

Supplementary material

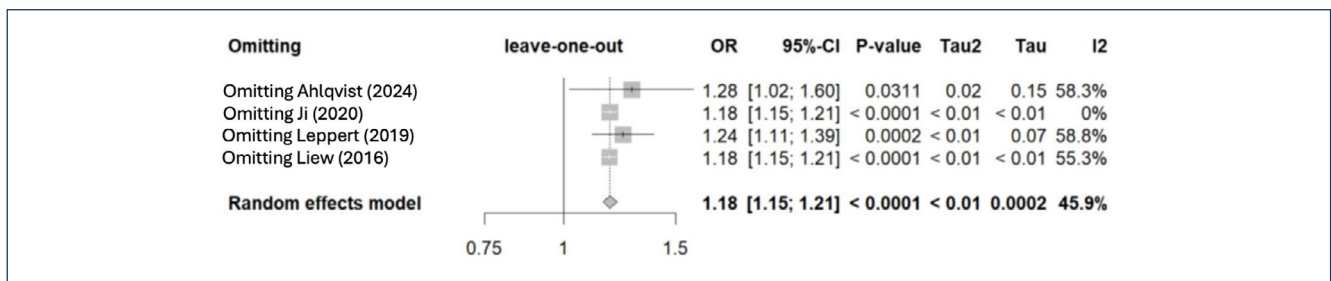


Figure S1. Leave-one-out analysis for the outcome of ASD diagnosis

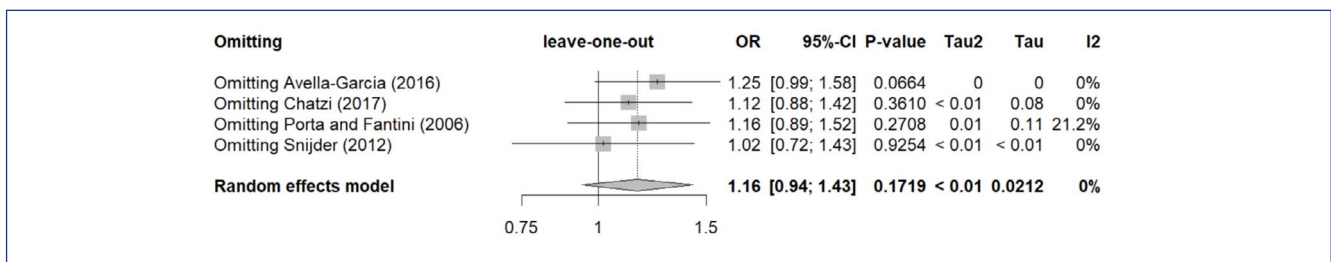


Figure S2. Leave-one-out analysis for the outcome of ASD symptoms

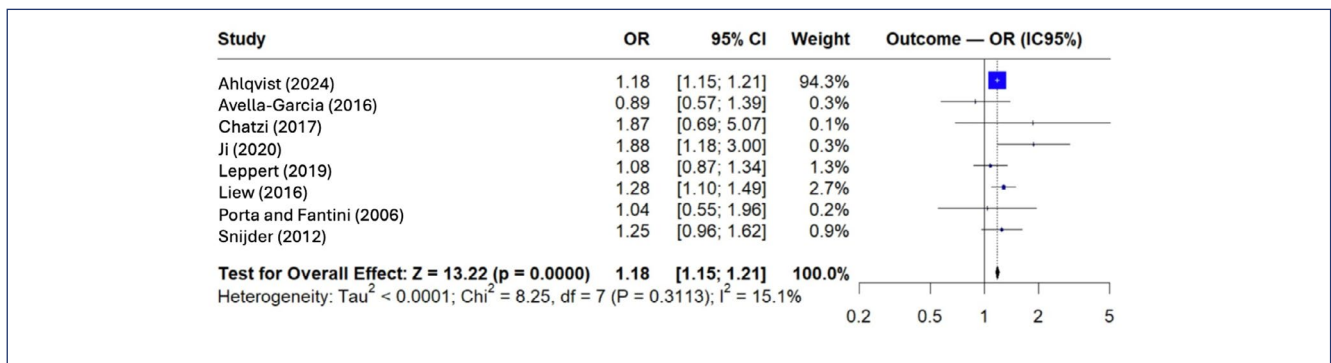


Figure S3. Pooled analysis of studies showed higher odds of autism disorder symptoms or diagnosis in the use of acetaminophen during pregnancy when compared with the not-exposed group

