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Postmarketing Analysis of Medicines

Methodology and Value of the Spanish Case-Control Study and Surveillance System in Preventing Birth Defects

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Abstract

There are many surveillance systems of congenital defects all over the world; several of them have developed specific approaches to generate and test selected hypotheses regarding human teratogens. However, to the best of our knowledge, none of them have a permanent and systematised programme for the study of the risk and safety of drugs.

The aim of this article is to describe the research programme on the potential effects of drugs in pregnancy followed by the Spanish Collaborative Study of Congenital Malformations (ECEMC), which is a permanent ongoing case-control study and surveillance system. The programme to analyse drugs includes a continuous and systematic study on the potential effects of medicines used during pregnancy. This programme has several characteristics that make it different from other current systems: (i) the collection of numerous datapoints (up to 312 per infant) in a case-control design; (ii) the use of a versatile and specific coding of birth defects; (iii) a specific programme for the continuous analysis of the

potential effects of each type of drugs used during pregnancy that has been developed specifically for the ECEMC methodology, including its dysmorphological coding system.

The description of the ECEMC's approach to surveillance of the effects of drug use during pregnancy may help researches in this area, particularly those using data from birth defects registries.

The recognition in 1961[1] that prenatal exposure to thalidomide altered embryonic development (teratogenic effect), initiated not only the review of the pre-requisites for drugs to be approved, but also the development of programmes for the surveillance of congenital malformations all over the world based on the registration of newborn infants with congenial anomalies.[2-6] The majority of these programmes are population-based,[4,5] whereas others are hospital-based and surveillance systems.[6] To the best of our knowledge, only three of these have a specific case-control design.[6,7] Most of these registries were organised under the hypothesis that the use of a new teratogenic drug increasing the frequency of some malformations would be detected by the registry.

Few teratogens, if any, have been identified only through these registries without a previous clinical suspicion, because observational studies of the teratogenic effects of drugs (as well as of other risk factors) used during pregnancy in humans are very difficult. This is due to several problems, including the fact that results from pre-clinical animal studies cannot be extrapolated to the human embryo due to genetic differences and the huge complexity of the human functional genome.[8,9] Other problems derive from the potential methodological biases that may be implicated in epidemiological studies, including sample size, selection of exposed and non-exposed infants, selection of the control (or reference) group and in the analyses performed.[10-15]

In 2003, Mitchell[16] commented that although there are individual programmes that have developed specific research approaches to generate and test selected hypotheses regarding human teratogens, none of them are focused on the "systematic and routine" study of the risk and safety of drugs. He concluded that a comprehensive surveillance system for teratogens is critically needed. Thus, the aim of this article is to describe the continuous research programme on the potential effects of drugs during pregnancy, conducted by the Spanish Collaborative Study of Congenital Malformations (ECEMC).

The ECEMC programme includes a systematic and continuous study on the effect (either of risk or safety) of drugs used during pregnancy, which is based on: (i) the collection of numerous datapoints in a case-control design; (ii) the use of a versatile and specific coding system of birth defects; and (iii) a specific programme for the ongoing surveillance of the potential effects of each type of drugs used during pregnancy, developed specifically for the ECEMC case-control methodology and its dysmorphological coding system.

1. Three Essential Characteristics the Spanish Collaborative Study of Congenital Malformations (ECEMC)

1.1 Case-Control Design and Data Collection

The ECEMC started in April 1976 as a continuous ongoing hospital-based, case-control study and surveillance system. This methodology is aimed not only at the surveillance of congenital anomalies, but also at investigating their characteristics, the clusters of congenital defects and their causes (either genetic or environmental).

The ECEMC network consists of two groups: the data-collecting ('peripheral') group and the 'co-ordinating' group. The peripheral group includes physicians in >80 collaborating hospitals throughout Spain who, being interested in the problem of con-

genital defects, altruistically collaborate with the ECEMC programme after having agreed to follow its common and strict methodology.

The collaborating paediatricians, who are trained to assess major and minor anomalies that are described in a common operational manual, examine the newborn infants just after birth and within the first 3 days of life to identify the cases and control newborns. Once these infants have been identified, described clinically and their data collected, the same paediatricians identify and interview the mothers of case and control infants (also during the first 3 days after delivery), using defined protocols to gather the same information (up to 312 different datapoints) in both groups of mothers.

Thus, when the paediatricians select the cases and control newborn infants, they are blinded to the different maternal and family data that they are going to collect. This information includes reproductive and family history, obstetrical data and exposures during pregnancy, such as acute and chronic maternal diseases, drugs, alcohol and tobacco, and maternal and paternal occupation, among others.

