



National Institute for Public Health
and the Environment
Ministry of Health, Welfare and Sport

An overview of the available data on the reproductive toxicity of molybdenum and its compounds

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RIVM letter report 2023-0450

Colophon

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DOI 10.21945/RIVM-2023-0450

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This investigation was performed by order, and for the account, of the Health Council of the Netherlands, within the framework of the project 'Literature reviews for use in advisory reports on substances'

Published by:
**National Institute for Public Health
and the Environment, RIVM**
P.O. Box 1 | 3720 BA Bilthoven
The Netherlands
www.rivm.nl/en

Synopsis

An overview of the available data on the reproductive toxicity of molybdenum and its compounds.

RIVM has examined existing scientific literature to find out what information is available regarding the potential reproductive toxicity associated with molybdenum and its compounds. These substances find application in various consumer products such as antifreeze, fertilisers and certain plastics. Additionally, professionals and industrial workers use them in products such as metalworking fluids.

The information gathered has been summarised. The Health Council of the Netherlands will use this summary to evaluate the potential adverse effects on reproduction. Furthermore, it will use it to provide recommendations to the Ministry of Social Affairs and Employment regarding substance classification. Manufacturers are obligated to safeguard their employees from substances that are toxic for reproduction, which is why substances of this nature are an important topic for the Ministry of Social Affairs and Employment.

Keywords: molybdenum, reproduction, fertility, developmental toxicity, sexual function

Publiekssamenvatting

Molybdeen en zijn verbindingen: een overzicht van de beschikbare informatie over mogelijke reproductietoxiciteit

Het RIVM heeft onderzocht wat er in de wetenschappelijke literatuur bekend is over de mogelijke schadelijke eigenschappen op de voortplanting van de stof molybdeen en de verbindingen daarvan. Deze stoffen worden gebruikt in verschillende consumentenproducten. Voorbeelden zijn antivriesproducten, kunstmest en sommige plastics. Professionele en industriële werknemers gebruiken ze in onder andere metaalbewerkingsvloeistoffen.

De beschikbare informatie is samengevat. De Gezondheidsraad gebruikt dit overzicht om de schadelijke effecten op de voortplanting te beoordelen. Ook gebruiken zij het overzicht om het ministerie van Sociale Zaken en Werkgelegenheid (SZW) te adviseren over de classificatie van de stof. Producenten moeten hun werknemers beschermen tegen stoffen die schadelijk zijn voor de voortplanting. Daarom heeft dit soort stoffen de aandacht van SZW.

Kernwoorden: molybdeen, reproductietoxiciteit, voortplanting, ontwikkelingstoxiciteit

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Summary

RIVM has carried out a literature search for information on the potential reproductive toxic properties of molybdenum and its compounds. Molybdenum compounds are used in a wide range of consumer products, for instance in anti-freeze products, paints and coatings, machine wash liquids and fertilizers. Professional and industrial workers use molybdenum compounds in welding and soldering products, pH regulators, metal working fluids and water treatment products.

Available animal and epidemiological data on effects of molybdenum or its compounds on sexual function and fertility, development and on or via lactation, published since 2012, were evaluated and summarized. These included a 90-day toxicity study (OECD 408), a dose-range finding study, a two-generation study (OECD 416), a 30-day testicular toxicity study and two prenatal developmental toxicity studies (OECD 414) with sodium molybdate, all performed in rats. Two 14-day drinking water studies were performed in mice, also treated with sodium molybdate. A 100-day drinking water study with mice was performed with an unspecified molybdenum compound, and molybdenum nanoparticles were investigated in a prenatal developmental toxicity study in mice. Further, three *in vitro* studies were included; two performed with ammonium molybdate and one with sodium molybdate. Most studies were considered of sufficient quality. For a number of studies the quality could not be assessed since only an abstract was available. In addition to the animal and *in vitro* studies, multiple studies with human data have been summarised. These include two longitudinal cohort studies, ten prospective cohort studies, and numerous case control and cross-sectional studies.

At the request of the Dutch Minister of Social Affairs and Employment, the Health Council of the Netherlands will use the overview of the literature to assess the reproduction toxic properties and to provide a recommendation for its classification.

1 Introduction

The aim of current research is to identify and summarize the available data from studies with laboratory models, test animals and humans on the substance molybdenum and its compounds. The focus of current literature review will be on the reproductive toxic properties of these substances.

The Health Council of the Netherlands published an advisory report on molybdenum compounds in 2013 (1). RIVM was requested by the Health Council of the Netherlands to perform a literature search and identify and summarize publications on reproductive toxicity of molybdenum and its compounds published from 2012 until present. Current RIVM-report does not include an assessment of the reported reproductive toxic properties of molybdenum and its compounds, nor does it provide a recommendation for classification of these substances based on the CLP-criteria (2).

At the request of the Dutch Minister of Social Affairs and Employment, the Health Council of the Netherlands will use the summaries to assess the reproductive toxic properties and to provide a recommendation for its classification. The assessment will be performed by the Health Council's Subcommittee on Classifying Reproductive Toxic Substances. This subcommittee falls under the Dutch Expert Committee on Occupational Safety, which focuses on health risks associated with occupational exposure of workers to chemicals.

The literature search strategy which forms the basis of current literature overview is presented in chapter 2. In chapter 3 the substance identity of molybdenum and its compounds is provided. Chapter 4 presents information on international classifications of molybdenum and its compounds. Available information on monitoring (*i.e.* environmental and biological exposure monitoring) and manufacture and use is presented in chapters 5 and 6, respectively. A summary of the (toxico)kinetics of molybdenum and its compounds is described in chapter 7. Chapter 8 describes an overview of the data on reproductive toxicity, which includes data on adverse effects on fertility and sexual function, adverse effects on development and adverse effects on or via lactation.

2 Literature search strategy

The Health Council of the Netherlands issued an advice on molybdenum in 2013 (1). That advice was based on (metallic) molybdenum (CAS # 7439-98-7), molybdenum trioxide (CAS # 1313-27-5) and sodium molybdate (CAS # 7631-95-0), the only compounds for which sufficient scientific literature was available at the time. For the present report, a literature search for publications on reproductive toxicity of molybdenum has been performed using various databases from 2012 up to August 2022. Additionally, publications on (toxico)kinetics and monitoring were searched for as well. Below the literature search strategy and its results is presented for the search until April 2021. An update of the search was performed in August 2022.

2.1 Embase

Table 1 presents the search terms and the results for the database Embase.

Table 1 Search strategy and result for Embase.

No.	Query	Results
#1	'molybdenum'/exp	17,196
#2	'molybdenum complex'/exp	1,180
#3	'molybd*'	27,836
#4	#1 OR #2 OR #3	27,836
#5	'prenatal exposure'/exp OR 'maternal exposure'/exp OR 'paternal exposure'/exp	28,190
#6	((('prenatal' OR 'maternal' OR 'paternal') NEAR/3 'expos*')):ti,ab	29,085
#7	'reproductive toxicity'/exp OR 'teratogenicity'/exp OR 'developmental toxicity'/exp OR 'ferotoxicity' OR 'embryotoxicity'/exp	36,242
#8	((('repro*' OR 'development*') NEAR/3 'toxic*')):ti,ab OR 'teratogen*':ti,ab OR 'reprotox*':ti,ab OR 'embryotox*':ti,ab	40,384
#9	#5 OR #6 OR #7 OR #8	101,209
#10	#4 AND #9	96
#11	'fertility'/exp OR 'lactation'/exp OR 'breast milk'/exp OR 'pregnancy'/exp OR 'parameters concerning the fetus, newborn and pregnancy'/exp OR 'infertility'/exp OR 'organogenesis'/exp	1,392,318
#12	'pregnancy outcome*':ti,ab OR 'pregnan*':ti OR 'fertilit*':ti OR 'infertilit*':ti OR 'subfertilit*':ti OR 'fecundit*':ti OR (((('differential' OR 'effect*' OR 'agent*') NEAR/3 'fertilit*')):ti,ab) OR (('breast' NEAR/3 'milk*')):ti,ab OR (('milk' NEAR/3 'secret*')):ti,ab) OR 'lactation*':ti,ab OR 'organogenes*':ti	427,958

No.	Query	Results
#13	#4 AND (#11 OR #12)	415
#14	'toxicokinetics'/exp OR 'toxicokinetic*':ti,ab	14,029
#15	'bioaccessib*' OR 'bioelut*':ti,ab	3,188
#16	((('environment*' OR 'human' OR 'biologic*') NEAR/3 'exposure monitor*')):ti,ab	123
#17	#4 AND (#14 OR #15 OR #16)	59
#18	'xenobiotic metabolism'/exp OR 'metal metabolism'/mj OR 'metabolism'/mj	231,380
#19	'metabolism':ti OR 'adme':ti,ab OR 'absorption distribution metabolism excretion':ti,ab	238,526
#20	#18 OR #19	441,561
#21	#20 AND [humans]/lim	172,545
#22	'murine'/exp OR 'experimental animal'/exp OR 'animal experiment'/exp OR 'leporidae'/exp OR 'rat':ti,ab OR 'rats':ti,ab OR 'mouse':ti,ab OR 'mice':ti,ab OR 'hamster*':ti,ab OR 'pig*':ti,ab OR 'monkey*':ti,ab OR 'rabbit*':ti,ab	5,465,637
#23	#20 AND #22	142,025
#24	#4 AND (#21 OR #23)	296
#25	#10 OR #13 OR #17 OR #24	779
#26	#25 AND [2012-2021]/py	361

2.2 PubMed

Table 2 presents the search terms and the results for the database Pubmed:
Table 2 Search strategy and result for Pubmed.

Search	Search terms	Items found
1	"Molybdenum"[Mesh] OR "molybd*"[tw]	20,451
2	"Prenatal Exposure Delayed Effects"[Mesh] OR "Maternal Exposure"[Mesh] OR "Paternal Exposure"[Mesh] OR "Organogenesis"[Mesh]	154,360
3	"prenatal exposure"[tw] OR "maternal exposure"[tw] OR "paternal exposure"[tw]	41,787
4	"Teratogens"[Mesh] OR "Toxicogenetics"[Mesh]	8,643
5	("reproductive tox*"[tw] OR "developmental toxicity"[tw] OR "fetotoxic*"[tw] OR "teratogen*"[tw] OR "reprotox*"[tw] OR "embryotox*"[tw])	29,793
6	#1 and (#2 or #3 or #4 or #5)	79
7	("Fertility"[Mesh] OR "fertility"[tw] OR "Lactation"[Mesh] OR "Milk, Human"[Mesh] OR "Milk"[Mesh:NoExp] OR "Pregnancy"[Mesh:NoExp] OR "Pregnancy Outcome"[Mesh] OR	1,110,491

Search	Search terms	Items found
	"infertility"[tw] OR "subfertility"[tw] OR "fecundity"[tw])	
8	("pregnancy outcome*[tw] OR "pregnan*[ti] OR "fertilit*[ti] OR "differential fertilit*[tw] OR "breast milk"[tw] OR "milk secret*[tw] OR "lactation"[tw] or "infertilit*[ti] OR "subfertil*[ti] OR "fecundit*[ti] OR "organogenes*[ti])	382,981
9	#1 and (#7 or #8)	450
10	("Toxicokinetics"[Mesh] OR "Toxicological Phenomena"[Mesh] OR "toxicokinetic*[tw] OR "bioaccessib*[tw] OR "bioelut*[tw])	460,333
11	("exposure monitor*[tw] AND ("environment*[tw] OR "human"[tw] OR "biologic*[tw]))	521
12	#1 and (#10 or #11)	229
13	("Molybdenum/metabolism"[Majr] OR "Metabolism"[Majr:NoExp] OR "metabolism"[ti] OR "adme"[tw] OR "absorption distribution metabolism excretion"[tw])	220,956
14	("rat"[tw] OR "rats"[tw] OR "mouse"[tw] OR "mice"[tw] OR "hamster*[tw] OR "pig"[tw] OR "pigs"[tw] OR "monkey*[tw] OR "rabbit*[tw] OR "human*[tw] OR "man"[tw] OR "men"[tw] OR "woman"[tw] OR "women"[tw] OR "child*[tw] OR "infant*[tw] OR "newborn*[tw] OR "fetus*[tw] OR "neonate*[tw])	23,163,797
15	#1 and #13 and #14	399
16	#6 or #9 or #12 or #15	1,075
17	#16 and 2012:2021[dp]	275

2.3 Scopus

The following search terms were used for the database Scopus:
 ((TITLE-ABS-KEY (*molybd*)) AND ((TITLE-ABS-KEY (('prenatal' OR 'maternal' OR 'paternal') W/3 'expos*')) OR (TITLE-ABS-KEY (('repro*' OR 'development*') W/3 'toxic*') OR 'teratogen*' OR 'reprotox*' OR 'embryotox*'))OR (TITLE-ABS-KEY ('pregnancy-outcome*' OR 'differential-fertilit*' OR ('breast' W/3 'milk') OR ('milk' W/3 'secret*') OR 'lactation')) OR (TITLE ('pregnan*' OR 'fertilit*' OR 'fecundit*' OR 'infertilit*' OR 'subfertilit*' OR 'organogenes*')))) OR ((TITLE-ABS-KEY (*molybd*))AND ((TITLE-ABS-KEY ('toxicokinetic*' OR 'bioaccessib*' OR 'bioelut*' OR (('environment*' OR 'human' OR 'biologic*') W/3 'exposure-monitor*')))OR (TITLE-ABS-KEY ('adme' OR 'absorption-distribution-metabolism-excretion') OR TITLE ('metabolism'))) AND (TITLE-ABS-KEY ('rat' OR 'rats' OR 'mouse' OR 'mice' OR 'hamster*' OR 'pig' OR 'pigs' OR 'monkey*' OR 'rabbit*' OR

'human*' OR 'man' OR 'men' OR 'woman' OR 'women' OR 'child*' OR 'infant*' OR 'newborn*' OR 'fetus*' OR 'neonate*'))) AND PUBYEAR > 2011

This resulted in 108 records.

2.4 Toxcenter

Table 3 presents the search terms and the results for the database Toxcenter:

Table 3 Search strategy and result for Toxcenter.

Query	Search terms	Number of records
L1	?MOLYBD?	50,693
L2	(PRENATAL OR MATERNAL OR PATERNAL)(3W)EXPOS?	54,888
L3	(REPRO? OR DEVELOPMENT?)(3W)TOXIC? OR TERATOGEN? OR REPROTO	114,430
L4	PREGNANCY-OUTCOME? OR DIFFERENTIAL FERTILIT? OR BREAST(3W)MILK OR MILK(3W)SECRET? OR LACTATION	43,557
L5	(PREGNAN? OR FERTILIT?)/TI	81,673
L6	TOXICOKINETIC? OR BIOACCESSIB? OR BIOELUT? OR (ENV OR BIOLOGIC?)(3W) EXPOSURE MONITOR?IRONMENT? OR HUMAN	27,036
L7	ADME OR ABSORPTION DISTRIBUTION METABOLISM EXCRETION OR METABOLISM/TI	136,721
L8	L1 AND (L2 OR L3 OR L4 OR L5)	300
L9	L1 AND (L6 OR L7)	375
L10	L9/HUM,ANI	32
L11	L8 OR L10	332
L12	L11 AND 2012-2020/PY	116

2.5 ECHA database

The ECHA database was searched for information on the 10 selected molybdenum compounds (see Chapter 3 and Annex A for the selection). These data are included in section 8.

In addition, the database was used to search for registration dossiers of molybdenum-containing substances, e.g. including reaction products and multi-constituent substances, that have information on reproduction toxicity in the registration dossier. These compounds are more complex structures and cannot be used for grouping with molybdenum compounds. Therefore, they are not taken into account for the selection. However, the reproduction toxicity data may be of interest for interpretation of data from molybdenum compounds and are included in Annex B.

2.6 Secondary sources

Secondary sources were consulted. These included e.g. IARC, SCOEL, WHO, IPCS, ATSDR, DFG; primarily consulted via echemportal¹. Also RIVM-reports and evaluations and the RIVM-website 'Risico's van stoffen'² were consulted as well.

2.7 Overall evaluation of results literature search

The obtained records were evaluated, duplicates were removed, and records were included if considered relevant based on title and abstract. Additionally, publications cited in the selected publications, but not selected during the primary search, were added if considered appropriate.

With respect to human health endpoints evaluated in current report (i.e. reproductive toxicity), this resulted in 14 studies for effects on sexual function and fertility, 22 studies for effects on development and 1 study for effects on or via lactation.

¹ <https://www.echemportal.org>

² <https://rvs.rivm.nl/>

3 Substance identification

3.1 Name and other identifiers of the substance

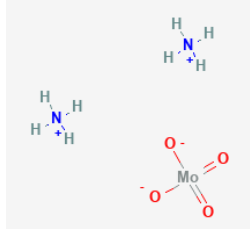
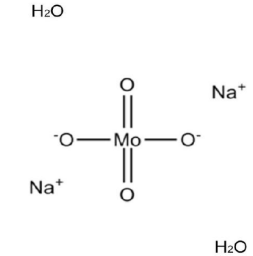
This overview summarizes information of molybdenum and a selection of molybdenum compounds. The main sources used were a report of The Dutch Expert Committee on Occupational Safety (a Committee of the Health Council of the Netherlands) (1), a report by the ATSDR (3), the Handbook of chemistry and physics (4) and ECHA's database of registrations (5). First, compounds with reproduction toxicity data were selected, which were: molybdenum, sodium molybdate and ammonium molybdate (VI). In addition, also molybdenum trioxide was selected based on available data on this substance before 2013. Molybdenum compounds with a similar solubility as one of the selected compounds were also selected. This resulted in three groups of compounds:

1. Molybdenum
2. Soluble compounds: ammonium molybdate (VI), sodium molybdate, ammonium paramolybdate, diammonium dimolybdate, ammonium tetrathio molybdate, potassium molybdate
3. Slightly soluble compounds: molybdenum trioxide, tetraammonium hexamolybdate, molybdenum sulfide (MoS_2) - roasted.

An overview of all molybdenum compounds considered and further explanation on the criteria for selection can be found in Annex A.

The identity of molybdenum and the selected molybdenum compounds is presented in Table 4, 5 and 6 below.

Table 4 Substance identity and information related to molecular and structural formula of molybdenum, ammonium molybdate and disodium molybdate (dihydrate).

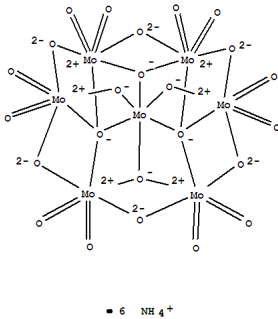
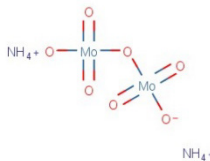
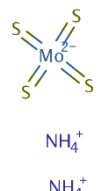
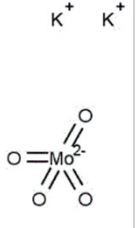
Name(s) in the IUPAC nomenclature or other international chemical name(s)	Molybdenum	Ammonium molybdate(VI)	Disodium dioxomolybdenumbis (olate)
Other names (usual name, trade name, abbreviation)		Diammonium molybdate; Ammonium orthomolybdate	Sodium molybdate, disodium molybdate
ISO common name (if available and appropriate)	N/A	N/A	N/A
EC/EINECS number (if available and appropriate)	231-107-2	236-031-3	231-551-7
EC name (if available and appropriate)	Molybdenum	Ammonium molybdate(VI)	Disodium molybdate
CAS number	7439-98-7	13106-76-8	7631-95-0; 10102-40-6 (dihydrate)
SMILES code (if available)	[Mo]	[NH4+].[NH4+].[O-][Mo](=O)(=O)[O-]	O.O.[O-][Mo](=O)(=O)[O-].[Na+].[Na+]
Molecular formula	Mo	(NH ₄) ₂ MoO ₄	Na ₂ MoO ₄ (Na ₂ MoO ₄ .2H ₂ O)
Structural formula	Mo		
Molecular weight or molecular weight range	95.96	196.01	205.92; 241.95 (dihydrate)

Name(s) in the IUPAC nomenclature or other international chemical name(s)	Molybdenum	Ammonium molybdate(VI)	Disodium dioxomolybdenumbis (olate)
Information on optical activity and typical ratio of (stereo) isomers (if applicable and appropriate)	N/A	N/A	N/A
Description of the manufacturing process and identity of the source (for UVBC substances only)	N/A	N/A	N/A
Degree of purity (%) (if relevant for the entry in Annex VI)	N/A	N/A	N/A

^a Substance consists for ≥ 45 – $\leq 96\%$ (w/w) of MoO_3 , and to a lesser extent SiO_2 , Mo suboxides, MoO_2 , iron molybdates, lead molybdate, arsenic oxide, copper molybdate, calcium molybdate and ammonium molybdates.

Table 5 Substance identity and information related to molecular and structural formula of ammonium paramolybdate, diammonium dimolybdate, ammonium tetrathio molybdate and dipotassium tetraoxomolybdate.

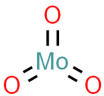
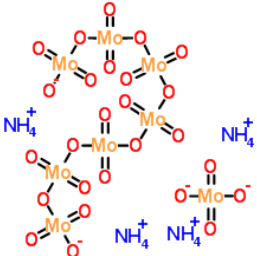
Name(s) in the IUPAC nomenclature or other international chemical name(s)	Hexaammonium heptamolybdate tetrahydrate	diammonium [(oxidodioxomolybdenio)oxy]molybdenumoylolate	Bisammonium tetrakis(sulfido) molybdate(2-)	Dipotassium tetraoxomolybdenum diuide
Other names (usual name, trade name, abbreviation)	Ammonium molybdate (VI) tetrahydrate; ammonium paramolybdate	Diammonium dimolybdate	Ammonium tetrathio molybdate	Dipotassium tetraoxomolybdate; potassium molybdate
ISO common name (if available and appropriate)	N/A	N/A	N/A	N/A
EC/EINECS number (if available and appropriate)	234-722-4	248-517-2	640-219-4	236-599-2
EC name (if available and appropriate)	N/A	Diammonium dimolybdate		Dipotassium tetraoxomolybdate
CAS number	12027-67-7;	27546-07-2	15060-55-6	13446-49-6

Name(s) in the IUPAC nomenclature or other international chemical name(s)	Hexaammonium heptamolybdate tetrahydrate	diammonium [(oxidodioxomolybdenio)oxy]molybdenumoylolate	Bisammonium tetrakis(sulfido) molybdate(2-)	Dipotassium tetraoxomolybdenum diuide
	12054-85-2 (tetrahydrate)			
SMILES code (if available)	[NH4+].[NH4+].[NH4+].[NH4+].[NH4+].[NH4+].[O-][Mo](=O)(=O)[O-].[O-][Mo](=O)(=O)[O-].[O-][Mo](=O)(=O)[O-].[O-][Mo](=O)(=O)[O-]	[NH4+].[NH4+].[O-][Mo](=O)(=O)[O-].[O-][Mo](=O)(=O)[O-]	[NH4+].[NH4+].[S]=[Mo-].[S]=[Mo-].[S]=[Mo-].[S]=[Mo-]	[K+].[K+].[O]=[Mo-].[O]=[Mo-].[O]=[Mo-].[O]=[Mo-]
Molecular formula	(NH ₄) ₆ Mo ₇ O ₂₄ (NH ₄) ₆ Mo ₇ O ₂₄ · 4H ₂ O	(NH ₄) ₂ Mo ₂ O ₇	H ₈ MoN ₂ S ₄	K ₂ MoO ₄
Structural formula				
Molecular weight or molecular weight range	1163.80 1235.86 (tetrahydrate)	339.95	260.28	238.14
Information on optical activity and typical ratio of (stereo) isomers (if applicable and appropriate)	N/A	N/A	N/A	N/A

Name(s) in the IUPAC nomenclature or other international chemical name(s)	Hexaammonium heptamolybdate tetrahydrate	diammonium [(oxidodioxomolybdenio)oxy]molybdenumoylolate	Bisammonium tetrakis(sulfido) molybdate(2-)	Dipotassium tetraoxomolybdenum diuide
Description of the manufacturing process and identity of the source (for UVBC substances only)	N/A	N/A	N/A	N/A
Degree of purity (%) (if relevant for the entry in Annex VI)	N/A	N/A	N/A	N/A

Table 6 Substance identity and information related to molecular and structural formula of molybdenum trioxide, tetraammonium hexamolybdate and molybdenum sulfide, roasted.

Name(s) in the IUPAC nomenclature or other international chemical name(s)	Molybdenum trioxide	Tetraammonium bis (dioxomolybdenumbis (olate))	Silicon(4+) trioxomolybdenum dioxidandiide
Other names (usual name, trade name, abbreviation)		Tetraammonium hexamolybdate; ammonium octamolybdate	
ISO common name (if available and appropriate)	N/A	N/A	N/A
EC/EINECS number (if available and appropriate)	215-204-7; 231-970-5 (molybdic acid; hydrate)	235-650-6	289-178-0
EC name (if available and appropriate)	Molybdenum trioxide; molybdic acid (hydrated forms)	Tetraammonium hexamolybdate	Molybdenum sulfide (MoS ₂), roasted
CAS number	1313-27-5; 7782-91-4, 25942-34-1 (molybdic acid hydrates)	12411-64-2	86089-09-0
SMILES code (if available)	O=[Mo](=O)=O	[NH4+].[NH4+].[NH4+].[NH4+].[O-].[Mo](=O)(=O)[O-].[O-].[Mo](=O)(=O)O[Mo](=O)(=O)O[Mo](=O)(=O)O[

Name(s) in the IUPAC nomenclature or other international chemical name(s)	Molybdenum trioxide	Tetraammonium bis (dioxomolybdenumbis (olate))	Silicon(4+) trioxomolybdenum dioxidandiide
		$\text{Mo}](=\text{O})(=\text{O})\text{O}[\text{Mo}](=\text{O})(=\text{O})\text{O}[\text{Mo}](=\text{O})(=\text{O})\text{O}[\text{Mo}](=\text{O})(=\text{O})[\text{O}^-]$	
Molecular formula	MoO_3 $\text{MoO}_3 \cdot \text{H}_2\text{O}$ (hydrate) $\text{MoO}_3 \cdot 2\text{H}_2\text{O}$ (dihydrate)	$(\text{NH}_4)_4\text{Mo}_8\text{O}_{26}$	UVCB ^a
Structural formula			
Molecular weight or molecular weight range	143.94	1255.66	
Information on optical activity and typical ratio of (stereo) isomers (if applicable and appropriate)	N/A	N/A	N/A
Description of the manufacturing process and identity of the source (for UVBC substances only)	N/A	N/A	N/A
Degree of purity (%) (if relevant for the entry in Annex VI)	N/A	N/A	N/A

^a Substance consists for ≥ 45 – $\leq 96\%$ (w/w) of MoO_3 , and to a lesser extent SiO_2 , Mo suboxides, MoO_2 , iron molybdates, lead molybdate, arsenic oxide, copper molybdate, calcium molybdate and ammonium molybdates.

3.2 Physico-chemical properties

The physico-chemical properties of molybdenum and its compounds are presented in Table 7, 8 and 9 below. The ECHA dissemination website (5) and the Handbook of chemistry and physics (4) were used as the primary source.

Molybdenum (Mo) is a naturally occurring metallic trace element found in natural minerals, but not as the free metal. Biologically, it is an important micronutrient in plants and animals, including humans. It is used widely in industry for metallurgical applications (3). Molybdenum has oxidation states from -2 to +6. Commonly encountered compounds are those of molybdenum in oxidation state +6 (Mo(VI), MoO₃, molybdates) and +4 (Mo(IV), MoS₂) (3).

Molybdenum (VI) anions include molybdate (MoO₄²⁻) and polymeric anions ('isopolymolybdates') of which the most common are heptamolybdate (Mo₇O₂₄⁶⁻) and octamolybdate (Mo₈O₂₆⁴⁻). These anions occur in sodium and ammonium salts, often hydrated, which are the common sources of molybdenum in commerce and industrial applications (3).

Under physiological conditions (pH >6.5), the molybdate anion, MoO₄²⁻, is the sole molybdenum species in aqueous media. Molybdenum compounds (e.g., molybdenum trioxide and polymolybdates) transform rapidly to the MoO₄²⁻ ion under environmentally relevant test conditions. Protonated forms, such as HMoO₄⁻ and H₂MoO₄, are found at pH <5. Molybdenum tends to be more mobile under alkaline conditions, but adsorption increases with decreasing pH (3).

Table 7 Summary of physicochemical properties of molybdenum and its selected compounds, part 1.

Substance	State of the substance at normal temperature and pressure	Melting/freezing point (at 101325 Pa)	Boiling point (at 101325 Pa)	Relative density (at 20°C)	Water solubility	Partition coefficient n-octanol/water
Molybdenum	Solid	2,623°C	4,639°C	10.18	Insoluble	N/A
Ammonium molybdate(VI)	Solid	>190°C ^b	N/A ^a	1.4	Soluble: 10 g/L	N/A
Disodium molybdate	Solid	687°C	N/A ^a	2.59	Soluble: 654 g/L	N/A
Ammonium paramolybdate	Solid	>90°C ^b	N/A ^a	2.86	Soluble: 206.5 g/L ^c	N/A
Diammonium dimolybdate	Solid, powder, white to greyish, odourless, inorganic.	>150°C ^b	N/A	2.97	Soluble: 228 g/L	N/A
Ammonium tetrathio molybdate^a	Solid	>100°C ^b	-	-	Very soluble	-
Dipotassium tetraoxomolybdate	Solid fine white powder	926°C	N/A ^b	3.09	Soluble: 183 g/100 g H ₂ O	N/A
Molybdenum trioxide	Solid, powder, white-yellow to bluish, odourless, inorganic.	802°C	1,155°C	4.66	Slightly soluble: 1 g/L	N/A
Tetraammonium hexamolybdate	Solid, crystalline, white, odourless, inorganic	>287°C ^b	N/A	3.74	Slightly soluble: 1 g/L	N/A
Molybdenum sulfide (MoS₂), roasted	Solid	801°C	1,150°C	4.48	Slightly soluble: 560 mg/L	N/A

^a No REACH registration dossier available^b Decomposes before melting.^c Solubility for CAS number 12054-85-2, according to the CRC handbook: 43 g/100 ml H₂O

Table 8 Summary of physicochemical properties of molybdenum and its selected compounds, part 2.

