Original Investigation

Association Between Use of Oral Fluconazole During Pregnancy and Risk of Spontaneous Abortion and Stillbirth

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IMPORTANCE Vaginal candidiasis is common during pregnancy. Although intravaginal formulations of topical azole antifungals are first-line treatment for pregnant women, oral fluconazole is often used despite limited safety information.

OBJECTIVE To study the association between oral fluconazole exposure during pregnancy and the risk of spontaneous abortion and stillbirth.

DESIGN, SETTING, AND PARTICIPANTS Nationwide register-based cohort study in Denmark, 1997-2013. From a cohort of 1 405 663 pregnancies, oral fluconazole–exposed pregnancies were compared with up to 4 unexposed pregnancies matched on propensity score, maternal age, calendar year, and gestational age (based on gestational age at first day of treatment with eligible controls surviving through this date). To test for confounding by indication, pregnancies exposed to intravaginal formulations of topical azoles were used as an additional comparator group.

EXPOSURES Filled prescriptions for oral fluconazole were obtained from the National Prescription Register.

MAIN OUTCOMES AND MEASURES Hazard ratios (HRs) for spontaneous abortion and stillbirth, estimated using proportional hazards regression.

RESULTS Among 3315 women exposed to oral fluconazole from 7 through 22 weeks' gestation, 147 experienced a spontaneous abortion, compared with 563 among 13 246 unexposed matched women. There was a significantly increased risk of spontaneous abortion associated with fluconazole exposure (HR, 1.48; 95% CI, 1.23-1.77). Among 5382 women exposed to fluconazole from gestational week 7 to birth, 21 experienced a stillbirth, compared with 77 among 21 506 unexposed matched women. There was no significant association between fluconazole exposure and stillbirth (HR, 1.32 [95% CI, 0.82-2.14]). Using topical azole exposure as the comparison, 130 of 2823 women exposed to fluconazole vs 118 of 2823 exposed to topical azoles had a spontaneous abortion (HR, 1.62 [95% CI, 1.26-2.07]); 20 of 4301 women exposed to fluconazole vs 22 of 4301 exposed to topical azoles had a stillbirth (HR, 1.18 [95% CI, 0.64-2.16]).

CONCLUSIONS AND RELEVANCE In this nationwide cohort study in Denmark, use of oral fluconazole in pregnancy was associated with a statistically significant increased risk of spontaneous abortion compared with risk among unexposed women and women with topical azole exposure in pregnancy. Until more data on the association are available, cautious prescribing of fluconazole in pregnancy may be advisable. Although the risk of stillbirth was not significantly increased, this outcome should be investigated further.

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JAMA. 2016;315(1):58-67. doi:10.1001/jama.2015.17844

Pregnant women are at increased risk of vaginal candidiasis, due to the increased secretion of sex hormones.[1] The prevalence of vaginal candidiasis among pregnant women is estimated to be 10% in the United States.[2] Although intravaginal formulations of topical azoles are first-line treatment during pregnancy, oral fluconazole is used in cases of recurrence, severe symptoms, or when topical treatment has failed[3] but also may be used as the first treatment by personal preference.[4]

Concern regarding the safety of fluconazole use in pregnancy has been raised after case reports have linked long-term, high-dose fluconazole treatment in pregnant women to a distinct pattern of craniofacial and skeletal birth defects.[5-8] Consequently, epidemiologic safety studies of the lower doses of fluconazole commonly used in pregnancy have focused on possible teratogenic effects and have not shown an association with birth defects overall or most specific birth defects.[9-14] Only 2 epidemiologic studies have investigated spontaneous abortion and stillbirth, suggesting that fluconazole is not associated with an increased risk; however, with a combined sample of 1512 fluconazole-exposed pregnancies, these studies may not have had sufficient power to detect even a moderately increased risk.[11,13]

In a nationwide register-based cohort study, we evaluated the association between oral fluconazole exposure during pregnancy and the risk of spontaneous abortion and stillbirth.

Methods

Study Design

The Medical Birth Register contains records of all Danish births with gestational age recorded in days[15] and primarily based on ultrasonography.[16] The National Patient Register contains records of all outpatient and inpatient hospital contacts in Denmark.[17] In records with diagnoses of abortive outcomes, gestational age is primarily recorded in days (89.4% in our study cohort; those recorded in weeks were converted into days by multiplying with 7) and based on ultrasonography or the first day of last menstrual period (eMethods in the Supplement). Based on the 2 registers, we identified a cohort of all pregnancies ending with a singleton live birth, stillbirth, spontaneous abortion, and other abortive outcomes (including ectopic pregnancy, hydatidiform mole, other abnormal products of gestation, or induced abortion) in Denmark between January 1, 1997, and December 31, 2013. Pregnancy onset was estimated by subtracting gestational age from the date of birth or abortive outcome. We excluded pregnancies with a missing or implausible gestational age and pregnancies with multiple records on overlapping dates. Using unique personal identifiers, individual-level data on maternal exposures and covariates were obtained from the National Prescription Register,[18] the National Patient Register,[17] the Central Person Register,[19] and Statistics Denmark and linked to the cohort (registers are described in eMethods in the Supplement). Many very early pregnancy losses are not recognized clinically, and we expected incomplete recording in the National Patient Register. Hence, including pregnancies with very early exposure to oral fluconazole in the study cohort, which is constructed by identifying pregnancy outcomes (in contrast to all initiated pregnancies), would have introduced immortal time bias—an artificially low risk of very early spontaneous abortions biasing the results downward. Therefore, ascertainment of exposure and pregnancy outcomes started in gestational week 7. Furthermore, we excluded women who filled oral azole antifungal prescriptions before pregnancy onset to account for delayed use and women diagnosed with fungal infection during hospitalization to account for unavailable data on inpatient antifungal drug treatment (eMethods in the Supplement). The study was approved by the Danish Data Protection Agency. Informed consent is not required for register-based research in Denmark.

