid and the negative feeling could be discussed. This discussing helped the core teams in their role of stimulating their colleagues to keep educating about folic acid.

A limitation of this study is that it was only carried out in four pharmacies, which all volunteered to participate in the project. This means they had some affinity with the subject and they felt they had time and people to work on the project. On the other hand, it is nowadays important (financially) for pharmacists in the Netherlands to be able to show which pharmaceutical care projects they performed per year. The gained experience of this project can be used for other projects as well. Besides, a motivated team is always necessary for introducing new interventions.

Since our responders are relatively high educated and relatively frequently employed we can question if the lower educated women are reached by this approach. Unfortunately this is also the group were education about FA is mostly needed.[4] Nevertheless we argue that FA education through OCs is a daily routine in the Dutch pharmacies this is a firm basis from which plans can be made to reach a broader population.

For generalising the project on folic acid to other health education topics, the idea of working with a core team is very useful. A core team of a pharmacist and at least one pharmacy technician means the expertise of an executive person who has an overview of the pharmacy in a broad perspective and the expertise of a team member who has insight in daily practice. In addition, pharmacy technicians' involvement in the whole process increases acceptation by the whole team and thus the chance of a successful intervention. Anderson and Raiyagury [11] investigated the views of pharmacists and technicians on folic acid education materials in the UK. They found that most pharmacists and technicians were comfortable advising regular customers but they would only raise sensitive issues like preventing neural tube defects by taking folic acid with customers they have a relationship

with. However, our study shows that Dutch pharmacists and technicians feel free to communicate this issue with all customers. Therefore we feel we should encourage the Dutch pharmacies to educate their clients, on non-sensitive issues as well as on sensitive issues.

We performed a follow-up after approximately one year to investigate the sustainability of the implementation of the intervention of these four pharmacies. We noticed that they all still performed the intervention as planned after the third research cycle.

In conclusion, a multiple intervention program is feasible in the daily practice of a pharmacy. Good communication between members of the core team and the other pharmacy team members is essential to overcome barriers that can threaten the intervention. Materials that every team member likes and feels comfortable about to work with will lead to less discussion and therefore the intervention has more opportunity to be continued. The target group is mainly positive about the intervention and by making this visible, for example by explicitly asking them, will lead to less threat of ending the intervention.

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Chapter 10

Improved periconceptional use of folic acid after patient education in pharmacies – promising results of a pilot study in the Netherlands

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Abstract

Background: Although most Dutch women plan their pregnancy, less then half begin using folic acid supplements before they become pregnant.

Objective: To assess the effect of the provision of unsolicited information through community pharmacies on the knowledge, attitudes and behaviour towards folic acid of women receiving oral contraceptives on prescription.

Methods: Seven of nine community pharmacies (four intervention and three 'reference' pharmacies) in a small Dutch city participated in the study. A sample of 880 women (600 from intervention and 280 from reference pharmacies) receiving oral contraceptives on prescription were sent a postal questionnaire. The questions covered knowledge of and attitude and future intentions to take folic acid. Women were also asked whether they had received a sticker and leaflet about folic acid (the interventions) during the previous year and about sources of information on folic acid. Demographic information requested included age and any previous pregnancies.

Results: Three quarters of women in the intervention group reported receiving a sticker about folic acid and 48.4% said they had received a leaflet. Women who received information about folic acid with their oral contraceptives knew more about folic acid and there were differences in attitudes towards and intended use of folic acid in relation to women in the reference group. Some of the differences were statistically significant and were particularly evident among women who had not previously had a child (nulligravidae) and those who were intending to become pregnant within the next 12 months.

Conclusion: This study showed that providing information about folic acid from pharmacies to women using oral contraceptives has the potential to increase awareness and use of folic acid among women planning to become pregnant.

Introduction

Periconceptional folic acid supplementation reduces the risk of neural tube defects by more than 50%.[1, 2] Therefore, Dutch women planning a pregnancy are advised to take folic acid tablets from four weeks before conception until eight weeks after.[3] Current and recent programmes to increase periconceptional folic acid use may leave at least 50% and as many as 80% unprotected from the beneficial effects of folic acid.[4, 5]

Since fortification of staple food with folic acid is currently not allowed in the Netherlands,[6] consumption of folic acid supplements is the only means of sufficiently increasing the daily intake of folic acid. Because over 80% of Dutch women plan their pregnancy,[7] there is an opportunity to plan folic acid consumption. However, the problem remains of how to reach this target group with information about folic acid before they attempt to get pregnant. Since approximately 70% of Dutch women take oral contraceptives at some time before their first pregnancy and therefore visit their pharmacy regularly, folic acid information could be provided in the community pharmacy. In the Netherlands, oral contraceptives are always obtained from a pharmacy and patients visit only one pharmacy.

We therefore developed a programme in Dutch community pharmacies which aimed to inform and motivate women using oral contraceptives to start taking folic acid supplements a few weeks before they attempt to become pregnant. The aim of the study was to evaluate the effect of the information on women's knowledge and attitudes and in particular among those planning a pregnancy.

Methods

The setting for the study was a small Dutch town (approximately 80,000 inhabitants). Of its nine community pharmacies, one was excluded because one of the researchers (RAJ) is employed there and another could not participate due to organisational changes. The remaining seven pharmacies all agreed to participate and were randomly assigned as intervention (n=4) or reference (n=3) pharmacies. The intervention took place from February to July 2002 and consisted of adding stickers about folic acid on the boxes of Oral contraceptives dispensed during the study period with the text: "Are you planning to have a baby? Ask for information about folic acid in your pharmacy." Additionally the intervention pharmacies were asked to give a leaflet about folic acid at least once to every woman with a prescription for oral contraceptives during the intervention period. The implementation of the intervention is described in detail elsewhere.[8] Six months after the intervention a random sample of women, who had received an oral contraceptive during the study period, received

Figure 1: The questionnaire (demographic questions not included).

Questions asked		Possible answers
1	In the last 12 months, have you seen a sticker about planning to have a baby and folic acid on one or more of your oral contraceptives boxes?	yes/no
2	In the last 12 months, have you received a leaflet about folic acid when you collected your oral contraceptives at your pharmacy?	yes/no
3	What have you heard or read about folic acid?	(open question)
4	When do you think is the best time to use folic acid if a women wants to become pregnant?	(open question)
5	How did you obtain this knowledge? (more than one answer possible)	media (TV/radio/newspaper/magazine) my GP/midwife.gynaecologist told me my pharmacist (technician) told me from a leaflet family/friends other (please state) ……..
6	Would you advise other women who wish to become pregnant to use folic acid?	yes/no/don't know
7	Do you plan to become pregnant within the next 12 months? If Yes	yes/no
8	Do you take folic acid or tablets that contain folic acid (like certain multivitamins)?	yes, because ……. no, but I intent to, because…… no, because……

a postal questionnaire via their pharmacist. Each pharmacist complied a list of women who had a prescription for oral contraceptives dispensed and drew a sample according to instructions from the research team. The researchers had no access to the lists so had no information about individual women. The intervention period was divided in three periods of six to eight weeks each, and women were sampled six months after each period in order to have the same timeframe between the oral contraceptive related visit to the pharmacy and receiving the questionnaire. For each intervention pharmacy, 50 women were randomly selected from each time period using a random number table (four pharmacies x three time periods x 50 women = 600 questionnaires). For each reference pharmacy 35 women were randomly selected from each time period. One of the reference pharmacies was not able to send the questionnaires to the last group of 35 women because it was taken over by a new

pharmacist. In total 280 (two pharmacies x three time periods x 35, plus one pharmacy x two time periods x 35) questionnaires were sent. After two weeks, pharmacies sent reminders to the non-responders.