Substance	Vapour pressure	Surface tension	Flash point	Flammability	Explosive properties	Self-ignition temperature
Molybdenum	N/A	N/A	N/A	Not flammable	Not explosive	Not auto-flammable
Ammonium molybdate(VI)	N/A	N/A	N/A	Not flammable	Not explosive	Not auto-flammable
Disodium molybdate	N/A	N/A	N/A	N/A	Not explosive	Not auto-flammable
Ammonium paramolybdate	N/A	N/A	N/A	N/A	Not explosive	Not auto-flammable
Diammonium dimolybdate	N/A	N/A	N/A	Not flammable	Not explosive	Not auto-flammable
Ammonium tetrathio molybdate^a	-	-	-	-	-	-
Dipotassium tetraoxomolybdate	N/A	N/A	N/A	Not flammable	Not explosive	N/A
Molybdenum trioxide	N/A	N/A	N/A	Not flammable	Not explosive	N/A
Tetraammonium hexamolybdate	N/A	N/A	N/A	Not flammable	Not explosive	Not auto-flammable
Molybdenum sulfide (MoS₂), roasted	N/A	N/A	N/A	N/A	Not explosive	Not auto-flammable

^a No REACH registration dossier available.

Table 9 Summary of physicochemical properties of molybdenum and its selected compounds, part 3.

Substance	Oxidising properties	Granulometry	Stability in organic solvents and identity of relevant degradation products	Dissociation constant (pKa)	Viscosity
Molybdenum	Non-oxidising	D50: 8.4 µm MMAD: 21.68 µm	N/A	N/A	N/A
Ammonium molybdate(VI)	Non-oxidising	53-150 µm	N/A	N/A	N/A
Disodium molybdate	Non-oxidising	D10: 34.5 µm D50: 143.1 µm D90: 295.9 µm	N/A	N/A	N/A
Ammonium paramolybdate	Non-oxidising	D10: 57.8 µm D50: 268.3 µm D90: 590.0 µm	N/A	N/A	N/A
Diammonium dimolybdate	Non-oxidising	D50: 189 µm MMAD: 27.46 µm	N/A	N/A	N/A
Ammonium tetrathio molybdate^a	-	-	-	-	-
Dipotassium tetraoxomolybdate	Non-oxidising	D10: 37.1 µm D50: 151.1 µm D90: 535.3 µm	N/A	N/A	N/A
Molybdenum trioxide	Non-oxidising	D50: 195 µm MMAD ^b : 3.83 µm (8.8%) and 29.88 µm (91.2%)	N/A	N/A	N/A
Tetraammonium hexamolybdate	Non-oxidising	D50: 3.4 µm MMAD ^b : 4.56 µm (31.5%) and 33.25 µm (68.5%)	N/A	N/A	N/A
Molybdenum sulfide (MoS₂), roasted	Non-oxidising	D10 = 29 µm D50 = 189.0 µm D85 = 500 µm	N/A	N/A	N/A

MMAD: Mass median aerodynamic diameter.

^a No REACH registration dossier available.^b percentage in parentheses indicates weighting factor of bimodal distribution.

4 International classifications

4.1 European Commission

Molybdenum trioxide has a harmonized classification for three hazard classes:

- Eye Irrit. 2 (H319: causes serious eye irritation)
- STOT SE 3 (H335: may cause respiratory irritation)
- Carc. 2 (H351: suspected of causing cancer)

The other selected molybdenum compounds and molybdenum do not have a harmonized classification under the European CLP regulation.

4.2 The Health Council

Molybdenum has been evaluated by the Health Council of the Netherlands in 2013 (1).

For molybdate compounds, the Subcommittee on the Classification of reproductive toxic substances recommended classifying molybdate compounds for adverse effects on fertility and sexual function in category 2 (suspected human reproductive toxicant), and labelling with H361f (suspected of damaging fertility). For the other molybdenum compounds assessed, no classification for effects on fertility could be proposed due to a lack of data.

Lack of appropriate data precluded the assessment of molybdenum compounds for effects on development and for effects during lactation.

4.3 Other countries

In Japan, several classifications are applicable for the selected molybdenum and molybdenum compounds (Table 10).

Table 10 Classification of molybdenum and molybdenum compounds in Japan.

Compound (CAS number)	GHS classification in Japan
Molybdenum ³ (7439-98-7)	Skin Irrit. 2 (H315: Causes skin irritation) Eye Irrit. 2 (H319: Causes serious eye irritation) STOT SE 3 (H335: May cause respiratory irritation)
Ammonium molybdate (VI) (13106-76-8)	-
Sodium molybdate (7631-95-0) ⁴	Acute Tox. 3 (H301: Toxic if swallowed) Skin Irrit. 2 (H315: Causes skin irritation) Eye Irrit. 2 (H319: Causes serious eye irritation) Muta. 2 (H341: Suspected of causing genetic defects) Carc. 2 (H351: Suspected of causing cancer) Repr. 2 (H361: Suspected of damaging fertility or the unborn child)

³ <https://www.nite.go.jp/chem/english/ghs/15-mhlw-0126e.html>

⁴ <https://www.nite.go.jp/chem/english/ghs/15-mhlw-0127e.html>

Compound (CAS number)	GHS classification in Japan
	STOT SE 3 (H335: May cause respiratory irritation) STOT RE 1 (H372: Causes damage to organs through prolonged or repeated exposure (systemic toxicity, testis)) STOT RE 2 (H373: May cause damage to organs through prolonged or repeated exposure (kidney))
Sodium molybdate dihydrate (10102-40-6)	-
Ammonium paramolybdate (12027-67-7) ⁵	Acute Tox. 4 (H302: Harmful if swallowed) Eye Irrit. 2 (H319: Causes serious eye irritation) Carc. 2 (H351: Suspected of causing cancer) Repr. 2 (H361: Suspected of damaging fertility or the unborn child) STOT SE 3 (H335: May cause respiratory irritation) STOT RE 2 (H373: May cause damage to organs through prolonged or repeated exposure (kidney))
Ammonium paramolybdate tetrahydrate (12054-85-2)	-
Diammonium dimolybdate (27546-07-2)	-
Ammonium tetrathiomolybdate (15060-55-6)	-
Potassium molybdate (13446-49-6)	-
Molybdenum trioxide (1313-27-5) ⁶	Eye Irrit. 2 (H319: Causes serious eye irritation) Carc. 2 (H351: Suspected of causing cancer) Repr. 2 (H361: Suspected of damaging fertility or the unborn child) STOT SE 3 (H335: May cause respiratory irritation) STOT RE 1 (H372: Causes damage to organs through prolonged or repeated exposure (respiratory organs, reproductive organs male)) STOT RE 2 (H373: May cause damage to organs through prolonged or repeated exposure (kidney))
Molybdic acid (7782-91-4)	-
Tetraammonium hexamolybdate (12411-64-2)	-
Molybdenum sulfide, roasted (86089-09-0)	-

⁵ <https://www.nite.go.jp/chem/english/ghs/15-mhlw-0128e.html>⁶ <https://www.nite.go.jp/chem/english/ghs/19-mhlw-2001e.html>

None of the 10 selected molybdenum compounds is currently classified for reproductive toxicity in Australia⁷. Molybdenum trioxide and its hydrated form molybdenic acid are classified in Australia for carcinogenicity (category 2), eye irritation (category 2A) and specific target organ toxicity - single exposure (category 3), in accordance with GHS.

In Germany, none of the selected molybdenum compounds is included in the list of additional CMR substances in the context of worker protection.⁸ However, molybdenum trioxide is classified in carcinogen category 3B by the German Research Foundation (DFG). Category 3 includes substances for which in vitro or animal studies have yielded evidence of carcinogenic effects that is not sufficient for classification of the substance in one of the other categories. Further studies are required before a final decision can be made. A MAK or BAT value can be established provided no genotoxic effects have been detected⁹.

According to the IARC, molybdenum trioxide is possibly carcinogenic to humans (Group 2B). The other selected molybdenum compounds have not been evaluated by IARC.

In the state of California, molybdenum trioxide is listed as a carcinogen since 19 March 2021¹⁰. The other selected molybdenum compounds are not included in this list.

The selected molybdenum compounds are not included in the Report on Carcinogens (14th edition)¹¹, and also not in the NIOSH carcinogen list¹².

⁷ <http://hcis.safeworkaustralia.gov.au/HazardousChemical/>

⁸ https://www.baua.de/DE/Angebote/Rechtstexte-und-Technische-Regeln/Regelwerk/TRGS/pdf/TRGS-905.pdf?__blob=publicationFile

⁹ https://series.publisso.de/sites/default/files/documents/series/mak/lmbv/Vol2020/Iss1/Doc001/mbwl_2020_d_eu.pdf (in German).

¹⁰ <https://oehha.ca.gov/proposition-65/chemicals/molybdenum-trioxide>

¹¹ <https://ntp.niehs.nih.gov/whatwestudy/assessments/cancer/roc/index.html#toc1>

¹² <https://www.cdc.gov/niosh/topics/cancer/npotocca.html>

5 Monitoring

5.1 Environmental exposure monitoring

Four methods to measure molybdenum are included in the NIOSH Manual of Analytical Methods¹³

- Method 7302 (6): Elements by ICP (Microwave Digestion). This method is for the analysis of metal and non-metal dust collected on MCE filters in the workplace.
- Method 7304 (7): Elements by ICP (Microwave Digestion). This method is for the analysis of metal and non-metal dust collected on PVC filters that are also used for gravimetric analysis.
- Method 7306 (8): Elements by Cellulosic Internal Capsule Sampler, using Inductively Coupled Plasma – Atomic Emission Spectrometry (ICP-AES). The working range of this method is 4×10^{-5} mg/m³ to 10 mg/m³ for each element in a 500-L air sample. This is simultaneous multi-elemental analysis, not compound-specific.

5.2 Biological exposure monitoring

Molybdenum can be measured in blood and urine by inductively coupled plasma-mass-spectrometry (ICP-MS). An example is given in Kim et al. (2018), who measured urinary trace metals, including molybdenum (9). The National Center for Health Statistics, part of the Centers for Disease Control and Prevention (CDC), uses ICP-MS with dynamic reaction cell (ICP-DRC-MS) to measure multiple elements in urine, including molybdenum¹⁴.

The NIOSH manual of Analytical Methods contains method 8200, describing a method to analyse nail samples from animals exposed to welding fume in laboratory controlled inhalation studies (10). It is useful for monitoring nail tissues of workers exposed to several metals simultaneously. It may also be used for the analysis of freeze-dried tissue samples (liver, lungs, etc.) from laboratory studies. This is a simultaneous multi-elemental analysis, but is not compound-specific.

A biological reference value of 150 µg/l in urine is derived by the Commission for the Investigation of Health Hazards of Chemical Compounds in the Work in Germany (11). Hays et al. (2016) calculated Biomonitoring Equivalent (BE), i.e. estimates of the concentration of a chemical or its metabolite in blood or urine that are consistent with defined exposure guidance values such as reference doses or tolerable daily intakes (12). BEs were calculated for molybdenum in plasma, whole blood, and urine associated with exposure guidance values set to protect against both nutritional deficits and toxicity. For protection against nutritional deficits, the BE values in plasma, whole blood and urine are 0.5, 0.45 and 22 µg/L, respectively. The BEs associated with toxicity range from 0.9 to 31 µg/L in plasma, 0.8-28 µg/L in whole blood and 200-7500 µg/L in urine (12).

¹³ <https://www.cdc.gov/niosh/nmam/chemicals.html>

¹⁴ https://wwwn.cdc.gov/nchs/data/nhanes/2011-2012/labmethods/uhm_g_met_heavy_metals.pdf

No biological monitoring guidance values were available from the Health and Safety Executive (13).

6 Manufacture and uses

Information on manufacture and use has been presented in the advice on molybdenum of the Health Council of the Netherlands in 2013 (1).

Molybdenum is used primarily in metallurgical applications, including as an alloying agent in cast iron, steel, and superalloys to enhance properties such as hardenability, strength, toughness, and wear- and corrosion-resistance. Molybdenum is commonly used in combination with other alloy metals like chromium, cobalt, manganese, nickel, niobium, and tungsten. The leading form of molybdenum used by industry, particularly in stainless steel production, is molybdenum trioxide (3).

Molybdenum is also used significantly as a refractory metal and molybdenum compounds in a variety of non-metallurgical chemical applications, such as catalysts, lubricants, and pigments (3).

Molybdenum and its compounds are registered under REACH for use as intermediate to manufacture other chemicals and for the manufacturing of amongst others paper products, plastic products, machinery and vehicles, fabricated metal products, and electrical, electronic and optical equipment. Industrial products include water treatment chemicals, anti-freeze products, metal working fluids, washing & cleaning products, biocides (e.g. disinfectants, pest control products), fertilizers, pH regulators, lubricants and greases, and paper and chemicals dyes. Consumer products reported under REACH include anti-freeze products, heat transfer fluids, fertilizers, water treatment chemicals and adsorbents (5).

7 (Toxico)kinetics

7.1 Animal and human data on (toxico) kinetics

A detailed description of the toxicokinetics of molybdenum compounds in humans and animals was provided in the advice of the Health Council of the Netherlands in 2013. Additional relevant information is presented below.

In an in vivo study in rats by Su et al. (2014) the permeability of the blood–brain barrier to a number of substances, among others molybdenum (molybdate; MoO_4^{2-}), was studied (14). They used an automatic online analytical system comprising a microdialysis sampling device, a minicolumn packed with nonfunctionalized poly(vinyl chloride) beads, and an ICP-MS for continuous in-vivo measurement of their dynamic variation in the extracellular space of the brains of living rats. It was found that a single intraperitoneal administration of 1 mg MoO_4^{2-} /kg bw led to observable blood-brain-barrier permeability. An increase in molybdenum in the brain was already detected 15 min after administration. Molybdenum concentrations reached a plateau 1.5 h post-administration and remained unchanged until 4 h post-administration (14).

In a 90-day study in Sprague Dawley rats the sub-chronic toxicity of sodium molybdate dihydrate, given in the diet for 90 days at dose levels of 0, 5, 17 or 60 mg Mo/kg bw/day was investigated. Among others, serum blood, liver and kidney samples were analysed for molybdenum, copper, zinc, manganese, iron, cobalt and selenium. High levels of molybdenum and copper were found in the serum, blood, liver and kidneys of rats treated with 60 mg Mo/kg bw/day (15).

A simulated in vitro gastro-intestinal model method was used to assess the bioaccessibility of among others molybdenum in ten nutritional supplement samples and three certified reference materials. The bioaccessible fraction of molybdenum in the gastric phase was in the range of 42–101% for molybdenum. The range for the gastrointestinal phase was 40–109% for molybdenum (16).

Data on bioaccessibility were available in the REACH registration dossier of molybdenum, available at the ECHA website¹⁵. Table 11 includes the key results of investigations on bioaccessibility, i.e. the dissolved concentrations of Mo (in $\mu\text{g/L}$) in the solutions and the fraction of molybdenum dissolved (% of Mo solid loaded which was dissolved). To increase the readability of this table, only the data after 24 hours is included and the standard deviations are not included.

¹⁵ <https://echa.europa.eu/nl/registration-dossier/-/registered-dossier/15524/7/2/1>

Table 11 Dissolved concentration of Mo in µg/L and fraction of loaded solid material that has dissolved (% , in parentheses). A loading of 0.1 g/L was used for all test items except MoO₃ (1 g/L for practical reasons). From ECHA website¹⁶

Material (test item)	GMB pH 7.4	PBS pH 7.2	ASW pH 6.5	ALF pH 4.5	GST pH 1.6
Mo	169 (0.17)	5,776 (6.1)	2,305 (2.4)	574 (0.58)	531 (0.50)
MoO ₂	189 (0.25)	339 (0.48)	699 (0.93)	177 (0.25)	61 (0.08)
MoO ₃	653,933 (95.3)	630,167 (95.1)	709,900 (106)	507,300 (77.2)	69,587 (10.4)
Na ₂ MoO ₄ ·2H ₂ O	35,300 (89.49)	29,693 (80.2)	32,057 (82.0)	23,150 (58.2)	32,220 (83.2)
(NH ₄) ₄ Mo ₈ O ₂₆	40,070 (67.8)	48,083 (86.5)	50,157 (88.0)	38,277 (66.7)	56,293 (94.0)
Roasted molybdenite concentrate (RMC)	70,900 (114.5)	64,700 (101.6)	not available	35,400 (55.6)	15,600 (24.8)

ALF: Artificial lysosomal fluid; ASW: artificial sweat solution; GMB: Gamble's solution; GST: Artificial gastric fluid; PBS: phosphate-buffered saline.

7.2 Additional information relevant for read-across / grouping

The list of molybdenum compounds is long and for most of the compounds data gaps exist. Therefore, read-across of information could be considered. Read-across shall be done with care, as the effects of molybdenum and molybdenum compounds can differ. The toxicity of most metals is associated to a large degree with the release of soluble metal ions, their uptake by the body and interaction at their target sites. Bioavailability is one of the major drivers of systemic toxicity, and is related to the water solubility of a chemical. Therefore, read-across between molybdenum compounds can only be applied if compounds have a similar water solubility.

Water solubility was therefore used to make a selection of molybdenum compounds for this overview. More information on the selection criteria and the resultant selected compounds is provided in Annex A.

Read-across was also applied by registrants as was observed in the REACH registration dossiers¹⁷. The categories as suggested by the registrants are presented in Table 12.

¹⁶ <https://echa.europa.eu/nl/registration-dossier/-/registered-dossier/17317/7/2/1>

¹⁷ <https://echa.europa.eu/nl/registration-dossier/-/registered-dossier/17317/7/2/1>

Table 12 Categories for grouping, as suggested by registrants of molybdenum and molybdenum compounds. From ECHA website¹⁸

Substance/Formula	CAS:	Properties	Read-Across-Grouping Long-term effects	Read-Across-Grouping Acute effects
Roasted Molybdenite Concentrate (UVCB)	86089-09-0	soluble molybdenum substances;	Grouped based on chemical similarity for long-term, local effects via inhalation (suspected carcinogenicity via inhalation).	Grouped for all acute effects (local and systemic).
Molybdenum Trioxide (MoO ₃)	1313-27-5	water solubility above ca. 100 mg/L; solubility in biological fluids 30-100%;		
Sodium Molybdate (Na ₂ MoO ₄)	10102-40-6	"high bioaccessibility"	Grouped for all long-term, systemic effects (all release MoO ₄ ²⁻ ion).	
Ammonium Dimolybdate ((NH ₄) ₂ Mo ₂ O ₇)	27546-07-2			
Ammonium Heptamolybdate ((NH ₄) ₆ Mo ₇ O ₂₄)	12027-67-7 & 12054-85-2			
Ammonium Octamolybdate ((NH ₄) ₄ Mo ₈ O ₂₆)	12411-64-2			
Calcium Molybdate (CaMoO ₄)	7789-82-4			
Iron Molybdate (Fe ₂ (MoO ₄) ₃)	13769-81-8			
Molybdenum (metal) (Mo)	7439-98-7	Poorly/hardly soluble molybdenum substances;	Grouped for all long-term, systemic effects (all release MoO ₄ ²⁻ ion).	Grouped for all acute effects (local and systemic).
Ferromolybdenum Slags (UVCB)	84144-95-6	water solubility well below ca. 10 mg/L; solubility in biological fluids well below 10%;		
Molybdenum Dioxide (MoO ₂)	18868-43-4	"negligible bioaccessibility"	Conservative read-across to "high bioaccessibility" group above.	Conservative read-across to "high bioaccessibility" group above.

¹⁸ <https://echa.europa.eu/nl/registration-dossier/-/registered-dossier/17317/7/2/1>

8 Toxicity for reproduction

8.1 Adverse effects on sexual function and fertility

8.1.1 *Animal studies*

In the advice of the Health Council of the Netherlands on molybdenum in 2013 various studies in animals on the effects of molybdenum compounds on sexual function and fertility were described (1).

Below, additional studies that were published after release of the advice of the Health Council are described. The studies in experimental animals on the effects of molybdenum compounds on sexual function and fertility are summarized in Table 13. In these studies, animals were exposed to the substance via the oral route. No inhalation or dermal studies were available.

Reproduction toxicity data for more complex structures containing molybdenum, such as reaction products, are included in Annex B. Data from these compounds cannot be used for grouping and read-across, but may still be of interest. Further details on the selection of compounds is included in Annex A.

Table 13 Summary of animal studies of molybdenum and its compounds on sexual function and fertility.

Reference	Species	Experimental period and design	Dose and route	General toxicity	Effects on reproductive organs or reproduction	Remarks
Murray et al., 2014 (15)	Sprague Dawley rats, males and females. N= 10/sex/dose. In addition N= 10/sex for the control and high dose were subject to a 60 day recovery period following 90 days of treatment.	OECD guideline 408 (repeated dose 90-day oral toxicity study in rodents). Additional analysis of oestrus cycles, various sperm parameters and detailed histopathology of reproductive organs. Serum blood, liver and kidney samples were analysed for molybdenum, copper, zinc, manganese, iron, cobalt and selenium.	0, 5, 17 and 60 mg Mo/kg bw/day, in the diet. Test item: Sodium molybdate dihydrate Purity: >99%	<ul style="list-style-type: none"> Decreased body weight, body weight gain, food conversion efficiency (both sexes) and renal histopathology (females only) at 60 mg Mo/kg bw/day. No adverse effects in the high dose animals after the 60-day recovery period, with the exception of reduced body weight gain in one male rat. 	No treatment-related adverse effects on reproductive organ weights or histopathology, oestrus cycles or sperm parameters at any dose level.	
Study report, 2016 (17)	Sprague Dawley rats, males and females. N = 10/sex/dose	Dose-range finding study for the two-generation study by Murray et al. (2019). Study duration: Males: 10 weeks before cohabitation, during the cohabitation period, and continued through	0, 3, 20 and 40 mg Mo/kg bw/day in diet or via drinking water. Test item: Sodium molybdate dihydrate Purity: >99%	<ul style="list-style-type: none"> Reduced bodyweight in males of the 40 mg Mo/kg bw/day exposure group via drinking water Reduced bodyweight in males and females of the 40 mg Mo/kg bw/day exposure group via diet. 	<i>Drinking water</i> <ul style="list-style-type: none"> Reduced pregnancy. Pregnancy in 10, 9, 9 and 6 rats in the 0, 3, 20 and 40 Mo/kg bw/day exposure groups. Rate of 6/10 was outside of the historical control average for 	GLP compliance: yes Only summary available, from REACH registration dossier. Original study report not available.

Reference	Species	Experimental period and design	Dose and route	General toxicity	Effects on reproductive organs or reproduction	Remarks
		to the day before euthanasia Females: 10 weeks before cohabitation, during the cohabitation when male diets and water were used, gestation, and lactation periods until Day 21 of lactation (rats that delivered a litter) or Day 25 of presumed gestation (rats that did not deliver a litter). Examination: sperm parameters, litter observations, postmortem examinations of parental animals and offspring, reproductive indices and offspring viability indices.		<ul style="list-style-type: none"> • Dose related increase in molybdenum levels in serum, liver and kidney of parental animals, indicating absorption of molybdenum from both diet and drinking water with levels generally higher from diet than from drinking water. • Dose related increase in molybdenum levels in serum and tissue (at termination on PND 22). Levels were generally higher from diet than from drinking water. 	<p>pregnancy at the testing facility.</p> <ul style="list-style-type: none"> • Reduction in the number of live born pups • Increase in still born pups. The stillborn pups were all in a single litter. Overall mean litter size did not differ between groups. No statistically significant differences when analysed using a Fisher's Exact Test. <p>No other effects on reproduction. <i>Diet</i> No effects on reproduction.</p>	Results primarily presented in a qualitative manner.
Murray et al., 2019 (18)	Sprague Dawley rats,	OECD guideline 416 (two generation	0, 5, 17, or 40 mg molybdenum (Mo)/kg	Decreased body weight, food consumption (males	<i>Drinking water</i> • Increase in average number of primordial	Well-performed study; GLP.

Reference	Species	Experimental period and design	Dose and route	General toxicity	Effects on reproductive organs or reproduction	Remarks
	males and females. N = 24/sex/dose	reproductive toxicity study) P-males: exposure for at least 10 weeks before cohabitation, during the cohabitation, and continuing through to the day of euthanasia (total 147-151 days) P-females: for at least 10 weeks before cohabitation, during the cohabitation, gestation, littering and post-partum periods (lactation period) and continuing through to the day of euthanasia (total 156-158 days). F1: during lactation, 10 weeks pre-mating, cohabitation, and continued through the day of euthanasia. Effect parameters as described in OECD 416.	bw/day in drinking water. Additional group: 40 mg Mo/kg bw/day via diet. Test item: Sodium molybdate dihydrate Purity: 99%	only) and water consumption at 40 mg/kg bw/day.	follicles in the left ovary of <u>parental</u> females at 17 mg/kg bw/day, and in the right, left, and combined ovaries in the <u>F1 generation</u> at 17 and 40 mg/kg bw/day. All values were within the historical control range. • A slight increase (not statistically significant) was observed in the percent of no head sperm in the group given 40 mg/kg bw/day. In both cases, they were largely attributable to one male each. Average values were within historical control range. <i>Diet</i>	

Reference	Species	Experimental period and design	Dose and route	General toxicity	Effects on reproductive organs or reproduction	Remarks
					<ul style="list-style-type: none"> • Increase in average number of primordial follicles in the left ovary of <u>parental</u> females and in the right, left, and combined ovaries in the <u>F1 generation</u>. All values were within the historical control range. • Increased percent of sperm with no head in the parental generation compared to the control value. 	
Khorami et al., 2020 (19)	Wistar rats, males. N = 6/dose	30-day testicular toxicity study. Male rats were treated for 30 consecutive days by oral gavage. The right testis was homogenized for biochemical assays (oxidative stress parameters superoxide dismutase	Controls, 0.05, 0.1, 0.2, and 0.4 mg/kg bw/day. Controls: distilled water. Test item: sodium molybdate Purity: not mentioned.	No data were provided on general toxicity.	<ul style="list-style-type: none"> • No adverse effects on sperm count, sperm viability, sperm morphology, sperm membrane integrity or sperm motility. • No effects on oxidative stress. • No adverse effects based on 	

Reference	Species	Experimental period and design	Dose and route	General toxicity	Effects on reproductive organs or reproduction	Remarks
		(SOD), catalase (CAT), malondialdehyde (MDA), and glutathione peroxidase (GPX)). The left testis was processed for histopathology. Parameters analysed included: sperm motility, sperm count, sperm viability, sperm abnormalities and sperm membrane integrity.			histopathological analysis.	
Zhai et al., 2013 (20)	ICR mice, males. N = 10/dose	Sub-acute study. Male mice received molybdenum in drinking water for 14 days. Sperm parameters, including the epididymis index, sperm motility, sperm count, and morphology were	0, 12.5, 25, 50, 100, or 200 mg/L, in drinking water, corresponding to 0, 2.5, 5, 10, 20 and 40 mg/kg bw/d ¹⁹ . Test item: sodium molybdate dihydrate Purity not mentioned.	Not reported.	<ul style="list-style-type: none"> Effects on sperm parameters at ≥ 20 mg/kg bw/day, compared with controls: decrease in epididymis index, decreased sperm motility, decreased sperm concentration and an increased 	Mice were maintained under GLP conditions.

¹⁹ Conversion from mg/L to mg/kg bw/day is based on an assumed water consumption of 5 ml for a mouse weighing 25 grams, based on the default values as reported in ECHA guidance R.8, version 2.1, Table 8-17.

Reference	Species	Experimental period and design	Dose and route	General toxicity	Effects on reproductive organs or reproduction	Remarks
		evaluated. Malondialdehyde (MDA), superoxide dismutase (SOD), and glutathione peroxidase (GPx) levels were determined in testes.			<p>sperm abnormality rate.</p> <ul style="list-style-type: none"> • Effects on sperm parameters at 5 mg/kg bw/day, compared with controls: increase in epididymis index, increased sperm motility, increased sperm concentration and a decreased sperm abnormality rate. • Decreased activity of SOD and GPx and increased activity of SOD at ≥ 20 mg/kg bw/day. • Increased SOD and GPx activity at 5 mg/kg bw/day. 	
Zhang et al., 2013 (21)	ICR mice, females. N=25/dose.	Female mice received molybdenum in the drinking water for 14 days. Ovaries were examined by electron	0, 5, 10, 20, or 40 mg/L, in drinking water, corresponding to 0, 1, 2, 4 and 8 mg/kg bw/d ²⁰ .	Not reported.	<ul style="list-style-type: none"> • Improvement of number of ovulations, and oocyte morphology at 1 mg/kg bw/day, 	Mice were maintained under GLP conditions. Not clear if other study elements were also

²⁰ Conversion from mg/L to mg/kg bw/day is based on an assumed water consumption of 5 ml for a mouse weighing 25 grams, based on the default values as reported in ECHA guidance R.8, version 2.1, Table 8-17.