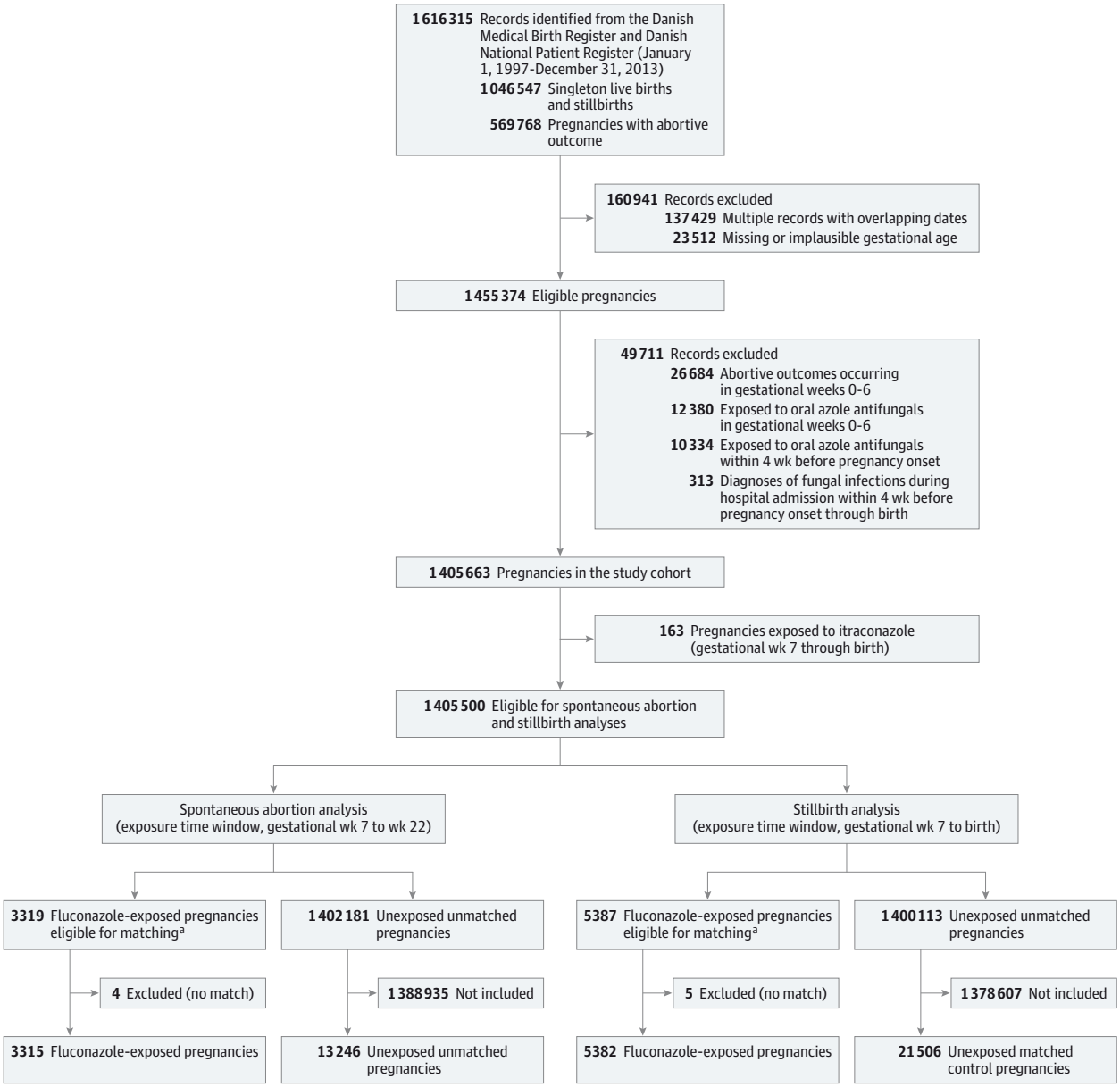
Oral Fluconazole–Exposed Pregnancies

Information on prescriptions for oral fluconazole (a prescription-only drug) was obtained from the National Prescription Register. The specific time windows of exposure and nonexposure in the analyses of spontaneous abortion and stillbirth were gestational week 7 to 22 and week 7 to birth, respectively (eFigure in the Supplement). Exposed person-time was defined to start on the date when the first prescription was filled; thereafter, women were considered exposed throughout follow-up. In the analysis of different doses of fluconazole, we calculated the total milligram amounts in the latest filled prescription. The predefined dose categories (150 to 300 mg and 350 to 5600 mg) were based on the standard treatment of vaginal candidiasis, which is one or two 150-mg doses of oral fluconazole, while higher doses are used for more complicated fungal infections, including recurrent vaginal candidiasis.[20]

Matched Control Pregnancies

To control for potential confounders, we matched each fluconazole-exposed pregnancy to up to 4 unexposed control pregnancies based on propensity scores, maternal age (5-year categories), calendar year, and gestational age (based on gestational age at first day of treatment for each exposed pregnancy; unexposed pregnancies eligible as matches were those who had survived through to this date of gestational age). The nearest neighbor matching algorithm was used (caliper width 0.2 of the standard deviation of the logit score).[21,22] The propensity score was estimated using logistic regression as the probability of fluconazole treatment given all maternal baseline characteristics at pregnancy onset and calculated for the specific exposure time window for spontaneous abortion and stillbirth (eTable 1 in the Supplement). Covariate balance in the matched cohorts was assessed by checking standardized differences between exposed and unexposed pregnancies. Any given characteristic was considered well-balanced if the standardized difference was less than 10%.