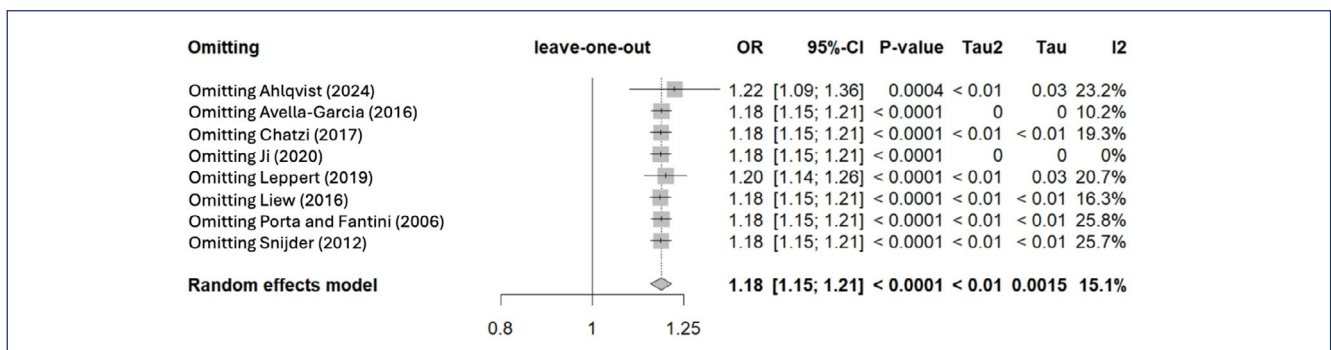


Figure S4. Leave-one-out analysis with the omission of individual studies showing low heterogeneity

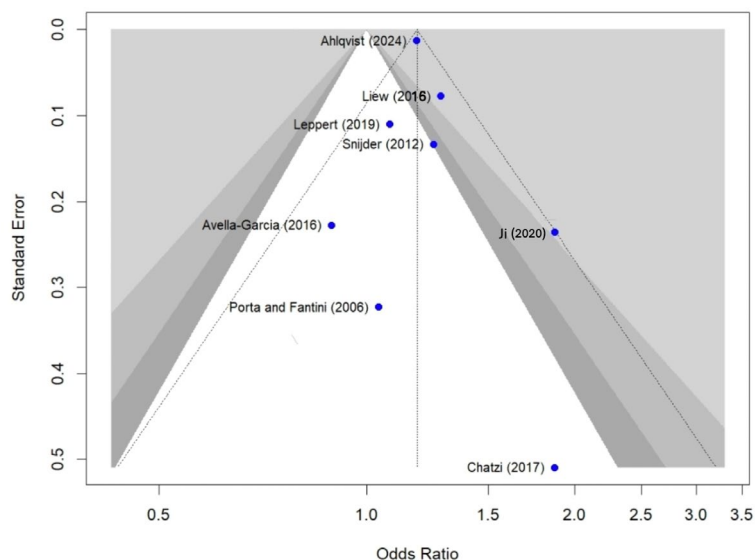


Figure S5. Funnel plot for the overall ASD identification (diagnosis and symptoms)