Reference	Species	Experimental period and design	Dose and route	General toxicity	Effects on reproductive organs or reproduction	Remarks
		microscopy. Oocyte quality was microscopically assessed. Biochemical indicators of oocyte oxidative stress were investigated. It is not clear in how many mice the various parameters were examined.	Test item: Sodium molybdate dihydrate Purity not mentioned.		<p>accompanied by increased GPx activities.</p> <ul style="list-style-type: none"> • Decreased ovary index, and increased rate of abnormal oocyte morphology at 8 mg/kg bw/day. • Morphologically abnormal ovarian mitochondria at 4 and 8 mg/kg bw/day. • Changes in antioxidant activity at higher dose levels: <ul style="list-style-type: none"> ○ Reduced superoxide dismutase activity and increased malondialdehyde contents at 4 and 8 mg/kg bw/day. ○ Decreased GPx activity at 8 mg/kg bw/day. 	performed or generated according to GLP.
Wang et al., 2016 (22)	Kunming mice, males. N= 20/dose	100-day study, no guideline. Male mice received low and high molybdenum diets	400 mg Mo/L drinking water for 100 days,	Decreased body weight, serum total protein and albumin, and superoxide dismutase.	<ul style="list-style-type: none"> • Reduction of sperm density and increased teratosperm. • Decreased superoxide dismutase, total 	Molybdenum compound not specified. The text was not consistent with the

Reference	Species	Experimental period and design	Dose and route	General toxicity	Effects on reproductive organs or reproduction	Remarks
		<p>(with normal or low copper levels) and additional molybdenum in drinking water for 100 days. Serum antioxidant status of superoxide dismutase (SOD), malondialdehyde (MDA), glutathione peroxidase (GSH-Px), and total antioxidant capacity (T-AOC) was analysed. Morphological changes in testicular tissue were examined with hematoxylin and eosin staining and ultrastructural changes were monitored by transmission electron microscopy.</p>	<p>corresponding to 80 mg/kg bw/day²¹. Test item: Molybdenum compound and purity not specified. Control group was included.</p>		<p>antioxidant capacity and increased malondialdehyde level in testicular tissue.</p> <ul style="list-style-type: none"> • Degenerated and atrophic germinal cells lumina of the tubules. • Morphological changes in many spermatogenic cells, including reduced amounts of chromatin, cellular nuclear volume loss, endoplasmic reticulum dilation, and nuclear membrane breakage or disappearance. Extensive vacuolization and swelling of the mitochondria of the spermatogenic cells and sperms. 	<p>figures, resulting in uncertainty in the data.</p>

²¹ Conversion from mg/L to mg/kg bw/day is based on an assumed water consumption of 5 ml for a mouse weighing 25 grams, based on the default values as reported in ECHA guidance R.8, version 2.1, Table 8-17

90-day rat study (Murray et al., 2014)

In a 90-day study in Sprague–Dawley rats the sub-chronic toxicity of sodium molybdate dihydrate, given via diet at dose levels of 0, 5, 17 or 60 mg molybdenum/kg bw/day, was investigated (15). The study complied with OECD Test Guideline (TG) 408, with additional examination of oestrus cycles and sperm count, motility, and morphology from OECD TG 416. The overall no-observed-adverse-effect level (NOAEL) was 17 mg molybdenum/kg bw/day, based on effects on body weight, body weight gain, food conversion efficiency and renal histopathology (females only) at 60 mg molybdenum/kg bw/day. No adverse effects were observed in the high dose animals after the 60-day recovery period, with the exception that male rats did not fully recover from reduced body weight. No treatment-related adverse effects on reproductive organ weights or histopathology, oestrus cycles or sperm parameters were observed at any dose level (Table 14 and 15).

Table 14 Length of oestrus cycles, number of cycles and number of females with irregular cycles among female rats in the 90 day toxicity study of sodium molybdate dihydrate^a (15)

Dose (mg Mo/kg bw/day)	Mean length of oestrus cycle, days (No. of rats)	Mean number of cycles (No. of rats)	Number of females with irregular cycles
0	4.0 ± 0.11 (19) ^b	4.3 ± 0.7 (19) ^b	1
5	4.1 ± 0.17 (10)	3.9 ± 0.7 (10)	1
17	4.0 ± 0.26 (10)	4.1 ± 0.9 (10)	1
60	4.1 ± 0.29 (20)	4.0 ± 0.9 (20)	1

^a Daily vaginal smears were taken from each female at approximately the same time each day and stage of oestrus determined for 3 weeks starting after completing 6 weeks of treatment.

^b Mean ± standard deviation (n).

A slight, but statistically significant decrease in the percentage of progressively motile sperm observed at 60 mg molybdenum/kg bw/day was considered by the study authors to be likely attributable to the control group having a value that approached the upper end of the range for historical controls (mean of 59.8 ± 16.2%). Serum blood, liver and kidney samples were analysed for molybdenum, copper, zinc, manganese, iron, cobalt and selenium; high levels of molybdenum and copper were found in the serum, blood, liver and kidneys of rats treated with 60 mg molybdenum/kg bw/day (15).

Table 15 Mean sperm motility and counts among male rats at the terminal sacrifice in the 90 day toxicity study of sodium molybdate dihydrate (10 animals per group) (15).

Dose (mg Mo/kg bw/day)	Right vas deferens ^a		Right cauda epididymis ^a		Right testis ^a	
	Motile sperm (%)	Progressively motile sperm (%)	Weight (g)	Sperm count (millions/g)	Weight (g)	Spermatid count (millions/g)
0	97.3 ±2.6	69.4 ±10.9	0.374 ±0.036	542.7 ±120.5	1.710 ±0.080	91.8 ±21.5
5	97.9 ±2.0	60.4 ±14.4	0.399 ±0.045	640.1 ±97.4	1.869 ±0.169	91.4 ±13.9
17	98.0 ±1.4	65.6 ±8.1	0.393 ±0.038	649.0 ±152.2	1.797 ±0.135	89.4 ±18.7
60	98.1 ±1.3	59.0 ±6.8 ^b	0.374 ±0.032	520.9 ±132.1	1.745 ±0.132	84.7 ±17.1

^a Mean ± SD

^b Significantly different from the control group (p < 0.05).

Dose-range finding study for the two-generation study (ECHA, 2016, study #003 and #004)

A dose-range finding study was performed to determine the dose levels for the two-generation study by Murray et al. (2019) (see below). Sprague-Dawley rats (10/sex/dose group) were administered sodium molybdate dihydrate at 0, 3, 20 or 40 mg Mo/kg bw/day via drinking water or via diet. Sperm parameters, litter observations, postmortem examinations of parental animals and offspring, reproductive indices and offspring viability indices were examined. The range-finder and the main study each included three dose levels via drinking water. The range-finder also included three dose levels via the diet; for the main study, only the top-dose was administered via the diet.

Test item related reduction of the body weight was observed in the males of the 40 mg Mo/kg bw/day exposure group via drinking water, and at the same dose level in the males and females exposed via diet. A dose related increase in molybdenum levels was observed in serum, liver and kidney of parental animals and in pups (no quantitative data available). This indicated absorption of molybdenum from both diet and drinking water with levels generally higher from diet than from drinking water in both parents and pups.

Pregnancy rate was reduced in the 40 Mo/kg bw/day drinking water exposure group and was outside the historical control average. A reduction in the number of live born pups with an increase in still born pups was observed upon treatment via drinking water. The stillborn pups were all in a single litter and the overall mean litter size did not differ between groups. No treatment-related effects were observed in any of the other parameters.

Based on the results of this range-finder study, doses as high as 40 mg Mo/kg bw/day were selected by the study authors for the evaluation in the full multigenerational study (17).

Two-generation rat study (Murray et al., 2019)

In a multigenerational study, performed according to OECD TG 416, groups of 24 male and 24 female Sprague-Dawley rats were administered sodium molybdate dihydrate at 0, 5, 17, or 40 mg molybdenum (Mo)/kg bw/day in the drinking water or 40 mg Mo/kg bw/day in the diet over two generations to assess reproductive toxicity (18). The dose levels were chosen based on a range-finder study in rats given sodium molybdate dihydrate in the drinking water or diet (see above). The drinking water route of administration was chosen as the main route of exposure for this study because it allowed the authors comparison with a study by Fungwe et al. (23) and because the range-finder study suggested a possible decrease in the pregnancy rate (6/10) at 40 mg Mo/kg bw/day in the drinking water, but not in the diet.

A statistically significant increase in the average number of primordial follicles was observed in the left ovary of parental females at 17 mg/kg bw/day (drinking water) and 40 mg/kg bw/day (diet). This was also observed in the F1 generation in the right, left, and combined ovaries at 17 and 40 mg/kg bw/day (drinking water) and 40 mg/kg bw/day (diet). All values were within the historical control range.

The percent of sperm with no head was statistically significantly increased in the parental generation given 40 mg/kg bw/day in the diet compared to the control value (see Table 17). A slight increase (not statistically significant) was observed in the percent of no head sperm in the group given 40 mg/kg bw/day in drinking water. In both cases, they were largely attributable to one male each. Average values were within historical control range.

No other adverse effect on reproductive function was observed at any dose level in either generation as indicated by no significant dose-related effect on oestrus cycles, sperm parameters, mating, fertility, gestation, litter size, pup survival or growth (Table 16, 17 and 18). Also no adverse effects on development were observed, as described in 8.2.1.

Systemic toxicity, including decreased body weight, food consumption (males only) and water consumption, was observed among both sexes given 40 mg Mo/kg bw/day in the diet. Serum levels of Mo and copper were increased in a dose-related manner (18).

Table 16 Reproductive performance of P and F1 generation male and female rats exposed to sodium molybdate dihydrate in the drinking water or diet (18).

	Dose Group (mg Mo/kg bw/day)				
	0	5 (water)	17 (water)	40 (water)	40 (diet)
P generation males					
Rats in cohabitation, N	24	24	24	23	24
Days in cohabitation, mean ± SD	2.9 ±2.0	3.2 ±1.7	2.8 ±1.9	2.8 ±1.1	2.7 ± 1.2
Mating Index, N/N (%) ^a	23/24 (95.8)	23/24 (95.8)	24/24 (100.0)	23/23 (100.0)	24/24 (100.0)
Fertility Index, N/N (%) ^b	22/23 (95.6)	22/23 (95.6)	23/24 (95.8)	22/23 (95.6)	19/24 (79.2)
F1 generation males					
Rats in cohabitation, N	24	24	22	24	24
Days in cohabitation, mean ± SD	2.7 ±2.0	2.5 ±1.7	2.3 ±1.0	2.9 ±2.4	3.0 ±2.2
Mating Index, N/N (%) ^a	24/24 (100.0)	24/24 (100.0)	22/22 (100.0)	22/24 (91.7)	23/24 (95.8)
Fertility Index, N/N (%) ^b	22/24 (91.7)	23/24 (95.8)	21/22 (95.4)	19/22 (86.4)	22/23 (95.6)
P generation females					
Rats evaluated, N	24	24	24	24	24
Oestrus cycles/14 days, mean ± SD	3.4 ±0.6	3.4 ±0.6	3.4 ±0.6	3.1 ±0.6	3.3 ±0.7
Females with persistent oestrus, N (%)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Females with persistent dioestrus, N (%)	0 (0.0)	1 (4.2)	0 (0.0)	2 (8.3)	0 (0.0)
Rats in cohabitation, N	24	24	24	24	24
Days in cohabitation, mean ± SD	2.9 ±2.0	3.2 ±1.7	2.8 ±1.9	3.0 ±1.2	2.7 ±1.2
Mating Index, N/N (%) ^a	23/24 (95.8)	23/24 (95.8)	24/24 (100.0)	24/24 (100.0)	24/24 (100.0)
Fertility Index, N/N (%) ^b	22/23 (95.6)	22/23 (95.6)	23/24 (95.8)	23/24 (95.8)	19/24 (79.2)
F1 generation females					
Rats evaluated, N	24	24	24	24	24
Oestrus cycles/14 days, mean ± SD	3.1 ±0.7	3.3 ± 0.5	3.3 ±0.6	3.1 ±0.6	3.2 ±0.6
Females with persistent oestrus, N (%)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Rats in cohabitation, N	1 (4.2)	0 (0.0)	1 (4.2)	1 (4.2)	1 (4.2)
Days in cohabitation, mean ± SD	24	24	24	24	24
Mating Index, N/N (%) ^a	24/24 (100.0)	24/24 (100.0)	24/24 (100.0)	24/24 (100.0)	23/24 (95.8)
Fertility Index, N/N (%) ^b	22/24 (91.7)	23/24 (95.8)	23/24 (95.8)	21/24 (87.5)	22/23 (95.6)

^a Number of rats that mated/number of rats in cohabitation.

^b Number of pregnancies/number of rats that mated.

Table 17 Semen evaluation of the P and F1 generation male rats exposed to sodium molybdate dihydrate in the drinking water or diet.

	Dose Group (mg Mo/kg bw/day)				
	0	5 (water)	17 (water)	40 (water)	40 (diet)
P generation females and litters					
Rats evaluated, N	24	24	24 ^a	23	24
Sperm density ^b cauda epididymis, mean ± SD	644 ± 377	675 ± 303	672 ± 391	756 ± 396	608 ± 314
Spermatid density ^c testes, mean ± SD	51 ± 51	52 ± 46	52 ± 47	54 ± 44	47 ± 34
% Motile sperm, mean ± SD	89 ± 7	89 ± 10	90 ± 6	88 ± 5	87 ± 14
% Normal morphology sperm, mean ± SD	96 ± 2.8	95 ± 5.2	94 ± 4.4	93 ± 10	91 ± 17
% Abnormal morphology sperm, mean ± SD	3.7 ± 2.8	3.9 ± 2.5	5.6 ± 4.4	6.8 ± 10	8.8 ± 17
% Detached head sperm, mean ± SD	2.2 ± 2.0	3.1 ± 3.9	2.8 ± 1.4	3.4 ± 5.0	5.1 ± 12
% No head sperm, mean ± SD	1.2 ± 0.9	1.1 ± 1.3	1.3 ± 1.3	2.6 ± 4.9	3.1 ± 5.5*
% Broken flagellum sperm, mean ± SD	0.4 ± 0.5	0.7 ± 0.7	0.9 ± 2.3	0.7 ± 0.9	0.9 ± 1.5
F1 generation females and litters					
Rats evaluated, N	23 ^d	24	22 ^{d,e}	24	24
Sperm density ^b cauda epididymis, mean ± SD	1652 ± 485	1649 ± 388	1747 ± 648	1784 ± 544	2011 ± 578
Spermatid density ^c testes, mean ± SD	406 ± 195	350 ± 116	316 ± 92	331 ± 150	280 ± 90
% Motile sperm, mean ± SD	68 ± 16	67 ± 10	64 ± 12	66 ± 18	68 ± 11
% Normal morphology sperm, mean ± SD	96 ± 1.9	95 ± 1.8	97 ± 2.2	97 ± 2.0	97 ± 2.4
% Abnormal morphology sperm, mean ± SD	3.9 ± 1.9	5.0 ± 1.8	3.1 ± 2.2	3.2 ± 1.9	3.2 ± 2.4
% Detached head sperm, mean ± SD	2.2 ± 1.1	3.0 ± 1.3	1.9 ± 1.7	1.9 ± 1.3	2.3 ± 1.8
% No head sperm, mean ± SD	1.5 ± 1.3	1.4 ± 0.8	1.1 ± 1.0	0.9 ± 0.8	0.9 ± 0.8
% Broken flagellum sperm, mean ± SD	0.2 ± 0.4	0.5 ± 0.7	0.2 ± 0.4	0.3 ± 0.6	0.04 ± 0.14

* Statistically significantly different from control value, $p < 0.05$.

^a Two males in the 17 mg Mo/kg bw/day group were excluded from the summaries of the sperm morphology because fewer than 200 sperm were available for evaluation. One male had zero sperm in the sample for morphology; the other had 10 sperm in the sample.

^b Sperm density expressed as million sperm/mL was calculated by dividing the sperm count by the volume in the image area and adjusting for dilution.

^c Spermatid density expressed as million spermatid/mL was calculated by dividing the spermatid count by the volume in the image area and adjusting for dilution.

^d Excludes one rat that was euthanized due to adverse clinical observations.

^e Excludes one rat that was discovered to be a female on PND 35.

Table 18 Natural delivery observations among P and F1 generation females exposed to sodium molybdate dihydrate in the drinking water or diet (18).

	Dose Group (mg Mo/kg bw/day)				
	0	5 (water)	17 (water)	40 (water)	40 (diet)
P generation females and litters					
Number pregnant/number tested, N (%)	22/24 (91.7)	22/24 (91.7)	23/24 (95.8)	23/24 (95.8)	19/24 (79.2)
Delivered litters, N (%)	22 (100.0)	22 (100.0)	23 (100.0)	23 (100.0)	19 (100.0)
Duration of gestation (days), mean ± SD	22.5 ± 0.5	22.6 ± 0.5	22.6 ± 0.5	22.6 ± 0.5	22.6 ± 0.5
Implantation sites/delivered litter, mean ± SD	15.8 ± 2.9	16.1 ± 1.9	16.6 ± 2.0	15.4 ± 2.4	15.9 ± 1.5
Gestation Index, % ^a	100	100	100	100	100
Litter size (live+stillbirths), mean ± SD	15.2 ± 3.0	15.2 ± 2.1	15.5 ± 1.9	14.6 ± 2.4	15.3 ± 1.7
Liveborn, mean ± SD	15.1 ± 3.0	15.1 ± 2.2	15.3 ± 2.0	14.5 ± 2.4	15.1 ± 1.8
Stillborn, N (%)	1 (0.3)	3 (0.9)	4 (1.1)	3 (0.9)	3 (1.0)
Viability Index, % ^b	98.5	99.1	99.4	97.9	99.3
Lactation Index, % ^c	98.2	98.8	99.1	99.4	99.6
% Male pups/litter on PND 0, mean ± SD	50.2 ± 15.6	50.6 ± 13.0	52.3 ± 14.0	53.0 ± 14.2	54.2 ± 14.8
% Male pups/litter on PND 21, mean ± SD	50.5 ± 15.8	50.5 ± 14.0	52.4 ± 13.8	52.8 ± 14.3	53.9 ± 15.4
F1 generation females and litters					
Number pregnant/number tested, N (%)	22/24 (91.7)	23/24 (95.8)	23/24 (95.8)	21/24 (87.5)	22/24 (91.7)
Delivered litters, N (%)	22 (100.0)	23 (100.0)	23 (100.0)	21 (100.0)	22 (100.0)
Duration of gestation (days), mean ± SD	22.9 ± 0.3	23.0 ± 0.2	22.9 ± 0.3	23.0 ± 0.3	22.9 ± 0.4
Implantation sites/delivered litter, mean ± SD	14.8 ± 1.4	15.5 ± 1.8	14.9 ± 3.4	15.1 ± 2.1	15.3 ± 2.4
Gestation Index, % ^a	100	100	100	100	100
Litter size (live+stillbirths), mean ± SD	14.3 ± 1.3	15.4 ± 2.2	14.3 ± 3.8	14.5 ± 1.9	14.9 ± 2.2
Liveborn, mean ± SD	14.1 ± 1.4	15.4 ± 2.2	14.0 ± 3.8	14.5 ± 1.9	14.9 ± 2.2
Stillborn, N (%)	4 (1.) ^d	1 (0.3)	6 (1.8)	0 (0.0)	0 (0.0)
Viability Index, % ^b	97.7	98.3	97.8	99.3	97.9
Lactation Index, % ^c	99.0	98.8	98.4	99.7	98.8
% Male pups/litter on PND 0, mean ± SD	47.8 ± 16.6	52.7 ± 17.8	53.7 ± 13.8	53.7 ± 12.2	54.3 ± 15.3
% Male pups/litter on PND 21, mean ± SD	47.5 ± 16.6	53.4 ± 17.3	53.1 ± 13.0	54.3 ± 12.5	53.8 ± 15.0

^a Number of rats with live offspring/number of pregnant rats.

^b Number of live pups on Day 3 postpartum/number of liveborn pups on Day 0 postpartum.

^c Number of live pups on Day 21 (weaning) postpartum/number of live pups on Day 3 postpartum.

^d SD as presented in publication by Murray et al. (2019). Part of the number is missing.

30-day male rat study (Khorami et al., 2020)

The effect of sodium molybdate on testicular toxicity was investigated in adult Wistar rats. Rats were treated by oral gavage with 0.05, 0.1, 0.2, and 0.4 mg/kg bw/day sodium molybdate (6 animals per dose group). Control animals received distilled water. Following 30 days of administration, animals were sacrificed for biochemical and histopathological assays.

No effects on sperm count, sperm viability, sperm morphology, sperm membrane integrity or sperm motility were observed upon treatment with sodium molybdate (Table 19). Also no effects of sodium molybdate were seen on oxidative stress parameters (Table 20). Histopathology of seminiferous tubules in the animals treated with sodium molybdate showed normal spermatocytes, spermatids and spermatozoa. Testicular pathology was ranked according to a classification by Johnsen²² with a score of 1-10, where 10 is complete spermatogenesis and perfect tubules and 1 is neither germ cells nor Sertoli cells present. All dose groups had a score 10. The level of aquaporin 9 expression, determined by immunohistochemistry staining of AQP9 in the testicular tissues, was not affected by sodium molybdate (Table 21) (19).

²² S.G. Johnsen, Testicular biopsy score count - a method for registration of spermatogenesis in human testis: normal values and result in 352 hypogonadal males, *Hormones* 1 (1970) 2-25. <https://doi.org/10.1159/000178170>

Table 19 Effect of sodium molybdate on sperm count, quality and motility in Wistar rats (19).

Sodium molybdate (mg/kg bw/day)	Sperm count (× million)	Viability (%)	Sperm morphology (%)	Membrane integrity (%)	Total motility (mean ± SEM)
0	55.22 ± 0.16	63.07 ± 0.95	84.66 ± 0.72	71.79 ± 1.79	69.95 ± 0.74
0.05	58.47 ± 0.99	65.30 ± 0.56	83.67 ± 0.74	71.87 ± 0.47	72.45 ± 0.46
0.1	54.98 ± 1.75	62.20 ± 0.88	84.41 ± 0.65	71.51 ± 1.74	71.22 ± 1.73
0.2	58.85 ± 1.05	65.23 ± 1.12	83.17 ± 0.92	67.92 ± 1.52	71.72 ± 1.59
0.4	53.81 ± 1.06	62.62 ± 0.91	86.06 ± 0.89	67.05 ± 1.05	72.22 ± 1.10

Values are expressed as mean ± SEM for six rats.

Table 20 Effect of sodium molybdate on antioxidant enzyme activities and MDA levels in the testicular tissue of Wistar rats (19).

Sodium molybdate (mg/kg bw/day)	CAT (U/mg protein)	SOD (U/mg protein)	GPX (U/mg protein)	MDA (nmol/mg protein)
0	22.58 ± 0.27	30.04 ± 0.33	44.31 ± 0.08	0.33 ± 0.01
0.05	23.02 ± 0.13	30.43 ± 0.13	44.74 ± 0.06	0.32 ± 0.02
0.1	22.29 ± 0.02	30.67 ± 0.41	44.08 ± 0.01	0.32 ± 0.01
0.2	23.14 ± 0.24	31.05 ± 0.27	44.77 ± 0.19	0.34 ± 0.01
0.4	22.32 ± 0.19	29.93 ± 0.07	44.52 ± 0.32	0.34 ± 0.01

CAT: catalase; SOD: superoxide dismutase; GPX: glutathione peroxidase; MDA: malondialdehyde.

Values are expressed as mean ± SEM for six rats.

Table 21. Effect of sodium molybdate on histopathological evaluation of testis and on AQP9 expression levels in healthy rats.

Sodium molybdate (mg/kg bw/day)	Johnsen's scores (mean ± S.E.M.)^a	Level of AQP9 expression (mean ± S.E.M.)
0	10 ± 0	3 ± 0
0.05	10 ± 0	3 ± 0
0.1	10 ± 0	3 ± 0
0.2	10 ± 0	3 ± 0
0.4	10 ± 0	3 ± 0

^a Classification on a scale of 1 to 10. Score 10 = Complete spermatogenesis and perfect tubules.

14-day male mouse study (Zhai et al., 2013)

The effects of molybdenum on sperm parameters and testicular oxidative stress were investigated in a sub-acute toxicity study (20). ICR mice were exposed to 0, 12.5, 25, 50, 100 or 200 mg/L sodium molybdate dihydrate in drinking water for 14 days (corresponding to 0, 2.5, 5, 10, 20 and 40 mg/kg bw/d based on ECHA guidance R.8). The results showed that the sperm parameters, including the epididymis index, sperm motility, sperm count, and morphology, increased by a moderate dose of molybdenum (5 mg/kg bw/day), but were negatively affected at high doses (≥ 20 mg/kg bw/day) (Table 22). Results for the abnormality rate were consistent with those findings, showing a decrease of abnormality at 5 mg/kg bw/day and an increase at 20 and 40 mg/kg bw/day. In addition, the changes of sperm parameters were accompanied with changes of the superoxide dismutase (SOD) activities, the glutathione peroxidase (GPx) activities, and the malondialdehyde (MDA) levels in testes (Table 23).

Table 22 Effects of molybdenum treatments on sperm parameters in mice (20).

Sodium molybdate dihydrate (mg/kg bw/day)	Epididymis Index $\times 10^{-3}$ ^a	Motility (%)	Concentration ($\times 10^6$/mL)	Abnormality rate** (% , n\geq200) ^b
0	2.37 \pm 0.06	62.82 \pm 0.03	5.01 \pm 0.08	28.23 \pm 1.79
2.5	2.53 \pm 0.08	67.18 \pm 0.08	5.90 \pm 0.03	24.21 \pm 0.87
5	2.70 \pm 0.05**	76.44 \pm 0.02**	6.98 \pm 0.11**	18.08 \pm 1.77**
10	2.20 \pm 0.07	55.31 \pm 0.13	4.35 \pm 0.02	29.60 \pm 3.47
20	2.00 \pm 0.11**	49.60 \pm 0.05*	3.93 \pm 0.05*	37.23 \pm 1.69**
40	1.97 \pm 0.08**	39.56 \pm 0.04**	3.68 \pm 0.02*	49.01 \pm 4.43**

^a Epididymis index: epididymis weight/body weight.

^b Abnormality rate: (no. sperm with abnormal morphology/ no. total spermatozoa) \times 100.

* P<0.05; **P<0.01

Data represent means \pm SD.

Table 23 Effects of molybdenum treatments on the SOD, GPx, and MDA levels of testes in mice (20).

Sodium molybdate dihydrate (mg/kg bw/day)	SOD activities (U/mg prot)	GPx activities (U/mg prot)	MDA contents (nM/mg prot)
0	221.49 \pm 4.43	36.32 \pm 0.31	0.9952 \pm 0.0178
2.5	225.85 \pm 4.07	42.52 \pm 1.39**	0.9862 \pm 0.0207
5	276.24 \pm 4.14**	44.41 \pm 1.95**	0.9594 \pm 0.0232
10	205.53 \pm 3.59	33.41 \pm 1.10*	1.2273 \pm 0.0245
20	190.20 \pm 3.19*	32.17 \pm 0.61**	1.3508 \pm 0.0270*
40	178.04 \pm 3.52*	31.06 \pm 0.21**	1.6129 \pm 0.0540**

* P<0.05; **P<0.01

Data represent means \pm SD.

14-day female mouse study (Zhang et al., 2013)

A 14-day toxicity test was performed to investigate the effects of sodium molybdate dihydrate on ovarian function. ICR adult female mice were exposed to molybdenum by free access to distilled water containing the sodium molybdate dihydrate at 0, 5, 10, 20, and 40 mg/L (corresponding to 0, 1, 2, 4 and 8 mg/kg bw/d) for 14 days. The levels of molybdenum in the standard diet are not provided. Compared to the control group, M II oocyte morphology, ovary index, and ovulation improved within the 1 mg/kg bw/day group, but were negatively affected by sodium molybdate dihydrate at 8 mg/kg bw/day (Table 24). These alterations accompanied the changes in superoxide dismutase (SOD), glutathione peroxidase (GPx), and malondialdehyde (MDA) levels in ovaries (Table 25). Morphologically abnormal ovarian mitochondria were observed at ≥ 4 mg/kg bw/day.

Table 24 Effects of molybdenum exposure on ovary parameters and MII oocyte morphology (21)

Sodium molybdate dihydrate in mg/L	Ovary index ($\times 10^{-3}$)*	No. ovulation	Abnormal rate/%**
0 (Control)	0.9985 \pm 0.0643	31.5000 \pm 2.7773	31.8542 \pm 2.3361
5 (1 mg/kg bw/d)	0.9807 \pm 0.0511	54.3500 \pm 2.8316 ^a	18.6753 \pm 0.8782 ^a
10 (2 mg/kg bw/d)	0.9854 \pm 0.0527	45.4500 \pm 2.6526 ^a	28.4928 \pm 1.9862
20 (4 mg/kg bw/d)	0.9533 \pm 0.0596	41.6000 \pm 3.2577 ^b	34.4304 \pm 3.0439
40 (8 mg/kg bw/d)	0.8394 \pm 0.0538 ^a	35.7368 \pm 3.6714	45.4962 \pm 3.3147 ^a

Mo: molybdenum; Data represent means \pm SD.

Compared with the control group, ^aP < 0.01, ^bP < 0.05.

* Ovary index = wet ovary weight / total wet body weight;

**Abnormal rate = (no. total ovulation - no. normal MII oocytes) / No. total ovulation.

Table 25 Effects of molybdenum exposure on the levels of SOD, GPx, and MDA in ovaries (21).

Sodium molybdate dihydrate (mg/L)	SOD activities (U/mg prot)	GPx activities (U/mg prot)	MDA contents (nM/mg prot)
0 (Control)	115.22 ± 14.32	32.68 ± 1.32	1.0394 ± 0.1282
5 (1 mg/kg bw/d)	118.56 ± 10.45	36.48 ± 2.03 ^b	0.9913 ± 0.0113
10 (2 mg/kg bw/d)	131.38 ± 11.52 ^b	39.57 ± 3.19 ^a	0.9756 ± 0.1428
20 (4 mg/kg bw/d)	100.71 ± 8.78 ^b	31.39 ± 2.44	1.6071 ± 0.1087 ^a
40 (8 mg/kg bw/d)	76.62 ± 6.40 ^a	22.61 ± 1.73 ^a	1.6808 ± 0.1886 ^a

Mo: molybdenum; SOD: superoxide dismutase; GPx: glutathione peroxidase; MDA: malondialdehyde. Data represent means ± SD.

Compared with the control group, ^a P < 0.01; ^b P < 0.05.

It was concluded that molybdenum affects oocyte quality possibly through regulating ovarian oxidative stress in a dose-dependent manner. The study authors concluded that it appears that Mo may improve ovarian function at a suitable concentration, which might be a candidate for the treatment of female infertility (21).

100-day male mouse study (Wang et al., 2016)

To evaluate the effects of dietary high molybdenum (HMo) and low copper (LCu) concentrations on reproductive toxicity of male Kunming mice, 80 mice were divided into 4 groups of 20. These groups were fed with the following: (1) normal control (NC) diet (NC group); (2) NC and HMo diets (HMo group); (3) LCu diet (LCu group); and (4) HMo and LCu diets (HMoLCu group). It was not specified which molybdenum compound was used. The HMo and HMoLCu groups were supplied with drinking water containing molybdenum at a concentration of 400 mg/L (corresponding to 80 mg/kg bw/day based on the ECHA guidance R.8). On the 50th and 100th day, antioxidant status was analysed and morphological changes in testicular tissue were evaluated. Results of the HMo group are presented below; any effects in the other groups are not reported here.