Figure 1. Numbers of Pregnancies Included in the Study Cohort and in the Analyses



[a] Matching 1:4 on gestational age, calendar year, and propensity score.

Pregnancy Outcomes

Cases of spontaneous abortion, defined as pregnancy loss from 7 through 22 gestational weeks, were identified through the National Patient Register and cases of stillbirth, defined as pregnancy loss from 23 weeks, were identified through the National Patient Register and the Medical Birth Register. The definition of stillbirth in Denmark changed during the study period; from April 2004, stillbirth was defined as pregnancy loss after 22 completed weeks, while prior to this the cutoff was after 28 completed weeks.[23] In our study, pregnancy losses from week 23 were considered stillbirths (eMethods in the Supplement).

Statistical Analysis

Proportional hazards regression with gestational age in days as the underlying time-scale was used to estimate hazard ratios (HRs) with 95% confidence intervals, comparing hazard rates of spontaneous abortion and stillbirth in exposed and matched control pregnancies. The final model included the matched cohorts for analyses of spontaneous abortion and stillbirth, respectively, with no additional adjustment. In the analysis of spontaneous abortion, follow-up was from the day of exposure or index date (from week 7 at the earliest) through gestational week 22. Censoring criteria were other abortive outcomes in the follow-up period. In the

analysis of stillbirth, follow-up was from gestational week 23 or from the day of exposure or index date (if this occurred after week 23) and censoring criteria were induced abortion or livebirth in the follow-up period. The proportional hazards assumption was assessed using a Wald test for the interaction between treatment status and the time scale; the assumption was fulfilled in all analyses. Analyses were conducted on individual pregnancies, and a woman could contribute with repeated pregnancies. We used the generalized-estimating-equation method (with the mother's identification number as a cluster) to take into account the possibility of correlation between several pregnancies in the same mother.

SAS version 9.4 (SAS Institute Inc) was used for all analyses. Differences were considered statistically significant when the 95% CIs did not overlap 1.0 and when $P < .05$ (2-sided test).

We conducted a number of preplanned sensitivity analyses. To investigate the possibility of confounding by indication, each oral fluconazole-exposed pregnancy was matched to 1 pregnancy exposed to topical azoles. Intravaginal formulations of clotrimazole or miconazole are considered safe, owing to minimal systemic absorption,[24,25] and represent first-line treatment of vaginal candidiasis during pregnancy.[3] They can also be purchased over the counter in Denmark, but only those filled as prescriptions are included in the National Prescription Register. The matching was based on propensity score and the gestational age at which the exposure occurred. Similarly, each fluconazole-exposed pregnancy was matched to 1 pivmecillinam-exposed pregnancy (a prescription-only penicillin used as first-line treatment of urinary tract infection during pregnancy in Denmark[26]) to control for unmeasured confounders related to infections treated with systemic anti-infectives during pregnancy. We also matched each fluconazole-exposed pregnancy to a pregnancy exposed to fluconazole in the year before the index pregnancy on propensity score, maternal age, calendar year, and gestational age to address the influence of time-independent characteristics of women using fluconazole. To control for time-independent factors, such as hereditary predisposition and unmeasured risk factors related to lifestyle, we analyzed the risk associated with fluconazole in a cohort of women who had at least 2 pregnancies with different treatment status. The within-mother analysis was adjusted for maternal age, parity, and calendar year (eMethods in the Supplement).

Additionally, to examine potential mechanisms, we assessed the risk of spontaneous abortion and stillbirth according to low (150-300 mg) vs high (350-5600 mg) fluconazole dose, within 2 weeks after exposure (which would presumably be consistent with an acute adverse effect) vs later, and restricted the exposure time window to week 7 to 10 weeks vs later. Follow-up started in week 7. The period of maximal susceptibility to teratogenic agents is gestational weeks 4 to 10[27]; an increased risk isolated to exposure in this period may suggest potential for malformation-related fetal death. The difference in the HRs in the sensitivity analyses was assessed using a Wald test for the interaction between treatment status and subcategory of exposure. We assessed HRs according to the timing of spontaneous abortion (gestational weeks 7 through 12, with exposure time-window in the same period and weeks 13 through 22, with exposure time window from week 7 to 22). In post hoc sensitivity analyses, estimates of spontaneous abortion were adjusted for filled prescriptions of different anti-infective agents during pregnancy, antihypertensive drugs before pregnancy onset, and hospital diagnosis of infections during pregnancy (eMethods in the Supplement).

We also analyzed the association between oral itraconazole exposure and spontaneous abortion to investigate a class effect (eMethods in the Supplement). Itraconazole is an azole antifungal agent also used for recurrent vaginal candidiasis but not recommended during pregnancy because of higher treatment doses.

Results

A total of 1 405 663 pregnancies were included in the study cohort (**Figure 1**). Compared with unexposed unmatched pregnant women, fluconazole-exposed pregnant women differed in a number of characteristics. However, these differences were eliminated by the matching procedure, in which controls were identified for all but 5 fluconazole-exposed women (0.09%) (**Table 1** and **Table 2**; eTable 2 in the Supplement).

Spontaneous Abortion

One hundred forty-seven spontaneous abortions occurred in the 3315 pregnancies exposed to fluconazole in weeks 7 through 22, and 563 spontaneous abortions occurred in the 13 246 unexposed matched control pregnancies. Fluconazole exposure, compared with nonexposure among matched control pregnancies, was associated with a significantly increased risk of spontaneous abortion (HR, 1.48; 95% CI, 1.23-1.77) (**Table 3**). A similar risk was observed comparing with unexposed unmatched pregnancies (HR, 1.49; 95% CI, 1.27-1.75). The mean gestational age of spontaneous abortion was 77 days for for fluconazole-exposed women and 76 days for unexposed matched control women.

