Case Report

Association of High-Dose Bifonazole Administration during Early Pregnancy and Severe Limb Reduction Defects in the Newborn

Nehama Linder,[1,2]* Mandola Amarilla,[1] Adriana Hernandez,[1] Tsippy Tamiri,[3] Lea Sirota,[2,4] Gil Klinger,[2,4] Itzhak Levy,[5] and Paul Merlob[1,2]

[1]Department of Neonatology, Rabin Medical Center, Petach Tikva, Israel
[2]Sackler School of Medicine, Tel Aviv University, Tel Aviv, Israel
[3]Division of Identification and Forensic Science, Israel Police National Headquarters, Jerusalem
[4]Department of Neonatal Intensive Care
[5]Pediatric Infectious Diseases Unit; Schneider Children's Medical Center of Israel, Petach Tikva, Israel

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BACKGROUND: Neonatal limb reduction defects may be caused by exposure to an external agent. The azole derivatives are used in the treatment of systemic and dermal mycoses. Their relative teratogenic risk is still controversial. **CASES:** We describe two newborns with severe limb defects who were exposed to high doses of oral (an unacceptable route) and/or intravaginal bifonazole during the entire first trimester of pregnancy. **CONCLUSION:** Although only two cases are insufficient to establish a relationship, our data suggest that maternal intake of bifonazole in early pregnancy poses a risk of morphogenic malformations. The literature suggests several possible mechanisms. *Birth Defects Research (Part A) 88:201–204, 2010.* © 2009 Wiley-Liss, Inc.

Key words: bifonazole; teratogenicity; limb defects

INTRODUCTION

Neonatal limb reduction defects may be caused by a genetic factor (primary defects) or occur secondary to exposure to an external agent, such as radiation, chemicals, or pharmacologic agents. Malformations caused by amniotic bands are included in the latter group.

Despite concerns about the effect of maternal drug use during pregnancy, according to the World Health Organization, the incidence in Europe is surprisingly high. Approximately 80% of medications taken during pregnancy are antiinfective agents; topical antifungals account for approximately 12% of this total (Shepard and Lemire, 2007). The azole derivates play an important role in the treatment of systemic and dermal mycoses. They are considered less toxic than other antifungal medications, but their relative teratogenic risk is controversial. In humans, data on limb defects in developing fetuses exposed to antifungal medications are sparse (Aleck and Bartley, 1997; Kazy et al., 2005, Leachman and Reed, 2006; Carter et al., 2008). The aim of this study is to describe two cases of limb malformations in newborns exposed to bifonazole during early pregnancy.

CASE REPORTS

Two infants born with severe limb defects were exposed to high doses of maternal oral and/or intravaginal bifonazole during the entire first trimester of pregnancy. In both cases, other possible explanations for the

*Correspondence to: Nehama Linder, Department of Neonatology, Rabin Medical Center, 2 Denmark St. Petach Tikva 49100, Israel.
E-mail: linderm@ netvision.net.il
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congenital malformations, including genetics (family pedigree, consanguinity), intrauterine infection, and exposure to environmental factors (e.g., radiation, chemicals, other pharmacological agents) were ruled out.

Patient 1

The first patient was the first child of healthy, non-consanguineous parents (maternal age, 20 years) of Ashkenazi Jewish origin. The family history was unremarkable and negative for congenital malformations. The mother did not smoke or drink alcohol or coffee, and she had no known infection during pregnancy. From the 6 to 16 weeks' gestation, to alleviate nausea she self-administered an unmarked syrup prepared by her pharmacist. An investigation after the birth of the child by the Division of Identification and Forensic Science of the Israel Police revealed that the syrup contained pure bifonazole. In addition, the mother had used topical clotrimazole to treat a vaginal yeast infection. In the 9th week of gestation, she exhibited anxiety restlessness, and sleep disturbances and was treated by her physician with an herbal preparation of valerian.

A male infant was born at 40 weeks' gestation with a birth weight of 2450 grams. Physical examination revealed severe skeletal malformations: amelia of the lower limbs, absence of the right forearm, absence of the left elbow, sacral dysgenesis, and bilateral 12th rib agenesis. There were no anomalies of the face or skull. The results of ultrasound and funduscopic examinations of the head, abdomen, kidneys and heart were normal, as were a brainstem auditory evoked response test and liver and kidneys function tests. On follow-up at age 2 years, physical and neurologic development were within normal limits. Speech and cognitive development were good (Fig. 1).

Figure 1. Patient 1 with severe limb reduction defects. [Color figure can be viewed in the online issue, which is available at www.interscience.wiley.com.]

Patient 2

The second patient was the first child of healthy, unrelated Sephardic Jewish parents (maternal age, 22 years). The prenatal and family histories were unremarkable. To treat a vaginal discharge, the mother had started to use vaginal suppositories of bifonazole (500 mg) on a daily basis before conception, and she continued to do so until 20 weeks' gestation, and then intermittently during the rest of her pregnancy. In addition, she was prescribed 10 ml (500,000 units per 5 ml) of oral nystatin syrup daily.