All the information on cases and control children, together with the number of total births by sex occurring in each hospital (the number of total births constitutes the denominator for the analysis of birth defects frequencies), is sent to the co-ordinating group on a monthly basis.

The co-ordinating group, located in Madrid, Spain, is composed of experts in congenital defects epidemiology, clinical teratology, dysmorphology, clinical genetics and cytogenetics; they perform the coding of congenital anomalies of the cases, the cytogenetic studies including fluorescent *in situ* hybridisation (FISH) techniques and, in collaboration with the participating paediatricians of the peripheral group, make the diagnosis of the cases and the epidemiological analyses.

1.1.1 Case Definition

Once the paediatricians have identified a newborn with birth defects, this is selected as a 'case', and all the detected anomalies, whether major and/or minor, are described in detail in the ECEMC's common protocols. In most instances, photographs, blood samples to perform the karyotypes with high-resolution bands and FISH techniques, imaging studies, pathology reports and results of other studies are also gathered, sent to the co-ordinating group, stored and made available for review. Blood spots from the cytogenetic samples are also stored in the ECEMC bio-bank.

1.1.2 Control Definition

For each case, the next non-malformed infant of the same sex born in the same hospital is selected as a control. Thus, the controls are selected from the same population as the cases and are representative of those who, had they developed malformations, would have been selected as cases. In Spain, 100% of the population is covered by the Public Health System (PHS); therefore, because 95% of the collaborating hospitals belong to the PHS, the ECEMC samples are representative of the Spanish population.

1.1.3 Drug Information and Coding

In the ECEMC protocols, apart from an open-ended question on drugs consumption in general, there are specific questions for the type of drugs belonging to each one of 13 main therapeutic groups, including over-the-counter medications. The collected information for each type of drug includes the brand name, daily doses, gestational age when the treatment occurred and duration of treatment. Through the brand name and the national code that each medicine has, we can identify their active principles for the analyses.

1.2 Coding System for Congenital Defects

The coding system followed in the ECEMC programme is highly sophisticated, since each child with any type of congenital anomaly is first analysed clinically and dysmorphologically, and the global pattern of anomalies is broken apart into all the recognisable pathogenetic patterns before being coded. Thus, to capture all this clinical information, a coding system with two levels and three sublevels was developed (table I).

The first level codes each individual anomaly (major and minor) detected in each child, without

Table I. An example of the clinical coding for infants with multiple congenital anomalies followed in the Spanish Collaborative Study of Congenital Malformations (ECEMC) Programme

Clinical description	Level 1	Level 2		
	codes for each defect	sublevel 1 (coding the global pattern of the child)	sublevel 2 (coding the specific diagnosis for the child)	sublevel 3 (code the different recognised patterns among the total defects of each child)
Holoprosencephaly	7421	Chromosomal syndrome	Trisomy 13	Holoprosencephay DFD
Microphthalmia	7441			Conotruncal DFD
Tetralogy of Fallot	7462B			More than three defects of the VACTERL DFD
Bilateral cleft lip and palate	7492			
Thoracic hemivertebrae	7568G			
Polycystic kidneys	7534B			
Absence of digits in the left hand	7552S			
Postaxial feet polydactyly	7550B			
Rokerbuton foot	7544C			

DFD = developmental field defect; **VACTERL** = vertebral anomalies, anal atresia, congenital cardiac disease, tracheoesophageal fistula, renal anomalies, radial dysplasia and other limb defects.

any limit of the number. To codify the different congenital defects, the 8th edition of the *International Classification of Diseases* (the only version available in 1976) was modified by adding up to three more digits, to obtain more specificity.

The second-level codes are separated into three sublevels. Sublevel 1 codes the global pattern of defects present in each child, indicating whether the child has an isolated defect, a sequence, a developmental field defect (DFD), an association, a complex, a particular type of syndrome (chromosomal, autosomal dominant, recessive, and others) or has a multiple congenital anomaly pattern of unknown cause and pathogenesis. Sublevel 2 codes the specific diagnosis corresponding to the pattern identified in sublevel 1. For instance, if sublevel 1 indicates a malformation sequence, the code in sublevel 2 would indicate the specific sequence (i.e. spina bifida sequence). Finally, sublevel 3 is used for children with multiple defect codes of any type. This sublevel 3, which has heuristic purposes, is used to code all well known (or suspected) pathogenetic patterns of anomalies that we are able to recognise among the whole group of congenital defects observed in each child.