Stillbirth

Twenty-one stillbirths occurred in the 5382 pregnancies exposed to fluconazole from week 7 to birth, and 77 stillbirths occurred in the 21 506 unexposed matched pregnancies. The HR for stillbirth in fluconazole-exposed pregnancies, compared with unexposed matched control pregnancies, was 1.32 (95% CI, 0.82-2.14) (Table 3). Compared with unexposed unmatched pregnancies, the HR was 1.44 (95% CI, 0.94-2.21).

Sensitivity Analyses

For spontaneous abortion, the HRs associated with fluconazole doses of 150 mg to 300 mg and 350 mg to 5600 mg were 1.47 (95% CI, 1.22-1.77) and 1.55 (95% CI, 0.94-2.58),

Table 1. Demographic Characteristics of Fluconazole-Exposed and Unexposed Pregnancies Before and After Matching, 1997-2013[a]

Characteristic	Exposure Time Gestational Week 7 to 22, No. (%)[b]				Exposure Time Gestational Week 7 to Birth, No. (%)[c]			
	Before Matching		After Matching		Before Matching		After Matching	
	Fluconazole-Exposed Pregnancies (n = 3319)	Unexposed Unmatched Pregnancies (n = 1 402 181)	Fluconazole-Exposed Pregnancies (n = 3315)	Unexposed Matched Control Pregnancies (n = 13 246)	Fluconazole-Exposed Pregnancies (n = 5387)	Unexposed Unmatched Pregnancies (n = 1 400 113)	Fluconazole-Exposed Pregnancies (n = 5382)	Unexposed Matched Control Pregnancies (n = 21 506)
Age at pregnancy onset, y								
≤24	874 (26.3)	280 954 (20.0)	873 (26.3)	3490 (26.3)	1258 (23.4)	280 570 (20.0)	1257 (23.4)	5017 (23.3)
25-29	1011 (30.5)	458 344 (32.7)	1011 (30.5)	4041 (30.5)	1704 (31.6)	457 651 (32.7)	1704 (31.7)	6816 (31.7)
30-34	908 (27.4)	427 281 (30.5)	907 (27.4)	3627 (27.4)	1542 (28.6)	426 647 (30.5)	1541 (28.6)	6163 (28.7)
35-39	440 (13.3)	192 078 (13.7)	439 (13.2)	1753 (13.2)	754 (14.0)	191 764 (13.7)	752 (14.0)	3004 (14.0)
≥40	86 (2.6)	43 524 (3.1)	85 (2.6)	335 (2.5)	129 (2.4)	43 481 (3.1)	128 (2.4)	506 (2.4)
Place of birth								
Denmark	2840 (85.6)	1 182 979 (84.4)	2838 (85.6)	11 491 (86.8)	4581 (85.0)	1 181 238 (84.4)	4579 (85.1)	18 506 (86.1)
Europe	194 (5.8)	94 548 (6.7)	193 (5.8)	682 (5.1)	358 (6.6)	94 384 (6.7)	356 (6.6)	1309 (6.1)
Rest of the world	285 (8.6)	124 654 (8.9)	284 (8.6)	1073 (8.1)	448 (8.3)	124 491 (8.9)	447 (8.3)	1691 (7.9)
County of residence								
Copenhagen	1106 (33.3)	515 493 (36.8)	1105 (33.3)	4536 (34.2)	1800 (33.4)	514 799 (36.8)	1800 (33.4)	7264 (33.8)
Sealand	467 (14.1)	173 967 (12.4)	466 (14.1)	1 758 (13.3)	754 (14.0)	173 680 (12.4)	751 (14.0)	2922 (13.6)
Central Jutland	709 (21.4)	307 882 (22.0)	708 (21.4)	2 707 (20.4)	1194 (22.2)	307 397 (22.0)	1194 (22.2)	4656 (21.6)
North Jutland	324 (9.8)	132 654 (9.5)	324 (9.8)	1306 (9.9)	510 (9.5)	132 468 (9.5)	509 (9.5)	1965 (9.1)
South of Denmark	713 (21.5)	272 185 (19.4)	712 (21.5)	2939 (22.2)	1129 (21.0)	271 769 (19.4)	1128 (21.0)	4699 (21.8)
Married or living together	997 (30.0)	329 511 (23.5)	994 (30.0)	3829 (28.9)	1471 (27.3)	329 037 (23.5)	1468 (27.3)	5747 (26.7)
Level of education								
Primary school	1049 (31.6)	358 677 (25.6)	1047 (31.6)	3975 (30.0)	1558 (28.9)	358 168 (25.6)	1556 (28.9)	5745 (26.7)
Secondary school	421 (12.7)	182 438 (13.0)	421 (12.7)	1573 (11.9)	648 (12.0)	182 211 (13.0)	648 (12.0)	2486 (11.6)
Vocational/short tertiary	1133 (34.1)	479 607 (34.2)	1133 (34.2)	4670 (35.3)	1842 (34.2)	478 898 (34.2)	1842 (34.2)	7339 (34.1)
Medium/long tertiary	716 (21.6)	381 459 (27.2)	714 (21.5)	3028 (22.9)	1339 (24.9)	380 836 (27.2)	1336 (24.8)	5936 (27.6)
Household income, yearly quintiles								
First	805 (24.3)	280 289 (20.0)	804 (24.3)	3235 (24.4)	1260 (23.4)	279 834 (20.0)	1258 (23.4)	5007 (23.3)
Second	769 (23.2)	280 320 (20.0)	768 (23.2)	2973 (22.4)	1132 (21.0)	279 957 (20.0)	1130 (21.0)	4310 (20.0)
Third	564 (17.0)	280 537 (20.0)	563 (17.0)	2191 (16.5)	1007 (18.7)	280 094 (20.0)	1006 (18.7)	4026 (18.7)
Fourth	580 (17.5)	280 532 (20.0)	580 (17.5)	2284 (17.2)	978 (18.2)	280 134 (20.0)	978 (18.2)	3879 (18.0)
Fifth	601 (18.1)	280 503 (20.0)	600 (18.1)	2563 (19.3)	1010 (18.7)	280 094 (20.0)	1010 (18.8)	4284 (19.9)

[a] Percentages may not sum to 100 because of rounding.

