**Supplementary Table 1. Folic acid antagonist exposures by case and control group, Slone Birth Defects Study, 1998-2015**

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
|  | **Periconceptional exposure** | |  | **First trimester exposure** | | | |
| **Drug** | **Controls**†  **N=10194** | **NTD Cases**  **N=402** |  | **Controls**†  **N=10205** | **Oral clefts‡**  **N=1284** | **Urinary tract defects**§  **N=2472** | **Cardiac defects¶**  **N=6095** |
| Unexposed during pregnancy | 10126 (99.3%) | 395 (98.3%) |  | 10126 (99.2%) | 1266 (98.6%) | 2441 (98.7%) | 6036 (99.0%) |
| *DHFR Inhibitor* | 16 (0.2%) | 0 (0.0%) |  | 26 (0.3%) | 3 (0.2%) | 9 (0.4%) | 10 (0.2%) |
| Trimethoprim | 10 (0.1%) | -- |  | 21 (0.2%) | 2 (0.2%) | 4 (0.2%) | 9 (0.1%) |
| Triamterene | 4 (<0.1%) | -- |  | 4 (<0.1%) | 1 (0.1%) | 3 (0.1%) | -- |
| Sulfasalazine | 1 (<0.1%) | -- |  | 1 (<0.1%) | -- | 1 (<0.1%) | -- |
| Methotrexate | -- | -- |  | -- | -- | 1 (<0.1%) | 1 (<0.1%) |
| Proguanil | 1 (<0.1%) | -- |  | -- | -- | -- | -- |
|  |  |  |  |  |  |  |  |
| *Any AED* | 53 (0.5%) | 7 (1.7%) |  | 53 (0.5%) | 15 (1.2%) | 21 (0.9%) | 49 (0.8%) |
| Older AED | 22 (0.2%) | 3 (0.8%) |  | 22 (0.2%) | 5 (0.4%) | 8 (0.3%) | 19 (0.3%) |
| Valproic Acid | 8 (0.1%) | 2 (0.5%) |  | 7 (0.1%) | 3 (0.2%) | 5 (0.2%) | 6 (0.1%) |
| Carbamazepine | 5 (<0.1%) | 1 (0.3%) |  | 5 (<0.1%) | 1 (0.1%) | 3 (0.1%) | 5 (0.1%) |
| Phenytoin | 5 (<0.1%) | -- |  | 6 (0.1%) | 1 (0.1%) | -- | 5 (0.1%) |
| Primidone | 2 (<0.1%) | -- |  | 2 (<0.1%) | -- | -- | -- |
| Phenobarbital | 2 (<0.1%) | -- |  | 2 (<0.1%) | -- | 1 (<0.1%) | 4 (0.1%) |
|  |  |  |  |  |  |  |  |
| Newer AED | 34 (0.3%) | 4 (1.0%) |  | 34 (0.3%) | 12 (0.9%) | 15 (0.6%) | 35 (0.6%) |
| Topiramate | 11 (0.1%) | 2 (0.5%) |  | 11 (0.1%) | 8 (0.6%) | 4 (0.2%) | 11 (0.2%) |
| Lamotrigine | 8 (0.1%) | 1 (0.3%) |  | 7 (0.1%) | 1 (0.1%) | 2 (0.1%) | 14 (0.2%) |
| Gabapentin | 9 (0.1%) | 1 (0.3%) |  | 10 (0.1%) | 2 (0.2%) | 5 (0.2%) | 5 (0.1%) |
| Levetiracetam | 7 (0.1%) | -- |  | 7 (0.1%) | 2 (0.2%) | 3 (0.1%) | 1 (<0.1%) |
| Oxcarbazepine | 1 (<0.1%) | -- |  | 1 (<0.1%) | -- | 1 (<0.1%) | -- |
| Pregabalin | 1 (<0.1%) | -- |  | 1 (<0.1%) | -- | 1 (<0.1%) | 2 (<0.1%) |
| Zonisamide | 1 (<0.1%) | -- |  | 1 (<0.1%) | 1 (0.1%) | -- | -- |
| AED NOS | -- | -- |  | -- | -- | -- | 2 (<0.1%) |

DHFR=dihydrofolate reductase inhibitor, AED=anti-epileptic drug, NOS=not otherwise specified, periconceptional=one lunar month pre-LMP through first lunar month).