The questionnaire asked if the woman had been pregnant before, and whether they were either currently pregnant or planning to become pregnant, and also asked the woman's age. There were questions about the respondents' knowledge (the beneficial effects of folic acid and when to take folic acid), attitude (would they advise other women to take folic acid) and behaviour (whether they were taking or would take folic acid when pregnant or attempting to conceive). The questionnaire also asked about sources of information about folic acid (see figure 1).

Data Analysis

P-values were calculated with chi-square tests. Sub analyses were performed on the questionnaires of women with definite plans to become pregnant. The level of significance was set as p of 0.05 or lower. The results are presented as percentages and percentage differences per group.

Table 1: Respondent characteristics and received intervention in the intervention and reference groups.

	Intervention group (N=579)		Reference group (N=266)		p-value (χ^2)
General variables	n	(%)	n	(%)	
Response (%)	364	(62.9)	164	(61.7)	0.74
Mean age (SD) (range)*	33.2	(3.40) (27-39)	32.6	(3.51) (22-39)	0.02*
Born in the Netherlands (%)	316	(91.5)	149	(90.9)	0.82
Education					
Low (%)	60	(16.5)	25	(15.2)	
Middle (%)	167	(45.9)	78	(47.6)	0.91
High (%)	127	(34.9)	56	(34.1)	
Employed (%)	273	(75.0)	127	(77.4)	0.55
Nulligravida (%)	133	(36.5)	88	(53.7)	0.00
received intervention					
Sticker on oral contraceptives	272	(74.7)	20	(12.2)	0.00
Leaflet	176	(48.4)	11	(6.7)	0.00

* t-test was used for calculating this p-value

Results

Of the total of 880 questionnaires 35 were returned because the woman was no longer traceable at that address, leaving 845 that were delivered. The response rates were 364/579 (62.9%) in the intervention group and 164/266 (61.7%) in the reference group. There were few missing responses, so the number of missing values was very low.

The groups were similar in many respects but those in the intervention group were slightly older and more likely to have been pregnant before than those in the reference group. In the intervention group, 74.7% claimed to have received a sticker on their oral contraceptives, and 48.4% said they had received a leaflet about folic acid with their oral contraceptives (table 1).

Data on the outcome measures (knowledge, attitude and (intended) behaviour) of the respondents are shown in table 2. Since gravidity differed between the groups, results are presented for nulligravidae and gravidae separately. There was a non-significant trend

Table 2: Comparison of outcome measures between nulligravidae and gravidae of the intervention and reference groups.

	Nulligravidae					Gravidae				
	Intervention (N=133)		Reference (N=88)			Intervention (N=231)		Reference (N=76)		
Knowledge	%	n	%	n	p-value (χ^2)	%	n	%	n	p-value (χ^2)
Prevents neural tube defect	35.3	(47)	33.0	(29)	0.72	55.4	(128)	43.4	(33)	0.07
Correct time period	15.1	(18)	10.4	(9)	0.34	22.3	(50)	16.4	(12)	0.28
Start before pregnancy	62.2	(74)	52.3	(45)	0.18	80.4	(180)	69.9	(51)	0.06
Information Sources										
Pharmacy	6.9	(9)	1.1	(1)	0.05	12.6	(29)	3.9	(3)	0.03
Leaflet	35.9	(47)	10.2	(9)	<0.01	42.9	(99)	28.9	(22)	0.03
Other health professionals	4.6	(6)	3.4	(3)	0.68	43.3	(100)	27.6	(21)	0.02
Would recommend folic acid to other women	55.1	(70)	43.4	(36)	0.10	69.9	(160)	62.2	(46)	0.22

Note: Percentages can vary due to a small number of missing values.

Table 3: Comparison of outcome measures between nulligravidae and gravidae of the intervention and the reference group for women who were pregnant or intending to become pregnant.

	Nulligravidae					Gravidae				
	Intervention (N=24)		Reference (N=13)			Intervention (N=20)		Reference (N=15)		
Knowledge	%	n	%	n	p-value (χ^2)	%	n	%	n	p-value (χ^2)
Prevention neural tube defect	62.5	(15)	61.5	(8)	0.95	65.0	(13)	53.3	(8)	0.49
Right period	37.5	(9)	15.4	(2)	0.16	45.0	(9)	6.7	(1)	0.01
Start before pregnancy	83.3	(20)	61.5	(8)	0.14	90.0	(18)	66.7	(10)	0.09
Use or planning to use										
Current use	62.5	(15)	30.8	(4)	0.02	40.0	(8)	26.7	(4)	0.42
Intended to start using	20.8	(5)	7.7	(1)		20.0	(4)	40.0	(6)	

Note: Percentages may vary due to a small number of missing values.

towards greater knowledge of the protective effects of folic acid in the intervention group. The between-group differences for intervention and reference groups were 2.3% for nulligravidae and 12.0% for gravidae. In the intervention group, a higher percentage could state the correct period of use (4.7% and 5.9% difference between the groups for nulligravidae and gravidae respectively). Most respondents knew that folic acid should be started before pregnancy (these groups included the respondents who stated the correct period of use).

The pharmacy (pharmacist or technician), leaflets and other professionals were reported more often as sources of information by women in the intervention group. Leaflets were mentioned by 146 (40%) of women in the intervention and 12% in the reference group. The pharmacy was mentioned by 38 (10%) of women in the intervention and 4 (2%) in the reference group. Apart from the source ‘other professionals” among the nulligravidae, these differences were all significant. It was not possible to determine whether the respondent was referring to the leaflet received as part of the intervention because the questionnaire did not specifically ask about that leaflet and any others. More women in the intervention group said they would recommend the use of folic acid to other women compared with the reference

group. The between-group differences for intervention and reference were 11.7% and 7.7% for nulligravidae and gravidae, although these differences were not statistically significant.

Among women planning to become pregnant in the next year (table 3), knowledge about beneficial effects and about correct period of use of folic acid was higher in the intervention group compared with the reference group. For knowing the correct period of use, this difference was significant among the gravidae. The percentages for knowledge in this subgroup were also higher compared to all responders (table 2). Current use of folic acid among women planning a pregnancy was higher in the intervention group compared with the reference group (62.5 vs. 30.8% in nulligravidae and 40.0% vs. 26.7% in gravidae). Among the nulligravidae planning a pregnancy, current use, intention to use, and not intending to use all differed significantly between the intervention and reference groups ($p=0.02$).

Discussion

Women who received information about folic acid with their oral contraceptives seemed to have greater knowledge about, a more positive attitude towards and higher intention to use folic acid compared with women who did not receive this information. The differences were most evident for nulligravidae and for those who intended to conceive within the next 12 months. Since in the Netherlands no wide spread preconception care is available, these two groups are considered the most difficult to reach. Women in the intervention group were three times more likely to mention a leaflet as a source of information about folic acid and five times more likely to mention the pharmacy as were women in the reference group. Our data strongly suggest that information about folic acid provided from community pharmacies with an opportunity for discussion with pharmacists and technicians can reach and inform women who are planning to become pregnant. Among these women, the use of folic acid and future intention to use folic acid was significantly higher in the intervention group. Although, as expected, the proportion of women who knew about folic acid was higher in the group that had been pregnant before, our results indicate that continued education is still needed among this group.