[b] For analyses of spontaneous abortion.

[c] For analyses of stillbirth.

respectively (P = .84 for difference between HRs) (Table 3). For stillbirth, the HRs associated with low and high dose were 0.99 (95% CI, 0.56-1.74) and 4.10 (95% CI, 1.89-8.90), respectively; the HRs were significantly different (P = .002). Table 3 shows the other sensitivity analyses, including the HRs within 2 weeks after exposure vs later, and exposure in gestational week 7 to 10 vs later; none of the HRs were significantly different. In the analysis of the timing of the outcome, the HRs were 1.32 (95% CI, 1.06-1.65) for early spontaneous abortion and 1.90 (95% CI, 1.39-2.60) for late spontaneous abortion.

In the sensitivity analyses with different comparator groups, oral fluconazole-exposed pregnancies were at significantly increased risk of spontaneous abortion compared with topical azole-exposed pregnancies (130/2823 vs 118/2823, respectively; HR, 1.62 [95% CI, 1.26-2.07]) and compared with pivmecillinam-exposed pregnancies (140/3018 vs 143/3018, respectively; HR, 1.44 [95% CI, 1.14-1.82]) (**Figure 2**). The HRs for spontaneous abortion were attenuated and not significantly increased in the comparison of pregnancies exposed to fluconazole during pregnancy with pregnancies exposed to fluconazole in the year before pregnancy onset (92/2338 vs 107/2338, respectively; HR, 1.23 [95% CI, 0.93-1.62]) (Figure 2) and in the within-mother analysis (69 cases in 1727 fluconazole-exposed pregnancies and 671 cases in 7975 previously unexposed

Table 2. Clinical Characteristics of Fluconazole-Exposed and Unexposed Pregnancies Before and After Matching, 1997-2013[a]