Table S1. Search strategy of the included studies

Database	Search strategy
Pubmed	["N-Acetyl-p-aminophenol" OR Acetamidophenol OR Acetaminophen OR Acetaminophen OR APAP OR "p-Acetamidophenol" OR "p-Hydroxyacetanilide" OR Paracetamol OR Hydroxyacetanilide OR "N-[4-Hydroxyphenyl]acetanilide" OR Algotropyl OR Acamol OR "Anacin-3" OR Anacin3 OR Anacin 3 OR Datril OR Panadol OR Tylenol OR Acetaco OR Acephen] AND ["Autism spectrum disorder" OR autistic OR autism OR ASD OR "asperger's syndrome" OR asperger OR "pervasive development disorders" OR PDD OR "childhood disintegrative disorder" OR "child development disorders, pervasive" OR "Pervasive Development Disorders" OR "kanner's syndrome" OR "kanners syndrome" OR "kanner syndrome"]
Embase and Cochrane	["N-Acetyl-p-aminophenol" OR Acetamidophenol OR Acetaminophen OR Acetaminophen OR APAP OR "p-Acetamidophenol" OR "p-Hydroxyacetanilide" OR Paracetamol OR Hydroxyacetanilide OR "N-[4-Hydroxyphenyl]acetanilide" OR Algotropyl OR Acamol OR "Anacin-3" OR Anacin3 OR Anacin 3 OR Datril OR Panadol OR Tylenol OR Acetaco OR Acephen] AND ["Autism spectrum disorder" OR autistic OR autism OR ASD OR "aspergers syndrome" OR asperger OR "pervasive development disorders" OR PDD OR "childhood disintegrative disorder" OR "child development disorders, pervasive" OR "Pervasive Development Disorders" OR "kanners syndrome" OR "kanner syndrome"]

Table S2. Summary of findings associated to autism and hyperkinetic disorder

Study (Author, Year)	Outcome	Exposure Level/Group	Cases (N) / Total (N)	Adjusted HR/OR (95% CI)
Liew (2016)	Autistic spectrum disorders (Overall)	Never used	401 / 358,257 person-years	1.00 (ref)
		Ever used	626 / 460,130 person-years	1.19 [1.04–1.35]
	Infantile autism (Overall)	Never used	139 / 355,987 person-years	1.00 (ref)
		Ever used	206 / 456,439 person-years	1.10 [0.89–1.37]
	Autism spectrum disorders with Hyperkinetic symptoms	Never used (ASD without HK)	295 cases [295 / cohort, ref for this specific analysis]	1.00 (ref)
		Ever used (ASD with HK)	412 cases [412 / cohort]	1.51 [1.19–1.92]
	Infantile autism with Hyperkinetic symptoms	Never used (Inf. Aut. without HK)	110 cases [110 / cohort]	1.00
		Ever used (Inf. Aut. with HK)	147 cases [147 / cohort]	1.55 [0.98–2.45]
	Asperger syndrome with Hyperkinetic symptoms	Maternal use (with HK)	Not specified in detail	Positively associated [Supporting Info Table S1]
	PDD-NOS with Hyperkinetic symptoms	Maternal use (with HK)	Not specified in detail	Positively associated [Supporting Info Table S1]
Ji (2020)	ADHD only	Cord unchanged acetaminophen (First tertile)	62 / 332	NA
		Cord unchanged acetaminophen (Second tertile)	75 / 332	2.26 [1.40–3.69]
		Cord unchanged acetaminophen (Third tertile)	120 / 332	2.86 [1.77–4.67]
	ASD only	Cord unchanged acetaminophen (First tertile)	12 / 332	NA
		Cord unchanged acetaminophen (Second tertile)	21 / 332	2.14 [0.93–5.13]
		Cord unchanged acetaminophen (Third tertile)	33 / 332	3.62 [1.62–8.60]
Ahlqvist et al. (2024)	Autism (Population-based model)	Ever-use vs. No use	5857 exposed cases / 185,909	HR 1.05 [1.02–1.08]
		Risk Difference at 10 years	NA	RD 0.09% [–0.01% to 0.20%]
	Autism (Sibling control model)	Ever-use vs. No use	3956 exposed cases / 1,773,747 [sibling cohort]	HR 0.98 [0.93–1.04]
		Risk Difference at 10 years	NA	RD 0.02% [–0.14% to 0.18%]
		Low dose (<166 mg/d) vs. No use	NA	HR 0.85 [0.68–1.08]
		High dose (≥430 mg/d) vs. No use	NA	HR 0.88 [0.68–1.14]