Although some of the differences between intervention and reference groups were significant, some were not. The largest limitation of the study, being a pilot study, was that it might have been under-powered to detect differences between sub-groups, especially because we felt it was necessary to present the results for the nulligravidae and gravidae separately, some of the cell sizes were small. All women in the intervention group should

have received both a sticker on their oral contraceptives and a leaflet about folic acid. Although the majority recalled the former, only around half said they had received the leaflet. Thus the delivery of the intervention was not uniform across the group. A small number of women in the reference group reported receiving a sticker and/or leaflet with their oral contraceptives. If this had influenced the results of the study, we believe our results are an underestimation of the possible reachable effect if all women in the intervention group had received full education. The response rate of over 60% was good, but another weakness of the study is that we have no information on non-responders to the questionnaire. Although it could be argued that women interested in the subject would be more likely to respond to the questionnaire, we do not believe that the differences between the groups could be attributed to this.

The most important outcome was whether women planning a pregnancy or in the early stages of pregnancy were actually taking folic acid. Our results indicate that positive effects for this outcome variable can be achieved. Among the nulligravidae we found a significant difference in use and intentional use. During their first pregnancy Dutch women usually visit their midwife or physician in week 10, when the neural tube is already closed and when information on folic acid comes too late. Involving pharmacists and technicians to inform and stimulate women to take folic acid before they become pregnant seems to add to the awareness and use of folic acid. Further studies with larger numbers of pharmacies are now needed to further evaluate the effects. In the Netherlands this approach will now be rolled out to pharmacies across the country and the effects will be measured on a larger scale. Furthermore we believe that this approach is also applicable in other countries and we would encourage others to test it.

Conclusion

Community pharmacies can provide targeted information to women who are planning to become pregnant. This study showed that providing information about folic acid from pharmacies to women using oral contraceptives has the potential to increase awareness and use of folic acid among women planning to become pregnant.

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Summary and General Discussion

Summary

Periconceptional exposure might result in a broad range of expected and unexpected effects. In this thesis, the risks and benefits of intrauterine exposure were studied for several drugs and drug groups (part A) and possibilities to overcome some limitations of current datasets were investigated (part B). Finally, some methods to educate women of childbearing age about these risks and benefits were studied (part C).

Part A: Drug use in pregnancy; studies on intrauterine exposure and foetal risk
To get some insight in the extent of drug use among pregnant women, the kind of drugs used during pregnancy and the safety of these drugs, we examined drug use during pregnancy in chapter 1. The interpretation of available studies is often hampered because it is unclear to what extent women have changed their drug choice in case of pregnancy. We therefore described drug use of pregnant women and comparable non-pregnant women based on pharmacy data and with reference to the Australian risk classification system. Of all prescriptions for non-pregnant women, 35% were safe in pregnancy (Australian classification A), of 14% the risk was unknown (B1, B2), 49% were potentially harmful drugs (B3, C, D, X), and for 3% no classification was available. For pregnant women these figures were 86, 3, 10 and 2%. We conclude that many drugs used by non-pregnant women should be avoided in pregnancy, and that pregnant women indeed do so. However, for some drug groups the available safe alternatives are limited.

Besides increasing risks on birth defects, intrauterine exposure can be beneficial as well, as is the case with folic acid (FA). The protective effect of FA on FA-sensitive anomalies is studied using data on births from the EUROCAT Northern Netherlands registry (NNL) (chapter 2). This study shows a protective effect of FA for the FA-sensitive anomalies as a whole group, but this effect is not significant anymore after adjusting for maternal age and year of birth. Furthermore, a significant effect is found for the heart anomalies in particular and the odds ratios (OR) for neural tube defects (NTD), urinary anomalies and limb reduction defects are, although not significant, indicative for a protective effect of FA. Therefore this study supports some of the positive findings of the effect of FA in other studies. If FA protects against certain birth defects, it is reasonable to assume intrauterine exposure to FA antagonists increases the risk on these defects. FA antagonists can be divided into dihydrofolate reductase inhibitors and antiepileptic drugs. In the same database as the previous study we studied these associations (chapter 3). We found a statistically significant effect on FA sensitive defects after exposure to an antiepileptic drug (OR=3.45,

95% CI:1.04-11.48) which is in agreement with findings of various other studies on the teratogenicity of antiepileptics. An association between dihydrofolate reductase inhibitors and FA sensitive defects was not found, nor for FA antagonists all together as one group. As shown in chapter 2 as well in many other studies, FA assumedly protects against heart anomalies as a group, with conotruncal defects and ventricular septal defects in particular. Many children born with Down syndrome (DS) suffer from heart defects, mostly ventricular septal defects, atrial septal defects and endocardial cushion defects. In chapter 4, we therefore investigated whether FA also shows a protective effect on heart defects in DS children. For this study we used data from the Birth Defects Study of the Slone Epidemiology Center. After adjustment for many possible confounders, no protective effect of FA was found on heart anomalies overall. The four subgroups of heart defects we investigated (conotruncal defects, ventricular septal defects, ostium secundum type atrial septal defects, and endocardial cushion defects) showed some variation in point estimates, but none of the findings was significant.

In the last chapter of this part, chapter 5, the association between clomiphene and hypospadias was examined. Clomiphene, used to induce ovulation, is chemically related to diethylstilbestrol (DES). DES is associated with vaginal cancer and infertility among daughters and with hypospadias among second generation offspring. Since clomiphene has a long half life, foetal exposure is possible and possible risks of this exposure on the occurrence of hypospadias are therefore explored. For penoscrotal hypospadias we found a significant increased OR (6.08; 95% CI 1.40-26.33); the ORs for the mild and moderate forms of hypospadias were not increased. We concluded that the effect of clomiphene on penoscrotal hypospadias is diluted when studying all hypospadias together as a group. Therefore, our study stresses the importance of studying birth defects on a detailed level as possible.

Part B: Improving data collection for epidemiologic studies on safety of drug exposure during pregnancy

In the second part of the thesis, some methods to improve or to extend current datasets are studied. To increase numbers to perform risk studies on, data of comparable registries can be combined. Within the EUROCAT network of 40 European registries, the availability of drug exposure data was investigated and described in chapter 6. From a questionnaire about their data sources on drug exposure and on drug coding, which was sent to all registry leaders, it became clear that obstetric records are the most frequently used sources of drug information and that most full members use the EUROCAT drug code. Only four registries

use the international ATC drug code. Furthermore, the available data on drug exposure during the first trimester available in the central EUROCAT database for the years 1996-2000 was summarised for 15 out of 25 responding full members. Percentages of cases with drug exposure (excluding vitamins/minerals) varied from 4.4% to 26.0% among different registries. The categories of drugs recorded varied widely between registries. We conclude that practices vary widely between registries regarding recording drug exposure information. The EUROCAT network has the potential to be an effective collaborative framework to contribute to post-marketing drug surveillance in relation to teratogenic effects, but work is needed to implement ATC drug coding more widely, and to diversify the sources of information used to determine drug exposure in each registry.

In chapter 7, the possibilities to collect data on non-malformed births as controls for the EUROCAT NNL cases are explored. Because of existing contacts, recruitment of non-malformed controls via pharmacies was examined first, but both possible methods led to selection and were therefore not suitable. As an alternative, recruitment via a midwife practice was explored. From this exploration it became clear that this strategy is feasible in a midwife practice and that the participating pregnant women were a representative sample of the practice. Furthermore, interviews with the participants showed they had a positive attitude towards the study. This was only a first step in collecting data on a representative sample in the northern Netherlands. Recruitment should be extended to other practices and hospital setting should be included as well. Furthermore, internal as well as external validity should be explored more thoroughly. Nevertheless, this first exploration gives reasons to be optimistic about the possibilities to include non-malformed births in the standard data collection in the future.