Characteristic	Exposure Time Gestational Week 7 to 22, No. (%)[b]				Exposure Time Gestational Week 7 to Birth, No. (%)[c]			
	Before Matching		After Matching		Before Matching		After Matching	
	Fluconazole-Exposed Pregnancies (n = 3319)	Unexposed Unmatched Pregnancies (n = 1 402 181)	Fluconazole-Exposed Pregnancies (n = 3315)	Unexposed Matched Control Pregnancies (n = 13 246)	Fluconazole-Exposed Pregnancies (n = 5387)	Unexposed Unmatched Pregnancies (n = 1 400 113)	Fluconazole-Exposed Pregnancies (n = 5382)	Unexposed Matched Control Pregnancies (n = 21506)
Fluconazole Exposure								
Cumulative dose 150-300 mg	2847 (85.8)		2844 (85.8)		4616 (85.7)		4611 (85.7)	
Gestational days at first prescription of fluconazole, median (IQR)	69 (51-111)		69 (51-111)		121 (60-192)		121 (60-192)	
Calendar Year of Delivery or Pregnancy Loss								
1997-1999	516 (15.5)	262 882 (18.7)	516 (15.6)	1955 (14.8)	689 (12.8)	262 709 (18.8)	689 (12.8)	2639 (12.3)
2000-2002	448 (13.5)	257 562 (18.4)	447 (13.5)	1800 (13.6)	621 (11.5)	257 389 (18.4)	619 (11.5)	2504 (11.6)
2003-2005	452 (13.6)	252 451 (18.0)	450 (13.6)	1775 (13.4)	653 (12.1)	252 250 (18.0)	651 (12.1)	2563 (11.9)
2006-2008	576 (17.4)	250 684 (17.9)	576 (17.4)	2299 (17.4)	960 (17.8)	250 300 (17.9)	960 (17.8)	3801 (17.7)
2009-2011	764 (23.0)	236 079 (16.8)	764 (23.0)	3120 (23.6)	1356 (25.2)	235 487 (16.8)	1356 (25.2)	5499 (25.6)
2012-2013	563 (17.0)	142 523 (10.2)	562 (17.0)	2297 (17.3)	1108 (20.6)	141 978 (10.1)	1107 (20.6)	4500 (20.9)
Pregnancy History								
Parity								
0	1424 (42.9)	632 404 (45.1)	1422 (42.9)	5896 (44.5)	2229 (41.4)	631 599 (45.1)	2227 (41.4)	9374 (43.6)
1	1061 (32.0)	470 875 (33.6)	1061 (32.0)	4339 (32.8)	1874 (34.8)	470 062 (33.6)	1874 (34.8)	7727 (35.9)
2	585 (17.6)	218 173 (15.6)	585 (17.6)	2143 (16.2)	916 (17.0)	217 842 (15.6)	915 (17.0)	3271 (15.2)
≥3	249 (7.5)	80 729 (5.8)	247 (7.5)	868 (6.6)	368 (6.8)	80 610 (5.8)	366 (6.8)	1134 (5.3)
History of spontaneous abortion	544 (16.4)	237 631 (16.9)	544 (16.4)	1908 (14.4)	889 (16.5)	237 286 (16.9)	889 (16.5)	3270 (15.2)
History of stillbirth	15 (0.5)	9069 (0.6)	15 (0.5)	41 (0.3)	25 (0.5)	9059 (0.6)	25 (0.5)	92 (0.4)
History of ectopic pregnancies	121 (3.6)	49 494 (3.5)	121 (3.7)	384 (2.9)	213 (4.0)	49 402 (3.5)	213 (4.0)	704 (3.3)
History of induced abortion	882 (26.6)	309 543 (22.1)	880 (26.5)	3134 (23.7)	1361 (25.3)	309 064 (22.1)	1357 (25.2)	4697 (21.8)
Medical History								
Diabetes mellitus	97 (2.9)	25 284 (1.8)	96 (2.9)	449 (3.4)	160 (3.0)	25 221 (1.8)	158 (2.9)	726 (3.4)
HIV infection	5 (0.2)	653 (<0.1)	4 (0.1)	17 (0.1)	8 (0.1)	650 (<0.1)	7 (0.1)	23 (0.1)
Immunodeficiency	7 (0.2)	801 (0.1)	6 (0.2)	30 (0.2)	8 (0.1)	800 (0.1)	8 (0.1)	35 (0.2)
Hospital admissions in past y, no.								
0	2455 (74.0)	1 117 211 (79.7)	2453 (74.0)	10 172 (76.8)	4057 (75.3)	1 115 609 (79.7)	4053 (75.3)	16 810 (78.2)
1-2	305 (9.2)	112 555 (8.0)	305 (9.2)	1053 (7.9)	473 (8.8)	112 387 (8.0)	473 (8.8)	1537 (7.1)
≥ 3	559 (16.8)	172 415 (12.3)	557 (16.8)	2021 (15.3)	857 (15.9)	172 117 (12.3)	856 (15.9)	3159 (14.7)
No. of outpatient hospital contacts in past y								
0	2054 (61.9)	972 910 (69.4)	2054 (62.0)	8283 (62.5)	3325 (61.7)	971 639 (69.4)	3324 (61.8)	13 398 (62.3)
1-2	533 (16.1)	195 988 (14.0)	530 (16.0)	1942 (14.7)	856 (15.9)	195 665 (14.0)	854 (15.9)	3112 (14.5)
≥3	732 (22.1)	233 283 (16.6)	731 (22.1)	3021 (22.8)	1206 (22.4)	232 809 (16.6)	1204 (22.4)	4996 (23.2)
No. of filled prescriptions in past 6 mo								
0	693 (20.9)	472 197 (33.7)	693 (20.9)	2694 (20.3)	1197 (22.2)	471 693 (33.7)	1197 (22.2)	4661 (21.7)
1-2	1429 (43.1)	617 080 (44.0)	1429 (43.1)	5653 (42.7)	616 257 (44.0)	2252 (41.8)	2252 (41.8)	8945 (41.6)
3-4	737 (22.2)	216 312 (15.4)	736 (22.2)	3050 (23.0)	215 873 (15.4)	1176 (21.8)	1176 (21.9)	4717 (21.9)
≥5	460 (13.9)	96 592 (6.9)	457 (13.8)	1849 (14.0)	96 290 (6.9)	762 (14.1)	757 (14.1)	3183 (14.8)

Abbreviation: IQR, interquartile range.

[a] Percentages may not sum to 100 because of rounding.

[b] For the analyses of spontaneous abortion.

[c] For the analyses of stillbirth.

Table 3. Primary Analysis and Sensitivity Analyses of Fluconazole Exposure in Pregnancy and Risk of Spontaneous Abortion and Stillbirth in a Matched Cohort

Analysis	No. of Events/ Total No. of Pregnancies	HR (95% CI)	*P* Value for Equal HRs[a]
Primary Analysis			
Spontaneous abortion[b]			
Fluconazole exposed	147/3315	1.48 (1.23-1.77)	
Unexposed matched control pregnancies	563/13 246	1 [Reference]	
Stillbirth			
Fluconazole-exposed	21/5382	1.32 (0.82-2.14)	
Unexposed matched control pregnancies	77/21 506	1 [Reference]	
Fluconazole Dose[c]			
Spontaneous abortion			
150-300 mg	132/2986	1.47 (1.22-1.77)	
350-5600 mg	15/345	1.55 (0.94-2.58)	.84
Unexposed matched control pregnancies	563/13 246	1 [Reference]	
Stillbirth			
150-300 mg	14/4831	0.99 (0.56-1.74)	
350-5600 mg	7/597	4.10 (1.89-8.90)	.002
Unexposed matched control pregnancies	77/21 506	1 [Reference]	
Time Since Last Fluconazole Exposure			
Spontaneous abortion			
Within 2 wk	70/3315	1.32 (1.03-1.69)	
After 2 wk	77/2256	1.65 (1.30-2.10)	.17
Unexposed matched control pregnancies	563/13 246	1 [Reference]	
Stillbirth			
Within 2 wk	1/5382	0,78 (0.11-5.55)	
After 2 wk	20/4431	1.37 (0.84-2.24)	.58
Unexposed matched control pregnancies	77/21 506	1 [Reference]	
Timing of Fluconazole Exposure in Pregnancy			
Spontaneous abortion			
Gestational wk 7-10	122/1665	1.45 (1.19-1.77)	
Gestational wk 11-22	25/1650	1.61 (1.08-2.41)	.64
Unexposed matched control pregnancies	563/13 246	1 [Reference]	
Stillbirth			
Gestational wk 7-10	4/1665	1.40 (0.51-3.83)	
Gestational wk 11-birth	17/3717	1.30 (0.77-2.21)	.90
Unexposed matched control pregnancies	77/21 506	1 [Reference]	
Timing of Spontaneous Abortion[d]			
Gestational wk 7-12			
Fluconazole-exposed pregnancies	97/1965	1.32 (1.06-1.65)	
Unexposed matched control pregnancies	398/7850	1 [Reference]	
Gestational wk 13-22			
Fluconazole-exposed pregnancies	50/2223	1.90 (1.39-2.60)	
Unexposed matched control pregnancies	165/11 483	1 [Reference]	

Abbreviation: HR, hazard ratio.