For example, if a child has a trisomy 13 (table I), the first level specifies the codes for each individual defect that this child has. In the first sublevel of level 2, the code indicates that the child has a chromosomal syndrome and in the second sublevel of level 2 code, indicates specifically that the child has trisomy 13. Finally, the third sublevel includes the codes of the different specific patterns of anomalies identified among all the congenital defects (coded in level 1) of this infant that are: the codes to describe the holoprosencephay DFD (which are arrhinencephaly, hypotelorism and cleft lip and palate), a cardiac DFD (the group of conotruncal defects), and that stating that the child had three or more defects that are part of the VACTERL (vertebral anomalies, anal atresia, congenital cardiac disease, tracheoesophageal fistula, renal anomalies, radial dysplasia, and other limb defects); in this example, these are cardiac defects, hemi-vertebrae, limb deficiencies and renal anomalies. In this heuristic classification, the categories can be easily modified when necessary or in the light of new knowledge.

The advantage of this clinical coding system is that it allows for the analysis, as discrete units, of different groups of defects that are considered pathogenetically related as well as their clinical spectra. Clinically, we have used this system to demonstrate that several patterns of anomalies, such as VACTERL,[17] diaphragmatic hernia and upper

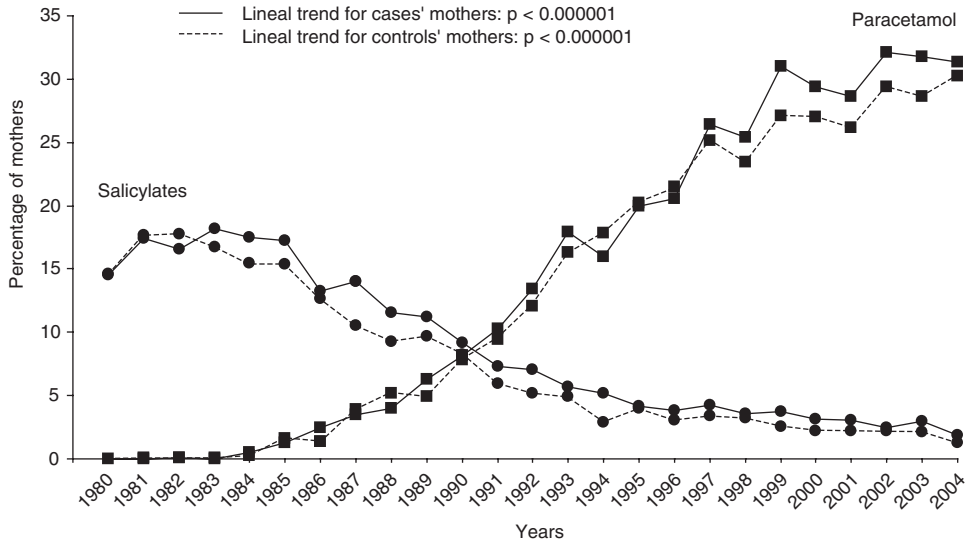

Fig. 1. Secular distribution of the use of both salicylates and paracetamol (acetaminophen) at any time during pregnancy.

limb deficiencies,[18] were, in reality, DFDs.[19-21] This coding system is also quite useful for the study of the teratogenic effects of particular types of drugs (or other teratogenic agents), since it can be analysed in specific groups of malformations or by the different code levels of our coding system, which also permit the analysis of the variability of the potential effect of each drug.

1.3 Systematic and Routine Study of Drugs Used during Pregnancy

The aforementioned characteristics of the ECEMC methodology allow the continuous surveillance and analysis of drugs used during pregnancy through three main systematic approaches.

1.3.1 Ongoing Surveillance of the Secular Use of Drugs during Pregnancy

As the design of the ECEMC methodology is based on case-control, the secular analysis of drugs used during pregnancy by the mothers of both cases and controls provides us with a large amount of useful information in at least three aspects:

1. An analysis of the temporal changes that may occur in the use of drugs during pregnancy. For example, using our data, figure 1 indicates that maternal use of salicylates by mothers of both cases and controls shows a decreasing lineal trend along time ($p < 0.000001$, respectively), while the use of paracetamol (acetaminophen) presents a significant increasing lineal trend also in both groups of mothers ($p < 0.000001$, respectively). Another example (figure 2) is the secular trend of maternal use of sex hormones, which shows a decreasing trend until 1996, followed by an increase in the use of these drugs from 1997 onwards. This last observation promoted the analysis of the causes for the observed tendencies, and it was possible to determine that they were mainly related to the increasing maternal age and the concomitant increase of assisted reproductive technology (ART) that have been observed during the last few years in our population.[22]