Part C: Implementing knowledge: folic acid education in pharmacy daily practice

After gathering new insights, this new knowledge should be communicated with the actual target population, e.g. women of reproductive age. To implement strategies to improve preconceptional FA use, we first investigated the current status of knowledge and use of FA among women that wish to become pregnant. Chapter 8 shows that, of the women that wish to become pregnant within six months, 69% knows about the protective effect of FA and 81% knows to start before conception and 64% knows both. These percentages drop with increasing term to wish to become pregnant. Among women planning their pregnancy within six months, 59% reports current use of FA compared with 6% among those wishing to conceive in 12-24 months. Furthermore, the multivariate model shows FA use is associated

with knowledge, a positive attitude and receiving oral information from a health care provider. Therefore, interventions to increase FA use should be aiming on these variables. A pilot study on the feasibility of implementing FA education via in pharmacies is presented in chapter 9. In four pharmacies a core team (one pharmacist and one or two technicians) was formed that was responsible for planning an intervention that minimally existed of adding a label about FA on the box of oral contraceptives (OC) and handing out a leaflet about FA at least once during the intervention period. Adjustments in the intervention were made based on experiences of the pharmacy team, on responses in the pharmacy and on the results of a questionnaire sent to women one week after they visited the pharmacy with an OC-prescription. This six weeks-cycle of planning, action, observation and reflection was repeated twice. The minimal intervention was carried out by all four pharmacies. Other activities like using posters or a display window, work with age limits or wearing buttons, differed per pharmacy. Of the target group, 44% was positive about the received sticker, 49% was neutral and 4% was negative. Besides, 56% of the target group stated that they appreciate the preventive information given through the pharmacy. We concluded that working with core teams seems to be a successful strategy and implementation of the intervention is feasible. By editing the intervention per research cycle during the core team meetings an optimal method can be reached that fits in the existing organisation and possible barriers can be overcome. The positive response of the target group motivated the core teams enormously. The effect of this intervention on the knowledge and use of FA among the target population is subsequently described in chapter 10. Three quarters of women in the intervention group reported receiving a sticker about FA and 48.4% said they had received a leaflet. Women who received information about FA with their oral contraceptives knew more about FA and there were differences in attitudes towards and intended use of FA in relation to women in the reference group (three pharmacies in the same region). Some of the differences were statistically significant and were particularly evident among women who had not previously had a child and those who were intending to become pregnant within the next 12 months. We could thus conclude that providing information about FA from pharmacies to women using OCs has the potential to increase awareness and use of FA among women planning to become pregnant within one year.

General Discussion

As stated in the General Introduction, pre-marketing information about the effects of a drug on the foetus originates mostly from animal studies. Consequently, information about the effect in humans can only be obtained if the drug is used by pregnant women after marketing. Therefore, it is of the utmost importance to monitor and register drug use and other environmental factors during pregnancy. Only then, teratogens can be discovered as soon as possible.

Some methodological considerations of studying teratogenicity

The perfect database to study drug-birth defect associations does not exist; every set of data has its own strengths and limitations. By knowing and addressing these strengths and limitations, the reliability of findings can be discussed openly and one can try to avoid obvious pitfalls. Because of the peculiarities of each dataset, it is necessary that associations are studied in databases in different populations and with different qualities. If the same association is found in different databases, as was the case with folic acid and for example heart defects, we can carefully conclude this association might be real. In the following paragraphs, some methodological aspects of studying teratogenicity encountered in part A of the thesis, are discussed.

Clustering data

The power to detect a significant finding depends on the number of cases and the exposure rate among controls.[1] Since specific birth defects are rare and exposure to a specific drug among women of childbearing age is often rare as well, studies on teratogenicity can be strongly limited by the power to detect a significant association. For example, in chapter 2 we could not confirm the general accepted protective effect of folic acid on neural tube defects due to our small numbers. To increase these numbers, exposure or outcome data can be clustered or whole datasets can be collapsed. Clustering malformations is common and mostly based on malformations within the same organ system, like heart defects, urogenital defects and so on. In chapters 2 and 3, birth defects were even clustered on a larger scale, namely on being sensitive to folic acid. However, despite biological logical clusters of defects per organ system or drugs on pharmacological similarities, drug-defect associations do not always follow this logic and therefore unexpected associations or lack of associations can be found. Furthermore, the risk of diluting an existing effect is increased by clustering, even with an already specific malformation as is shown in chapter 5 with

hypospadias: only for the severe form penoscrotal hypospadias an effect is found which was diluted if all hypospadias cases were investigated together. Therefore, if no effect is found after clustering, like the lack of association between dihydrofolate reductase inhibitors and folic acid sensitive anomalies (chapter 3), conclusions from the data should be drawn with care.

If data from several birth defect registries are collapsed, like in the EUROCAT Central Database, more possibilities arise to investigate specific exposures-malformations associations. On the other hand, data sources, data collection, data handling, and data coding, should be as similar as possible in all registries involved to guarantee the reliability and validity of studies using these data. Whether this is indeed the case should be subject of study itself before using the collapsed data for risk assessment purposes.

Use of pharmacy data for exposure information

Except for chapter 4, the studies in part A of the thesis use pharmacy dispensing data for exposure information. Although Dutch community pharmacy data are an accurate source of drug use of non-hospitalized patients,[2,3] dispension date and recorded dose are only an estimation of actual exposure. However, with EUROCAT NNL, prescription drugs reported by pharmacies are presented to women and it is asked whether she actually took the drug, in what period, for how long etc. She is also asked why the drug was prescribed. This procedure increases the internal validity compared with only pharmacy data.

Recall bias and selection bias -or- issues related to selecting controls

Since specific birth defects occur rarely, the case-control design is often used in studies on teratogenicity. Choosing controls is a big challenge in this design. One can choose between malformed controls, i.e. children with other birth defects than the defect of interest in the study, and non-malformed controls. The concern that mothers of infants with a birth defect recall more events and details about exposure than mothers of a healthy infant is discussed extensively in literature,[4-8] but these concerns are largely theoretical and difficult to investigate because the lack of a golden standard. To minimize the potential recall bias, malformed controls can be used.[9,10] But if the selected malformed controls also have a relation with the exposure, selection bias can occur.[11,12] An interesting example of such bias did occurr in a study of Lakos and Czeizel.[13] In their study with spina bifida and anencephaly as controls, no association between antiepileptics and clefts was found. Later studies showed a relation between antiepileptics and neural tube defects. This example illustrates how selection bias might lead to an underestimation of the actual risk of the exposure under

investigation. Since observed risks of drugs under investigation are often small, even a small amount of selection bias might produce an estimate close to null leading to dismiss an actually existing risk.

Improving data collection methods for drug safety studies

Discussing limitations of current datasets is only a starter. Subsequently, it should be investigated if and how to overcome these limitations. After all, a study can only be as good as the database used. If structured data collection is already available, it is important to investigate possibilities to improve or extend these.