[a] The difference in the HRs in the sensitivity analyses was assessed using a Wald test for the interaction between treatment status and subcategory of exposure.

[b] With adjustment for potential confounders (including use of oral or topical metronidazole or topical clindamycin, use of any systemic antibiotics, inpatient or outpatient diagnosis of infection, use of pivmecillinam or sulfamethizole, all in pregnancy, or use of antihypertensives before pregnancy onset), the hazard ratio was 1.48, with 95% CIs of 1.23 or 1.24 to 1.77 or 1.78.

[c] Number of exposed pregnancies in the 2 dose categories may exceed the number of exposed pregnancies in the main analysis because a pregnancy can be exposed to more than 1 dose.

[d] Only for spontaneous abortion for which the follow-up period (week 7-22) is divided into 2 different strata. Exposed pregnancies can contribute to both analyses (spontaneous abortion in gestational weeks 7-12 and in gestational weeks 13-22); therefore, these numbers exceed the total number of exposed pregnancies in the main analysis.

pregnancies; adjusted HR, 1.34 [95% CI, 0.94-1.92]). In post hoc sensitivity analyses, additional adjustment for potential confounders had no effect on the HRs for spontaneous abortion (Table 3).

In itraconazole-exposed pregnancies, 7 of 131 women had spontaneous abortions, vs 34 of 524 in the matched control pregnancies (HR, 1.16 [95% CI, 0.51-2.60]) (eTable 3 in the Supplement).

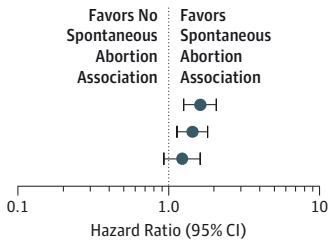
Discussion

In this nationwide cohort in Denmark, oral fluconazole use in pregnancy was associated with a significantly increased risk of spontaneous abortion. The risk of stillbirth was not significantly increased, but this outcome was relatively rare and the results were therefore imprecise.

Figure 2. Sensitivity Analyses of the Association Between Fluconazole-Exposed Pregnancies and the Risk of Spontaneous Abortion and Stillbirth Using Matched Control Pregnancies Exposed to Anti-infective Agents

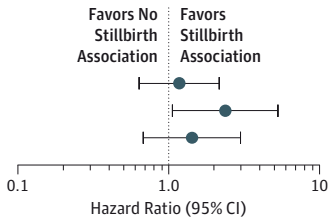
A Spontaneous abortion

	Fluconazole-Exposed (Gestational Week 7-22)		Matched Control Pregnancies		
	Spontaneous Abortion, No.	Total No.of Pregnancies	Spontaneous Abortion, No.	Total No.of Pregnancies	Hazard Ratio (95% CI)
Fluconazole vs topical azole antifungals	130	2823	118	2823	1.62 (1.26-2.07)
Fluconazole vs pivmecillinam	140	3018	143	3018	1.44 (1.14-1.82)
Fluconazole vs fluconazole in the year before pregnancy onset	92	2338	107	2338	1.23 (0.93-1.62)



B Stillbirth

	Fluconazole-Exposed (Gestational Week 7-Birth)		Matched Control Pregnancies		
	Stillbirth, No.	Total No.of Pregnancies	Stillbirth, No.	Total No.of Pregnancies	Hazard Ratio (95% CI)
Fluconazole vs topical azole antifungals	20	4301	22	4301	1.18 (0.64-2.16)
Fluconazole vs pivmecillinam	17	4357	9	4357	2.38 (1.06-5.32)
Fluconazole vs fluconazole in the year before pregnancy onset	15	3853	13	3853	1.43 (0.68-3.00)



Abortions have been observed in animals exposed to fluconazole, but at doses many times higher than those recommended for humans; thus, this effect may be attributable to toxic overdosing.[28] Fluconazole works by inhibiting the fungal CYP51 enzyme, essential for ergosterol synthesis in the cell membrane.[28] Fluconazole can (while much less potently) interfere with human CYP450 enzymes, which are also expressed during in utero development, and a potential adverse drug effect could operate through this mechanism of action.[29] Two previous epidemiologic studies have investigated the prevalence of spontaneous abortion and stillbirth associated with fluconazole. One study found no significantly increased risk of spontaneous abortion (odds ratio [OR], 1.21 [95% CI, 0.67-2.21]) or stillbirth (OR, 0.36 [95% CI, 0.03-3.90]) in 226 first-trimester exposed pregnancies.[11] Another study found no increased risk of stillbirth among 1286 first-trimester exposed pregnancies (OR, 1.10 [95% CI, 0.40-3.50]); spontaneous abortion was not investigated.[13] In comparison, our study included 5382 women exposed to fluconazole during pregnancy, which adds substantially to the existing safety data. Furthermore, our study design implemented survival analyses that took gestational age into account, which is essential when investigating spontaneous abortion.[30]