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Avella-Garcia et al. (2016)	CAST all children (ever-exposed vs. non-exposed)	CAST males (ever-exposed)	751 (total exposed males with data)	b 0.63 (0.09–1.18)
	CAST males (frequency of use)	CAST females (ever-exposed)	716 (total exposed females with data)	b -0.51 [-0.98–0.05]
Chatzi 2017 (RHEA Cohort from Alemany 2021)	Autistic Spectrum Symptoms (CBCL 6/18)	Sporadic use vs. Never use	272 (sporadic exposed males)	b 0.56 (0.001–1.11)
		Persistent use vs. Never use	21 (persistent exposed males)	b 1.91 (0.44–3.38)
Porta 2006 (GASPII Cohort from Alemany 2021)	Autistic Spectrum Symptoms (PDP subscale of CBCL1½–5)	Prenatal acetaminophen exposure	345 (cohort size)	OR ~1.06 (0.77–1.45) [Visual est. from Alemany Fig. 1A]
		Prenatal acetaminophen exposure	153 (cohort size)	OR ~1.15 (0.81–1.62) [Visual est. from Alemany Fig. 1A]

Note: For Liew et al. (2015), the "Total (N)" for "Cases (N) / Total (N)" refers to person-years for overall ASD/Infantile Autism, and case counts within the entire cohort for hyperkinetic subtypes where specific denominators for exposed/unexposed for that specific outcome were not provided. For Ji et al. (2019), the "Total (N)" represents the number of participants within that specific tertile or detection group for the cord biomarker. For Ahlqvist et al. (2024), "Cases (N) / Total Exposed (N)" reflects the number of observed neurodevelopmental cases within the exposed group (or sibling cohort for sibling analysis) as given in Figure 3 of the paper, not the total number of individuals in the exposed group. The primary finding from their sibling analysis is the adjusted HR. For Avella-Garcia et al. (2016), "Total Exposed (N)" refers to the "Nc" value, which is the number of subjects with acetaminophen exposure, neurodevelopment outcome, and potential confounders available for that specific analysis. "NA" denotes not applicable or not directly reported as a specific count in the original paper for that specific result type. For Chatzi 2017 (RHEA Cohort) and Porta 2006 (GASPII Cohort), the reported ORs are visual estimations from the forest plots in Figure 1A of Alemany et al. (2021) meta-analysis.

Table S3. Summary of findings of other neurodevelopmental and related outcomes.