2. The comparison of secular distributions observed among mothers of cases and controls (as in figures 1 and 2). This observation permits us to determine whether or not both distributions overlap along the years, giving clues to perform an in-depth analysis to identify if the study drug may bear a risk for birth defects. Figures 1 and 2 show that the use of the drugs for case and control mothers is quite similar (particularly for the use of paracetamol) for all years since the beginning (1980) through to the year 1996, when the situation changed. Since 1996, the use of

paracetamol and sex hormones has been more frequent in mothers of cases than in those of controls. The subsequent analysis showed that the increased use of these drugs was related to older maternal ages. The observed increased use in mothers of cases is influenced by both the increased risk of some congenital defects that older mothers have and the increased risk for some birth defects observed in children conceived by ART,[23,24] which is a technology frequently used by older mothers. Thus, from the comparison of secular distributions observed among mothers of cases and controls, it is also possible to obtain clues on the potential effects of other agents. The identification of factors related to the drugs and congenital defects is most important because they are confounder factors that should be considered in the final adjusted analysis of the risks.

3. Rapid identification of clues on the potential risks and safety of new drugs. For instance, figure 3 shows the secular maternal use of newer anticonvulsant drugs (i.e. lamotrigine, gabapentine) in the data of the ECEMC. Although the numbers are still too small, the observation of a higher use by mothers of cases than of controls prompted the individual analyses of data from the nine exposed cases and the three exposed control infants. The results show that four of nine cases were prenatally exposed to monotherapy at different doses and throughout pregnancy, presenting the following defects: one case with syndactyly (exposed to lamotrigine), one case with pylonidal sinus plus sacrococcygeal tail (exposed to gabapentin), one case with balanic hypospadias (exposed to lamotrigine) and one case with Ebstein anomaly (exposed to lamotrigine). The other five cases were prenatally exposed to anticonvulsant polytherapy: two to lamotrigine and topiramate; one to lamotrigine plus valproic acid; one to carbamazepine plus topiramate; and one to carbamazepine plus gabapentin. Four of these five newborns had multiple congenital anomaly patterns, including major and minor defects. These patterns were concordant with those observed in cases with embryofetopathy by anticonvulsant drugs. The remaining infant, who was exposed to lamotrigine and valproic acid, had trisomy 21. None of these five cases had neural tube defects, which may be due to prenatal diagnosis of these defects with subsequent termination of pregnancy. Among the three exposed controls, one was exposed to monotherapy and the other two to polytherapy. Thus, although the results do not show any association because the sample size is quite small, a specific ongoing surveillance for these drugs has been established to quickly detect an

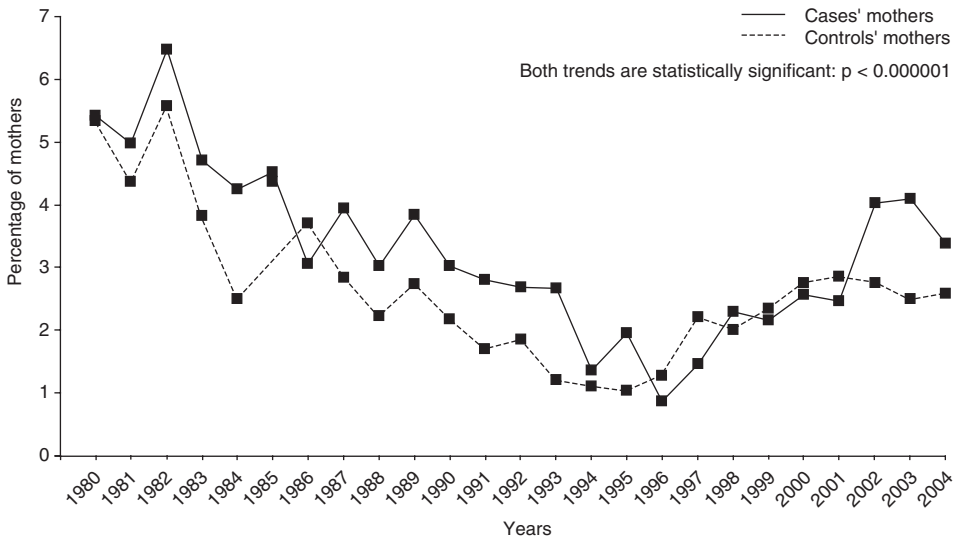

Fig. 2. Secular distribution of the use of sex hormones at any time during pregnancy.