The EUROCAT Central Database

Luckily, much effort has been put into data collection in order to study prevalences and aetiology of birth defects. A collaboration of over 40 registries as the EUROCAT network provides researchers with enormous opportunities to conduct studies in this field. In this network, much effort has been made to synchronise collecting and coding of birth defects in every registry.[14,15] Hence, this database is very suitable for studying prevalences of different birth defects in different regions in Europe. Furthermore, it has been shown that etiologic studies can also be performed with these data since basic characteristics as maternal age and smoking are present in the database or can be collected from the individual registries. In chapter 6 we explored if this database is suitable for drug safety studies as well. At the moment, drug exposure data vary widely between registries and drugs are coded in drug groups instead of using a coding for each individual drug. However, at least half of the registries within the network collect information on drug use in the first trimester. Therefore the EUROCAT Central Database is a resource for post-marketing drug surveillance since the infrastructure of data collection is already there. With the introduction of drug specific coding and enlargement of sources of information, performing drug safety studies will be possible in the future.

Collecting data on non-malformed births

Ideally, both malformed and non-malformed controls are available to assure both recall and selection bias are dealt with. Currently, EUROCAT NNL only collects data on malformed births. But, as shown in chapter 7, it is possible to extend this data collection with non-malformed births as well. Besides the issue of selection bias, having data on non-malformed controls also enables researchers to investigate items that could otherwise not be investigated. For example, in etiologic studies, infants with chromosomal anomalies are

often selected as controls, but what if these are subject of study and selected as cases? Especially with increasing attention for gene-environment interactions, the need for non-malformed controls will increase as well. After the extension of data collection with the parental questionnaire and pharmacy data in 1997,[16] the current extension with non-malformed controls will lead to the availability of a unique dataset with unique and extensive data to perform drug safety studies in pregnancy. Therefore it is extremely important that EUROCAT NNL is given the opportunity to continue and extend data collection.

Dissemination of knowledge

Not only should we study safety of intrauterine exposure and publish the results in international scientific journals, the scientific society should also take responsibility in informing health care providers and the general population about new findings. Strategies to do so need to be developed and implemented as soon as possible in order to prevent future infants to be exposed unnecessary to potentially harmful drugs or to prevent unnecessary termination of a wanted pregnancy after a possible risky exposure.

Tools for health care providers

To provide health care providers with guidance in the decision whether or not to prescribe a certain drug during pregnancy, several classification systems are developed that place drugs in risk groups regarding their known or suspected adverse effects on the unborn child. Examples of these systems include the FDA (Food and Drug Administration) Classification, the Swedish Classification, and the Australian Classification. The drawback is that different sources of information are used by these systems and these different sources give different results which are translated in different risk categories. In fact, the three risk classification systems differ hugely in the classification of drugs: only 26% of the drugs have same classification in three systems.[17] Furthermore, evidence is often scarce thus many drugs are classified as unknown to be safe or harmful,[18] which is hardly informative for health care providers in terms of advising them what to do. Additionally, for a risk-benefit discussion with a patient, health care providers need more sophisticated information than only a classification code. With all these limitations, the usefulness of such classification systems is doubtful.

Educating the target population

Last but certainly not least, the target population, i.e. women of childbearing age, should be educated about (anti)teratogenic risks. Since the foetus is most vulnerable in the first weeks

of pregnancy, when women often are not aware of the existence of the pregnancy, educating is difficult in terms of when and who. Ideally, a women informs her health care providers she wants to become pregnant. At that moment she can be informed about risks and benefits. Although providing preconception counselling by general practitioners in the Netherlands is under exploration,[19] no widespread and consequent preconception care is available in the Netherlands at this moment. Furthermore, not all pregnancies are planned. Luckily, in the Netherlands, most patients are only visiting one pharmacy. Pharmacies can use this contact to pro-active educate women of childbearing age. This education can be given to women who collect their oral contraceptives (chapter 9), or it can be related to other drugs used by women of childbearing age. As shown in this thesis, pharmacies are very capable to educate women of childbearing age about exposure risks during pregnancy. Although pro-active education is relatively new for the pharmacy teams, they have the knowledge, the infrastructure and patient contacts to do so. Therefore they should not be overlooked but stimulated to play this new role and helped to implement long lasting strategies.

Perspectives and final remarks

From the previous it is clear studying and communicating teratogenic risks will remain an issue of great concern. There is still a huge lack of knowledge about how and why some drugs act as a teratogen while others with a comparable chemical structure do not. It is also unclear why the risk on a specific defect can be increased by a drug while the risk on other defects on the same organ system or originating from the same cells are not. Thus, studying teratogenicity of drugs is only just past childhood but still far from adulthood. Nevertheless, it is therefore of utmost importance to continue monitoring pregnancies. Resources should stay available to continue monitoring. Not only would this increase numbers and thus the power and possibilities to investigate rare exposures, also new drugs will come into the market and their safety needs to be investigated as well.

From this perspective, the change in regulation of drug approval might stimulate future continuation of pregnancy monitoring. Strom proposes to, instead of fully approve a drug after phase three studies, to conditionally approve a new drug on the market after phase three and introduce an compulsory post marketing follow up period before fully approve the drug.[20] For increase of knowledge about drug safety in pregnancy, this obligation of collecting post-marketing data might be a first step in the right direction, although information about the safety of a drug during pregnancy requires special attention. Pharmaceutical companies follow exposed pregnancies in special pregnancy registries. For this purpose,

they can use the guidelines from the European Agency for the Evaluation of Medical Products (EMEA) or the FDA on post-authorisation data collection on safety of a drug used during pregnancy.[21,22] Nevertheless, it would be most useful if, following reasoning of obligated post-marketing surveillance, not every drug is investigated separately by its producing company. Instead, resources should be combined to invest in central databases where knowledge and means are combined and all required variables are collected centrally and studies can include not only single drug exposure but also multiple exposures or where several drugs in the same therapeutic class can be compared.

For educating the target population, i.e. women of childbearing age, about drug safety issues during pregnancy, health care providers need information and tools to implement this education in their daily practice. Therefore, scientific findings need to be 'translated' into clinical useful information like guidelines. Unfortunately, scientists are driven to publish results, for example because that is how they are rated. Nevertheless, because of the possible impact of a publication, they should consider carefully whether a study is indeed informative and of high enough quality to be published. Furthermore, due to the need to publish, science is a very competitive field. But because of the scarcity of data, less competition and more co-operation is needed in teratogenicity research.

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Samenvatting

Inleiding

Heel lang werd gedacht dat de placenta het ongeboren kind kon beschermen tegen alle mogelijke schadelijke invloeden van buitenaf. Ondanks de eerdere berichtgevingen over het schadelijke effect van straling en de mazelen op het ongeboren kind, maakte het thalidomide-drama (Softenon®) rond 1960 de wereld pijnlijk duidelijk dat die placenta toch niet de perfecte barrière vormde. Wereldwijd zijn duizenden kinderen geboren met ontbrekende of onderontwikkelde ledematen en andere aangeboren afwijkingen omdat hun moeder thalidomide had gebruikt in de eerste weken van de zwangerschap.

Deze tragedie was het begin van het onderzoek naar aangeboren afwijkingen en het begrip ‘teratogeen’ werd geïntroduceerd. Hiermee wordt bedoeld dat een geneesmiddel of andere stof waaraan de zwangere is blootgesteld tijdens de zwangerschap, aangeboren afwijkingen kan veroorzaken bij het ongeboren kind. Het woord teratogeen is afgeleid van de griekse woorden teras (monster) en gennan (voortbrengen). In de loop van de jaren werd duidelijk dat teratogeniteit veel breder is dan zichtbare afwijkingen bij de geboorte. Schadelijke blootstelling in de baarmoeder kan ook leiden tot een miskraam, een doodgeboren kind, vroeggeboorte, laag geboortegewicht, groeivertraging, zowel voor als na de geboorte, gedragsstoornissen, en functionele stoornissen van bepaalde organen.