A female infant was born at 40 weeks' gestation with a birth weight of 2974 grams. Physical examination revealed left ulnar and radial hypoplasia, left phocomelia, and a left preauricular sinus. Echocardiography revealed a small atrial septal defect. Findings of head and renal ultrasound, funduscopic, brainstem auditory evoked response, liver function, and kidney function tests were normal. There were no skeletal abnormalities apart from the limb defect. On follow-up at age 2 years, physical and neurologic development were within normal limits.

DISCUSSION

We describe two patients with severe limb reduction defects whose mothers were treated with high doses of the antimycotic drug bifonazole during at least the first trimester of pregnancy. The duration of exposure was 10 weeks in the first case and several months in the second.

Studies have shown that azole derivates cross the placenta in humans. Following the administration of certain drugs, either orally (fluconazole and ketoconazole) or topically (bifonazole, clotrimazole, econazole, miconazole, and sulconazole), plasma and tissue levels were found to be above half maximal inhibitory concentration (IC50) values (Kragie et al., 2002; Turner et al., 2002). In a report on five cases of high-dose (>400 mg/day) fluconazole administration during the first trimester, one study documented a distinctive pattern of multiple malformations characteristic of Antley-Bixler syndrome (craniosynostosis, short or absent thumbs and hallux, digital synostosis, and joint contractions), different from the severe limb reduction defects noted here. The only other azole derivatives associated with limb disruptions in humans were itraconazole (one case of right-hand dysplasia) and ketoconazole (one case of a nonspecified limb anomaly).

There are several studies of the administration of lower doses of azole derivatives, such as those prescribed for vaginal fungal infections, which were associated with fewer adverse outcomes (Briggs et al., 1996; Pursley et al., 1996; Aleck and Bartley, 1997; King et al., 1998;

Reardon et al., 2000; Fukami et al., 2005; Lopez-Rangel and Van Allen, 2005; Leachman and Reed, 2006; Reproductive Toxicology Center, 2009c). However, recent data on the vaginal use of a combination of azole derivatives (metronidazole and miconazole) increase the risk for poly-syndactyly sixfold (Kazy et al., 2005; Reproductive Toxicology Center, 2009f).

It is noteworthy that bifonazole specifically is administered as a topical preparation. Although the oral route is not recommended, the mother in the first case was prescribed the drug as a syrup by her pharmacist.

Besides the dose and route, the timing and duration of administration appear to play a crucial role in the teratogenic response. Animal studies show a significant incidence of axial and appendicular skeletal defects, but only when the antimycotic drug was administered during the gestational period of limb development (Rosa et al., 1987; Lee et al., 1992; Briggs et al., 1996; King et al., 1998; Tiboni, 2006; Shepard and Lemire, 2007; Carter et al., 2008; Reproductive Toxicology Center 2009a, 2009b, 2009c, 2009d, 2009e). In several reports, in utero exposure to high doses of fluconazole had no adverse effect (Briggs et al., 1996; Aleck and Bartley, 1997; Jick, 1999; Reardon et al., 2000; Reproductive Toxicology Center, 2009c), and bifonazole administered orally in high doses to animals did have embryotoxic and fetotoxic effects. Therefore, it is noteworthy, that in our first case, bifonazole was used from 6 to 16 weeks' gestation, and in the second case, from before conception to week 20, continuously. Both cases were exposed to bifonazole on a daily basis between 4 and 12 weeks' gestation, which are known to be the weeks of limb development.

The mechanisms underlying the effects of bifonazole on fetal skeletal development remain unclear. Studies have shown that the azole derivatives, though inexpensive and broadly available, have the potential to inhibit steroidogenesis, adversely affecting endocrine and reproductive system function and sexual behavior (Zahn, 2003). The antifungal effect of the azole derivatives is by inhibition of sterol C-14 alpha demetilation. In the genetic abnormality of Antley-Bixler syndrome, decreased lanosterol 14 demethylase (CYP 51) is found, which is similar to what appears in azole embriopathy (Byskov et al., 1995; Stromstedt et al., 1998; Zhang et al., 2002; Zahn, 2003; Cotman et al., 2004; Reproductive Toxicology Center, 2009c).

Alternatively, cholesterol is a precursor of steroid hormones and bile acids and has long been recognized as an important structural component of cellular membranes and myelin. It is believed to play a crucial role in human embryogenesis. Several inherited disorders have been linked to enzyme defects in the cholesterol biosynthetic pathway, such as Smith-Lemli-Opitz syndrome, X-linked dominant chondrodysplasia, punctata, congenital hemidysplasia, ichthyosiform erythema, and limb defects (Aleck and Bartley, 1997; Reardon et al., 2000; Waterham, 2002; Zhang et al., 2002; Reproductive Toxicology Center, 2009g). Specific impairments in cholesterol biosynthesis may have contributed to the development of the pathologic skeletal features in our cases as well.

It is impossible to establish a causal relationship between bifonazole intake during pregnancy and severe limb reduction defects in the newborn based on only two cases. Nevertheless, this report suggests that the administration of bifonazole in the first trimester (the period of limb development), especially by an unacceptable route (oral) and for a prolonged period, poses a risk of multiple morphogenic and congenital malformations, including internal organ, skeletal, and skin abnormalities (Rosa et al., 1987; Briggs et al., 1996; Aleck and Bartley, 1997; Kragie et al., 2002; Shepard and Lemire, 2007). Further studies should examine the preliminary results of an association between limb defects and high-dose bifonazole administration during early pregnancy.

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