The results showed that administration of HMo decreased sperm density and increased the rate of teratosperm occurrence (no absolute data presented; text and figures not consistent). A significant increase in MDA content and a decrease in SOD and T-AOC contents in testicular tissue was observed in the high molybdenum group (Table 26).

Table 26 Determination of SOD, MDA, T-AOC and GSH-Px in the testicular tissues of mice ($n = 6$, mean \pm SD) (22).

Day	Variable	NC group	HMo group
50	SOD (U/mg prot)	72.28 \pm 2.39	66.96 \pm 2.58 ^a
	MDA (nmol/mg prot)	25.11 \pm 3.55	29.70 \pm 4.59
	T-AOC (U/mg prot)	2.35 \pm 0.24	2.12 \pm 0.22 ^a
	GSH-Px (U/mg prot)	51.10 \pm 3.41	47.68 \pm 3.45
100	SOD (U/mg prot)	71.77 \pm 3.58	65.91 \pm 3.56 ^a
	MDA (nmol/mg prot)	24.65 \pm 2.52	29.35 \pm 3.73 ^a
	T-AOC (U/mg prot)	2.23 \pm 0.20	1.98 \pm 0.19 ^a
	GSH-Px (U/mg prot)	49.25 \pm 4.36	46.34 \pm 4.04

SOD: superoxide dismutase, MDA; malondialdehyde, T-AOC: total antioxidant capacity; GSH-Px: glutathione peroxidase; NC: normal control; HMo: high molybdenum diet.

^a $p < 0.05$: compared with the NC group.

^b $p < 0.01$: compared with the NC group.

Histopathological examination of testicular tissue showed slight histological alterations in animals treated with molybdenum. Degenerated and atrophic germinal cells were detected in the atrophic lumina of the tubules in the HMo group.

In the NC group, spermatogenic cells and sperms showed normal features as observed under transmission electron microscopy. In the HMo group, many spermatogenic cells showed morphological changes, including reduced amounts of chromatin, cellular nuclear volume loss endoplasmic reticulum dilation, and nuclear membrane breakage or disappearance. The mitochondria of the spermatogenic cells and sperms showed extensive vacuolization, were swollen and were less dense than those of the controls. (22).

8.1.2 Human data

In the advice of the Health Council of the Netherlands on molybdenum in 2013 effects of molybdenum on semen quality (sperm count, sperm concentration, percent motile sperm, and sperm morphology), and metals in blood among men recruited through fertility clinics ($N=219$) were reported (1). Meeker et al. (2010) found molybdenum-dependent decreases in sperm concentration and normal morphology, when adjusted for age, current smoking, and the impact of multiple metals on semen quality simultaneously for sperm concentration and sperm morphology (24). In a subsequent study in the same group of men a significant inverse trend between molybdenum concentrations in blood and testosterone levels was found. In addition an interaction between high molybdenum levels and low zinc levels was observed.

Additional studies on the effects of molybdenum on sexual function and fertility in humans are presented below.

The studies in humans on the effects of molybdenum compounds on sexual function and fertility are summarized in Table 27; studies on pregnancy-related health outcomes in mother are summarized in Table 28.

Table 27 Summary of human studies of molybdenum and its compounds on sexual function and fertility

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
Female sexual function and fertility – Cross-sectional studies					
<ul style="list-style-type: none"> • Syrkasheva et al., 2021 (25) • Cross-sectional study • Moscow, Russia • 30 women (aged 18-39 years), residents of Moscow for the last 5 years • Couples who applied for assisted reproductive technology (ART) • 2017 to 2018 • Inclusion criteria: <ul style="list-style-type: none"> - no contraindications for ART - normal karyotype of both spouses - absence of severe male factor (100% teratozoospermia, absolute asthenozoospermia, all types of azoospermia) 	<ul style="list-style-type: none"> • Blood samples taken on day of transvaginal puncture • Concentration ($\mu\text{g/L}$) of molybdenum and 30 other elements determined using ICP-MS • LOD/LOQ not reported 	<ul style="list-style-type: none"> • ART included ovarian stimulation with gonadotropin releasing hormone antagonists, transvaginal ovarian puncture, and in vitro oocyte fertilization • blood sample 14 days after embryo transfer with measurements of: <ul style="list-style-type: none"> - Human chorionic gonadotropin ($\beta\text{-hCG}$) - antimullerian hormone (AMH) - free thyroxine (T4_{free}) • Clinical pregnancy defined as registration of the embryo's heartbeat 5 weeks after transfer. • Other outcome variables: <ul style="list-style-type: none"> - number of previous pregnancies - gynaecological diseases: endometriosis, myoma, inflammatory diseases of pelvic organs 	<ul style="list-style-type: none"> • Molybdenum was detected in all blood samples (median 0.705, range 0.4 to 1.15 $\mu\text{g/L}$) • Molybdenum concentration increased with increasing age ($r=0.384$; $p=0.036$) • No associations between molybdenum concentration and any of the health outcome parameters 	<ul style="list-style-type: none"> • Analyses not adjusted for potential confounders • No interpretation possible regarding absolute concentration levels of molybdenum in blood 	<p>Results of analyses for molybdenum not provided; in tables a selection of metals with statistically significant results only</p>

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<p>- age 18 through 39 years</p> <p>- BMI 19–25 kg/m²</p> <p>• Exclusion criteria:</p> <p>- use of donor gametes or surrogacy</p> <p>- obtaining ≤3 oocytes on day of transvaginal ovarian puncture</p>		<p>- primary or secondary infertility and duration</p> <p>- features of the ovarian stimulation protocol: duration of stimulation and total dose of gonadotropins</p> <p>- parameters of oogenesis and early embryogenesis, e.g. number of blastocysts obtained</p> <p>- levels of β-hCG, AMH and T4free in blood sample</p> <p>Statistical analysis:</p> <ul style="list-style-type: none"> • Pearson’s correlation coefficients and Mann-Whitney U test between molybdenum concentration and continuous health outcomes • Chi-square test for categorical outcomes 			
Male sexual function and fertility – Prospective cohort studies					
<p>Ashrap et al., 2020 (26)</p> <p>Prospective cohort study</p> <p>Mexico City, Mexico</p>	<p>Women: interview-based questionnaires at 3 visits during</p>	<p>Early adolescence fasting serum samples analysed for: oestradiol, testosterone, inhibin B, sex hormone-binding globulin (SHBG) and</p>	<p>Prenatal urinary molybdenum concentrations (µg/L): GM 19.5, median 25.7, IQR 12.7-42.9</p> <p>Peripubertal children’s molybdenum concentrations (µg/L): GM 46.6, median 50.2, IQR 33.7-67.1</p>	<p>Results not adjusted for exposure to other metal(loid)s.</p>	

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<p>997 women recruited in first trimester of pregnancy and their sons 1997 - 2004</p> <p>Inclusion criteria: not planning to leave the area within 5 years; no history of infertility, diabetes, or psychosis; not consuming alcoholic beverages daily during pregnancy; no addiction to illegal drugs; no diagnosis of a high-risk pregnancy; being pregnant with singleton.</p>	<p>pregnancy, spot urine samples in 3rd trimester (n=212). Boys: spot urine samples in early adolescence (n=118, 8-14 years). Urinary molybdenum and 13 other metal(loid) concentrations measured with ICP-MS. LOD 2.9 µg/L</p>	<p>dehydroepiandrosterone sulphate (DHEA-S)</p> <p>Physical examination (sexual maturation) in early and late adolescence: Tanner staging of genital and pubic hair development and testicular volume assessed by two trained paediatricians.</p> <p>Statistical analyses: - multivariable linear regression to assess associations between urinary molybdenum and hormone levels - generalized estimating equation (GEE) to explore associations between urinary molybdenum and sexual maturation parameters - all analyses were adjusted for child age, BMI z-score, and urinary specific gravity (SG) as measure of urinary dilution, but not for other metal(loid)s</p>	<p>Differences (%) in peripubertal hormone concentrations associated with IQR increase in molybdenum concentration <u>in utero</u>:</p> <ul style="list-style-type: none"> - Oestradiol -1.1 (-10.0, 8.7) - Testosterone 51.3 (19.1, 92.4), remained statistically significant after correction for multiple testing - Inhibin B 2.9 (-6.9, 13.8) - SHBG -1.1 (-10.3, 9.0); - DHEA-S -0.1 (-12.5, 14.2) <p>No differences (%) in peripubertal hormone concentrations associated with IQR increase in molybdenum concentration <u>peripubertal</u>:</p> <ul style="list-style-type: none"> - Oestradiol -0.9 (-10.0, 9.3) - Testosterone -18.1 (-38.2, 8.6) - Inhibin B -2.1 (-12.3, 9.3) - SHBG 3.5 (-7.0, 15.1) - DHEA-S 5.5 (-8.8, 22.2) <p>No associations between in utero and peripubertal molybdenum concentrations and (changes in) genital development, pubic hair development, and testicular volume.</p> <p>Sensitivity analyses:</p>		

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
			<p>In prepubertal boys (n=94), difference (%) in testosterone concentrations associated with IQR increase in molybdenum concentration in utero 119.4 (31.5, 266). Similar estimates with and without adjustment for BMI or household SES.</p>		
Male sexual function and fertility – Cross-sectional studies					
<ul style="list-style-type: none"> Guzikowski et al., 2015 (27) Cross-sectional study 34 men (26-42 years) from primary infertile couples in the rural area of Opole, Poland January-June 2009 Inclusion criteria: <ul style="list-style-type: none"> regular unprotected intercourse for at least 12 months without conception 	<ul style="list-style-type: none"> Semen samples were collected after abstinence period of 5 days Concentration of molybdenum and 8 other metals determined using ICP-MS with time-of-flight analyser LOD/LOQ not reported 	<ul style="list-style-type: none"> Semen quality according to WHO criteria: <ul style="list-style-type: none"> volume pH (not reported) sperm count sperm motility sperm morphology Statistical analysis <ul style="list-style-type: none"> Pearson's correlation coefficients between molybdenum concentration and semen quality parameters Cluster analysis using all metals 	<ul style="list-style-type: none"> Concentrations of molybdenum were correlated with the concentrations of all other metals ($p < 0.05$) except zinc 23 men with one or more deviating values for sperm quality parameters: <ul style="list-style-type: none"> sperm concentration $< 20 \times 10^6/\text{ml}$ $< 50\%$ motile sperm $< 15\%$ normal forms 11 men with sperm quality parameter values in the normal range Pearson's correlation coefficients for molybdenum concentration with semen parameters (N=34): <ul style="list-style-type: none"> Sperm count: $r = 0.32$ ($p > 0.05$) Sperm motility: $r = 0.19$ ($p > 0.05$) Sperm morphology: $r = 0.15$ ($p > 0.05$) 	<ul style="list-style-type: none"> Confounding is possible since no other factors were considered in crude correlation analyses 	<ul style="list-style-type: none"> Statistical analysis and reporting are poor; some descriptions in the results section do not match the tables Unclear whether association with motility can be attributed to molybdenum (due to correlations)

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<ul style="list-style-type: none"> - no previous fertility treatment - no known causes of infertility in patient's partner 		<ul style="list-style-type: none"> - Euclidean distance between objects as a measure of similarity - Ward's method was used as an agglomerative hierarchical clustering procedure - Multivariate techniques of principal component analysis and cluster analysis were employed to identify possible sources of the selected metals in the semen of the two groups of patients • Level of significance: $p \leq 0.05$ 	<ul style="list-style-type: none"> • Factors of correlations between metal-to-metal and semen count-to-motility-to-morphology - Sperm count: $r=0.3732$ ($p=0.115$) - Sperm motility: $r=0.5451$ ($p=0.016$) - Sperm morphology: $r=0.2763$ ($p=0.252$) Multivariate techniques of principal component analysis and cluster analysis were elaborated for metals other than molybdenum (Zn and Cd) 		with other metals)
<ul style="list-style-type: none"> • Lewis & Meeker, 2015 (28) • Cross-sectional study • United States (general population) • 484 men, aged 18-55 years, participating in NHANES 	<ul style="list-style-type: none"> • Urine samples were collected • Concentration of molybdenum ($\mu\text{g/L}$) and 5 other metals (+3 metals in serum) 	<ul style="list-style-type: none"> • Blood samples were collected • Serum testosterone was measured with HPLC-MS/MS • Statistical analysis - Associations between urinary molybdenum concentration and log-transformed serum 	<ul style="list-style-type: none"> • Urinary molybdenum detected in all 484 urine samples (median 46.05, IQR 23.50-76,70) Association between urinary molybdenum modelled <i>continuously</i> and testosterone: - Unadjusted -2.67 (CI -5.66, 0.41); $p=0.089$ - Adjusted -4.26 (CI -7.70, -0.69); $p=0.020$ 		<ul style="list-style-type: none"> • Distribution and other descriptive results on serum testosterone levels not provided

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<ul style="list-style-type: none"> • 2011-2012 • 	<ul style="list-style-type: none"> determined using ICP-MS • LOD 0.99 µg/L • 	<p>testosterone concentration assessed using multivariable linear regression models</p> <ul style="list-style-type: none"> - Models were done using molybdenum concentration [1] continuously (µg/L); [2] in quartiles; and [3] in quintiles (as sensitivity analysis); the latter two to explore potential non-linear relationships - All measures of association were expressed as % change in serum testosterone concentration associated with a doubling (100% increase) in molybdenum concentration and adjusted for age, BMI, income, race, serum cotinine, and urinary creatinine • 	<p>(% change associated with a doubling increase in molybdenum concentration)</p> <ul style="list-style-type: none"> • Association between urinary molybdenum in <i>quartiles</i> and testosterone: <ul style="list-style-type: none"> - Results from adjusted models are presented as Figure and not as exact numbers. ±10% lower serum testosterone in Q2, Q3, and Q4 compared with Q1 of urinary molybdenum level. P for inverse trend 0.107 • Association between urinary molybdenum in <i>quintiles</i> and testosterone: p for inverse trend 0.020. More details on regression estimates not provided. • 		
<ul style="list-style-type: none"> • Skalnaya et al., 2015 (29) • Cross-sectional study • Orenburg, Russia • 148 men 	<ul style="list-style-type: none"> • Semen samples were collected according to current WHO recommendations 	<ul style="list-style-type: none"> • Spermogram analysis according to WHO manual using the recommended normal ranges - ejaculate volume - absolute and relative sperm count - sperm motility 	<ul style="list-style-type: none"> • Inverse association ($p < 0.05$) between molybdenum concentration and seminal liquid volume • molybdenum concentration was not significantly ($p > 0.05$) associated with <ul style="list-style-type: none"> - sperm count - sperm concentration 	<ul style="list-style-type: none"> • Analyses not adjusted for potential confounders • No interpretation possible regarding absolute concentration levels 	<ul style="list-style-type: none"> • Reporting is poor with some essential information missing Data from tables (a.o.)

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<ul style="list-style-type: none"> • Study period not reported, but < 2016 • Other details and inclusion/exclusion criteria not provided 	<ul style="list-style-type: none"> • Concentration ($\mu\text{g/mL}$ of ejaculate) of molybdenum and 19 other metals determined using ICP-MS • LOD/LOQ not reported 	<ul style="list-style-type: none"> - sperm vitality • Statistical analysis: Spearman's rank correlation coefficients between molybdenum concentration and semen quality parameters. 	<ul style="list-style-type: none"> - sperm motility - sperm vitality 	of molybdenum in semen	correlations between molybdenum concentrations and semen quality parameters) were missing
<ul style="list-style-type: none"> • Zeng et al., 2015 (30) • Cross-sectional study • Wuhan, China • 394 men blindly and randomly selected from 2090 men • April 2011 – May 2012 • Inclusion criteria: <ul style="list-style-type: none"> - Presenting at reproductive centre for semen analysis • Exclusion criteria: <ul style="list-style-type: none"> - Azoospermia, orchiditis, epididymitis, 	<ul style="list-style-type: none"> • Single spot urine samples • Concentration of molybdenum and 12 other metals determined using ICP-MS • LODs ranged from 0.001 to 0.29 $\mu\text{g/L}$ • Expressed as creatinine-adjusted urinary concentration ($\mu\text{g/g}$ creatinine) 	<ul style="list-style-type: none"> • Semen samples provided and analysed according to WHO guidelines: <ul style="list-style-type: none"> - semen volume - sperm count - sperm concentration (million/mL) - sperm motility (% A+B motile sperm) - sperm normal morphology (%) - sperm abnormal head (%) • Statistical analysis: <ul style="list-style-type: none"> - Multivariable logistic regression analysis for associations between quartiles of creatinine- 	<ul style="list-style-type: none"> • Molybdenum was detected in all urine samples, (median 42, range 8–425 $\mu\text{g/g}$ creatinine) and associated with almost all other metals • Sperm concentration <20 million/mL: n=46 • Sperm motility <50% motile: n=222 • Sperm count <40 million: n=38 • Control group with all the three parameters \geqreference values: n=169 • No significant associations between quartile of molybdenum concentration ($\mu\text{g/g}$ creatinine) and below-reference semen quality parameters 		

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
vesiculitis, vasectomy, undescended testicle, injury of testis, hernia repair complicated by testicular atrophy, and endocrine disease (e.g., diabetes, thyroid, or adrenal disorders).	<ul style="list-style-type: none"> Quartiles of molybdenum concentration Q1: <28.99 Q2: 28.99-41.63 Q3: 41.64-68.46 Q4: >68.46 µg/g creatinine 	<p>adjusted urinary molybdenum concentrations and sperm concentration, motility, count, and morphology dichotomized using WHO reference values</p> <p>- All models adjusted for age, abstinence time and smoking status</p>	<ul style="list-style-type: none"> Odds Ratios and 95% confidence intervals are presented as Figure and not as exact numbers P-values for trend are provided: <ul style="list-style-type: none"> Sperm concentration: p=0.58 Sperm motility: p=0.29 Sperm count: p=0.52 No significant associations between quartile of molybdenum concentration (µg/g creatinine) and semen morphology parameters Regression coefficients and 95% confidence intervals are presented as Figure and not as exact numbers. P-values for trend are provided: <ul style="list-style-type: none"> Sperm normal morphology: p=0.59 Sperm abnormal head: p=0.54 		
<ul style="list-style-type: none"> Wang et al., 2016 (31) Cross-sectional study Wuhan, China 1052 men of subfertile couples (mean age 32 years) visiting 	<ul style="list-style-type: none"> Two spot urine samples several hours apart (mean: 4.4 ± 3.7 h) Concentration (µg/L) of molybdenum and 17 other metals 	<ul style="list-style-type: none"> Serum hormones in blood samples drawn between 08:30 and 11:30 AM (n=511): <ul style="list-style-type: none"> oestradiol follicle stimulating hormone (FSH) luteinizing hormone (LH) 	<ul style="list-style-type: none"> Molybdenum was detected in all urine samples <ul style="list-style-type: none"> First sample: median 68, IQR 44-106 µg/L Second sample: median 67, IQR 36-103 µg/L No associations between quartiles of average molybdenum concentration and levels of serum reproductive 		

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<p>reproductive center for semen analysis</p> <ul style="list-style-type: none"> • March –June 2013 • Exclusion criteria: <ul style="list-style-type: none"> - Azoospermia, self-reported health conditions that may affect male reproductive health or urinary metals excretion, occupational exposure to metals 	<p>determined using ICP-MS</p> <ul style="list-style-type: none"> • LOQ not specified (but somewhere between 0.0013 to 0.29 µg/L) • Geometric mean concentrations were calculated from the results of the 2 samples and grouped into quartiles 	<ul style="list-style-type: none"> - sex hormone-binding globulin (SHBG) - total testosterone (T) - derived measures: total T/LH ratio, free androgen index, free T • Sperm characteristics in semen samples collected after 2-7 days of abstinence: <ul style="list-style-type: none"> - spermatozoa apoptosis (n=460), reported as % necrotic, % apoptotic, and % viable - DNA integrity by neutral comet assay (n=516): tail DNA %, tail length and tail distributed moment • Statistical analysis: <ul style="list-style-type: none"> - Associations between quartiles of molybdenum level (averaged over 2 samples) and markers of male reproductive health outcomes assessed using multivariable linear regression models, adjusted 	<p>hormones (oestradiol, FSH, LH, SHBG)</p> <ul style="list-style-type: none"> • Total T/LH ratio inversely associated with quartiles of average molybdenum concentration (FDR-adjusted p for trend 0.02), also when simultaneously adjusted for confounders and multiple metals: <ul style="list-style-type: none"> <25th: 0.00 (Reference) 25th–50th: –5.6% (–19%, 6.2%) 50th–75th: –8.9% (–25%, 5.1%) >75th: –16% (–34%, –1.0%) P for trend 0.03 • Dose-response association between average molybdenum concentration and total T/LH ratio: <ul style="list-style-type: none"> - P for overall association: 0.01 - P for non-linear association 0.20 <p>No associations between quartiles of average molybdenum concentration and apoptosis markers or sperm DNA integrity parameters (Annexin V+/PI-spermatozoa, PI+ spermatozoa, Annexin V-/PI-spermatozoa)</p> <ul style="list-style-type: none"> • No associations between quartiles of molybdenum concentration and 		

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
		for: age, BMI, smoking status, daily cigarette consumption, and urinary creatinine - False-discovery rate (FDR) correction to account for multiple testing - Restricted cubic spline functions to assess dose-response associations with reference values set to median for associations found in multivariable analysis	comet parameters (Tail%, Tail length, Tail distributed moment) • Analyses with molybdenum concentrations from separate urine samples did not yield different findings		
<ul style="list-style-type: none"> • Zhou et al., 2016 (32) • Cross-sectional study • Wuhan, China • 207 men of subfertile couples visiting reproductive centre for semen analysis • March – June 2012 - • Exclusion criteria: <ul style="list-style-type: none"> - Azoospermia, orchiditis, epididymitis, vesiculitis, 	<ul style="list-style-type: none"> • Single spot urine samples • Concentration of molybdenum and 12 other metals determined using ICP-MS • LOD 0.004 µg/L • Expressed as creatinine-adjusted urinary concentration 	<ul style="list-style-type: none"> • Semen samples were collected after abstinence time ranging from ≤ 2 to ≥ 6 days • Neutral comet assay to assess sperm DNA damage: <ul style="list-style-type: none"> - percent DNA tail - tail length - tail distributed moment • Statistical analysis: Multivariable linear regression analysis to assess dose-response relationships between quartiles of creatinine-adjusted urinary 	<ul style="list-style-type: none"> • Molybdenum detected in all urine samples (median 39 µg/g creatinine) • No associations between quartiles of molybdenum concentration and comet assay parameters (Percent DNA tail: p for trend 0.42; Tail length: p=0.83; Tail distributed moment: p=0.36) 		

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
vasectomy, undescended testicle, varicocele, injury of testis, and hernia repair complicated by testicular atrophy	(µg/g creatinine) • Grouped into quartiles Q1: <26.77 Q2: 26.77-38.94 Q3: 38.95-58.60 Q4: >58.60	metal levels and comet assay parameters Adjustment for: - age - BMI - smoking status - abstinence time			
<ul style="list-style-type: none"> • Branch et al. (2021) (33) • Cross-sectional study • 356 men (aged 19-51 years) participating in Longitudinal Investigation of Fertility and the Environment (LIFE) Study • Michigan and Texas, USA • 2005-2009 • Inclusion criteria: - men from couples who were 	<ul style="list-style-type: none"> • Urine sample collected upon completion of baseline interview • Concentration (µg/L) of molybdenum and 14 other metal(loid)s determined using ICP-MS • LOD not reported • Expressed as creatinine-adjusted 	<ul style="list-style-type: none"> • Semen samples were collected twice at home: (1) day after enrolment interview; (2) approximately 1 month later (mean abstinence time 4 days) • Semen analysis included quantification of 7 endpoints: <ul style="list-style-type: none"> - total sperm count ($\times 10^6$/ejaculate) - semen volume (mL) - sperm concentration ($\times 10^6$/mL) - next day motility (%) - traditional morphology (%) 	<ul style="list-style-type: none"> • Molybdenum was detected in all urine samples (median=46.65 µg/L or 38.36 µg/g creatinine) • Analysis [i]: molybdenum concentration was only selected for inclusion for sperm motility (adjusted beta=+0.26% per µg/L molybdenum) • Analysis [ii]: no association between molybdenum and motility: beta=0.07 (95%CI -0.3 to 0.44), adjusted for race/ ethnicity, study site and urinary levels of As, Cr, Pb, Tl, Sn, W, and U • Analysis [iii]: no associations between molybdenum concentration and any of the semen endpoints 	<ul style="list-style-type: none"> • Cross-sectional data; no information on historical exposure to molybdenum 	<ul style="list-style-type: none"> • Unclear how data from the two separate semen samples per participant were treated in analyses

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<p>discontinuing contraception in an attempt to achieve pregnancy</p> <ul style="list-style-type: none"> - at least 18 years of age - married or in a committed relationship - able to communicate in English or Spanish - without physician-diagnosed infertility • Earlier publication describing study population and data collection methods in LIFE: Buck Louis et al. (2011) (34) 	<p>urinary concentration ($\mu\text{g per g creatinine}$)</p>	<ul style="list-style-type: none"> - DNA fragmentation index (%) - high DNA stainability (%) • Statistical analysis: <ul style="list-style-type: none"> [i] penalized LASSO regression models to identify and select metal(loid)s most likely to be predictive of each semen quality endpoint and potential confounders (using log-transformed molybdenum concentration)[ii] unpenalized multivariable linear regression models with metal(loid)s and confounders selected in the LASSO regression only - Potential confounders: abstinence time; age; race/ethnicity; alcohol consumption; BMI; education; household income; fathered previous pregnancy; urinary creatinine; current smoking status (serum cotinine); study site 	<ul style="list-style-type: none"> - total sperm count: -0.03 (-0.63 to 0.58) - semen volume: $+0.03$ (-0.08 to 0.14) - sperm concentration: -0.13 (-0.54 to 0.28) - next day motility: $+0.21$ (-0.12 to 0.54) - traditional morphology: -0.30 (-2.35 to 1.74) - DNA fragmentation index: $+0.73$ (-0.75 to 2.20) - high DNA stainability: -0.04 (-0.81 to 0.72) 		

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
		[iii] multivariable linear regression models for log-transformed molybdenum concentration and all semen endpoints, adjusted for confounders but NOT for other metal(loid)s			

Female sexual function and fertility – Cross-sectional studies

Syrkasheva et al. (2021) studied associations between the concentrations of molybdenum (among other metals) in blood and parameters of ART treatment in 30 subfertile women in Moscow, Russia (25). Associations between blood molybdenum concentration and reproductive outcomes were evaluated by calculating Pearson's correlation coefficients and using Mann-Whitney U or chi-square tests, without adjustment for other factors. No significant ($p < 0.05$) associations were found between concentrations of molybdenum and outcome parameters considered (including levels of β -hCG, AMH, and free T4 in blood, clinical pregnancy, number of previous pregnancies, gynaecological diseases, primary or secondary infertility, features of the ovarian stimulation protocol, and parameters of oogenesis and early embryogenesis). No results of the (all non-significant) associations for molybdenum were provided in the tables.

Male sexual function and fertility – Prospective cohort studies

In a prospective cohort study in Mexico City, **Ashrap et al.** collected urine samples from 212 women during their third trimester of pregnancy and their sons ($n=118$) at age 8–14 years (26). The prenatal mean urinary molybdenum concentration was 19.5 $\mu\text{g/L}$, and the boys' peripubertal mean concentration was 46.6 $\mu\text{g/L}$. Associations between urinary concentrations of molybdenum and sex hormones in early adolescence, and indicators of sexual maturation in early and late adolescence were studied. An interquartile range (IQR) increase in in utero molybdenum concentrations was associated with 51% (95% CI 19.1-92.4) higher testosterone concentrations (119% (31.5-266) in 94 boys who were prepubertal). No associations were found with oestradiol, Inhibin B, SHBG, and DHEA-S. Peripubertal molybdenum concentrations were not associated with any of the measured hormone concentrations. No association were observed between in utero or peripubertal molybdenum concentrations and (changes in) genital development, pubic hair development, and testicular volume either.

Male sexual function and fertility – Cross-sectional studies

Guzikowski et al. (2015) studied cross-sectional associations between molybdenum and other metal concentrations in semen and sperm count, motility, and morphology in 34 men (aged 26-42 years) from primary infertile couples in the rural area of Opole, Poland (27). Of these, 23 men had at least one sperm quality parameter below the reference value (sperm concentration $< 20 \times 10^6/\text{ml}$, $< 50\%$ motile sperm, and/or $< 15\%$ normal forms). No statistically significant ($p < 0.05$) correlations (Pearson's r) were found between molybdenum concentration and the three sperm quality parameters. These results were not adjusted for other covariables.

Lewis & Meeker (2015) studied the associations between urinary molybdenum (and other metals) and serum testosterone in 484 men aged 18-55 years from the NHANES general population cohort in the USA in 2011-2012 (28). An inverse association was found between urinary molybdenum concentration and testosterone (4.26% (95% CI: 0.69-7.70) lower serum testosterone concentration with a doubling (100% increase) in urinary molybdenum concentration), adjusted for age, BMI, income, race, serum cotinine, and urinary creatinine. When

the molybdenum concentration was modelled in quintiles in a multivariable linear regression model, a p-value for trend of 0.02 was found.

Skalnaya et al. (2015) studied the correlations between the concentrations of molybdenum (among other metals) in semen and with sperm quality in 148 volunteers in Orenburg, Russia (29). The authors reported an inverse association (Spearman's r , $p < 0.05$) between molybdenum concentration and seminal liquid volume, whereas no statistically significant associations were found with sperm count, sperm concentration, sperm motility, and sperm vitality. No results of measurements were provided, tables are missing, and results were not adjusted for other factors.

Zeng et al. (2015) studied the associations between urinary level of molybdenum (among other metals) and below-reference semen quality parameters in 394 men presenting for semen analysis at a reproductive centre in Wuhan, China (30). No associations (p for trend > 0.05) were found between quartiles of molybdenum spot urine concentration ($\mu\text{g/g}$ creatinine) and below-reference sperm concentration, sperm motility, sperm count, and sperm morphology in multivariable logistic regression analyses adjusted for age, abstinence time, and smoking status. Molybdenum was not retained in analytical models including multiple metals.