†Note: Among controls, one mother was exposed to both a DHFR inhibitor and an AED: (triamterene and phenobarbital).

Seven controls were exposed to more than one AED: Three controls were exposed to both an older and a newer AED: (valproic acid and levetiracetam), (carbamazepine and levetiracetam), (primidone and lamotrigine); Four controls were exposed to two newer AEDs in the peri period: two to (topiramate and levetiracetam), (topiramate and gabapentin), (gabapentin and lamotrigine) – This group of multiply exposed controls is identical for both the periconceptional and first trimester exposure windows

‡Note: Among oral cleft cases, two cases were exposed to more than one AED: one case mother was exposed to four AEDs in T1, one older and three newer AEDs: (valproic acid, topiramate, levetiracetam and zonisamide), another case was exposed to both an older and a newer AED: (phenytoin and topiramate)

§ Note: Among urinary tract defect cases, one mother was exposed to both a DHFR inhibitor and an AED in T1: (triamterene and valproic acid), two cases were exposed to both an older and a newer AED (valproic acid and topiramate ), (valproic acid and gabapentin), one case was exposed to two older AEDs in T1: (valproic acid and carbamazepine), one case was exposed to two newer AEDs in T1: (levetiracetam and oxcarbazepine)

**¶** Note: Among cardiac cases, one mother was exposed to both a DHFR inhibitor and an AED in T1: (trimethoprim and anticonvulsant NOS), 5 were exposed to both an older and a newer AED: (phenytoin and topiramate)**,** (valproic acid and topiramate), (phenytoin and gabapentin), (phenytoin and lamotrigine) (carbamazepine and lamotrigine); one case was exposed to two older AEDs in T1: (carbamazepine and phenobarbital)

**Supplementary Table 2. Association between folic acid antagonists and malformation subgroups, Slone Birth Defect Study, 1998-2015**

|  |  | **DHFR Inhibitors** | | | **AED** | | |
| --- | --- | --- | --- | --- | --- | --- | --- |
| **Outcome** | **Unexposed** | **Any DHFR Inhibitor** | **Crude OR**  **(95% CI)** | **Adj. OR**  **(95% CI)†** | **Any AED** | **Crude OR**  **(95% CI)** | **Adj. OR**  **(95% CI)†** |
| **Controls (N=10194)** | 10126 (99.3%) | 16 (0.2%) | REF | REF | 53 (0.5%) | REF | REF |
| **Any NTD (N=402)** | 395 (98.3%) | 0 (0.0%) | --- | --- | 7 (1.7%) | 3.4(1.5,7.5) | 2.8(1.2,6.2)‡ |
|  |  |  |  |  |  |  |  |
| **Oral Clefts** (N=1284) | 1266 (98.6%) | 3 (0.2%) | 0.9(0.2,3.0) | --- | 15 (1.2%) | 2.3(1.3,4.0) | 1.9(1.1,3.5) |
| Isolated Oral Clefts (N=1086) | 1070 (98.5%) | 2 (0.2%) | 0.7(0.1,2.9) | --- | 14 (1.3%) | 2.5(1.4,4.5) | 2.1(1.1,3.9) |
|  |  |  |  |  |  |  |  |
| **Urinary Defects** (N=2472) | 2441 (98.7%) | 9 (0.4%) | 1.4(0.7,3.1) | 1.5(0.7,3.2) | 21 (0.9%) | 1.6(1.0,2.7) | 1.5(0.9,2.5) |
| Isolated Urinary Defect (N=1598) | 1581 (98.9%) | 6 (0.4%) | 1.5(0.6,3.6) | 1.5(0.6,3.8) | 11 (0.7%) | 1.3(0.7,2.6) | 1.3(0.7,2.5) |
| Non-Obstructive Urinary Defect (N=1526) | 1504 (98.6%) | 7 (0.5%) | 1.8(0.8,4.2) | 1.8(0.8,4.3) | 15 (1.0%) | 1.9(1.1,3.4) | 1.8(1.0,3.2) |
| Isolated Non-Obstructive Urinary Defect (N=934) | 920 (98.5%) | 6 (0.6%) | 2.5(1.0,6.2) | 2.7(1.1,6.5) | 9 (1.0%) | 1.9(0.9,3.8) | 1.8(0.9,3.7) |
|  |  |  |  |  |  |  |  |
| **Cardiac Defects** (N=6095) | 6036 (99.0%) | 10 (0.2%) | 0.6(0.3,1.3) | 0.6(0.3,1.3) | 49 (0.8%) | 1.6(1.1,2.3) | 1.4(0.9,2.1) |
| Isolated Cardiac Defect (N=4670) | 4626 (99.1%) | 8 (0.2%) | 0.7(0.3,1.5) | 0.7(0.3,1.5) | 37 (0.8%) | 1.5(1.0,2.3) | 1.4(0.9,2.1) |
|  |  |  |  |  |  |  |  |
| Conotruncal Defects (N=1090) | 1076 (98.7%) | 1 (0.1%) | -- | --- | 13 (1.2%) | 2.3(1.3,4.2) | 2.0(1.1,3.8) |
| Isolated Conotruncal Defect (N=859) | 849 (98.8%) | 0 (0.0%) | --- | --- | 10 (1.2%) | 2.3(1.1,4.4) | 2.0(1.0,4.0) |
| VSD (N=2888) | 2866 (99.2%) | 1 (0.0%) | -- | --- | 20 (0.7%) | 1.3(0.8,2.2) | 1.2(0.7,2.1) |
| Isolated VSD (N=2387) | 2370 (99.3%) | 1 (0.0%) | --- | --- | 16 (0.7%) | 1.3(0.7,2.3) | 1.2(0.7,2.2) |
| Other CHD (N=3528) | 3494 (99.0%) | 9 (0.3%) | 1.0(0.5,2.1) | 0.9(0.4,2.0) | 26 (0.7%) | 1.4(0.9,2.3) | 1.2(0.8,2.0) |
| Isolated Other CHD (N=2496) | 2472 (99.0%) | 7 (0.3%) | 1.1(0.5,2.5) | 1.0(0.4,2.3) | 18 (0.7%) | 1.4(0.8,2.4) | 1.2(0.7,2.1) |