Bestuderen van de veiligheid van geneesmiddelgebruik tijdens de zwangerschap

Het algemene advies luidt dat het beter is geen geneesmiddelen te gebruiken tijdens de zwangerschap tenzij het niet anders kan. Toch weten we dat ongeveer 80% van de zwangeren ten minste één geneesmiddel gebruikt. De noodzaak om meer kennis over de veiligheid van dit gebruik behoeft dus geen nadere uitleg. Voordat een nieuw geneesmiddel op de markt wordt gebracht, wordt de mogelijke teratogeniteit getest op dieren. Helaas zijn deze resultaten niet zonder meer te vertalen naar mensen. Dit betekent dat in de praktijk de teratogeniteit van veel geneesmiddelen pas zal blijken als het middel al op de markt is en gebruikt wordt door zwangeren. Gelukkig is er de afgelopen decennia veel energie gestopt in het verzamelen van gegevens die nodig zijn om relaties tussen geneesmiddelen en aangeboren afwijkingen te onderzoeken, zoals de EUROCAT registratie van aangeboren afwijkingen in Groningen. Ondanks dat hier inmiddels gegevens over duizenden zwangerschappen zijn verzameld, zijn de aantallen om relaties tussen een specifiek geneesmiddel en een specitieke afwijking te onderzoeken vaak nog te klein. Door geneesmiddelen met een vergelijkbare werking of verschillende aangeboren afwijkingen te groeperen kunnen deze aantallen wat groter gemaakt worden. Hierdoor neemt de

statistische power van de studie toe. Het blijkt echter dat stoffen die chemisch sterk op elkaar lijken niet altijd een vergelijkbaar teratogeen effect hebben. Ook blijkt dat een geneesmiddel een relatie kan hebben met een specifieke aangeboren afwijking, terwijl een dergelijke relatie met een verwante afwijking ontbreekt. Door geneesmiddelen of afwijkingen te groeperen om de power van de studie te vergroten kunnen bestaande relaties verdwijnen in de massa en op die manier over het hoofd gezien worden. Een andere methode om de aantallen - en daarmee de power van een studie - te vergroten is het samenvoegen van informatie uit verschillende registraties. EUROCAT Noord Nederland participeert bijvoorbeeld met ongeveer 40 andere registraties in het Europese EUROCAT netwerk.

Voorlichten van vrouwen in de vruchtbare leeftijd over mogelijke risico's en gunstige effecten van blootstelling tijdens de zwangerschap

De eerste acht weken na bevruchting zijn het meest cruciaal in de ontwikkeling van het embryo. Omdat het een paar weken duurt voordat een zwangerschap bekend is, zelfs als deze is gepland, bestaat het risico dat de foetus wordt blootgesteld aan een teratogeen middel. Bovendien zijn er geneesmiddelen die na gebruik heel lang in het lichaam aanwezig zijn. Een embryo kan hieraan dan zelfs worden blootgesteld wanneer zo'n middel al voor de zwangerschap is gebruikt. Het is dus belangrijk om vrouwen in de vruchtbare leeftijd al voor een mogelijke zwangerschap te informeren over eventuele risico's. Vrouwen komen echter pas bij een arts of verloskundige wanneer ze al geruime tijd zwanger zijn. Omdat ongeveer 70% van de vrouwen voor de eerste zwangerschap de anticonceptiepil gebruikt, kan de apotheek zich proactief opstellen en deze vrouwen bij het afhalen van de pil informatie geven over wat te doen wanneer ze stopt met de pil om zwanger te worden. Dus ondanks het gebrek aan georganiseerde en wijdverspreide preconceptiezorg in Nederland is het mogelijk om via de apotheek veel vrouwen voor te lichten over mogelijke teratogene risico's of juist over bijvoorbeeld het gunstige effect van foliumzuur op het voorkomen van bepaalde aangeboren afwijkingen. Om hierop in te kunnen springen moeten apotheken strategieën ontwikkelen om deze vorm van proactieve voorlichting te implementeren in de dagelijkse praktijk.

Opbouw van het proefschrift

In dit proefschrift zijn zowel gunstige als ongunstige effecten van blootstelling aan verschillende middelen voor het kind bestudeerd (deel A). In het tweede deel van het proefschrift (deel B) zijn enkelen methoden om de beperkingen van momenteel beschikbare

datasets te verkleinen uitgewerkt. In het laatste deel (deel C) is vervolgens onderzocht hoe vrouwen in de vruchtbare leeftijd voorgelicht kunnen worden over voor- en nadelen van het gebruik van verschillende middelen rond de conceptie.

Samenvatting

Deel A: studies naar blootstelling tijdens de zwangerschap en risico's voor het ongeboren kind.

Om inzicht te krijgen in de mate van geneesmiddelgebruik onder zwangeren, de soort geneesmiddelen die tijdens de zwangerschap worden gebruikt en de veiligheid van deze geneesmiddelen, is het geneesmiddelgebruik onder zwangeren bestudeerd in hoofdstuk 1. Hierin is het geneesmiddelgebruik van zwangere vrouwen en vergelijkbare maar niet zwangere vrouwen beschreven, waarbij we ons hebben gebaseerd op apotheek- gegevens. De gebruikte geneesmiddelen werden geclassificeerd als veilig, onbekend risico en mogelijk schadelijk, met behulp van het Australische risico-classificatiesysteem. Van alle geneesmiddelen die werden gebruikt door de niet-zwangeren was 35% veilig tijdens de zwangerschap (classificatie A), was van 14 % was het risico onbekend (classificatie B1 en B2), en 49% potentieel schadelijk voor het ongeboren kind (B3, C, D, X). De overige 3% van de geneesmiddelen kon niet geclassificeerd worden. Voor de geneesmiddelen gebruikt door zwangeren waren deze getallen 86, 3, 10 en 2%. De conclusie van onze studie is dat veel geneesmiddelen die worden gebruikt door vrouwen in de vruchtbare leeftijd die niet zwanger zijn, vermeden zouden moeten worden tijdens de zwangerschap. Het gebruik van deze middelen onder zwangeren ligt inderdaad veel lager ligt. Een kanttekening hierbij is dat er voor een kleine groep geneesmiddelen geen veilige alternatieven beschikbaar zijn.

Behalve een verhoging van het risico op aangeboren afwijkingen kunnen sommige middelen die rond de conceptie worden gebruikt ook juist gunstige effecten hebben op het ongeboren kind. Een voorbeeld hiervan is foliumzuur. Het beschermende effect van foliumzuur op verschillende aangeboren afwijkingen is bestudeerd in de database van EUROCAT Noord Nederland (hoofdstuk 2). We vonden een significant beschermend effect van foliumzuur op aangeboren hartafwijkingen en aanwijzingen dat foliumzuur beschermt tegen neurale buisdefecten (open ruggetje en anencefalie), urineweg afwijkingen en ledemaat reductie defecten, al waren deze drie effecten niet statistisch significant. De resultaten van onze studie ondersteunen het positieve effect van foliumzuur zoals gevonden in andere studies. Wanneer foliumzuur het risico op aangeboren afwijkingen verlaagt, is het redelijk om aan te nemen dat stoffen met een tegengestelde werking, de zogenaamde

foliumzuur antagonisten, het risico op deze afwijkingen verhoogt. Foliumzuur antagonisten kunnen verdeeld worden in twee groepen: dihydrofolaat reductaseremmers en bepaalde anti-epileptica. In dezelfde EUROCAT Noord Nederland database hebben we het effect van het gebruik van deze foliumzuur antagonisten in de eerste weken van de zwangerschap onderzocht (hoofdstuk 3). We vonden een statistisch significant verhoogd risico van anti-epileptica op de foliumzuur gevoelige afwijkingen (odds ratio=3,45, 95% BI: 1,04-11,48) wat overeenkomt met resultaten van andere studies. Voor de dihydrofolaat reductaseremmers werd geen verhoogd risico gevonden, noch voor alle foliumzuur antagonisten samen als één groep.