Wang et al. (2016) studied associations between urinary level of molybdenum (among other metals) and markers of male reproductive health in 1052 men of subfertile couples in Wuhan, China (31). Outcomes included spermatozoa apoptosis ($n=460$), sperm DNA-damage ($n=516$) and sex hormones in blood serum ($n=511$). Quartiles of average geometric mean molybdenum concentration ($\mu\text{g/g}$ creatinine) from two repeated urine samples were analysed in multivariable linear regression models, adjusted for age, BMI, smoking, and urinary creatinine and false-discovery rate. A lower total testosterone/luteinizing hormone ratio was associated with a higher level of molybdenum (p for trend 0.02). When also adjusting for other urinary metal levels, this ratio was 5.6%, 8.9%, and 16% lower for the 2nd, 3rd, and 4th quartiles of molybdenum concentration, respectively, compared to the first quartile (p for trend 0.03). No statistically significant ($p < 0.1$) associations with molybdenum were found for apoptosis markers and comet parameters. Findings did not change when molybdenum concentrations were modelled as continuous variables in a cubic spline analysis with possibly non-linear associations.

Zhou et al. (2016) studied associations between urinary level of molybdenum (among other metals) and sperm DNA damage in 207 men from subfertile couples in Wuhan, China (32). No statistically significant (p for trend > 0.3) associations were observed between quartiles of molybdenum spot urine concentration ($\mu\text{g/g}$ creatinine) and comet assay parameters (%DNA tail, tail length, and tail distributed moment) in multivariable models adjusting for age, BMI, smoking status, and abstinence time.

Branch et al. (2021) assessed associations between concentrations of molybdenum (among other metal(loid)s) in urine and 7 measures of semen quality among 356 reproductive-aged men from the LIFE-study recruited from 16 US counties between 2005–2009 (33). Semen quality endpoints were total sperm count, semen volume, sperm concentration, next day motility, traditional morphology, % DNA fragmentation index, and % high DNA stainability. The urinary molybdenum concentration in this general population sample was relatively low (median 47 µg/L). In multivariable linear regression models for log-transformed molybdenum concentration without taking into account other metal(loid)s, no statistically significant ($p > 0.05$) associations with semen quality endpoints were observed when adjusting for lifestyle and other potential confounders. Taking into account all 15 metal(loid)s under study, penalized LASSO regression models were fitted to identify and select metal(loid)s most likely to be predictive of each semen quality endpoint. Molybdenum concentration was only selected for inclusion in a subsequent unpenalized multi-metal(loid) and confounder-adjusted linear regression analysis on sperm motility, resulting in a beta coefficient of 0.07 (95% CI: -0.3, 0.44) % per µg/g Mo; $p = 0.72$).

Table 28 Summary of human studies on effects of molybdenum and its compounds on pregnancy-related health outcomes in mother

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
Prospective cohort studies					
<ul style="list-style-type: none"> • Bommarito et al., 2019 (35) • Nested case-control study • Boston MA, USA • 2006-2008 • 383 women recruited from the LIFECODES birth cohort • 28 cases: singleton preterm births with preeclampsia • 355 controls: unmatched singleton births without preeclampsia • Inclusion criteria: <ul style="list-style-type: none"> - having urine samples from the 3rd study visit (35 weeks gestation) available for metals analysis - at least 18 years of age 	<ul style="list-style-type: none"> • Urine samples from the 3rd study visit (35 weeks gestation) • Concentration ($\mu\text{g/L}$) of molybdenum determined using ICP-MS • LOD 0.30 $\mu\text{g/L}$ 	<ul style="list-style-type: none"> • Preeclampsia was defined (according to ACOG guidelines): <ul style="list-style-type: none"> - elevated maternal blood pressure ($> 140\text{mmHg}$ systolic and/or $> 90\text{mmHg}$ diastolic) - proteinuria ($> 300 \text{ mg/24 h}$ or a protein/creatinine ratio > 0.20) after 20 weeks gestation • Plasma biomarkers of angiogenesis (predictors of preeclampsia): <ul style="list-style-type: none"> - Measurement of circulating maternal soluble fms-like tyrosine kinase-1 (sFlt-1) and placental growth factor (PlGF) in plasma samples collected at the 3rd study visit - sFlt-1/PlGF ratio (i.e., a ratio of antiangiogenic to angiogenic activity) may more accurately predict preeclampsia than either biomarker alone 	<ul style="list-style-type: none"> • Molybdenum was detected in all urine samples • Median (IQR) concentration ($\mu\text{g/L}$) <ul style="list-style-type: none"> - 45.6 (36.2, 69.6) in 28 cases with preeclampsia - 51.3 (37.1, 68.8) in 355 controls without preeclampsia • Adjusted association between molybdenum and preeclampsia: HR=0.47 (0.21, 1.04); p=0.06 (adjusted for specific gravity of molybdenum, smoking during pregnancy, race, educational attainment, insurance status, infant sex, assisted reproductive technology, calcium supplementation, pre-pregnancy BMI and gestational age at study visit) • Adjusted associations between molybdenum and the percent change (95% CI) in circulating maternal angiogenic biomarkers: <ul style="list-style-type: none"> - sFlt-1: 0.69 (-9.49, 12.0); p=0.90 - PlGF: 4.99 (-6.09, 17.4); p=0.39 - sFlt-1/PlGF Ratio: -5.72 (-18.9, 9.60); p=0.44 		Preeclampsia is not a recognised reproductive outcome

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<p>- seeking prenatal care before 15 weeks gestation - intended delivery at the Brigham and Women’s Hospital in Boston MA, USA</p>		<ul style="list-style-type: none"> • Statistical analyses <ul style="list-style-type: none"> - Relationship between molybdenum and preeclampsia was estimated using multivariable Cox proportional hazard models - Expressed as hazard ratio (HR) and 95% CI associated with an IQR increase of urinary molybdenum - Relationships between molybdenum and log-transformed maternal angiogenic biomarkers were assessed using multivariable linear regression models <ul style="list-style-type: none"> - Expressed as % change (95% CI) associated with an IQR increase of urinary molybdenum - Urinary molybdenum concentrations were log-transformed in all analyses - Urinary trace metals were investigated as a mixture using PCA 	<p>(adjusted for specific gravity of molybdenum, smoking during pregnancy, race, educational attainment, insurance status, infant sex, assisted reproductive technology, calcium supplementation, pre-pregnancy BMI and gestational age at study visit)</p> <ul style="list-style-type: none"> • Including or excluding BMI in models did not yield different findings • Molybdenum was not included in principal components analyses 		

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
		<p>(applied to the set of 13 urinary trace metals with >30% detection to produce principal components)</p> <p>- Inverse probability weighting based on preterm birth case status was applied in all analyses to account for the disproportionate number of preterm births</p>			
<ul style="list-style-type: none"> Wang et al., 2020 (36) Panel study nested within larger birth cohort, Shanghai, China 234 healthy pregnant women without diagnosis of mental diseases or undergoing lithium augmentation treatment N= 222, 186, and 139 for the 	<p>Maternal peripheral blood samples collected at three prenatal care visits in the first (17.68 ± 1.10 weeks), second (26.33 ± 1.23 weeks), and third (32.48 ± 1.26 weeks) trimesters. Blood metal concentrations including molybdenum</p>	<p>Maternal pre-pregnancy weight and height self-reported at baseline visit. Weight during pregnancy measured at the hospital. Pregnancy diseases and childbirth information available through hospital records.</p> <p>Statistical analyses: Linear mixed-effects models with random intercept to estimate associations between metal concentrations and gestational weight gain (GWG) or GWG rate over the whole pregnancy. P-values</p>	<p>GWG 2.98 ± 3.10 kg in the first, 7.79 ± 2.99 kg in the second, 10.61 ± 3.63 kg in the third follow-up, 13.76 ± 1.40 kg during overall pregnancy. Corresponding GWG rates 0.17 ± 0.17, 0.30 ± 0.11, 0.33 ± 0.11 and 0.24 ± 0.17 kg/week.</p> <p>Association (β, 95% CI) of molybdenum concentrations with GWG and GWG rate, respectively: 0.024 (-0.066,0.114); 0.003 (-0.002,0.007).</p> <p>Trimester-specific analyses restricted to metals that were associated with overall GWG (rate), thus not performed for molybdenum.</p>	<p>Potential selection bias: participants who withdrew from the cohort before delivery were excluded.</p>	<ul style="list-style-type: none"> Gestational weight gain is not a recognised reproductive /developmental outcome

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<p>early second, second, and third trimester of pregnancy, respectively.</p>	<p>determined by ICP-MS. Demographic and lifestyle characteristics assessed via interview at baseline visit. Food intake estimated with FFQ.</p> <p>Molybdenum concentrations (median, P25, P75; µg/L) at early second, second, and third trimester, respectively: 5.13 (1.91, 5.46); 5.13 (4.92, 5.33)*; 5.15(4.95, 5.36)*. *: significantly different from early second trimester</p>	<p>adjusted for multiple testing using the false discovery rate.</p> <p>Robust generalized linear regression to estimate associations among circulating metals and GWG at each follow-up, and overall pregnancy GWG.</p> <p>Adjustment for gestational age (crude model) and maternal age, gestational age, pre-pregnancy BMI, energy intake in pregnancy, education, household income, physical activity, passive smoking, parity, gestational anaemia, gestational diabetes mellitus, and infant gender (adjusted model).</p>			

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<ul style="list-style-type: none"> • Mandiá et al. (2021) (37) • Prospective cohort study • 170 nursing mothers: <ul style="list-style-type: none"> - 70 full-term - 100 pre-term • University Clinical Hospital of Santiago de Compostela, Spain • Period 2018-2019 • Inclusion criteria: <ul style="list-style-type: none"> - maternal age > 18 years • Exclusion criteria: <ul style="list-style-type: none"> - chronic disease - taking nutrient supplements 	<ul style="list-style-type: none"> • Human milk samples (5–10 mL) obtained from 70 mothers of full-term newborns: <ul style="list-style-type: none"> - colostrum during the first 3–4 days of lactation - intermediate milk up to 7–10 days of lactation - later mature milk (up to 6 months after birth) • Human milk samples (5–10 mL) obtained from 100 mothers of premature newborns: <ul style="list-style-type: none"> - later mature milk (up to 6 months after birth) 	<ul style="list-style-type: none"> • Questionnaire including <ul style="list-style-type: none"> - Excessive maternal weight gain during pregnancy: ≥ 16 kg - Gestational hypertension • Statistical analysis: <ul style="list-style-type: none"> - ANOVA and Bonferroni tests for differences in molybdenum concentrations between groups - Regression models of molybdenum concentration (dependent variable) on selected medical factors (independent variables; either categorical or continuous) - Unclear whether data from all mothers and all samples were pooled - Adjustment unclear 	<ul style="list-style-type: none"> • Mean concentrations of molybdenum in milk from full-term mothers: <ul style="list-style-type: none"> - colostrum: 1.88 (SD 1.20) $\mu\text{g/L}$ - intermediate milk: 1.22 (SD 1.98) $\mu\text{g/L}$ - later mature milk: 0.96 (SD 1.16) $\mu\text{g/L}$ ($p < 0.001$ lower than colostrum) • Mean concentrations of molybdenum in milk from pre-term mothers: <ul style="list-style-type: none"> - later mature milk: 0.70 (SD 1.17) $\mu\text{g/L}$ (unclear whether this was significantly lower than 0.96 in later mature milk from full-term mothers?) • Associations with medical factors (using all samples from all 170 mothers?!): <ul style="list-style-type: none"> - Excessive maternal weight gain during pregnancy: $\text{beta} = -0.07$ ($p = 0.024$); thus molybdenum levels were on average lower in milk from mothers who had excessive maternal weight gain - Gestational hypertension: $\text{beta} = 0.008$ ($p > 0.999$); thus no correlation 	<ul style="list-style-type: none"> • Authors denote regression models on determinants as 'multivariate analysis', but is unclear whether adjusted for the other determinants and/or potential confounders. In that case, risk of confounding bias 	<ul style="list-style-type: none"> • Most outcomes are not recognised as reproductive /developmental outcomes • The design is referred to as "Prospective case series", which does not sound like a conventional design and it remains unclear whether is (partly) a case-control study (premature vs. full-term)? • Definition of pre-term

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
	<ul style="list-style-type: none"> • Concentration (µg/L) of molybdenum determined using ICP-OES (inductively coupled plasma optical emission spectrophotometry) • Detection limit not reported • (Also 5 minerals, 8 other essential trace elements and 21 toxic trace elements determined) 				<p>not provided (presumably <37 weeks of gestation?)</p> <ul style="list-style-type: none"> • Timing of later mature milk sample not clear (up to 6 months after birth?) • Statistical significance in tables is puzzling; unclear what was tested exactly. Hard to believe many of the p-values • In analysis of determinants (Table 5), it looks like all data

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
					<p>were pooled, thus including 3 samples for the same mother without controlling for dependency</p> <ul style="list-style-type: none"> • The latter is not a prospective analysis; more a cross-sectional analysis and exposure was determined <i>after</i> the endpoints
Case-control studies					
<ul style="list-style-type: none"> • Elongji-Moyene et al., 2016 (38) • Case-control study 	<ul style="list-style-type: none"> • 24-hour urine samples were collected (for the cases) 	<ul style="list-style-type: none"> • Preeclampsia: inpatients with in the 2nd or 3rd trimester of pregnancy with: 	<ul style="list-style-type: none"> • Molybdenum detected in all urine samples • Urinary molybdenum concentrations: 	<ul style="list-style-type: none"> • Differences in urinary molybdenum and excreted amount between cases and controls 	<ul style="list-style-type: none"> • Pregnancy hypertension is not a recognised

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<ul style="list-style-type: none"> • Kinshasa, Democratic Republic of Congo • Study period: 2011 (1 period during the rainy season and 1 period during the dry season) • Pregnant women having had at least one prior pregnancy • Cases: 88 women hospitalized with preeclampsia • Controls: 88 healthy pregnant women from the antenatal clinic 	<p>during hospitalisation)</p> <ul style="list-style-type: none"> • Concentration of molybdenum ($\mu\text{g/L}$) was determined using ICP-MS • LOD not reported, (but $<0.66 \mu\text{g/L}$) • Total amount of molybdenum excreted per 24 h ($\mu\text{g/day}$) calculated as urinary molybdenum concentration ($\mu\text{g/L}$) \times daily urine volume (L/day) <p>Molybdenum concentration and excreted amount expressed as geometric mean and 25th and 75th percentiles</p>	<ul style="list-style-type: none"> - hypertension (systolic blood pressure >140 mm Hg and diastolic blood pressure >90 mm Hg after at least 15 min of rest); AND - proteinuria (positive dipstick test or >300 mg proteins/24 h), with or without oedema - absence of neurological disease or brain injury • Statistical analysis <ul style="list-style-type: none"> - Differences in v concentration and excretion between cases and controls: one-way ANOVA on log-transformed values - Two-way ANOVA followed by Tukey's post hoc tests were performed to disentangle the separate and joint effects of preeclampsia and season - Significance level: $p < 0.05$ 	<ul style="list-style-type: none"> - Cases: GM=19.2 (IQR: 10.7-35.6) $\mu\text{g/L}$ - Controls: GM=13.3 (IQR: 7.2-29.8) $\mu\text{g/L}$ - 1.4-fold difference, $p=0.023$ • Amount of excreted molybdenum: <ul style="list-style-type: none"> - Cases: GM=16.3 (IQR: 7.4-31.1) $\mu\text{g/day}$ - Controls: GM=15.1 (IQR: 8.0-33.8) $\mu\text{g/day}$ - 1.1-fold difference, $p=0.64$ • Urinary concentration and daily excretion of molybdenum were higher in the rainy season ($n=67$) than in the dry season ($n=109$), regardless of case-control status 	<p>groups using ANOVA did not take into account potential confounders</p>	<p>developmental outcome</p>

Prospective cohort studies

Bommarito et al. (2019) studied the association between urinary molybdenum level during gestation, preeclampsia and plasma biomarkers of angiogenesis in a case-control study (28 cases and 355 controls nested within the LIFECODES birth cohort in Boston MA, USA (35). A borderline significant ($p=0.06$) inverse association between urinary molybdenum and preeclampsia was found; HR=0.47 (95% 0.21, 1.04) for an IQR increase of urinary molybdenum concentration. No statistically significant associations between molybdenum and maternal angiogenic biomarkers in blood plasma (circulating soluble fms-like tyrosine kinase-1 [sFlt-1], placental growth factor [PlGF], and the sFlt-1/PlGF ratio) were found. The endpoints under study are not recognised reproductive or developmental outcomes.

Wang et al., 2020 conducted a panel study nested within larger birth cohort in Shanghai. They aimed to investigate associations between maternal blood metal concentrations and gestational weight gain (GWG) across pregnancy in 234 healthy pregnant women (26). Maternal peripheral blood samples were collected at three prenatal care visits in the first ($n=222$), second ($n=186$) and third ($n=139$) trimesters of pregnancy. Linear mixed-effects models with adjustment for several confounders showed no significant associations of molybdenum concentrations with GWG and GWG rate. This study might suffer from selection bias as participants who withdrew from the birth cohort before delivery were excluded.

Mandiá et al. (2021) studied concentrations of molybdenum (among other elements) in human milk from nursing mothers from Santiago de Compostela, Spain and their associations with selected medical factors (37). Human milk samples were obtained at three time points from 70 mothers of full-term newborns: colostrum during the first 3–4 days of lactation, intermediate milk up to 7–10 days of lactation and later mature milk up to 6 months after birth. In addition, samples of later mature milk were also obtained from 100 mothers of premature newborns. A questionnaire included medical factors of the mother during pregnancy. Concentrations of molybdenum in milk from full-term mothers decreased as lactation progressed. Mean molybdenum concentrations in later mature milk were 0.96 and 0.70 $\mu\text{g/L}$ from mothers of full-term and preterm newborns, respectively. Molybdenum concentrations were on average 0.70 $\mu\text{g/L}$ lower ($p=0.024$) in milk from mothers who had an excessive maternal weight gain (≥ 16 kg) during pregnancy. No correlation ($p>0.999$) was found between molybdenum concentration and gestational hypertension. The endpoints under study are not recognised developmental outcomes.

Case-control studies

In a case-control study, **Elongi-Moyene et al. (2016)** measured molybdenum concentrations in 24-hour urine from 88 women with preeclampsia and 88 healthy pregnant women in Kinshasa, Democratic Republic of Congo to investigate the association between preeclampsia and exposure to molybdenum (among other metals) (38). The geometric mean urinary molybdenum concentration in cases with preeclampsia (19.2; IQR 10.7–35.6 $\mu\text{g/L}$) was significantly ($p=0.023$) higher than in controls (13.3; IQR 7.2–29.8 $\mu\text{g/L}$). The daily urinary

excretion was 275 mL (23 %) lower in cases than in controls. The geometric mean amount of excreted molybdenum did not differ significantly ($p=0.64$) between cases (16.3 $\mu\text{g}/\text{day}$) and controls (15.1 $\mu\text{g}/\text{day}$). Urinary concentration and daily excretion of molybdenum were higher in the rainy season than in the dry season, regardless of case-control status. Preeclampsia is not a recognised developmental outcome.

Li et al. (2022) performed a case-control study to investigate the association between blood serum trace elements (including molybdenum) in women and the risk of endometrial disease as a proxy for infertility (39). The study included 302 women (20-40 years) diagnosed with endometrial disease and 302 women without endometrial disease, recruited from the same hospital and age-matched to the cases. Diagnoses included endometritis (20.2%), polyps (39.4%), intrauterine adhesion (9.3%), uterine fibroids (15.9%), or a combination of two or more conditions (15.2%). Serum concentrations of Se, Mo, Cu, and Zn were determined by ICP-MS. Associations between trace elements and endometrial diseases were analyzed using restricted cubic splines and conditional logistic regression, adjusted for age, BMI, education, parity, passive smoking, and basal sex hormone levels. The former analysis showed an increasing risk of endometrial diseases with increasing molybdenum concentrations until the 75th percentile, whereas the ORs for quartiles of molybdenum concentration compared to the 1st quartile were 1.06 (0.76, 1.49), 1.23 (0.89, 1.72) and 1.18 (0.85, 1.65) for the 2nd, 3rd and 4th quartiles, respectively.

8.1.3 *Other relevant information*

In addition to the in vivo animal studies and studies in humans, two in vitro studies with ovarian cells of fragments are available. Below, these two additional studies are described, and summarized in Table 29.

Table 29 Summary table of other studies relevant for sexual function and fertility.

Reference	Species	Study design	Dose or concentration	Observations	Remarks
Kolesarova et al., 2011 (40)	Slovakian White gilts (young female pigs) N=32	In vitro assay with ovarian granulosa cells. Cells were incubated with ammonium molybdate for 18 hours. The secretion of growth factor IGF-I was assessed by radioimmunoassay (RIA) and expression of cyclin B1 and caspase-3 by immunocytochemistry.	0, 0.09, 0.17, 0.33, 0.5, 1.0 mg /mL Test item: ammonium molybdate Purity: no data	<ul style="list-style-type: none"> Decreased IGF-I release at 0.5 and 1.0 mg/mL. Increased expression of cyclin B1 at all concentrations. Increased Caspase-3 expression at 0.5 and 1.0 mg/mL. 	The study authors conclude that molybdenum affects ovarian functions, secretory activity, proliferation and apoptosis of granulosa cells through hormonal and intracellular substances such as cyclin B1 and caspase-3.
Roychoudhury et al., 2014 (41)	Wistar rat, females. N=25	Ovarian fragments of 2 mm size were incubated for 24 h in culture plates containing ammonium molybdate. The secretion activity (progesteron, 17 β -estradiol and IGF-I) was measured.	0, 90, 170, 330 and 500 μ g/ml. Substance: ammonium molybdate Purity: no data	<ul style="list-style-type: none"> Dose-dependent decrease (at the concentrations of 90, 170 and 500 μg/ml) in release of 17β-estradiol and IGF. Progesterone release was not affected. 	Publication is a rapid communication of 3 pages. The study authors conclude that the data contribute to new insights regarding the mechanism of action of Mo on rat ovarian functions.

In vitro study with porcine ovarian cells (Kolesarova et al., 2011)

An in vitro study was performed to examine the secretory activity of porcine ovarian granulosa cells after experimental ammonium molybdate administration and to outline the potential intracellular mediators of the effects of molybdenum. Ovarian granulosa cells were incubated with ammonium molybdate for 18 hours at 0.09, 0.17, 0.33, 0.5 or 1.0 mg/mL, while the control group received no molybdenum. The secretion of IGF-I and expression of cyclin B1 and caspase-3 was assessed.

IGF-I release was decreased by addition of ammonium molybdate at the concentrations of 0.5 and 1.0 mg/mL. The expression of cyclin B1 was stimulated at all concentrations. Caspase-3 expression was also stimulated at the concentrations of 0.5 and 1.0 mg/mL. (40).

In vitro study with rat ovarian fragments (Roychoudhury et al., 2014)

An in vitro study was performed to examine the secretion activity (progesterone, 17 β -estradiol and insulin-like growth factor-I) of rat ovarian fragments after molybdenum (Mo) addition. Rat ovarian fragments were incubated with ammonium molybdate at the concentrations 90, 170, 330 and 500 μ g/ml for 24 h and compared with control group without Mo addition. Release of progesterone (P4), estradiol (17 β -estradiol) and insulin-like growth factor I (IGF-I) by ovarian fragments was assessed.

Data show that P4 release by ovarian fragments was not affected by ammonium molybdate addition at all the concentrations used (90-500 μ g/ml). However, addition of ammonium molybdate was found to cause a concentration-dependent decrease (at the concentrations of 90, 170 and 500 μ g/ml) in release of 17 β -estradiol by ovarian fragments in comparison to control. Also, addition of ammonium molybdate significantly inhibited IGF-I release at all the concentrations (90-500 μ g/ml) used in the study (41).

8.2 Adverse effects on development

8.2.1 Animal studies

In the advice of the Health Council of the Netherlands on molybdenum in 2013, various studies in animals on the effects of molybdenum compounds on development were described (1).

Below additional studies that were published after release of the advice of the Health Council are described.

The in vivo studies in experimental animals on the effects of molybdenum compounds on development are summarized in Table 30. In these studies, animals were exposed to the substance via the oral route. No inhalation or dermal studies were available.

Reproduction toxicity data for more complex structures containing molybdenum, such as reaction products, are included in Annex B. Data from these compounds cannot be used for grouping and read-across, but may still be of interest. Further details on the selection of compounds is included in Annex A.

Table 30 Summary table of animal studies on adverse effects on development.

Reference	Species	Experimental period and design	Dose and route	General toxicity	Effects on development	Remarks
Jay Murray et al., 2014 (42)	Sprague Dawley rats. N= 25 time-mated females / dose.	OECD guideline 414 (prenatal developmental toxicity study). Serum blood, placenta, liver and kidney samples were analysed for molybdenum, copper, zinc, manganese, iron, cobalt and selenium.	0, 3, 10, 20 and 40 mg Mo/kg bw/day, in the diet. The corresponding sodium molybdate dihydrate concentrations in the diet were 0, 100, 338, 675 and 1350 ppm, respectively. Substance: Sodium molybdate dihydrate Purity: 99.9%	<ul style="list-style-type: none"> • No treatment-related adverse effects observed. • Dose-dependent increases in molybdenum in serum, placentae, liver and kidneys, accompanied with significant increases in serum and tissue copper levels. 	<ul style="list-style-type: none"> • No treatment-related adverse effects on development. 	Study under GLP
Murray et al., 2019 (18)	Sprague Dawley rats, males and females. N = 24/sex/ dose	OECD guideline 416 (two generation reproductive toxicity study) P-males: exposure for at least 10 weeks before cohabitation, during the cohabitation, and continuing through to the day of euthanasia (total 147-151 days) P-females: for at least 10 weeks before cohabitation, during the cohabitation, gestation, littering	0, 5, 17, or 40 mg molybdenum (Mo)/kg bw/day in drinking water. Additional group: 40 mg Mo/kg bw/day via diet. Test item: Sodium molybdate dihydrate	<ul style="list-style-type: none"> • Decreased body weight, food consumption (males only) and water consumption at 40 mg/kg bw/day. 	No adverse effects on development.	Well-performed study; GLP.

Reference	Species	Experimental period and design	Dose and route	General toxicity	Effects on development	Remarks
		and post-partum periods (lactation period) and continuing through to the day of euthanasia (total 156-158 days). F1: during lactation, 10 weeks pre-mating, cohabitation, and continued through the day of euthanasia. Effect parameters as described in OECD 416.	Purity: 99%			
Study report, 2020 (17)	Sprague-Dawley rats. Non-pregnant females, n=6/group.	14-day toxicity study to test for tolerance of sodium molybdate dihydrate by oral gavage. Preliminary study to the OECD 414 study as described below (Study report, 2021). Administration: 14 days (where tolerated) Parameters: viability, clinical signs, body weights, body weight gains, food consumption, bioanalysis, organ weights, and macroscopic observations.	300, 600, and 1000 mg/kg bw/day (equivalent to 120, 240, and 400 mg Mo/kg bw/day, respectively), by oral gavage. Test item: Sodium molybdate Purity: no data	<i>300 mg/kg bw/day</i> <ul style="list-style-type: none"> adverse clinical observations (dehydration, hunched posture, erect fur, cold to touch) Reduced bw (21%) Reduced food consumption (40%) <i>600 mg/kg bw/day</i> <ul style="list-style-type: none"> All euthanized at day 7 (adverse clinical observations, 25% reduction bw, 64% reduction food consumption) <i>1000 mg/kg bw/day</i> <ul style="list-style-type: none"> 4/6 died; 2/6 euthanized 	Developmental toxicity not evaluated.	

Reference	Species	Experimental period and design	Dose and route	General toxicity	Effects on development	Remarks
Study report, 2020 (17)	Sprague Dawley rats. N= 10 pregnant females / dose.	Dose range finder study for the OECD 414 study described below (Study report, 2021). Administration: GD 6-20, daily treatment. Controls: control group (base diet) and pair-fed control group.	300 and 400 mg/kg bw/day, in the diet. Equivalent to 120 and 160 mg Mo/kg bw/day. Substance: Sodium molybdate dihydrate Purity: no data	Effects in both treatment groups: <ul style="list-style-type: none"> • Reduced maternal body weight (14-23% on GD20) and body weight gain (41-68%) • Reduced corrected body weights (terminal body weight at GD21 minus the gravid uterine weight) (16-23%) • Reduced food consumption in both groups. • Reduced absolute and relative liver weights 	<ul style="list-style-type: none"> • Reduced gravid uterine weights at 300 mg/kg bw/day (9%, not statistically significant) and 400 mg kg bw/day (28%) • No treatment-related effect as observed in pre- or post-implantation loss, live foetuses per litter, sex ratio or average litter size. • Reduced total, male and female foetal weight at 300 and 400 mg /kg bw/day (84%, 72% and 73% respectively) • No effect on anogenital distance or external appearance. 	Only study summary available, and no absolute data.
Study report, 2021 (43)	Sprague Dawley rats, females.	OECD guideline 414 (prenatal developmental toxicity study). Effect parameters as described in OECD 414.	Nominal: 0, 200 and 300 mg/kg bw/day (corresponding to	<ul style="list-style-type: none"> • Dose-dependent moderate to marked maternal toxicity at both dose levels, including: 	<u>Caesarean section (GD21)</u> <ul style="list-style-type: none"> • Dose-dependent reductions in foetal 	Study under GLP. Only study summary

Reference	Species	Experimental period and design	Dose and route	General toxicity	Effects on development	Remarks
	N = 24/group	Additionally to the female rats sacrificed on gestation day 21, 48 female rats were assigned to 2 littering groups (control group and high dose group) of 24 rats which were allowed deliver naturally and raise their young to weaning.	0, 80 and 120 mg Mo/kg bw/day). Actual dose levels, based on food consumption and body weight: 94.1 and 125.7/128.5 mg Mo/kg bw/day. Exposure via diet. Substance: Sodium molybdate Purity: no data	<ul style="list-style-type: none"> ○ adverse clinical observations ○ reductions maternal weight gain (27.1% and 49.8% lower than control) ○ reduced food intake (11% and 25%) over the administration period ○ reduced corrected (for uterine content) body weight at gestation Day 21 (12.4 and 23.7% lower than control). ● Reduced liver weights ● Test item-related microscopic changes in the kidney: tubular regeneration and mononuclear cell infiltration, more prevalent at 300 mg/kg bw/day ● Test item-related microscopic changes in the liver: hepatocellular hypertrophy and glycogen accumulation (both dose levels) and karyocytomegaly and vacuolation (300 mg/kg bw/day). 	<p>weight (~11% and 22% at 200 and 300 mg/kg bw/day)</p> <ul style="list-style-type: none"> ● Reduced total placental weight per litter, compared to controls: 11% at 200 mg/kg bw/day (not significant), 24% at 300 mg/kg bw/day. ● No effect on the incidence of external, visceral and skeletal foetal malformations and variations in the treated animals. <p><u>Littering animals</u></p> <ul style="list-style-type: none"> ● Lower mean pup weights (combined sexes) in the 300 mg/kg bw/day group, compared to controls, at each interval measured (days 0, 4-preculling, 4- 	available, and no absolute data. Reduction in anogenital distances was considered attributable to the marked reduction in foetal weight and marked maternal toxicity, according to the authors of the registration dossier.