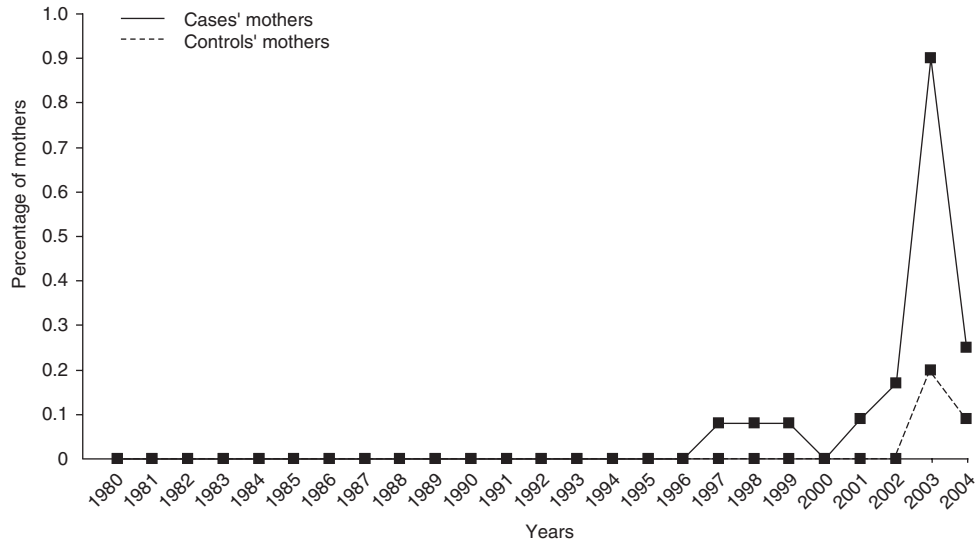

Fig. 3. Secular distribution of the use of new anticonvulsant drugs during pregnancy.

increased risk of birth defects, by using the approaches described in the next section.

1.3.2 Continuous and Systematic Analysis of the Safety Risk of the Drugs

The routine ongoing study of the effects of different drugs during pregnancy is performed through a system that was developed by the ECEMC's group specifically for its methodology and coding system. As that system was designed to calculate odds ratios, it is called the 'risks programme'. It is possible to select not only the drug to be studied, but also any other factor included in the protocols, such as the level of the study drug (i.e. by therapeutic class, brand name or active principle), the weeks of gestation when the exposure occurred and if the selected exposure was to mono- or polytherapy. It is also possible to select the non-exposed cases and controls (i.e. not exposed to the drug to be studied, not exposed to any drugs belonging to the same therapeutic group of the one selected for the analysis or not exposed to any type of drugs). The system also permits the selection of the type of defects to be studied (i.e. all types of congenital defects as a group or one by one, a particular group of defects, any of the subgroups of the coding system) and the ability to make exclusions (excluding syndromes, cases with known cause etc.). Finally, it is possible to select the group of controls to be included in the analysis (i.e. the corresponding control to each case, all the controls born in the same regions as the cases, all the controls).

We use the risks programme for different objectives, including:

- to quickly obtain global information on the relationship between the different drugs and birth defects and its concordance with observed secular trends of the use of these drugs during pregnancy, as shown in figure 1, figure 2 and figure 3;
- to have rough information on the routine and ongoing analysis of drugs and birth defects associations, as a surveillance of drug-effect;
- to have an ongoing surveillance of new drugs;
- to generate hypotheses on the potential effect of the different drugs used during pregnancy;
- to serve as the first step to quickly test previous hypotheses generated either by the literature, our ongoing study of the secular trend in the use of drugs, or medical or social alarms.[25-30]

1.3.3 The Adjusted Analysis of a Particular Drug-Effect Association

An adjusted analysis of a particular drug-effect association is the final calculation for some of the

results obtained in any of the previous approaches. This is performed through different logistic regression analyses (particularly conditional) controlling all the factors identified as possible confounders.

2. Limitations and Strengths of the ECEMC System

The limitations of the ECEMC system are mainly related to the period selected to identify the infants with congenital defects, which encompasses the first 3 days of life. Thus, the study of the effects of prenatal exposure to different drugs (or other potential risk factors) is limited to structural dysmorphological abnormalities and not to embryo-fetal lethality or to developmental disorders. However, even if these limitations affect the study of a particular type of drug, they do not affect the structure and design of our research system.