Zowel in hoofdstuk 2 van dit proefschrift als in vele andere studies werd een beschermend effect gevonden van foliumzuur op aangeboren hartafwijkingen, met name van de conotruncale defecten en de ventrikelseptumdefecten. Veel kinderen met Down syndroom hebben een hartafwijking, waarbij atriumseptumdefecten, atrio-ventrikulair septumdefecten en ventrikelsepteumdefecten het meest voorkomen. In hoofdstuk 4 is daarom met data van het Slone Epidemiology Center te Boston, VS, onderzocht of foliumzuur ook beschermt tegen hartafwijkingen bij kinderen met het syndroom van Down. We vonden geen effect.

In het laatste hoofdstuk van dit eerste deel is de relatie bestudeerd tussen het gebruik van clomifeen dat voor de zwangerschap wordt gebruikt om de eisprong op te wekken en het optreden van hypospadie. Dit is een afwijking bij jongetjes waarbij het uiteinde van de plasbuis niet op het puntje van de eikel zit maar lager op de penis. Clomifeen lijkt chemisch gezien sterk op diethylstillbestrol (DES). DES is bekend vanwege de sterk verhoogde risico's op vaginale kanker en infertiliteit onder dochters van de DES gebruiksters. DES is in de literatuur in verband gebracht met hypospadie bij kleinzonen van de DES gebruiksters. Hoewel clomifeen voor de zwangerschap wordt gebruikt blijft het lang aanwezig in het lichaam van de moeder waardoor het theorethisch mogelijk is dat het ongeboren kind aan de stof wordt blootgesteld. In onze studie vonden we een significant verhoogd risico (odds ratio=6,08; 95% BI: 1,40-26,33) op penoscrotale hypospadie, de meest ernstige vorm. Omdat we voor de minder ernstige vormen geen verhoogd risico vonden, zagen we vanwege de aantallen geen verhoogd risico wanneer we naar alle hypospadie gevallen als één groep keken. Deze studie benadrukt daarom het belang van het bestuderen van aangeboren afwijkingen op het meest gedetailleerde niveau.

Deel B: het verbeteren van dataverzameling voor epidemiologische studies naar de veiligheid van geneesmiddelen tijdens de zwangerschap.

In dit tweede deel van het proefschrift zijn enkele methoden om de nu beschikbare datasets te verbeteren of uit te breiden bestudeerd. Zoals eerder is aangegeven kunnen aantallen binnen studies worden vergroot door data van vergelijkbare registraties samen te voegen. Hierbij is het belangrijk dat verschillende registraties hun data op vergelijkbare manieren verzamelen en coderen. De beschikbaarheid van de geneesmiddelendata binnen het EUROCAT netwerk van 40 Europese registraties is onderzocht en beschreven in hoofdstuk 6. Door middel van een vragenlijst naar alle hoofden van die registraties werd informatie verzameld over de bron van hun geneesmiddelendata en over het vastleggen van deze informatie. Hieruit kwam naar voren dat verloskundige gegevens de meest frequente bron van informatie vormen en dat de meeste registraties de EUROCAT geneesmiddelcodering hanteren. Slechts vier gebruiken de veel gedetaileerdere internationale ATC-codering voor geneesmiddelen. Daarna werd voor 15 van de 25 responderende 'full members' de beschikbaarheid informatie over geneesmiddelengebruik in het eerste trimester tussen 1996 en 2000 bestudeerd in de centrale EUROCAT database. Percentages cases die waren blootgesteld aan geneesmiddelen varieerde van 4,4% tot 26,0% tussen deze registraties. Ook varieerde de soort geneesmiddelgroepen die vastgelegd waren sterk tussen de registraties. We concluderen daarom dat verschillende registraties in de praktijk geneesmiddelendata zeer verschillend hanteren. Het EUROCAT netwerk heeft desalniettemin de mogelijkheid om als effectief samenwerkingsverband bij te dragen aan post-marketing surveillance van geneesmiddelen in relatie tot teratogene risico's. Hiervoor dient wel het gebruik van ATC coderingen breder te worden ingevoerd en zullen meer informatiebronnen voor geneesmiddelengebruik moeten worden aangewend.

In hoofdstuk 7 worden de mogelijkheden om data te verzamelen over kinderen zonder aangeboren afwijkingen die kunnen dienen als controles voor de EUROCAT cases onderzocht. Vanwege de bestaande contacten werd eerst de mogelijkheid om de controles via de apotheek te recruteren onderzocht. De twee mogelijke routes binnen de apotheek bleken tot selectie te leiden en dus lijkt het recruteren via de apotheek niet de geschikte strategie. Als alternatief werd het recruteren van kinderen via de verloskundige onderzocht. Deze methode blijkt uitvoerbaar en de deelnemende zwangere vrouwen zijn een goede steekproef van de hele praktijk. Uit interviews met de deelnemers bleek dat deze een hele positieve houding hadden ten aanzien van het onderzoek. Hiermee is een eerste stap gezet in het verzamelen van een representatieve groep uit heel noord Nederland, de regio waarin EUROCAT de cases verzameld. De dataverzameling moet nu worden uitgebreid naar

andere verloskundigenpraktijken en naar de ziekenhuizen. Daarna moeten zowel de interne als externe validiteit grondig onderzocht worden. Desalniettemin geeft deze eerste stap redenen om optimistisch te zijn over de mogelijkheid om in de toekomst standaard de benodigde informatie te verzamelen over kinderen zonder aangeboren afwijkingen.

Deel C: implementeren van de kennis in de dagelijkse praktijk: foliumzuurvoorlichting in de apotheek.

Wanneer nieuwe kennis is verworven, moet dit gecommuniceerd worden met de uiteindelijke doelgroep: vrouwen in de vruchtbare leeftijd. Om strategieën te implementeren en daarmee het gebruik van foliumzuur te verbeteren hebben we in hoofdstuk 8 onderzocht wat de huidige kennis en het gebruik van foliumzuur is onder vrouwen die binnen bepaalde tijd zwanger willen worden. De resultaten laten zien dat 69% van de vrouwen die binnen zes maanden zwanger willen worden weet dat foliumzuur beschermt tegen aangeboren afwijkingen, 81% weet dat je al voor de conceptie moet beginnen met foliumzuur, en 64% weet beide. Deze percentages nemen af wanneer de periode waarin een vrouw zwanger wil worden langer wordt. Een vergelijkbare trend is te zien wanneer gevraagd werd naar het huidige gebruik van foliumzuur: van de vrouwen die binnen 6 maanden zwanger willen worden geeft 59% aan op dit moment foliumzuur te gebruiken tegenover 6% van de respondenten die over 12-24 maanden zwanger willen worden. Uit het multivariate model waarin gecorrigeerd wordt voor variabelen die mogelijk invloed kunnen hebben op de onderzochte relatie, blijkt dat het gebruik van foliumzuur samenhangt met kennis, een positieve attitude en het hebben ontvangen van mondelinge voorlichting van een medewerker in de gezondheidszorg.