Reference	Species	Experimental period and design	Dose and route	General toxicity	Effects on development	Remarks
					<p>postculling, 7, 14, 18, and 21 postpartum).</p> <ul style="list-style-type: none"> ○ 19.2% lower pup weight at birth ○ 9.4% lower pup weight at day 21 postpartum ○ Male and female pups similarly affected. • 6% reduction in adjusted male foetal anogenital distance at 300 mg/kg/day 	
Mohamed et al., 2020 (44)	Mice, strain and number of animals per dose not specified.	<p>Molybdenum nanoparticles with a size less than 100 nm were orally administered to pregnant mice from the 1st up to the 17th day of pregnancy or from the 9th up to the 17th day of pregnancy.</p> <p>Examinations: External (morphological) malformations and skeletal abnormalities.</p>	<p>Oral dose levels of 0, 500 or 750 mg/kg bw were used. Route of exposure not further specified. Test item: Mo nanoparticles (< 100 nm). Purity: purity was confirmed by X-ray diffraction, no quantitative data.</p>	<ul style="list-style-type: none"> • Decreases in the maternal body weight. • DNA damage and elevated expression of levels of p53 gene. • Increased expression levels of E-Cad and N-Cad genes that control skeleton development. 	<ul style="list-style-type: none"> • Decreased number and length of fetuses. • Increased incidence of skeletal abnormalities (reduced ossification and chondrification). 	No information on the species and number of animals and lack of data on the test item.

Prenatal developmental toxicity study (Jay Murray et al., 2014)

In a study performed in accordance with OECD TG 414 the developmental toxicity of sodium molybdate dihydrate, given in the diet to Sprague Dawley rats, was tested. Dose levels of 0, 3, 10, 20 and 40 mg Mo/kg bw/day (in the diet) were administered from gestation day (GD) 6 to GD 20. The dose levels were chosen based on a range-finder study in rats given sodium molybdate dihydrate. No treatment-related maternal effects or developmental toxicity were observed up to 20 mg Mo/kg bw/day (the highest dose tested) in this range-finder study.

In the main study, no adverse effects were observed at any dose level on the dams, or on embryofoetal survival, foetal bodyweight, or development, with no increase in malformations or variations (Table 31, 32 and 33). Significant increases in serum and tissue molybdenum and copper levels were observed but no toxicity related to these was observed (42).

Table 31 Summary of uterine implantation data (GD20) from rats exposed to dietary sodium molybdate dihydrate on days 6–20 of gestation.

		Dose Group (mg Mo/kg bw/day)				
		0	3	10	20	40
Number of females with live foetuses ^a		25	25	25	24	25
Total number of corpora lutea		360	364	354	345	364
Number of corpora lutea per female	Mean	14.4	14.6	14.2	14.4	14.6
	SEM	0.5	0.6	0.4	0.4	0.5
Total number of implantations		317	322	327	319	313
Number of implantations per female	Mean	12.7	12.9	13.1	13.3	12.5
	SEM	0.5	0.3	0.3	0.4	0.4
Total number of pre-implantation losses		43	42	27	26	51
Mean % per group		11.9	11.5	7.6	7.5	14.0
Total number of post-implantation losses		9	10	13	14	6
Mean % per group		2.8	3.1	4.0	4.4	1.9
Number of live foetuses per female	Mean	12.3	12.5	12.6	12.7	12.3
	SEM	0.5	0.3	0.3	0.4	0.4
Number of live foetuses as % of implantations	%	97.2	96.9	96.0	95.6	98.1
Total number of live foetuses		308	312	314	305	307 ^b
Number of males		152	141	162	155	149
Number of females		156	171	152	150	157
Foetal weight (sexes combined) (g)	Mean	4.02	4.04	4.02	4.02	4.04
	SEM	0.06	0.05	0.04	0.07	0.07
Foetal weight (males) (g)	Mean	4.13	4.16	4.12	4.13	4.14
	SEM	0.06	0.06	0.05	0.07	0.06
Foetal weight (females) (g)	Mean	3.90	3.95	3.91	3.91	3.95
	SEM	0.06	0.05	0.04	0.06	0.08

^aOne foetus inadvertently not sexed. ^b at scheduled termination.

Table 32 Occurrence of external, visceral and skeletal malformations and variations in the fetuses of rats exposed to sodium molybdate dihydrate (42).

	Dose Group (mg Mo/kg bw/day)				
	0	3	10	20	40
Total number of fetuses examined	308	312	314	305	307
Total number of litters examined	25	25	25	24	25
External defects					
Number of fetuses examined ^a	307	312	313	303	307
Number showing malformations	0	1	0	0	1
Number of litters affected	0	1	0	0	1
Number showing variations	0	0	0	0	0
Number of litters affected	0	0	0	0	0
Fresh visceral body-only defects					
Number of fetuses examined	150	151	151	152	152
Number showing malformations	0	0	0	0	0
Number of litters affected	0	0	0	0	0
Number showing variations	7	4	11	2	5
Number of litters affected	5	3	8	2	4
Bouin's head defects					
Number of fetuses examined	150	151	151	152	152
Number showing malformations	0	0	2	6	0
Number of litters affected	0	0	2	3	0
Number showing variations	0	0	0	2	1
Number of litters affected	0	0	0	1	1
Skeletal defects					
Number of fetuses examined	158	161	163	153	155
Number showing malformations	1	1	4	2	1
Number of litters affected	1	1	4	2	1
Number showing variations	54	69	69	65	64
Number of litters affected	18	21	22	19	22

^a In dose groups 0, 10 and 20 mg Mo/kg bw/day, 1, 1, and 2 fetuses, respectively were not examined externally but did have visceral or skeletal examination.

Table 33 Individual incidences of foetal malformations by dose group, dam and foetus of rats exposed to sodium molybdate dihydrate.

Dose group (mg Mo/kg bw/day)	Dam and foetus numbers	Findings
0	Dam 18, F 7	Short left rib and cartilage agenesis
3	Dam 29, F 12	Umbilical hernia
	Dam 36, F 3	Thoracic vertebral centrum, bipartite ossification and bipartite cartilage
10	Dam 51, F 10	Dilated nasal sinus
	Dam 60, F 12	Dilated nasal sinus
	Dam 63, F 8	Thoracic vertebral centrum, bipartite ossification and bipartite cartilage
	Dam 67, F 3	Thoracic vertebral centrum, bipartite ossification and bipartite cartilage
	Dam 68, F 1	Thoracic vertebral centrum, bipartite ossification and bipartite cartilage
	Dam 64, F 11	Rib 13, right, cartilage agenesis
20	Dam 78, F 1, F 4, F 5	Dilated nasal sinus
	Dam 85, F 8, F 10	Dilated nasal sinus
	Dam 100, F 10	Dilated nasal sinus
	Dam 79, F 5	Thoracic vertebral centrum, bipartite ossification and bipartite cartilage
	Dam 99, F 9	Rib 13, right, cartilage agenesis
40	Dam 125, F 7	Tail short and curly; rib 7 branched; rib 11 cartilage branched, and thoracic

Two-generation rat study – sodium molybdate dihydrate (Murray et al., 2019)

In a multigenerational study, performed according to OECD TG 416, groups of 24 male and 24 female Sprague-Dawley rats were administered sodium molybdate dihydrate at 0, 5, 17, or 40 mg molybdenum (Mo)/kg bw/day in the drinking water or 40 mg Mo/kg bw/day in the diet over two generations to assess reproductive toxicity. The dose levels were chosen based on a range-finder study in rats given sodium molybdate dihydrate in the drinking water or diet (see above). The drinking water route of administration was chosen as the main route of exposure for this study because it allowed the authors comparison with a study by Fungwe et al.(23) and because the range-finder study suggested a possible decrease in the pregnancy rate (6/10) at 40 mg Mo/kg bw/day in the drinking water, but not in the diet.

No adverse effect on development was observed at any dose level in either generation (Table 34 and 35). Also no adverse effects on reproduction were observed, as described in 8.1.1.

Systemic toxicity, including decreased body weight, food consumption (males only) and water consumption, was observed among both sexes given 40 mg Mo/kg bw/day in the diet. Serum levels of Mo and copper were increased in a dose-related manner (18).

Table 34 Developmental landmarks among the F1 and F2 generation pups exposed to sodium molybdate dihydrate in the drinking water or diet (18).

	Dose Group (mg Mo/kg bw/day)				
	0	5 (water)	17 (water)	40 (water)	40 (diet)
F1 generation pups					
Litters evaluated, N	22	22	23	23	19
Average day of eye opening ^a , mean ± SD	14.7 ± 0.9	14.6 ± 0.7	14.6 ± 0.9	14.4 ± 0.8	14.8 ± 0.7
Average day of hair growth ^a , mean ± SD	8.0 ± 0.0	8.0 ± 0.0	8.0 ± 0.0	8.0 ± 0.0	8.0 ± 0.0
Average day of incisor eruption ^a , mean ± SD	11.6 ± 0.8	11.7 ± 0.6	11.6 ± 0.9	11.2 ± 0.9	11.9 ± 0.9
Average day of pinna unfolding ^a , mean ± SD	3.5 ± 0.6	3.5 ± 0.7	3.4 ± 0.5	3.2 ± 0.4	3.3 ± 0.5
Average day of preputial separation ^a , mean ± SD	44.6 ± 1.7	44.8 ± 2.6	44.7 ± 1.9	44.2 ± 2.3	45.0 ± 2.0
Average day of vaginal patency ^a , mean ± SD	31.0 ± 1.5	31.0 ± 1.4	31.8 ± 2.2	31.2 ± 2.7	31.3 ± 2.0
F2 generation pups					
Litters evaluated, N	22	23	23	21	22
Average day of eye opening ^a , mean ± SD	13.0 ± 0.9	13.3 ± 0.6	13.1 ± 0.6	12.8 ± 0.9	13.3 ± 0.7
Average day of hair growth ^a , mean ± SD	7.0 ± 0.0	7.0 ± 0.0	7.0 ± 0.0	7.0 ± 0.0	7.0 ± 0.0
Average day of incisor eruption ^a , mean ± SD	10.1 ± 0.8	10.3 ± 1.1	10.3 ± 1.3	9.8 ± 0.7	10.3 ± 1.1
Average day of pinna unfolding ^a , mean ± SD	2.3 ± 0.4	2.3 ± 0.4	2.3 ± 0.5	2.2 ± 0.4	2.2 ± 0.4
Male anogenital distance on PDD 4, mean ± SD	4.77 ± 0.62	4.97 ± 0.47	4.89 ± 0.72	4.50 ± 0.51	4.70 ± 0.42
Female anogenital distance on PDD 4, mean ± SD	2.30 ± 0.21	2.30 ± 0.22	2.13 ± 0.28	2.16 ± 0.28	2.18 ± 0.24

^a The average day postpartum that at least 50% of the pups tested in the litter had the developmental measure present.
PDD: postdelivery day

Table 35 Litter observations among F1 and F2 generations exposed to sodium molybdate dihydrate in the drinking water or diet.

	Dose Group (mg Mo/kg bw/day)				
	0	5 (water)	17 (water)	40 (water)	40 (diet)
F1 litters					
Litters with one or more liveborn, N	22	22	23	23	19
Live litter size on PND 0, mean ± SD	15.1 ± 3.0	15.1 ± 2.2	15.3 ± 2.0	14.5 ± 2.3	15.1 ± 1.8
Live litter size on PND 21, mean ± SD	14.6 ± 3.0	14.8 ± 2.2	15.1 ± 1.9	14.1 ± 2.4	14.9 ± 1.8
Pup weight/litter on PND 0, g, mean ± SD	6.5 ± 0.7	6.7 ± 0.5	6.7 ± 0.7	6.8 ± 0.6	6.4 ± 0.6
Pup weight/litter on PND 21, g, mean ± SD	42.0 ± 8.4	42.6 ± 5.4	42.4 ± 7.2	45.4 ± 7.6	41.3 ± 4.9
F2 litters					
Litters with one or more liveborn, N	22	23	23	21	22
Live litter size on PDD 0, mean ± SD	14.1 ± 1.4	15.4 ± 2.2	14.0 ± 3.8	14.5 ± 1.9	14.9 ± 2.2
Live litter size on PDD 21, mean ± SD	13.6 ± 1.1	15.0 ± 2.0	13.5 ± 3.4	14.4 ± 1.8	14.4 ± 1.7
Pup weight/litter on PDD 0, g, mean ± SD	7.0 ± 0.6	6.8 ± 0.5	7.1 ± 0.8	7.0 ± 0.5	6.6 ± 0.4
Pup weight/litter on PDD 21, g, mean ± SD	48.0 ± 6.7	43.9 ± 6.4	48.6 ± 11.2	47.4 ± 8.2	45.6 ± 5.0

PDD: post delivery day

PND: post natal day.

Prenatal developmental toxicity study (Study report, 2021)

A prenatal developmental toxicity study (OECD 414) with sodium molybdate was performed in rats. Preliminary to this study, a dose range finder study and oral gavage tolerance study were performed. In the dose range finder study, pregnant rats were exposed to 300 and 400 mg/kg bw/day (equivalent to 120 and 160 mg molybdenum/kg bw/day, respectively) sodium molybdate via diet, from GD6-20. Excessive toxicity was observed at 400 mg/kg bw/day and therefore 300 mg/kg bw/day was selected as the high dose. The tolerance study of sodium molybdate via oral gavage demonstrated marked toxicity at 300 mg/kg bw/day and mortality at higher doses. The substance was less tolerated via gavage as compared to diet and the OECD 414 study was therefore performed via diet.

In the OECD 4141 study, Sprague Dawley rats were given 0, 200 or 300 mg/kg bw/day (0, 80 or 120 mg /kg bw/day expressed as Mo) of sodium molybdate via diet at GD 6-20 to investigate developmental toxicity. On gestation day 21, part of the females rats were euthanized (caesarean section animals). The other part was assigned to two littering groups (controls and high dose) to allow delivery and weaning of pups (littering animals).

Dose-dependent moderate to marked maternal toxicity was observed at both dose levels, including adverse clinical observations, reductions in maternal weight gain and food intake over the administration period, and reduced corrected (for uterine content) body weight at gestation Day 21. Liver weights were reduced and test item-related microscopic changes were present in the kidney and liver.

Total placental weight per litter was reduced at 300 mg/kg bw/day, compared to controls. Relative weights were not significantly different. In the caesarian section animals, dose-dependent reduction in foetal weight was observed. In littering animals, mean pup weights (combined sexes) were significantly lower in the highest dose group than in the control group. No effects were seen on viability indices, lactation indices, litter sex ratio and litter size. The mean anogenital distance was reduced in males 300 mg/kg bw/day compared to control, and after adjustment for foetal body weight. It is noted that standard deviations were high. The incidence of external, visceral and skeletal foetal malformations and variations was not affected by sodium molybdate. The slight differences in the ossification status of foetuses in the 300 mg/kg bw/day group were confirmed as transient by skeletal examination of pups at Day 21 post-partum, and are consistent with the reduced foetal weight, associated with the marked maternal toxicity observed at this dose level (43).

Mouse study on development (Mohamed et al., 2020)

A study was undertaken to estimate the possible induction of genotoxicity and foetal abnormalities, especially foetal malformations and skeletal abnormalities by molybdenum nanoparticle administration in mice. Molybdenum nanoparticles with a size less than 100 nm were suspended in deionized distilled water and were orally administered to pregnant mice at dose levels of 500 or 750 mg/kg bw from the 1st up to the 17th day of pregnancy (groups 1 and 3, respectively) or from the 9th up to the 17th day of pregnancy (groups 2 and 4, respectively).

Oral administration of molybdenum nanoparticles resulted in significant decreases in the maternal body weight, the number and length of foetuses (no quantitative data) as well as skeletal abnormalities, mainly less ossification and less chondrification (Table 36).

Administration of molybdenum nanoparticles also caused DNA damage induction (as measured by Comet assay) and elevated expression of levels of p53, a gene involved in maintaining the genomic stability and cell differentiation in both maternal and foetus tissues. The expression levels of E-Cad and N-Cad genes that control skeleton development were increased in the tissues of female mice administered Mo nanoparticles and their foetuses (44).

Table 36 Percent of skeletal abnormalities in 18 day old mice foetuses in different groups (44).

Group defects	Control (%)	G1 (%)	G2 (%)	G3 (%)	G4 (%)
Less or unossification of frontal, parietal and inter parietal bones	0	70	50	100	0
Cartilaginous portion of parietal and inter parietal bones	0	20	10	50	0
Partial ossification of both vertebral column, ribs, and sternum	0	0	40	100	0
Less chondrification of nasal, sternal portion of ribs, subscapula, joints, carpus, tarsus	10	80	40	100	10
Less ossification of humerus, radius, and ulna	0	10	10	90	20
Partial of unossified of metacarpus, phalanges	0	0	25	90	40
Less ossification of femur, tibia, fibula	0	10	50	100	10
Partial or unossified of metatarsus, phalanges	0	30	50	90	40

G1: mice administered Mo nanoparticles at the dose level 500 mg/kg bw from the 1st up to the 17th day of pregnancy; **G2:** mice administered Mo nanoparticles at the dose level 500 mg/kg bw from the 9th up to the 17th day of pregnancy; **G3:** mice administered Mo nanoparticles at the dose level 750 mg/kg bw from the 1st up to the 17th day of pregnancy; and **G4:** mice administered Mo nanoparticles at the dose level 750 mg/kg bw from the 9th up to the 17th day of pregnancy

8.2.2 *Human data*

In the advice of the Health Council of the Netherlands on molybdenum in 2013 various studies in humans on the effects of molybdenum compounds on development were described.

Below additional studies that were published after release of the advice of the Health Council are described.

The studies in humans on the effects of molybdenum compounds on development are summarized in Table 37.

Table 37 Summary of human studies of molybdenum on development

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
Prospective cohort studies					
<ul style="list-style-type: none"> • Shirai et al., 2010 (45) • Prospective cohort study • Tokyo, Japan • 78 pregnant women visiting obstetric outpatient hospital clinic, without clinical signs of any diseases • 2007-2008 	<p>Single spot urine sample at 9-40 gestational weeks, during regular maternal health check-ups.</p> <p>Urinary concentrations of molybdenum and 9 other metals measured by ICP-MS, creatinine-corrected. LOD 5.9 µg/g creatinine</p>	<p>Birthweight (kg), birth length (cm), and head circumference (cm) of newborns at time of delivery.</p> <p>Statistical analysis: - correlation analysis (Pearson's r) - multivariable linear regression analysis, with adjustment for gestational age, sex of newborn, birth order, maternal BMI (maternal height for analysis of birth length), maternal age, maternal or paternal smoking, and other urinary metal concentrations</p>	<p>Urinary molybdenum concentrations (µg/g creatinine): GM (GSD) 79.0 (1.72), range 10.3-369</p> <p>No correlations or associations between urinary molybdenum concentrations and birthweight, birth length, and head circumference (correlation and regression coefficients not reported).</p>	<p>Potential residual confounding by e.g. alcohol intake or nutrition</p> <p>Measurement in urine sample only once and at variable stages of pregnancy.</p>	<p>Population: recruitment not clearly described and inclusion and exclusion criteria only globally described.</p>
<ul style="list-style-type: none"> • Vázquez-Salas et al., 2014 (46) • Prospective cohort study • Morales State, Mexico • 147 women of reproductive age 	<ul style="list-style-type: none"> • Urine samples collected during each trimester of pregnancy • Concentration of molybdenum (µg/L) determined in 	<ul style="list-style-type: none"> • Neurodevelopment: • Follow-up visits at 1, 3, 6, 12, 18, 24 and 30 months: <ul style="list-style-type: none"> - Bailey's scale (psychomotor (PDI) and mental (MDI) indexes) - Anthropometry - History of breastfeeding - Diet information 	<ul style="list-style-type: none"> • Molybdenum detected in all urine samples • Median molybdenum concentration levels: <ul style="list-style-type: none"> - First trimester: 38.5 µg/L; 48.9 µg/g creat. - Second trimester: 39.6 µg/L; 59.1 µg/g creat. 	<ul style="list-style-type: none"> • Overadjustment by one or more of the potential confounders cannot be ruled out. 	

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<p>and their infants randomly selected from 294 eligible women</p> <ul style="list-style-type: none"> January 2001 - June 2005 Postnatal follow-ups were set at first, third, and sixth months of life and after that every 6 months until 30 months of age <p>• Inclusion criteria:</p> <ul style="list-style-type: none"> urine sample available in at least one trimester of pregnancy at least 5 of 7 follow-up visits between 1 and 30 months after birth singleton pregnancy birth weight ≥ 2 kg mother aged >15 years 	<p>duplicate using electrothermal atomic absorption spectrometry</p> <ul style="list-style-type: none"> LOD 0.2 $\mu\text{g/L}$ CV (duplicate samples) 2.4% Molybdenum concentration also expressed as $\mu\text{g/g}$ creatinine 	<p>• Statistical analysis:</p> <ul style="list-style-type: none"> Associations between molybdenum concentration in each trimester of pregnancy and infant neurodevelopment (PDI and MDI separately) estimated using multivariable generalized mixed effect models with age at follow-up as random effect Molybdenum concentrations in each trimester assessed as [1] $\mu\text{g/L}$; [2] $\mu\text{g/L}$ with adjustment for creatinine concentration; and [3] $\mu\text{g/g}$ creatinine Adjustment for potential confounders (fixed effects): gestational age, parity, maternal age, education, occupation, and IQ, birth weight, type of birth, sex of child, breastfeeding, quality of home environment at 6 months of age, and prenatal DDE exposure 	<ul style="list-style-type: none"> Third trimester: 37.7 $\mu\text{g/L}$; 58.6 $\mu\text{g/g}$ creat. Inverse associations between urinary molybdenum concentration in <u>third</u> trimester and PDI, expressed as change of index by doubling of molybdenum concentration: <ul style="list-style-type: none"> [1] -0.49 (95% CI -0.1, 0.03) [2] -0.54 (95% CI -1.1, -0.002) [3] -0.57 (95% CI -1.1, -0.1) Adding blood lead levels during pregnancy to the multivariable models (n=64) did not change the results. No associations between urinary molybdenum concentrations in first and second trimester and PDI No associations between urinary molybdenum concentrations in any trimester and MDI 		

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<ul style="list-style-type: none"> • Exclusion criteria: <ul style="list-style-type: none"> - maternal history of chronic illness or treated with anticonvulsants - breastfeeding at time of recruitment - diagnosis of severe cerebral atrophy, early birth, neonatal death, congenital hypothyroidism, cleft lip and palate, or perinatal asphyxia - subjects with just one follow-up visit <p>Final study population: Random subsample of eligible study participants</p>					
<ul style="list-style-type: none"> • Bloom et al., 2015 including corrigendum (47, 48) • Prospective cohort study • Michigan (4 counties) and 	<ul style="list-style-type: none"> • Spot urine samples were collected from both mothers and fathers before conception 	<ul style="list-style-type: none"> • Baseline questionnaire (administered at home by research nurse) on demographics, health-related behaviours, medical history and reproductive histories 	<ul style="list-style-type: none"> • Molybdenum detected in all urine samples • Mothers (n=215): <ul style="list-style-type: none"> - 1st tertile 1.86-17.91 µg/L - 2nd tertile 17.91-50.44 µg/L - 3rd tertile 50.44-256.69 µg/L • Fathers (n=213): <ul style="list-style-type: none"> - 1st tertile 5.22-32.30 µg/L 	<ul style="list-style-type: none"> • 	<ul style="list-style-type: none"> • Strong point of the study is use of preconception exposure, also of fathers

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<p>Texas (12 counties), USA</p> <ul style="list-style-type: none"> • Study period: from planning of pregnancy until delivery • 235 couples with singleton pregnancies from 501 couples planning pregnancy, participating in the Longitudinal Investigation of Fertility and the Environment (LIFE) study Couples recruited from general population with presumed exposure to persistent organic pollutants • Inclusion criteria: <ul style="list-style-type: none"> - committed heterosexual relationship - women aged 18-40 years 	<ul style="list-style-type: none"> • Concentrations of molybdenum ($\mu\text{g/L}$) and 20 other elements determined using ICP-MS • LOD not reported (but $<1.86 \mu\text{g/L}$) • Concentrations of molybdenum divided into tertiles, separately for mothers and fathers 	<ul style="list-style-type: none"> • Gestational age at delivery (days since ovulation; determined on basis of daily urine measurements [Fertility Monitor]) • Health parameters studied: <ul style="list-style-type: none"> - Gestational age (days) - Birth weight (kg) - Birth length (cm) - Head circumference (cm) - Ponderal index: indicator of foetal growth proportionality, defined as $100 \times \text{birth weight} / \text{birth length}^3$ - Infant sex • Statistical analysis <ul style="list-style-type: none"> - Multivariable linear regression analysis for gestational age, birth weight, birth length, head circumference, and ponderal index as continuous outcomes - Cox-proportional hazards analysis for gestational age - Log-binomial models for infant sex 	<ul style="list-style-type: none"> - 2nd tertile 32.30-75.62 $\mu\text{g/L}$ - 3rd tertile 75.6-268.822 $\mu\text{g/L}$ • No statistically significant ($p > 0.05$) associations between maternal or paternal molybdenum concentrations before conception and any of the outcome parameters under study • Interaction between infant sex and continuous paternal molybdenum concentration for head circumference: <ul style="list-style-type: none"> - boys: -0.57 (95% CI -1.11, -0.03) cm - girls: 0.10 (95% CI -0.42, 0.62) cm • Gestational age ($n=195$); Hazard Ratios: <ul style="list-style-type: none"> - Molybdenum in mother's urine T2: 0.90; T3: 0.92; p for trend 0.802 - Molybdenum in father's urine T2: 0.80; T3: 1.52; p for trend 0.425 • Birth weight ($n=232$), Beta's: <ul style="list-style-type: none"> - Molybdenum in mother's urine T2: -63.35; T3: +68.41; p for trend 0.947 - Molybdenum in father's urine T2: -45.05; T3: -137.05; p for trend 0.354 		<ul style="list-style-type: none"> • Study participants presumed to be at risk of environmental exposure, but most values were relatively low compared to US population •

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<p>- men aged ≥ 18 years</p> <p>- English or Spanish speaking</p> <p>- no use of injectable contraceptive within 12 months</p> <p>- menstrual cycle length of 21–42 days</p> <ul style="list-style-type: none"> • Exclusion criteria - couples with sterilized partner or prior infertility diagnosis 		<p>- Both maternal and paternal urinary molybdenum concentration in tertiles (simultaneously in models); first tertile is reference category</p> <p>- Adjustment for maternal age, difference between maternal and paternal ages, maternal and paternal smoking, income, race, total serum lipids (a proxy for persistent organic pollutants), and creatinine</p> <p>- Due to missing values, Markov Chain Monte Carlo-based multiple-imputation procedure was implemented under an assumption of 'missing at random'</p> <p>- Analyses were repeated using log-transformed continuous molybdenum concentrations instead of tertiles to detect linear trends</p> <ul style="list-style-type: none"> • 	<ul style="list-style-type: none"> • Birth length (n=231), Beta's: <ul style="list-style-type: none"> - Molybdenum in mother's urine T2: -0.09; T3: $+0.61$; p for trend 0.922 - Molybdenum in father's urine T2: $+0.01$; T3: 0.69; p for trend 0.877 • Head circumference (n=182), Beta's: <ul style="list-style-type: none"> - Molybdenum in mother's urine T2: $+0.19$; T3: $+0.44$; p for trend 0.541 - Molybdenum in father's urine T2: -0.58; T3: -0.80; p for trend 0.309 • Ponderal index (n=231), Beta's: <ul style="list-style-type: none"> - Molybdenum in mother's urine T2: -3.79; T3: -3.66; p for trend 0.927 - Molybdenum in father's urine T2: -3.04; T3: $+0.59$; p for trend 0.459 • Newborn sex (n=233), SSR (ratio of male to female): <ul style="list-style-type: none"> - Molybdenum in mother's urine T2: 1.13; T3: 1.13; p for trend 0.136 - Molybdenum in father's urine T2: 1.14; T3: 1.09; p for trend 0.869 		

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<ul style="list-style-type: none"> • Howe et al., 2020 (49) • Prospective cohort study • Los Angeles, CA, USA • 2015 – 2019 • Population: 262 participants in the Maternal and Developmental Risks from Environmental and Social Stressors (MADRES) study, i.e. pregnant women recruited at one of four prenatal care providers in LA, mainly lower-income Hispanic populations • Exclusion criteria: pregnancy ≥ 20 wks of gestation at recruitment, < 18 years of age, hiv positive, physical, 	<p>Spot urine samples collected during first study visit (median gestational age 13.1 weeks)</p> <p>Concentrations of molybdenum and 9 other metals measured by ICP-MS</p>	<ul style="list-style-type: none"> - Birth weight for gestational age and sex, z-scores based on a 2017 US reference (Airs, et al. 2019) - Birth weight measures obtained from medical records; if missing (n=22) based on information from mother - Gestational age estimates using ultrasound or observation at birth (physician's estimate). <p>Statistical analysis:</p> <ul style="list-style-type: none"> - primary analysis focused on combination of 7 metals, excluding molybdenum. - secondary exploratory analysis included molybdenum and all other metals, but excluded 3 participants with unusually low molybdenum concentrations - associations between metal mixture and outcome analysed using Bayesian kernel machine regression 	<p>Urinary molybdenum concentrations (urine specific gravity corrected, $\mu\text{g/L}$): median 56.8, IQR 42.9-80.7</p> <ul style="list-style-type: none"> • Posterior inclusion probability for molybdenum in secondary exploratory analysis was 0.41, which ranked in fifth place of importance. No dose-response relation was observed for molybdenum concentration and birth weight z-scores. 	<p>Only subjects with complete covariate information were included.</p>	<p>Focus of this study on mixtures of metals.</p> <ul style="list-style-type: none"> • Impoverished urban study population, probably above average at risk of exposure and intra-uterine growth retardation