The strengths of the ECEMC programme and the study of the teratogenic effects of drugs in pregnancy are based in their design and in the way of controlling or minimising several methodological biases, particularly in the following aspects: (i) the selection of cases and control infants; (ii) the collection of data and exposures; and (iii) their analyses.

2.1 To Control Biases in the Design Stage and Selection of Cases and Controls

The design for the methodology to obtain the information includes different aspects that minimise potential biases of this stage. These are:

- the selection of cases is based on the fact that they have any type of congenital defect, major or minor, and not because they presented with a particular type of defect or group of defects;
- the paediatricians examine all newborns in their hospitals and identify case and control children before they identify their mothers. Therefore, when the paediatricians identify the cases and controls, they are blinded to any familial or maternal characteristic and prenatal exposure;
- the collection of data is not focused to any particular study, but is a permanent and systematised registry to gather data independent of aetiology or causality;
- the collected variables are always the same for malformed children (regardless of the type of congenital defects they present) and their controls;
- the data on drugs are not collected to test any particular hypothesis, but they are part of the pool of data that should be collected, according to established protocols.

2.2 To Control the Biases in the Collection of Data

- The information on case and control newborns is gathered by the collaborating paediatricians, who examine all newborn infants within the first 3 days of life, identifying and describing the different congenital defects and other clinical characteristics.
- The collaborating paediatricians are trained to identify all major and minor congenital defects at birth. A list of the minor defects that should or should not be considered is included in the ECEMC operational manual.
- To gather the maternal and familial information, the same paediatrician interviews the case and control mothers. This implies that if a paediatrician introduces any bias in his/her way of interviewing the mothers, this bias will be the same for the mothers of both case and control infants. Thus, this bias will not affect the results.
- The maternal interview is performed during the first 3 days after birth. In this situation, mothers who have just delivered a malformed child are not psychologically predisposed to answer questions regarding what they did during the recent pregnancy. The opposite occurs with mothers having delivered healthy infants during the same period. In fact, they are happy to accept the interview and collaborate actively. Consequently, the control mothers answer more accurately than case mothers, and the potential bias of this difference (only occurring when the information is gathered soon after birth) will go in the direction of the null hypothesis.[31]

2.3 To Control the Biases in the Analysis of Data

- As the system includes information on many datapoints (up to 312) for each case and control pair, it is possible to study different risk factors to control many potential confounders during the analysis.
- The system includes >35 000 case-control pairs that increases each year by about 1100–1300 pairs.
- The coding of each malformed infant permits not only the selection of different types of malformed newborns, or specific groups of defects, but also exclusions, when necessary, of different types of children (i.e. those having a known genetic or environmental cause with different types of defects) and control infants.

3. Differences with Other Programmes

All the aforementioned characteristics make this programme different from other current systems. First, population-based studies usually collect data from hospital records and from other different sources. This implies that many different physicians and/or other health professionals generated all the data from cases (including major and minor congenital defects). This may introduce a degree of heterogeneity because of disparities in the ability of different physicians to detect some defects. Second, the clinical analyses are mostly based on codes that are performed by different people who are not connected and do not have the level of specificity of the ECEMC coding system, at least for several clinical patterns of defects. Third, the data on cases are collected by different professionals within each hospital and it is not known if these professionals are blinded to the maternal information when they register the defects of malformed children. Lastly, population-based studies assume that all the data have the same level of accuracy.

Regarding other current case-control studies, as far as we know, two of them[32,33] have the same type of design that the ECEMC has, in the sense that they are hospital-based and case-control studies, in the way they identify the cases and control infants and in the collection of data. However, the ECEMC has three main characteristics that have not been described in the other two programmes: the in-depth clinical analysis performed in the ECEMC and the two-level coding system, the amount of information (up to 312 datapoints) collected in the ECEMC programme (either for cases or controls) and the design of a permanent and systematic programme to study the potential effects of drugs used during pregnancy.

4. Conclusion

To conclude, it should be emphasised that the human teratogenic effect of different drugs can only be identified after they have been used during pregnancy and infants have been born with congenital malformations. Thus, what we need is a continuous, ongoing system that allows us to identify the safety or the potential teratogenic effects of the different drugs rapidly enough to avoid severe consequences, such as the affectation of many children and the unjustified termination of pregnancies just because the mothers took some drug. The description of the continuous and systematic Spanish system may help researchers in this area, and constitute valuable tools for clinicians, PHSs and regulatory agencies, such as the US FDA and the European Medicines Agency.

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