To address confounding by vaginal candidiasis in a sensitivity analysis, we used an active comparator group of pregnancies exposed to intravaginal formulations of azole antifungals used for the same indication. This analysis did not support confounding by indication as an explanation for the results observed in the main analysis of spontaneous abortion. However, confounding by severity of vaginal candidiasis might be present, since pregnant women treated with oral fluconazole may have more severe infection. It has been described that candida can ascend from the vagina and cause intrauterine candidiasis, which may result in prematurity and fetal loss.[31-35] The incidence of diagnosed intrauterine candida infection is unknown, but there are fewer than 100 cases reported in the literature; this is therefore unlikely to explain the association observed in our study. However, it remains a possibility that severe vaginal candida infection alone could result in prematurity or pregnancy loss, as has been proposed for bacterial vaginosis.[36] On the other hand, many patients with vaginal candidiasis prefer the convenience of oral fluconazole with no local adverse effects,[4] and the use of oral fluconazole vs topical azoles may therefore reflect not only severity of infection but also personal preferences. Late pregnancy losses have been associated with bacterial vaginosis,[37] intrauterine infections, or severe acute or chronic maternal illness (hypertension and diabetes).[38] However, when we adjusted our analyses for use of different anti-infective agents, diagnosis of infections (in hospitals), or antihypertensives, the estimates were similar to those in the main analysis of spontaneous abortion. This suggests that co-occurrence of discovered infections or treated hypertension among fluconazole-exposed women does not explain the observed increased risk of spontaneous abortion.

In the analysis comparing fluconazole exposure during pregnancy with pregnancies exposed to fluconazole in the year before pregnancy and in the within-mother analysis, the HRs for spontaneous abortion were somewhat attenuated and, while greater than 1.0, not statistically significant. These results could indicate that residual confounding from either family or lifestyle factors (such as smoking or alcohol consumption) or hereditary predisposition may explain the increased risk observed in the main analysis of spontaneous abortion. However, both these analyses were based on a lower number of exposed cases than the main analysis, so the results could also reflect statistical imprecision.

In women using high doses of fluconazole (350-5600 mg), we found no further increased risk of spontaneous abortion

(in fact, the association was not statistically significant) but a significantly elevated risk of stillbirth. However, the high-dose category was heterogeneous and the analyses were based on few exposed cases (n = 15 spontaneous abortion and n = 7 stillbirth exposed to high doses) and hence should be interpreted with caution. Analyses of potential mechanisms did not suggest that our results were explained by an acute toxic effect or implicate fatal teratogenesis. The small number of pregnancies exposed to itraconazole precludes comment on whether the association may be related to a class effect.

Our study has a number of strengths. The registry-based design allowed assembly of a nationwide cohort of all registered births and abortions for a 17-year period, with independent ascertainment of exposure and outcome. We used filled prescriptions for exposure ascertainment, and nondifferential nonadherence would bias our results toward no effect of the drug. However, because fluconazole is primarily prescribed as a single dose of 150 mg or 2 doses of 150-mg therapy, we would expect a high degree of medication adherence compared with long-term therapy. Validation studies have reported a near-complete (>99.5%) recording of births in the Medical Birth Registry[23] and a 99% positive predictive value for diagnoses of spontaneous abortions recorded in the National Patient Register.[39]

However, the study also has limitations. The National Patient Register, like any other large database of hospital care, is unlikely to capture very early spontaneous abortions, many of which are not recognized clinically.[40] To minimize any influence of this fact on the investigated association, we used a study design in which ascertainment of exposure and outcomes started in gestational week 7; hence, our results are not generalizable to fluconazole exposure or spontaneous abortion outcomes occurring in weeks 0 through 6. Given that Denmark has free and universal health care, we expect that almost all spontaneous abortions occurring after gestational week 7 are managed by hospital specialists. However, in the case of unrecognized early pregnancy, spontaneous abortion may be mistaken for a late menstrual period. If pregnancies were on average recognized earlier in fluconazole-exposed women than in unexposed women, then bias toward increased risk would be introduced, given the relatively higher risk of spontaneous abortion in early pregnancy. Likewise in cases of recognized pregnancy, if fluconazole-exposed women experiencing spontaneous abortion more often sought medical attention than unexposed women, then spontaneous abortion would be more likely to be diagnosed among these women, which would again bias the results toward an increased risk with fluconazole exposure. However, we believe that differential misclassification of outcome in fluconazole-exposed women vs unexposed women is unlikely, especially given that our analyses using active drug users as comparators (topical azole and pivmecillinam) were consistent with the main analysis of spontaneous abortion.

Conclusions

In this nationwide cohort study in Denmark, use of oral fluconazole in pregnancy was associated with a statistically significant increased risk of spontaneous abortion compared with risk among unexposed women and women with topical azole exposure in pregnancy. Until more data on the association are available, cautious prescribing of oral fluconazole in pregnancy may be advisable. Although the risk of stillbirth was not significantly increased, this outcome should be investigated further.

ARTICLE INFORMATION

Author Contributions: Dr Mølgaard-Nielsen had full access to all of the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.
Study concept and design: All authors.
Acquisition, analysis, or interpretation of data: All authors.
Drafting of the manuscript: Mølgaard-Nielsen.
Critical revision of the manuscript for important intellectual content: Svanstrom, Melbye, Hviid, Pasternak.
Statistical analysis: Mølgaard-Nielsen.
Obtained funding: Mølgaard-Nielsen, Pasternak.
Study supervision: Melbye, Hviid, Pasternak.

Conflict of Interest Disclosures: All authors have completed and submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest and none were reported.

Funding/Support: This study was supported by the Danish Medical Research Council.

Role of the Funder/Sponsor: The Danish Medical Research Council had no role in the design and conduct of the study; collection, management, analysis, and interpretation of the data; preparation, review, or approval of the manuscript; and decision to submit the manuscript for publication.

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