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<p>mental or cognitive disability, multiple gestation, incarceration, no urine sample at first visit, missing covariate information</p> <ul style="list-style-type: none"> Other publication in the same cohort: Howe et al. (2021), different health outcome and fewer participants 		<p>Directed acyclic graphs (DAGs) were used to identify potential confounders: recruitment site, self-reported maternal age, pre-pregnancy BMI, race by ethnicity and birth place, and smoke exposure during pregnancy, as well as measured pregnancy anemia and urinary arsenobetaine (as marker of fish consumption).</p>			
<ul style="list-style-type: none"> Kim et al., 2020 (50) Prospective cohort study Boston, MA, USA 2006-2008 390 participants in LIFECODES birth cohort study, i.e. pregnant women planning hospital delivery, enrolled before week 15 of 	<p>Concentrations of molybdenum and 16 other metals in urine samples, collected at median 26 range 20-32) weeks of pregnancy, measured with ICP-MS and corrected for</p>	<p>Parameters of foetal growth, measured by ultrasound at weeks 26 (median, range 20-32) and 35 (median, range 30-40), following guidelines of ACOG:</p> <ul style="list-style-type: none"> - Abdominal circumference (mm) - Head circumference (mm) - Femur length (mm) - Estimated foetal weight (EFW) from these measures, following Hadlock formula; 	<p>Molybdenum concentrations all above LOD. Molybdenum specific-gravity-corrected concentrations (ppb), weighted for case control design, median (IQR): 51.3 (37.1-69.7).</p> <p>Single metal models adjusted differences in z-scores for repeated measures of foetal growth associated with IQR increase in urinary molybdenum (β (95% CI)): Femur length 0.15 (-0.05, 0.35)</p>	<p>Ultrasounds at weeks 26 and 35 (visits 3 and 4) were taken at participant's request or when abnormality suspected → availability of ultrasound measurements was selective (sampling bias).</p>	

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<p>pregnancy and participating in up to four study visits</p> <ul style="list-style-type: none"> • Inclusion criteria: pregnancy resulting in preterm birth (n=130, almost all occurrences) or in at term birth (n=352, randomly selected in 3:1 ratio) originally selected for nested case-control study • Exclusion criteria: no urine sample from third study visit (=26 weeks of pregnancy) available. 	<p>urine specific gravity LOD 0.30 pbb Demographics, lifestyle factors, medical and pregnancy history obtained by questionnaire.</p>	<p>- Z-scores based on gestational age at scan, with all singleton pregnancies in the hospital in 2006-2012 as reference.</p> <p>- Birth weight (g), birth length (cm), and placental weight (g) (in subset).</p> <p>Statistical analysis:</p> <p>- Linear mixed effect models for associations between metals and repeated outcome measures (at 26 weeks, 35 weeks, birth),</p> <p>- Linear regression for associations with birth weight, birth length, and placental weight.</p> <p>- Co-variables (in all adjusted models): urine specific gravity, maternal age, race/ethnicity, education, pre-pregnancy BMI, type of insurance, self-reported use of alcohol and tobacco, assisted reproduction, gestational age at time of ultrasound, gestational age at delivery (when</p>	<p>Head circumference -0.05 (-0.22, 0.13) Abdominal circumference -0.02 (-0.21, 0.17) EFW + birth weight 0.02 (-0.14, 0.18) No associations with birth weight, birth length, and placental weight in adjusted linear regression analyses for single metals.</p> <p>Multi-metal models adjusted differences in z-scores for repeated measures of foetal growth associated with IQR difference in urinary molybdenum (β (95% CI)): Femur length 0.30 (0.08, 0.52) Head circumference 0.16 (-0.04, 0.36) Abdominal circumference 0.11 (-0.11, 0.33) EFW + birth weight 0.12 (-0.07, 0.31) No significant associations with birth weight, birth length, and placental weight in adjusted linear regression analyses for multi metals. Birth weight 19.9 (-74.5, 114.3); Birth length 0.16 (-0.34, 0.66); Placenta weight -0.78 (-44.8, 43.3). Adjusted for other metals, specific gravity, maternal age, pre-pregnancy BMI, race/ethnicity, education, infant</p>	<p>Measurement of metals in urine only at one time point at varying pregnancy durations, which might impact metal concentrations due to metabolic changes.</p>	

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
		<p>appropriate), and metal co-exposure (in multi-metal models)</p> <ul style="list-style-type: none"> - Inverse probability weighting (IPW) to account for case-control selection - Sensitivity analyses (amongst others) on missing data (multiple imputation by chained equation method) 	<p>sex, and gestational age at time of ultrasound or delivery (EFWZ + BWZ). Unadjusted results similar.</p> <p>Single metal models adjusted differences in z-scores for repeated measures of foetal growth associated with IQR difference in urinary molybdenum (β (95% CI)): FLZ 0.15 (-0.05, 0.35); HZC -0.05 (-0.22, 0.13); AZC -0.02 (-0.21, 0.17); EFWZ + BWZ 0.02 (-0.14, 0.18). Birth weight 19.9 (-74.5, 114.3); Birth length 0.16 (-0.34, 0.66); Placenta weight -0.78 (-44.8, 43.3). Adjusted for specific gravity, maternal age, pre-pregnancy BMI, race/ethnicity, education, infant sex, and gestational age at time of ultrasound or delivery (EFWZ + BWZ).</p> <p>Sensitivity analyses showed similar associations with slightly attenuated effect estimates.</p>		
<ul style="list-style-type: none"> • Howe et al., 2021 (51) • Prospective cohort study 	Spot urine samples collected during first study visit	Mid-pregnancy foetal growth measures evaluated at 18-22 weeks (median 20.4) of	Urinary molybdenum concentrations (urine specific gravity corrected, $\mu\text{g/L}$): median 57.4, IQR 44.3-81.1	Only subjects with complete covariate information were included.	Focus of this study on mixtures of metals.

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<ul style="list-style-type: none"> Los Angeles, CA, USA 2015 – 2019 Population: 193 participants in the Maternal and Developmental Risks from Environmental and Social Stressors (MADRES) study, i.e. pregnant women recruited prior to routine anatomy ultrasound scan at one of four prenatal care providers in LA, mainly lower-income Hispanic populations Exclusion criteria: pregnancy ≥ 20 wks of gestation at recruitment, < 18 years of age, hiv positive, physical, mental or cognitive disability, multiple 	<p>(median gestational age 12.4 weeks)</p> <p>Concentrations of molybdenum and 9 other metals measured by ICP-MS</p>	<p>pregnancy and obtained from medical records:</p> <ul style="list-style-type: none"> - Abdominal circumference - Head circumference - Biparietal diameter - Femur length - Estimated foetal weight (EFW) <p>EFW was main outcome in statistical analysis.</p> <p>Statistical analysis:</p> <ul style="list-style-type: none"> - primary analysis focused on combination of 6 metals, including molybdenum. - secondary exploratory analysis included molybdenum and all other metals - associations between metal mixture and outcomes analysed using Bayesian kernel machine regression - metals with high-ranking posterior inclusion probabilities further analysed with linear regression models <p>Directed acyclic graphs (DAGs) were used to identify</p>	<p>Posterior inclusion probabilities for molybdenum ranked highest in both primary (0.631) and secondary analysis (0.485) for EFW. Setting other metals to their median, an increase in molybdenum concentration from the 25th to 75th percentile was associated with a 0.114 (95% CI: 0.019, 0.247) SD higher EFW, equivalent to a ~ 7.4 g higher EFW. Visually, this association was attenuated at higher levels of barium (Ba), but the p value for interaction between molybdenum and Ba was 0.22 in linear regression analysis.</p> <p>Based on posterior inclusion probabilities, molybdenum consistently contributed most to the associations with all other fetal growth parameters as well. An interquartile change in molybdenum concentration was associated with a 0.30 (95% CI: 0.05, 0.56) SD difference in head circumference. Visual attenuation of the positive association of molybdenum with head circumference at higher levels of Ba was confirmed in linear regression analysis (p for interaction 0.03).</p>		<p>Impoverished urban study population, probably above average at risk of exposure and intra-uterine growth retardation.</p>

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<p>gestation, incarceration, no urine sample at first visit, missing covariate information, low molybdenum concentration (n=2).</p> <p>Other publication in the same cohort: Howe et al. (2020), different health outcome and more participants</p>		<p>potential confounders: recruitment site, gestational age at ultrasound, self-reported maternal age, pre-pregnancy BMI, race by ethnicity and birth place, education, infant sex, parity, prenatal vitamin use, and smoke exposure during pregnancy, as well as measured urinary arsenobetaine (as marker of fish consumption).</p>			
<ul style="list-style-type: none"> • Karakis et al., 2021 (52) • Prospective cohort study • Negev desert, Israel • Dec 2011 – Mar 2013 plus 5.4-6.9 years of follow-up • 111 mothers and their singleton 	<ul style="list-style-type: none"> • Urine samples collected just prior to delivery • Concentration (ppb) of molybdenum and 24 other metals determined using ICP-MS 	<ul style="list-style-type: none"> • Medical records prepared by local hospital and/or HMO personnel provided information on: <ul style="list-style-type: none"> - preterm delivery - small-for-gestational age (SGA) - congenital malformations - behavioural/developmental disorders 	<ul style="list-style-type: none"> • Molybdenum o concentration in urine (ppb): P40=19.64; P60=34.76 GM 7.23 (95% CI: 3.86, 13.55) • Associations between molybdenum concentration in quintiles and clinical outcome: adjusted Relative Risks (p-values) <ul style="list-style-type: none"> - preterm delivery: 1.32 (0.129) - congenital malformations: 0.89 (0.655) 	<ul style="list-style-type: none"> • 	<ul style="list-style-type: none"> • Explorative analysis • Relatively small size and hence limited statistical power for detecting relevant associations

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<p>newborns of Bedouin-Arab origin, recruited at obstetrics emergency department of Soroka University Medical Center (SUMC)</p> <ul style="list-style-type: none"> • Inclusion criteria: <ul style="list-style-type: none"> - member of 'Clalit' health maintenance organization (HMO) - ≥18 years of age - urine sample collected prior to birth - newborn survived birth hospitalization 	<ul style="list-style-type: none"> • LOQ=0.01 ppb 	<ul style="list-style-type: none"> - other disorders during follow-up • Statistical analysis: <ul style="list-style-type: none"> - metal concentrations ranked into quintiles - associations with health outcomes analysed using Poisson regression analysis, adjusted for maternal age, parity, newborn gender, and preterm birth 	<ul style="list-style-type: none"> - behavioural or developmental disorders: 1.86 (0.016) - Malformations: 0.89 (0.655) - No results reported for SGA 		<ul style="list-style-type: none"> • Molybdenum concentrations analysed as number of quintile; absolute concentration level not taken into account
<ul style="list-style-type: none"> • Mandiá et al. (2021) (37) • Prospective cohort study • 170 nursing mothers: <ul style="list-style-type: none"> - 70 full-term - 100 pre-term • University Clinical Hospital of 	<ul style="list-style-type: none"> • Human milk samples (5–10 mL) obtained from 70 mothers of full-term newborns: <ul style="list-style-type: none"> - colostrum during the first 	<ul style="list-style-type: none"> • Questionnaire including birth weight of newborn • Statistical analysis: <ul style="list-style-type: none"> - ANOVA and Bonferroni tests for differences in molybdenum concentrations between groups - Regression models of molybdenum concentration 	<ul style="list-style-type: none"> • Mean concentrations of molybdenum in milk from full-term mothers: <ul style="list-style-type: none"> - colostrum: 1.88 (SD 1.20) µg/L - intermediate milk: 1.22 (SD 1.98) µg/L - later mature milk: 0.96 (SD 1.16) µg/L (p<0.001 lower than colostrum) • Mean concentrations of molybdenum in milk from pre-term mothers: 	<ul style="list-style-type: none"> • Authors denote regression models on determinants as 'multivariate analysis', but is unclear whether adjusted for the other determinants and/or potential confounders. In 	<ul style="list-style-type: none"> • The design is referred to as "Prospective case series", which does not sound like a conventional design and

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<p>Santiago de Compostela, Spain</p> <ul style="list-style-type: none"> • Period 2018-2019 • Inclusion criteria: <ul style="list-style-type: none"> - maternal age > 18 years • Exclusion criteria: <ul style="list-style-type: none"> - chronic disease - taking nutrient supplements 	<p>3–4 days of lactation</p> <ul style="list-style-type: none"> - intermediate milk up to 7–10 days of lactation - later mature milk (up to 6 months after birth) • Human milk samples (5–10 mL) obtained from 100 mothers of premature newborns: <ul style="list-style-type: none"> - later mature milk (up to 6 months after birth) • Concentration ($\mu\text{g/L}$) of molybdenum determined using ICP-OES (inductively coupled plasma optical emission) 	<p>(dependent variable) on selected medical factors (independent variables; either categorical or continuous)</p> <ul style="list-style-type: none"> - Unclear whether data from all mothers and all samples were pooled • - Adjustment unclear 	<ul style="list-style-type: none"> - later mature milk: 0.70 (SD 1.17) $\mu\text{g/L}$ (unclear whether this was significantly lower than 0.96 in later mature milk from full-term mothers?) • Associations with birth weight (using all samples from all 170 mothers?!): Beta = -0.17 ($p=0.002$); thus molybdenum levels were on average lower in milk from mothers who had delivered heavier babies. Unit unclear; $\mu\text{g/mL}$ per g unit of birth weight? 	<ul style="list-style-type: none"> • that case, risk of confounding bias 	<ul style="list-style-type: none"> • it remains unclear whether is (partly) a case-control study (premature vs. full-term)? • Definition of pre-term not provided (presumably <37 weeks of gestation?) • Timing of later mature milk sample not clear (up to 6 months after birth?) • Statistical significance in tables is puzzling; unclear what was tested

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
	spectrophotometry) • Detection limit not reported • (Also 5 minerals, 8 other essential trace elements and 21 toxic trace elements determined)				exactly. Hard to believe many of the p-values • In analysis of determinants (Table 5), it looks like all data were pooled, thus including 3 samples for the same mother without controlling for dependency • The latter is not a prospective analysis; more a cross-sectional analysis and exposure

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
					was determined <i>after</i> the endpoints
<ul style="list-style-type: none"> • McKeating et al., 2021 (53) • Prospective cohort study • Adelaide, Australia • 128 pregnant women • May 2009 - July 2013 • Inclusion criteria: <ul style="list-style-type: none"> - Pregnant women attending first antenatal visit - aged >18 years 	<ul style="list-style-type: none"> • Urine and blood plasma samples were collected at 18-week visits at the clinic • Concentrations of molybdenum ($\mu\text{g/L}$ in plasma and ng/L in urine) and 36 other elements were determined using ICP-MS • LOD/LOQ not reported 	<ul style="list-style-type: none"> • Neonatal data were collected at delivery: <p>This resulted in</p> <ul style="list-style-type: none"> - 13 preterm birth (<37 weeks) - 10 small for gestational age (SGA / lowest 10%) - 18 low placental weight (lowest 25%) - 87 healthy infants (controls) <ul style="list-style-type: none"> • Statistical analysis <ul style="list-style-type: none"> - Associations between element levels and neonatal outcomes: <ul style="list-style-type: none"> - One-way ANOVA for differences in urinary and plasma molybdenum in the 4 groups followed by post-hoc pairwise comparisons 	<ul style="list-style-type: none"> • <u>Plasma</u> concentration (mean\pmSD in $\mu\text{g/L}$) <ul style="list-style-type: none"> - Preterm birth: 0.71 ± 0.15 - Low placental weight: $0.6\pm 0.15^*$ - Small for gestational age: 0.72 ± 0.13 - Controls: 0.89 ± 0.32 - * $p=0.0001$ for difference with controls • <u>Urine</u> concentration (mean\pmSD in ng/L) <ul style="list-style-type: none"> - Preterm birth: 4.12 ± 2.1 - Low placental weight: 4.56 ± 2.56 - Small for gestational age: 3.34 ± 0.98 - Controls: 4.66 ± 2.34 <p>No statistically significant ($p>0.05$) differences between infants with adverse outcomes and controls</p>	<ul style="list-style-type: none"> • Differences in urinary and plasma molybdenum between groups using ANOVA was not taking into account potential confounders 	<ul style="list-style-type: none"> • Plasma samples were not available for all participants • Small sample sized of adverse neonatal outcomes • Placental weight is not a recognised developmental outcome

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<ul style="list-style-type: none"> • Tung et al. (2022)(54) • Prospective cohort study • Rhode Island, US • 192 mother-infant pairs recruited from hospital in Rhode Island into Rhode Island Child Health Study (RICHS) • Oversampling of term infants born large for gestational age and small for gestational age • 2010 – 2011 • Inclusion criteria: <ul style="list-style-type: none"> - mothers ≥ 18 years without life-threatening medical complications - infants born free of life-threatening medical complications or congenital or 	<p>Placenta parenchyma tissue biopsied approximately 2 cm from cord insertion site and free of maternal decidua within 2 hours of delivery. Placental levels of molybdenum and 23 other trace elements analyzed with ICP-MS.</p>	<p>Newborn neurobehavioral performance assessed with NICU Network Neurobehavioral Scale (NNNS), administered by certified psychometrists 24 – 72 hours after birth. NNNS score patterns categorized into 5 profiles of which profile 5 indicates most atypical neurobehavioral performance.</p> <p>Statistical analysis:</p> <ul style="list-style-type: none"> - associations between individual metals and NNNS profiles (profile 5 vs. other profiles) assessed with multivariable logistic regression models, adjusted for infant sex, maternal age, maternal race, pre-pregnancy BMI, education, and smoking status during pregnancy - quantile g-computation for association between mixture of 8 metals (including molybdenum) and NNNS profile 5 vs. other profiles 	<p>Placental molybdenum concentration (ng/g): mean 6.76, median 6.58, IQR 5.85-7.42</p> <p>Adjusted odds ratios for NNNS profile 5 per doubling of placental molybdenum concentration approximately 2 with large 95% CI including unity.</p> <p>Molybdenum played a very small role in association between metal mixture and NNNS profiles</p>	<p>As the RICHS cohort focused on aberrant fetal growth, over half of the included infants were born small or large for gestational age, but analyses were not adjusted for birth weight category.</p>	

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
chromosomal abnormalities					
Case-control studies					
<ul style="list-style-type: none"> • Yan et al., 2017 (55) • Case-control study • Shanxi Province and Hebei Province, China • 2003-2007 • Cases: 191 women with a pregnancy complicated by a neural tube defect (live births, stillbirths, and pregnancy terminations) • Controls: 261 women who delivered full-term healthy infants in same birthing hospital, loosely matched on county/city of residence and last menstrual period 	<p>Unpainted maternal hair sections, grown from 1 month before to 2 months after conception (assuming hair growth rate of 1 cm per month) collected</p> <p>Molybdenum and 8 other essential trace metals measured with ICP-MS. LOD not reported</p>	<p>NTD subtypes: anencephaly (n=85), spina bifida (n=79), encephalocele (n=24), and unspecified (n=3).</p> <p>Statistical analysis: Comparison of hair molybdenum concentrations between groups: Mann-Whitney U test</p> <p>Unconditional multivariable logistic regression with dichotomized molybdenum concentrations based on median in controls as cut-off value and correction for covariables. Dose-response analysis by estimating adjusted ORs for quartiles of molybdenum concentrations based on quartiles in controls .</p> <p>Co-variables: maternal age, occupation, education,</p>	<p>Molybdenum concentrations in maternal hair (median, IQR in ng/mg hair) and adjusted odds ratios (95% CI):</p> <p>Total NTDs (n=191) cases: 0.071 (0.062-0.084) controls: 0.075 (0.063-0.088) P-value 0.032 Adjusted OR: 0.64 (0.42-0.98)</p> <p>Anencephaly (n=85) Cases: 0.071 (0.062-0.084) Controls: 0.075 (0.063-0.088) P-value 0.070 Adjusted OR: 0.51 (0.28-0.94)</p> <p>Spina bifida (n=79) Cases: 0.071 (0.063-0.082) Controls: 0.075 (0.063-0.088) P-value 0.039 Adjusted OR: 0.54 (0.31-0.94)</p> <p>Encephalocele (n=24) cases: 0.074 (0.062-0.091)</p>	<p>Differences between cases and controls for several co-variables, that were adjusted for in the analyses (maternal education, history of previous birth defects, fever or flu during early pregnancy, and active or passive smoking)</p> <p>Subjects were from regions geographically far apart.</p> <p>No multivariable analyses with co-exposure to other metals.</p> <p>To maximize the sample size,</p>	<p>Moment of hair sampling is unknown. Probably 2 months post-conception, as the authors assume that hair grows 3 cm in 3 months' time and they state they analysed hair grown in the period 1 month pre-conception until 2 months post-conception. Unexpected inverse associations observed between</p>

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
		<p>gravidity, history of previous birth defects, fever or flu during early pregnancy, alcohol consumption, periconceptual folate supplementation, active or passive smoking during periconceptual period (collected by face-to-face interview within first week after the end of pregnancy).</p>	<p>controls: 0.075 (0.063-0.088) P-value 0.786 Adjusted OR: 0.74 (0.29-1.94)</p> <p>Dose-response analysis showed decreasing trends in adjusted ORs (95% CI) with increasing quartiles of molybdenum concentrations for total NTD, anencephaly, and spina bifida.</p> <p>Total NTDs (n=191) Q1: 1.00 (ref) Q2: 1.08 (0.65-1.79) Q3: 0.95 (0.57-1.61) Q4: 0.58 (0.33-1.02)</p> <p>Anencephaly (n=85) Q1: 1.00 Q2: 0.95 (0.43-2.09) Q3: 0.66 (0.29-1.48) Q4: 0.35 (0.14-0.87)</p> <p>Spina bifida (n=79) Q1: 1.00 Q2: 1.15 (0.55-2.42) Q3: 0.79 (0.36-1.72) Q4: 0.38 (0.16-0.92)</p> <p>Encephalocele (n=24)</p>	<p>matched pairs were separated for the analysis with unconditional logistic regression.</p>	<p>molybdenum concentrations in maternal hair and NTDs in offspring.</p>

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
			Q1: 1.00 Q2: 0.77 (0.19-3.17) Q3: 0.36 (0.07-1.71) Q4: 1.02 (0.27-3.89)		
<ul style="list-style-type: none"> • Deyssenroth et al., 2018 (56) • Case-control study • Rhode Island, US • 2009-2013 • 195 mother-infant pairs selected from Rhode Island Child Health Study (RICHS) • Inclusion criteria: <ul style="list-style-type: none"> - mothers ≥18 years - infants without congenital or chromosomal abnormalities - complete molecular profile (placental RNA-Seq) and metal exposure data available • Cases: infants small for gestational age 	Maternal toenail clippings following hospital discharge (average time to collection 2.8 months (range, 0.3–7.1 months) postpartum) Concentration of Mo and 18 other metals analysed using ICP-MS.	Anthropometrics from structured reviews of medical records. Statistical analyses: Associations between molybdenum concentration and SGA status assessed using logistic regression. Metal mixture indices associated with SGA status derived with weighted quantile sum (WQS) regression; robustness of the major drivers of SGA status assessed with Bayesian kernel machine regression (BKMR). All regression models adjusted for infant gender, maternal ethnicity, maternal BMI, and maternal smoking status during pregnancy.	Mean (SD; min-max) molybdenum toenail concentrations ($\mu\text{g/g}$ dry weight): 0.018 (0.032; 0.002-0.366). Adjusted OR (95% CI) for association between log unit increase in molybdenum concentration and SGA (approximated from figure): 0.8 (0.4-1.2) WQS multi-metal index weight for molybdenum near zero, indicating low variable importance in driving the association of metal mixtures with SGA, corroborated in BKMR analysis.		

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<p>(SGA, <10% percentile)</p> <ul style="list-style-type: none"> • Controls: infants born appropriate for gestational age (AGA), matched to cases on gender, gestational age, and maternal age 					
<ul style="list-style-type: none"> • Hou et al., 2019 (57) • Nested case-control study • Guangxi Province, China • 2015-2016 • Participants in Guangxi Birth Cohort Study • Cases: 246 women with low birth weight children • Controls: 409 women with normal birth weight children, matched on maternal age, infant gender, 	<p>Concentrations of molybdenum and 21 other metals measured with ICP-MS in serum samples collected during prenatal examination.</p>	<p>Health outcome: birth weight - low birth weight (cases) < 2500 g - normal birth weight: 2500-4000 g</p> <p>Measures obtained from medical records database.</p> <p>Statistical analysis: Single metals associated with LBW using conditional logistic regression with quartiles of molybdenum concentration, adjusted for pre-pregnancy BMI, alcohol consumption pre-pregnancy, passive smoking during pregnancy, gravidity, and parity.</p> <p>Restricted cubic splines (RCS) to assess dose-</p>	<p>Serum molybdenum concentration ($\mu\text{g/L}$), median (IQR): - cases: 1.18 (0.90; 1.64)</p> <p>- controls: 1.07 (0.89; 1.37)</p> <p>Adjusted OR (95%CI) per quartile of molybdenum (based on distribution among controls):</p> <ul style="list-style-type: none"> - Q1: ($\leq 0.90 \mu\text{g/L}$): 0.75 (0.48-1.16) - Q2: (0.91-1.07 $\mu\text{g/L}$): 0.48 (0.30-0.77) - Q3: (1.08-1.37 $\mu\text{g/L}$): 0.71 (0.45-1.10) - Q4: ($>1.37 \mu\text{g/L}$): 1.00 (ref) <p>- P-trend: 0.018.</p> <p>Stratification according to gender, gestational age (\leq vs. >13 weeks), maternal age (\leq vs. >28 y) resulted in similar ORs.</p>	<p>Pre-pregnant BMI distribution different ($p=0.001$) between cases and controls, with more cases being underweight (BMI <18.5), but adjusted for in the analysis.</p> <p>Gestational age at delivery lower in cases than controls (35.5 versus 39.1 y, $p<0.001$), as expected with LBW.</p>	

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<p>gestational age at sample collection, and enrolment hospital in a 1:2 ratio</p> <ul style="list-style-type: none"> Exclusion criteria: multiple pregnancy, gender information missing, serum sample missing 		<p>response of molybdenum with LBW, adjusted for pre-pregnancy BMI.</p> <p>Multi-metal exposure analysed by elastic net regression followed by conditional logistic regression, adjusted for same potential confounders.</p>	<p>RCS analysis: no dose-response relationship between serum molybdenum concentration and LBW.</p> <p>In multi-metal analysis, 15 metals including molybdenum were associated with LBW: OR 5.41 (2.81-9.40)</p>		
Cross-sectional studies					
<ul style="list-style-type: none"> • Ovayolu et al., 2020 (58) • Cross-sectional study • Gaziantep, Turkey • November 2017 - July 2018 • Cases: 36 women with foetuses with neural tube defects (NTDs) • Controls: 39 women with unaffected foetuses, matched for maternal BMI and gestational 	<p>Amniotic fluid collected during amniocentesis in gestational weeks (mean (SD)):</p> <ul style="list-style-type: none"> - cases 21.6 (6.6) - controls 19.6 (2.4); P=0.096. <p>Concentrations of molybdenum and 13 other metals determined by ICP-MS.</p>	<p>Diagnosis of NTD with ultrasonographic examinations in pregnancy weeks 16-37.</p> <p>Statistical analyses: Differences in molybdenum concentrations between cases and controls tested with Student's t test.</p> <p>Co-variables assessed, but not matched or adjusted for: maternal age, parity, gravidity, previous births, abortion, frequency of sea food consumption, presence</p>	<p>Molybdenum concentration ($\mu\text{g/L}$; mean (SD)):</p> <ul style="list-style-type: none"> - cases 1.11 (1.06); - controls 2.47 (1.92); <p>P<0.001</p>	<p>Cases were younger than controls (27.1 vs 31.3 years, p=0.014) and less frequently had a history of abortion (0.2 vs 0.5, p=0.036).</p>	<p>No multivariable analysis with adjustment for potential confounders or co-exposure to other metals.</p>

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<p>weeks, who underwent amniocentesis because of age-related risk or increased risk in triple test.</p> <ul style="list-style-type: none"> Exclusion criteria: age <18 years, pregnancies conceived through artificial reproductive techniques, previous pregnancy affected by NTD, chronic diseases, drug use, non-use of folic acid in early weeks of pregnancy, and obstetric complications. 	LOD not reported.	of dental amalgam, and smoking/passive smoking status.			
<ul style="list-style-type: none"> Pi et al., 2019 (59) Cross-sectional study Shanxi Province, China 	Approximately 6 g of tissue from fetal portion of placenta sampled after delivery or	Diagnosis of OFC by county healthcare workers through physical examinations and prenatal ultrasound scans.	Placental molybdenum concentrations (ng/g dry weight; median (IQR)) in cases and controls: 35.9 (31.7–41.8) and 32.1 (27.3–37.0), respectively; P<0.001.	Placental molybdenum concentrations may not reflect concentrations during the critical	Analyses not adjusted by folic acid use because folic acid use was similar (ca.