Hoofdstuk 9 geeft de resultaten van het vooronderzoek naar de uitvoerbaarheid van het invoeren van foliumzuurvoorlichting in de apotheek. In vier apotheken zijn kernteams gevormd, bestaande uit één apotheker en één of twee assistenten, die verantwoordelijk waren voor de planning van de interventie. Deze moest minimaal bestaan uit het plakken van een sticker op de pil met de tekst “Kinderwens? Vraag informatie over foliumzuur in uw apotheek” en het minimaal één keer tijdens de onderzoeksperiode uitdelen van folders. Op basis van ervaringen van het apotheekteam, reacties van klanten en de resultaten van de vragenlijsten die naar de pilgebruikers was gestuurd kon de interventie worden aangepast. Deze zesweekse cyclus van planning, actie, observatie en reflectie werd twee keer herhaald om zo per apotheek tot een optimale interventie te komen. De minimale interventie werd door alle vier apotheken uitgevoerd. Andere methoden, zoals het ophangen van posters, het dragen van buttons, het inrichten van een etalage en het hanteren van een leeftijdsgrens

voor de sticker en folder verschilde per apotheek. Van de respondenten op een vragenlijst onder de doelgroep was 44% positief over de ontvangen sticker, 49% neutraal en 4% negatief. Daarnaast gaf 56% aan dat ze het waardeerden dat de apotheek preventie informatie verstrekte. Wij concludeerden dat het werken met een kernteam een succesvolle strategie was en dat het invoeren van de interventie in de apotheek uitvoerbaar is. Door de interventie cyclusgewijs te verbeteren tijdens de kernteambijeenkomsten kan een optimale methode worden bereikt die past in de bestaande organisatie en kunnen mogelijke hindernissen worden weggenomen. De positieve reactie van de doelgroep bleek zeer motiverend te zijn voor de kernteams.

Het effect van deze interventie op de kennis en het gebruik van foliumzuur in de doelgroep is beschreven in hoofdstuk 10. Driekwart van de vrouwen in de interventiegroep rapporteert een sticker te hebben ontvangen en 48% een folder. Vrouwen die informatie over foliumzuur hadden ontvangen met hun anticonceptiepil hadden meer kennis over foliumzuur en een positievere attitude ten aanzien van foliumzuur vergeleken met vrouwen uit de referentiegroep. Daarnaast was de intentie om foliumzuur te gaan gebruiken groter. De verschillen het duidelijkst onder de vrouwen die nog nooit zwanger waren geweest en onder vrouwen die aangaven binnen 12 maanden zwanger te willen worden. We kunnen hieruit dus concluderen dat het verstrekken van foliumzuurinformatie via de pil het bewustzijn over en het gebruik van foliumzuur doet toenemen onder vrouwen die binnen een jaar zwanger willen worden.

Conclusie

Het is duidelijk dat het bestuderen van en communiceren over teratogene risico's een gebied blijft dat aandacht behoeft. Er is een groot gebrek aan kennis over hoe en waarom sommige geneesmiddelen teratogene eigenschappen hebben terwijl andere, met een vergelijkbare structuur en werking, niet. Net zoals het vaak niet duidelijk is waarom het risico op een specifieke afwijking toeneemt na gebruik van een bepaald geneesmiddel terwijl dit middel geen effect lijkt te hebben op een verwante afwijking in hetzelfde orgaan. We kunnen dus stellen dat het onderzoek naar teratogeniteit nog nauwelijks uit de kinderschoenen is. De enige oplossing is het blijven investeren in het monitoren van zwangeren waarbij alle mogelijke relevante informatie zo goed mogelijk wordt vastgelegd zodat studies zeer gedetailleerd kunnen worden uitgevoerd. Niet alleen nemen zo de aantallen toe en dus de mogelijkheid om specifieke relaties te onderzoeken, ook komen er voordurend nieuwe middelen op de markt waarvan de veiligheid bestudeerd zal moeten worden. Hoewel

wetenschappers in een competitieve wereld werken, moeten ze in het veld van de teratogeniteit juist meer samenwerken vanwege de schaarsheid van de data.

Daarnaast moet er geïnvesteerd worden in de communicatie over deze risico's. Hierbij dienen wetenschappelijke bevindingen zo snel mogelijk te worden 'vertaald' naar klinisch bruikbare informatie zoals richtlijnen. Immers, artsen, apothekers en verloskundigen hebben deze informatie nodig om aanstaande zwangeren voldoende te kunnen informeren. De toekenning van een risico zoals in het Australische risico classificatiesysteem is een goede suggestie hoewel nu nauwelijks bruikbaar. De meeste geneesmiddelen zijn momenteel immers geclassificeerd als 'mogelijk schadelijk' of 'onbekend risico' wegens gebrek aan meer informatie. Tot slot geldt dat de voorlichting bij voorkeur wordt gegeven aan vrouwen voor de conceptie. Deze groep is moeilijk te bereiken en wisselt bovendien voortdurend van samenstelling. Investeren in de communicatie houdt dus ook in dat aandacht wordt besteed aan methodieken om deze doelgroep acitef te benaderen en de nieuwe informatievoorzieningen te implementeren in de dagelijkse praktijk van de arts, apotheker of iedere andere zorgverlener die in dit veld een rol kan spelen.

Dankwoord

Ook al staat er maar één naam op de kaft, een proefschrift schrijf je niet alleen. In de afgelopen jaren heb ik met veel mensen mogen samenwerken van wie ik allemaal heb kunnen leren. Het is dus ook niet meer dan logisch dat er een lange lijst van mensen is die ik voor hun inbreng wil bedanken.

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Curriculum Vitae

Willemijn Meijer was born in Meppel on February 4, 1976. In 1994 she started studying Pharmacy in Groningen where she received her Master's in pharmacoepidemiology in 1999. Her master thesis about the association between ovulation inducing drugs and birth defects was written in Århus, Denmark. In 2001 she received her Pharmaceutical degree. During her studies, Willemijn was appointed as teaching assistant several times to teach fourth grade students research methodology. She also had several assistantships at the Science Shop. Furthermore, she worked in a pharmacy in Groningen for a few months.

In September 2001, Willemijn started as a researcher at Social Pharmacy, Pharmacoepidemiology and Pharmacotherapy in Groningen on a project about introducing a folic acid intervention in daily practice of pharmacies. In January 2002 she was appointed a PhD student at the same group to study drug use before and during pregnancy and the risk on birth defects. In 2004, she also worked at the Eurocat Northern Netherlands registry of congenital anomalies for seven months. As a part of the PhD project, Willemijn visited the Slone Epidemiology Center of the Boston University, US, between November 2005 and February 2006 to study whether folic acid protects against heart defects among children with Down syndrome. This all resulted in her thesis titled "Drug safety in pregnancy. Studying and communicating teratogenic risks" which she will defend on December 8, 2006. Since August 2006, Willemijn has been working as a research associate at the Pharmo Institute for drug outcomes research in Utrecht.

Willemijn is a member of the Royal Dutch Pharmaceutical Society (KNMP), the Netherlands Epidemiological Society (VvE) and the International Society for Pharmacoepidemiology (ISPE).