† Adjusted for maternal education, pre-pregnancy BMI, first trimester infection, smoking, alcohol use and pregnancy intention

‡ Adjusted for maternal education, pre-pregnancy BMI, periconceptional infection, smoking and alcohol use

DHFR=dihydrofolate reductase inhibitor, AED=anti-epileptic drug, NTD=neural tube defect, VSD=ventricular septal defect, CHD=congenital heart defect, periconceptional=one lunar month pre-LMP through lunar month 1.

**Supplementary Table 3. Anti-epileptic drug exposure - stratified by indication for use, Slone Birth Defect Study, 1998-2015**

| **AED exposure** | **Controls** | **Any NTD** | **Crude OR**  **(95% CI)** | **Any Oral Cleft** | **Crude OR**  **(95% CI)** | **Any Urinary** | **Crude OR**  **(95% CI)** | **Any Cardiac** | **Crude OR**  **(95% CI)** |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Unexposed | 10126 (99.3%) | 395 (98.3%) | REF | 1266 (98.6%) | REF | 2441 (98.7%) | REF | 6036 (99.0%) | REF |
| Any AED | 53 (0.5%) | 7 (1.7%) | 3.4 (1.5,7.5) | 15 (1.2%) | 2.3 (1.3,4.0) | 21 (0.9%) | 1.6 (1.0,2.7) | 49 (0.8%) | 1.6 (1.1,2.3) |
| Epilepsy indication | 27 (0.3%) | 4 (1.0%) | 3.8 (1.0,11.0) | 7 (0.5%) | 2.1 (0.9,4.8) | 9 (0.4%) | 1.4 (0.6,2.9) | 22 (0.4%) | 1.4 (0.8,2.4) |
| Non-Epilepsy indication | 26 (0.3%) | 3 (0.7%) | 3.0 (0.6,9.7) | 8 (0.6%) | 2.5 (1.1,5.4) | 12 (0.5%) | 1.9 (1.0,3.8) | 27 (0.4%) | 1.7 (1.0,3.0) |