Study (Author, Year)	Outcome	Exposure Level/Group	Cases (N) / Total (N)	Adjusted HR/OR (95% CI)
Leppert 2019	ADHD symptoms (observational association)	Acetaminophen use during pregnancy	Not specified / 7486 (N for observational analysis)	RR 1.45 (1.18–1.78)
	Maternal ADHD PRS associated with acetaminophen use in late pregnancy	1-SD increase in ADHD PRS	Not specified / 7921 (mothers)	OR 1.11 (1.04–1.18)
Snijder 2012	Cryptorchidism	No mild analgesic use during 14–22 weeks gestation	Not specified / cohort of 3184 boys	1.00 (ref)
		Mild analgesic use during 14–22 weeks gestation	17 / 480 (16.8% of 2864 women reported use)	2.12 (1.17–3.83)
	Cryptorchidism	No paracetamol use during 14–22 weeks gestation	Not specified / cohort of 3184 boys	1.00 (ref)
		Paracetamol use during 14–22 weeks gestation	15 / 448 (16.1% of 2864 women reported use)	1.89 (1.01–3.51)
Ahlqvist et al. (2024)	ADHD (Population-based model)	Mild analgesic use (any period)	7 / 22 cases with hypospadias	No significant association (due to small case number)
	ADHD (Sibling control model)	Ever-use vs. No use	12714 exposed cases / 185,909	HR 1.07 (1.05–1.10)
		Risk Difference at 10 years	NA	RD 0.21% (0.08%–0.34%)
Avella-Garcia et al. (2016)	Hyperactivity/Impulsivity symptoms (ADHD-DSM-IV)	Ever-use vs. No use	8526 exposed cases / 1,773,747	HR 0.98 (0.94–1.02)
		Risk Difference at 10 years	NA	RD -0.02% [-0.21% to 0.15%]
	Intellectual Disability (Sibling control model)	Ever-use vs. No use	1350 exposed cases / 1,773,747	HR 1.01 (0.92–1.10)
		Risk Difference at 10 years	NA	RD 0.00% [-0.10% to 0.13%]
Chatzi 2017 (RHEA Cohort from Alemany 2021)	Hyperactivity/Impulsivity symptoms (ADHD-DSM-IV)	Ever-exposed vs. Non-exposed	1382 (exposed with data)	IRR 1.41 (1.01–1.98)
	K-CPT commission errors	Ever-exposed vs. Non-exposed	1255 (exposed with data)	IRR 1.10 (1.03–1.17)
	K-CPT detectability	Ever-exposed vs. Non-exposed	1255 (exposed with data)	b -0.07 [-0.12–0.02]
	K-CPT omission errors	Persistent use vs. Never use	48 (persistent exposed with data)	IRR 1.29 (1.02–1.64)
	K-CPT commission errors (females)	Persistent use vs. Never use	27 (persistent exposed females)	IRR 1.32 (1.05–1.66)
	K-CPT detectability (females)	Persistent use vs. Never use	27 (persistent exposed females)	b -0.18 [-0.36–0.00]
	BSID (Mental and Psychomotor Development) at 1 year	Ever-exposed vs. Non-exposed	955 (exposed with data)	b 0.75 [-0.75–2.25]
	MCSA (Cognitive and Motor Development) at 5 years	Ever-exposed vs. Non-exposed	828 (exposed with data)	b -0.21 [-1.70–1.28]
	CPSCS (Social Competence) at 5 years	Ever-exposed vs. Non-exposed	828 (exposed with data)	b -1.15 [-3.16–0.87]
	ADHD Symptoms (CBCL 6/18)	Prenatal acetaminophen exposure	345 (cohort size)	OR ~1.15 (0.82–1.62) [Visual est. from Alemany Fig. 1B]
Porta 2006 (GASPII Cohort from Alemany 2021)	ADHD Symptoms (CBCL1½–5)	Prenatal acetaminophen exposure	153 (cohort size)	OR ~1.15 (0.79–1.69) [Visual est. from Alemany Fig. 1B]

Note: For Leppert et al. (2019) observational findings, specific case numbers for exposed/unexposed groups were not provided, only the relative risk for the outcome. For Snijder et al. (2012), the "Total (N)" for exposure groups represents the number of cases of cryptorchidism found among mothers who reported using mild analgesics during the specified period. For Ahlqvist et al. (2024), "Cases (N) / Total Exposed (N)" reflects the number of observed neurodevelopmental cases within the exposed group (or sibling cohort for sibling analysis), not the total number of individuals in the exposed group. The primary finding from their sibling analysis is the adjusted HR. For Avella-Garcia et al. (2016), "Total Exposed (N)" refers to the "Nc" value, which is the number of subjects with acetaminophen exposure, neurodevelopment outcome, and potential confounders available for that specific analysis. "NA" denotes not applicable or not directly reported as a specific count in the original paper for that specific result type. For Chatzi 2017 (RHEA Cohort) and Porta 2006 (GASPII Cohort), the reported ORs are visual estimations from the forest plots in Figure 1B of Alemany et al. (2021) meta-analysis.