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<ul style="list-style-type: none"> • January 2003 through December 2016 • Rural population • Cases: 103 newborns and terminated fetuses with orofacial clefts (OFC) • Controls: 206 newborns without congenital malformations, randomly selected from 509 non-malformed newborns with available placental tissues, matched to cases by mother's residence, date of last menstrual period, and newborn sex. 	<p>pregnancy termination</p> <p>Placental concentrations of molybdenum and 5 other metals assessed with ICP-MS. Analysis staff blinded to case or control status.</p>	<p>Face-to-face interviews by local health care workers based on structured questionnaire to assess information on potential confounders.</p> <p>Statistical analyses: Differences between cases and controls in median molybdenum concentrations tested with Mann-Whitney U-test. Association between above/below median molybdenum concentrations and OFC estimated with unconditional logistic regression, adjusted for maternal age, BMI, farming occupation, influenza or fever, passive smoking, alcohol drinking during periconceptional period, gestational age, and history of pregnancy affected by birth defects.</p> <p>Dose-response analysis comparing ORs for tertiles of molybdenum concentration</p>	<p>Associations between above (≥ 33.6 ng/g) vs. below (< 33.6 ng/g) median molybdenum concentrations and risk of OFCs: crude OR 2.20 (1.36, 3.58), adjusted OR 1.42 (0.78, 2.59).</p> <p>ORs for tertiles of molybdenum concentration: < 30.1 ng/g: reference value $30.1-36.6$ ng/g: crude OR 2.30 (1.22, 4.32), adjusted OR 1.98 (0.95, 4.13) ≥ 36.6 ng/g: crude OR 3.14 (1.68, 5.87), adjusted OR 1.46 (0.67, 3.21) P for trend 0.001 (crude) and 0.354 (adjusted).</p>	<p>period of neural tube development, because tissue sampling occurred mostly in 2nd or 3rd trimester.</p> <p>Potential overadjustment by gestational age, which should not be included as a confounder</p> <p>No adjustment for co-exposure to other metals</p>	<p>50%) in the case and control groups.</p>

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<ul style="list-style-type: none"> • Troisi et al., 2019 (60) • Cross-sectional study • Three hospitals in Southern Italy • Study period January 2011 to December 2013 • Cases: 111 pregnant women diagnosed with foetal malformations or foetal chromosomal abnormalities, recruited during second trimester termination of pregnancy • Exclusion criteria: age > 40 y, twin pregnancy, women committed to carrying the pregnancy to term, TORCH (Toxoplasma, Rosolia, 	<p>Fasting blood samples collected in 2nd trimester immediately before termination of pregnancy and before any drug administration (cases) or during the routine scan (controls).</p> <p>Serum concentrations of molybdenum and 43 other metal(loid)s determined with ICP-QMS.</p> <p>Clinical history and demographics assessed by questionnaire and complete obstetric visit at enrolment.</p>	<p>Foetal malformations or chromosomal abnormalities assessed with ultrasound examination or karyotype, confirmed by postmortem autopsy by expert pathologist or after pediatric examination.</p> <p>Statistical analysis: Differences between cases and controls tested with independent t-test with Bonferroni correction (adjusted $\alpha=0.0006$).</p> <p>Comparison of metal distribution between the two case groups and controls using Principal Component Analysis (PCA) and Partial Least Squares Discriminant Analysis (PLS-DA).</p>	<p>No differences in natural logarithm (mean \pm SD) of serum molybdenum concentrations ($\mu\text{g/L}$) between controls (-3.09 ± 1.10), CNS group (-2.48 ± 0.57), and other malformations group (-4.12 ± 1.10), $p>0.0006$.</p> <p>PCA: no aggregation of subjects. PLS-DA: molybdenum was 9th among the 15 metal(oids) most important in distinguishing the CNS group from the other malformations group</p>	<p>Analyses were not adjusted for potential confounders.</p>	<p>Low number of cases in the CNS group.</p> <p>No information provided on the types of malformations .</p>

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<p>Citomegalovirus, Herpes) complex infection, or CNS defects with a known genetic cause. Subdivision into two groups: all CNS malformations with unknown aetiology (n=17) and all other malformations or chromosomal abnormalities (n=94).</p> <ul style="list-style-type: none"> • Controls: 90 women with normally developed fetuses at the same week of pregnancy, recruited during second trimester routine anomaly scan. 					
<ul style="list-style-type: none"> • Manduca et al., 2020 (61) • Cross-sectional study 	<ul style="list-style-type: none"> • Hair samples from the mothers were 	<ul style="list-style-type: none"> • Birth outcomes and defects: 	<ul style="list-style-type: none"> • 95-percentile molybdenum levels (95% CI) in ppm 2015, n=502: 0.26 (0.21-0.32) 2016, n=78: 0.22 (0.12-0.41) 		<ul style="list-style-type: none"> • In general, the text is hard to follow with

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<ul style="list-style-type: none"> • Gaza strip, Palestine Study period: 2015, 2016 and 2018-2019 • Population: 644 mother-child pairs from singleton births - Babies born in maternity hospital • Exclusion criteria: <ul style="list-style-type: none"> - Not enough hair at birth - Known history of birth defects within the family 	<ul style="list-style-type: none"> taken before delivery • Concentration of molybdenum determined using ICP-MS • Expressed as ppm (=mg/kg) • LOD/LOQ not reported • This should reflect the environmental exposure of the woman to toxicants/teratogens in the last 4–5 months • Mothers' hair was analysed in 2018–2019 for <ul style="list-style-type: none"> - 17 mothers with a normal baby - 23 with BD 	<ul style="list-style-type: none"> - Preterm birth (PB) is any delivery from the 28th to the 36th week - Low birth weight (LBW) at term are born at 37 or more weeks of gestation and of less than 2.5 kg - Neural tube defect, renal, gastro-intestinal, heart, abdominal, cleft lip/palate, multiple, all not further specified. • Statistical analysis: <ul style="list-style-type: none"> - Most focus on exposure to war attacks and other determinants of the adverse birth outcomes other than (specifically) molybdenum and other metal concentrations in hair - Comparison of 95-percentile values of molybdenum concentration in 2015, 2016 and 2018-2019 with reference cohort outside war areas - Comparison of the median molybdenum concentration in 	<p>2018-2019, n=64: 0.08 (0.07–0.21) (Reference: 0.03–1.00)</p> <ul style="list-style-type: none"> • Median molybdenum levels (IQR) in ppm <ul style="list-style-type: none"> 2015, n=502: 0.06 (0.04–0.11) 2016, n=78: 0.06 (0.03–0.08) 2018-2019, n=64: 0.02 (0.0–0.04) Kruskal-Wallis test, $p < 0.001$ • No statistically significant ($p > 0.1$) differences in % of PB, LBW and birth defects between births in 2016 and 2018-2019 • Direct comparison of molybdenum concentration between subgroups of birth outcomes (BD, PB, LBW) was not reliable because of small numbers. 		<p>in parts unintelligible English and lack of a clear structure</p> <ul style="list-style-type: none"> • No direct comparisons of molybdenum levels in hair and birth outcomes and defects and hence not informative on relationship between molybdenum and adverse birth outcomes

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
	<p>- 24 with a preterm baby</p> <ul style="list-style-type: none"> • Exposure was further evaluated based on questionnaires assessing environmental exposure to metals, and documentation on war damages and weaponry used 	<p>the mothers delivering in 2015 (n=502), in 2016 (n=78) and in 2018–2019 (n=64) with non-parametric Kruskal-Wallis test</p> <p>- Comparison of molybdenum concentration between subgroups of birth outcomes (BD, PB, LBW) was not reliable because of small numbers.</p> <ul style="list-style-type: none"> • 			
<ul style="list-style-type: none"> • Gomez Roig et al. (2021) (62) • Cross-sectional study • Barcelona, Spain • Study period not reported • 167 mother-infant pairs • Pregnant women (3rd trimester) recruited from 2 maternal-foetal 	<ul style="list-style-type: none"> • After delivery, a full-thickness section (0.5–1 cm x 3–4 cm thick) of a peripheral site of the placenta was taken • Concentration (µg/L) of molybdenum and 21 other elements 	<ul style="list-style-type: none"> • Prenatal ultrasound examinations (with colour Doppler imaging) at beginning of third trimester • Estimated foetal weight centiles calculated using local reference curves: <ul style="list-style-type: none"> - SGA: small for gestational age foetuses <10th percentile (n=71) - AGA: normal growth foetuses >10th percentile (n=96) 	<ul style="list-style-type: none"> • Molybdenum detected in >80% of the placenta samples • Molybdenum concentration: mean 0.01 (SD 0.03); median 0.01 (P25–P75 0.01–0.02) mg/kg • No differences in molybdenum concentration detected between SGA and AGA foetuses and between normal and abnormal placental function parameters: <ul style="list-style-type: none"> • - AGA n=96, molybdenum mean 0.01 (SD 0.01) mg/kg 	<ul style="list-style-type: none"> • Analyses for molybdenum were not adjusted for potential confounders 	<ul style="list-style-type: none"> • Exposure was determined later (after delivery) than health outcome (at third trimester) Dichotomisation of foetal weight (and artery PI) is

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<p>and neonatal medicine clinics</p> <ul style="list-style-type: none"> • Inclusion criteria: <ul style="list-style-type: none"> - healthy pregnant women - aged ≥ 18 years - with singleton pregnancy - planning delivery at either of the 2 clinics • Exclusion criteria: <ul style="list-style-type: none"> - no gestational check-ups - no placental samples available 	<p>determined using ICP-OES (inductively coupled plasma optical emission spectrophotometry)</p> <ul style="list-style-type: none"> • Limit of detection 0.0013 mg/kg 	<ul style="list-style-type: none"> • Placental function was assessed by : <ul style="list-style-type: none"> - Uterine artery Pulsatility Index (PI) - Umbilical artery PI - Middle cerebral artery PI • Statistical analysis: <ul style="list-style-type: none"> - groups (SGA vs. AGA and dichotomised artery PI) compared using Student's t-test - multivariable logistic regression model with the most significant variables in the unadjusted analyses was estimated using a forward variable selection method based on Likelihood Ratio (considered for each of the elements) 	<ul style="list-style-type: none"> • - SGA n=71, molybdenum mean 0.01 (SD 0.04) mg/kg • Student's t-test, p for difference 0.89 • Placental molybdenum concentration and artery PI: <ul style="list-style-type: none"> • - Abnormal uterine artery Doppler (PI>P95) <ul style="list-style-type: none"> • Yes: n=34, mean molybdenum 0.01 (SD 0.01) mg/kg • No: n=120, mean molybdenum 0.01 (SD 0.04) mg/kg • - Abnormal umbilical artery Doppler (PI>P95) <ul style="list-style-type: none"> • Yes: n=8, mean molybdenum 0.01 (SD 0.01) mg/kg • No: n=148, mean molybdenum 0.01 (SD 0.03) mg/kg • - Abnormal middle cerebral artery Doppler (PI<P5) <ul style="list-style-type: none"> • Yes: n=8, mean molybdenum 0.01 (SD 0.01) mg/kg • No: n=148, mean molybdenum 0.01 (SD 0.03) mg/kg • p>0.05 • Molybdenum was not selected for multivariable logistic regression analyses 		<p>probably not optimal for analyses in terms of statistical power</p> <ul style="list-style-type: none"> • 71 out of 167 fetuses (43%!) were SGA (<P10) while foetal weight was not part of the selection criteria. This may be due to the source population from neonatal medicine clinics <p>In Methods is stated that molybdenum concentration</p>

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
					s were log-transformed but in Tables comparisons between groups are presented as mean with SD
<ul style="list-style-type: none"> • Tian et al. (2021)(63) • Cross-sectional study • Shanxi province, China • 2003-2016 • Cases: 273 women with NTD-affected pregnancies • Controls: 477 women with non-malformed fetuses or newborns who delivered at the same birthing hospital, matched on last menstrual period (± 4 weeks) 	<p>Fasting and non-fasting blood samples: in cases taken during pregnancy, at termination of pregnancy, or at delivery; in controls initially taken at delivery, but in later study years at similar gestational age (± 4 weeks) as cases.</p> <p>Blood serum concentrations of molybdenum</p>	<p>Diagnosis of NTD by county healthcare workers through physical examinations and prenatal ultrasound scans.</p> <p>NTD subtypes: 97 anencephaly, 127 spina bifida, 29 encephalocele, and 20 NTDs with other malformations</p> <p>Statistical analysis: - Difference in median molybdenum concentrations between cases and controls tested with Mann-Whitney U test. - Associations between molybdenum concentrations and NTDs examined using multilevel mixed-effects</p>	<p>Median (IQR) molybdenum concentrations (ng/mL): Controls 2.66 (2.03-3.27) All NTD cases 2.51 (1.43-3.07), P=0.002 Anencephaly 2.50 (1.31-3.02), P=0.03 Spina bifida 3.12 (1.31-4.47), P=0.02</p> <p>Multilevel mixed-effects logistic regression adjusted ORs (95% CI) for tertiles of molybdenum concentrations with T1 as reference: All NTD: T2: 0.48 (0.26-0.90) T3: 0.54 (0.29-1.00)</p> <p>Anencephaly: T2: 1.04 (0.41-2.65) T3: 0.45 (0.17-1.18)</p>		<p>Number of gestational weeks (at which most of the blood samples were collected) differed substantially between cases (62.3% <28 weeks) and controls (90.7% ≥ 37 weeks).</p> <p>Population partly overlaps with that of Yin, 2019.</p>

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
	<p>and 9 other metals analyzed by ICP-MS.</p> <p>Sociodemographic characteristics and lifestyle collected via face-to-face interviews with structured questionnaire.</p>	<p>logistic regression (to adjust for heterogeneity in molybdenum by region and calendar year)</p> <p>- Joint effects of metal co-exposure analysed by Bayesian kernel machine regression (BKMR).</p> <p>All multivariable models adjusted for maternal age, BMI, education, gestational weeks, sex of the fetus, periconceptional folic acid use, and maternal flu or fever.</p>	<p>Spina bifida:</p> <p>T2: 0.41 (0.20-0.86) T3: 0.52 (0.54-1.05)</p> <p>Multi-metal analysis (BKMR): ORs for IQR increase in molybdenum concentration 0.89 (0.83-0.96), 0.87 (0.80-0.94), and 0.85 (0.78-0.93) when remaining nine elements were set at 75th, 50th, and 25th percentiles, respectively.</p>		
<ul style="list-style-type: none"> • Yin et al., 2020 (64) • Cross-sectional study • Shanxi Province, China • January 2003 through December 2016 • Rural population • Cases: 408 newborns and terminated 	<p>Approximately 6 g of tissue from foetal portion of placenta sampled after delivery or pregnancy termination</p> <p>Placental concentrations of molybdenum and 5 other</p>	<p>Diagnosis of NTD by county healthcare workers through physical examinations and prenatal ultrasound scans.</p> <p>Face-to-face interviews by local health care workers based on structured questionnaire to assess information on potential confounders.</p> <p>Statistical analyses:</p>	<p>Placental molybdenum concentrations (ng/g-dry weight; median (IQR)) in cases and controls: 41.3 (32.8–51.2) and 32.8 (26.8–39.7), respectively;; P<0.001. Similar concentration patterns for anencephaly and spina bifida cases separately.</p> <p>Associations between above (≥ 35.7 ng/g) vs. below (< 35.7 ng/g) median molybdenum concentrations and risk of NTDs: crude OR 3.07 (2.36–3.99); adjusted OR 3.73 (2.74–5.07). Similar</p>	<p>Placental molybdenum concentrations may not reflect concentrations during the critical period of neural tube development, because tissue sampling occurred mostly in 2nd or 3rd trimester.</p>	

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<p>foetuses with neural tube defects (NTD)</p> <ul style="list-style-type: none"> • Controls: 593 foetuses or newborns without structural malformations found by foetal ultrasound scan or identified at birth or at pregnancy termination from the same hospital, matched to cases by mother's residence and date of last menstrual period, including controls selected for orofacial cleft cases (see Pi et al., 2019). • Exclusion criteria: cases and controls with insufficient placental tissue or incomplete key information. 	<p>metals assessed with ICP-MS. Analysis staff blinded to case or control status.</p>	<p>Differences in molybdenum concentrations between cases and controls tested with Mann-Whitney U test. Association above/below median or for quartiles of molybdenum concentrations and NTD risk estimated with multivariable logistic regression, adjusted for gestational age at delivery/pregnancy termination, maternal occupation, maternal education, parity, history of birth defects, fever or flu during early pregnancy, periconceptional folic acid supplementation, maternal passive smoking.</p> <p>Bayesian kernel machine regression (BKMR) model used to quantify and visualize effects of overall metal exposure and individual components (including molybdenum) within the context of overall joint</p>	<p>results for anencephaly and spina bifida cases separately.</p> <p>OR (95% CI) per quartile molybdenum concentrations (ng/g): <28.5: reference value 28.5–35.7: crude 1.27 (0.86–1.89); adjusted 1.38 (0.88–2.15) 35.7–44.1: 1.88 (1.28–2.76); adjusted 2.20 (1.42–3.40) ≥ 44.1: crude 6.58 (4.45–9.73); adjusted 9.84 (6.18–15.67). P for trend for crude and adjusted ORs <0.001</p> <p>Similar results for anencephaly and spina bifida cases separately.</p> <p>No effects of molybdenum in BKMR model in which all other metals were also included.</p> <p>Sensitivity analyses to investigate potential residual confounding because of correlation between gestational age and molybdenum concentrations, as cases were mostly electively terminated pregnancies at earlier gestational stage: multivariable analyses repeated in subset of gestational age-matched cases and</p>	<p>Potential overadjustment by gestational age, which should not be included as a confounder</p>	

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
		exposure, adjusted for the same potential confounders.	controls and after exclusion of the gestational age-matched subset resulted in similar results as overall analysis.		
<ul style="list-style-type: none"> • Yin et al., 2020 (65) • Cross-sectional study • Beijing, Shandong, and Shanxi), China • October 2010 - January 2019 • Cases: 130 women with fetuses or newborns with orofacial clefts (OFCs): <ul style="list-style-type: none"> - 76 cleft lip with cleft palate (CLP) - 44 cleft lip only (CLO) - 10 cleft palate only (CPO) 11 cases were complicated with other system malformations (syndromic). 	Venous maternal blood samples collected during pregnancy or after birth. Concentrations of molybdenum and 5 other essential trace elements in serum analysed by ICP-MS.	<p>OFCs confirmed at delivery or elective termination of pregnancy after prenatal diagnosis of malformation.</p> <p>Statistical analysis:</p> <ul style="list-style-type: none"> - Difference in median molybdenum concentrations between cases and controls tested with Mann-Whitney U test. - Associations between molybdenum concentrations and risk for OFCs examined using multilevel mixed-effects logistic regression (to adjust for heterogeneity in molybdenum by region). - Joint effects of metal co-exposure analysed by Bayesian kernel machine regression (BKMR). 	<p>Median (IQR) molybdenum concentrations (ng/mL) differed between cases and controls ($p < 0.01$):</p> <p>Controls: 2.816 (2.392–3.496) Total OFCs: 2.378 (1.757–2.938)</p> <p>CLP: 2.413 (1.835–2.970)</p> <p>CLO: 2.327 (1.727–2.930)</p> <p>Adjusted ORs (95% CI) for tertiles of molybdenum concentrations with T1 as reference:</p> <p>Total OFCs</p> <p>T2: 0.37 (0.20–0.66)</p> <p>T3: 0.28 (0.15–0.54)</p> <p>P for trend < 0.01</p> <p>CLP</p> <p>T2: 0.42 (0.20–0.89)</p> <p>T3: 0.35 (0.15–0.80)</p> <p>P for trend 0.009</p> <p>CLO</p> <p>T2: 0.32 (0.14–0.77)</p> <p>T3: 0.27 (0.11–0.67)</p> <p>P for trend 0.004</p>	No adjustments for maternal age, occupation, or history of birth defects because these variables did not differ statistically significantly between cases and controls, although the history of birth defects differed (6.2% in cases vs. 1.9% in controls).	<p>Number of gestational weeks at blood sample collection differed substantially between cases (31.5% <28 weeks) and controls (78.1% \geq37 weeks).</p> <p>Population partly overlaps with that of Pi, 2019.</p>

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
<ul style="list-style-type: none"> • Controls: 260 women with non-malformed fetuses or infants. matched on province or city and first day of last menstruation (± 4 months) 		<ul style="list-style-type: none"> - All multivariable models adjusted for gestational weeks, maternal education, flu or fever, periconceptional folic acid supplementation, parity, and passive smoking. 	<p>Similar associations after adjustment for sample collection period and exclusion of 11 cases of syndromic OFCs.</p> <p>Multi-metal analysis (BKMR): molybdenum serum concentrations inversely associated with risk of OFCs.</p>		
<ul style="list-style-type: none"> • Zhao et al., 2021 (66) • Cross-sectional study • Hangzhou, China • January – December 2016 • 220 pregnant women during second trimester • Inclusion criteria: <ul style="list-style-type: none"> - singleton foetus in 22-28 weeks of gestation at enrolment - willing to deliver in study hospital, participate in face-to-face interviews, complete ultrasound 	<ul style="list-style-type: none"> • Spot urine samples collected at 24.9 ± 0.8 weeks • Concentrations ($\mu\text{g/L}$) of molybdenum and copper (Cu) determined using ICP-MS • LOD $0.031 \mu\text{g/L}$ • Creatinine level was also determined 	<ul style="list-style-type: none"> • Prenatal ultrasound scanning in 22-26 weeks of gestation. Foetal biometric parameters: <ul style="list-style-type: none"> - abdominal circumference - biparietal diameter - femur length - head circumference - estimated foetal weight (calculated using Hadlock's formula) • Birth weight and infant sex retrieved from medical records • Structured questionnaires used to collect information on potential confounders • Statistical analysis: 	<ul style="list-style-type: none"> • All samples showed molybdenum concentration $> \text{LOD}$ • Median concentration (P25–P75) was: <ul style="list-style-type: none"> - $50.3 (30.6–76.0) \mu\text{g/L}$ (unadjusted) - $61.5 (47.0–87.8) \mu\text{g/g creatinine}$ (adjusted) • Associations between creatinine-adjusted molybdenum concentration and foetal growth parameters as beta (95% CI): representing the change in mean outcome variable per one increase in ln-transformed (creatinine-adjusted) molybdenum concentration <ul style="list-style-type: none"> - abdominal circumference (cm): $-0.34 (-0.63 \text{ to } -0.04)$; $p=0.025$ - biparietal diameter (cm): $+0.02 (-0.06 \text{ to } +0.09)$; $p=0.680$ 		<p>No other metals were taken into account.</p>

Study design and population	Exposure assessment	Health assessment	Results	Bias/confounding	Remarks
examinations, and provide urine sample at prenatal care visits in second trimester		<ul style="list-style-type: none"> - Urinary molybdenum concentration was corrected by creatinine level, and then ln-transformed as continuous variable - Multivariable linear regression models to estimate associations between molybdenum concentration and foetal biometric parameters and birth weight, adjusted for maternal age, education, household income, parity, pre-pregnancy BMI, second hand smoke in pregnancy, gestational age at ultrasound scanning, and foetal sex - Potential effect modification by urinary copper level explored by stratification on median Cu level and calculating the p for multiplicative interaction 	<ul style="list-style-type: none"> - femur diaphysis length (cm): 0.00 (-0.06 to 0.06); p=0.921 - head circumference (cm): -0.01 (-0.26 to 0.23); p=0.917 - estimated foetal weight (g): -18.2 (-40.5 to 4.2); p=0.110 • No association between molybdenum concentration and birth weight (g): +6.8 (-103.6 to 117.2); p=0.904 • In pregnant women with Cu levels below the median value, associations between molybdenum concentration and abdominal circumference (-0.55, -1.13 to 0.04) and estimated foetal weight (-42.3, -87.4 to 2.8) were stronger than in women with Cu levels from the median upwards (-0.08, -0.57 to 0.42 and -3.8, -40.2 to 32.6, respectively), with p-values for interaction of 0.340 and 0.222. 		

Prospective cohort studies

Shirai et al. (2010) evaluated the associations between maternal exposure to 10 metals including molybdenum and birth weight, birth length, and head circumference in 78 pregnant women visiting the obstetrics outpatient clinic of a hospital in Tokyo, Japan, in the period 2007-2008 (45). Recruitment and in-and exclusion criteria were only globally described. Metal concentrations were measured in single spot urine samples collected at 9-40 gestational weeks during regular maternal health check-ups. The mean (SD) molybdenum concentration was 79.0 (1.72) $\mu\text{g/g}$ creatinine. No correlations or associations were observed between urinary molybdenum concentrations and birthweight, birth length, and head circumference.

Vázquez-Salas et al. (2014) studied the associations between prenatal exposure to molybdenum and infant neurodevelopment during the first 30 months of life in a random subsample of 147 mother-child pairs who participated in a prospective cohort study in Morelos, Mexico (46). The concentration of molybdenum was determined in urine samples during each trimester of pregnancy with median values ranging from 48.9 to 59.1 $\mu\text{g/g}$ creatinine. The psychomotor and mental indexes of Bailey's scales were administered at 1, 3, 6, 12, 18, 24 and 30 months of age. In multivariable generalized mixed effect models, inverse associations were found between urinary molybdenum concentration in the third trimester and the psychomotor index. A doubling of molybdenum concentration was associated with a 0.57 lower index (95% CI 0.1 to 1.1; $p=0.03$), adjusted for prenatal dichlorodiphenyldichloroethylene (DDE) exposure at first trimester of pregnancy, gestational age, parity, maternal age, education, and IQ, birthweight, type of birth, sex of child, breastfeeding, and quality of home environment. No significant associations were observed for molybdenum concentrations in the first and second trimester or with the mental index of the Bailey's scale.

Bloom et al. (2015) assessed the impact of preconceptional parental urinary molybdenum concentrations and perinatal outcomes in 235 couples with singleton pregnancies from the LIFE prospective cohort in Texas and Michigan, USA (47, 48). Median molybdenum concentrations were 31.43 $\mu\text{g/L}$ and 48.9 $\mu\text{g/L}$ in maternal and paternal urine, respectively. Multivariable linear and log-binomial regression models included tertiles of molybdenum concentration of both mothers and fathers, and were adjusted for maternal age, the difference between maternal and paternal ages, maternal and paternal smoking, income, race, total serum lipids (as a proxy for persistent organic pollutants) and creatinine. No statistically significant ($p>0.05$) associations were found between maternal or paternal molybdenum concentration in urine and gestational age, birth weight, birth length, head circumference, ponderal index, and infant sex.

In a prospective cohort study, **Howe et al. (2020)** investigated whether prenatal exposure to mixtures of heavy metals including molybdenum were associated with lower birth weight for gestational age, in a predominantly lower-income Hispanic pregnancy cohort in Los Angeles (US) (49). Ten metals were measured in spot urine samples of 262 pregnant women participating in the MADRES cohort. Urine samples

were collected in the first half of pregnancy (median gestational age 13.1 weeks). The median (IQR) urinary molybdenum concentration was 56.8 (42.9-80.7) $\mu\text{g/L}$. All analyses used a combination of metals, in which the contribution of molybdenum was limited.

In a study on a sub-cohort selected from a larger birth cohort study in the USA, **Kim et al. (2020)** investigated whether exposure to metals, including molybdenum, negatively impacts intra-uterine growth (50). The study included 130 (= almost all) women who experienced a preterm delivery and 352 women with at term births, originally selected for a nested-control study. Molybdenum and 16 other metals were measured in urine samples, which were collected at a median of 26 weeks of pregnancy. Median (IQR) molybdenum concentration, specific-gravity-corrected and weighted for case-control design, was 51.3 (37.1-69.7) ppb. Metal concentrations were associated with various parameters of foetal growth assessed by ultrasound at 26 weeks (median) and 35 weeks (median) of pregnancy using linear mixed effects models, adjusted for multiple confounders. Linear regression analyses were used for associations with birthweight, birth length, and placental weight. In single metal analysis, no associations were observed between molybdenum concentration and any of the outcomes. In models with additional adjustment for other metals, molybdenum concentrations were associated with differences in femur length z-scores: β (95% CI) = 0.30 (0.08, 0.52) and head circumference z-scores: 0.16 (-0.04, 0.36).

In the MADRES prospective cohort study, **Howe et al. (2021)** evaluated the associations between the same exposure and foetal size at mid-pregnancy in a subset of 195 participants who enrolled prior to the routine ultrasound scan (51). The methods of measurement and analyses were the same as above, with a median (IQR) urinary molybdenum concentration of 57.4 (44.3-81.1) $\mu\text{g/L}$. Of the six and 10 metals evaluated simultaneously in the primary and secondary analysis, respectively, molybdenum ranked highest as predictor of estimated foetal weight (EFW) and other foetal growth parameters evaluated (abdominal circumference, biparietal diameter, femur length, head circumference). Setting other metals to their median, an increase in molybdenum from the 25th to 75th percentile was associated with a 0.114 (95% CI: 0.019, 0.247) SD higher EFW, equivalent to a ~ 7.4 g higher EFW, a 0.30 (95% CI: 0.05, 0.56) SD difference in head circumference.

Karakis et al. (2021) performed an exploratory analysis using a cohort of 111 pregnant women of Arab-Bedouin origin and their offspring in Negev desert, Israel (52). Associations between molybdenum (among other metals) in urine sampled just prior to delivery and adverse perinatal outcomes and other health problems of the offspring up to six years after birth were studied. Molybdenum concentration (in quintiles) was associated with behavioural/developmental disorders as reported in medical records: RR=1.86 ($p=0.016$) associated with a one-step higher urine molybdenum quintile. Molybdenum concentration was not significantly ($p>0.05$) associated with preterm delivery, congenital malformations, or other disorders.

Mandiá et al. (2021) studied concentrations of molybdenum (among other elements) in human milk from nursing mothers from Santiago de Compostela, Spain and their associations with birth weight (37). Human milk samples were obtained at three time points from 70 mothers of full-term newborns: colostrum during the first 3–4 days of lactation, intermediate milk up to 7–10 days of lactation and later mature milk up to 6 months after birth. In addition, samples of later mature milk were also obtained from 100 mothers of premature newborns. A questionnaire included medical factors of the mother during pregnancy and birth weight of the newborn. Concentrations of molybdenum in milk from full-term mothers decreased as lactation progressed. Mean molybdenum concentrations in later mature milk were 0.96 and 0.70 µg/L from mothers of full-term and preterm newborns, respectively. Molybdenum was inversely associated with birth weight ($p=0.002$).

In an Australian prospective cohort study among pregnant women, **McKeating et al. (2020)** applied elemental metabolomics in plasma and urine to identify associations between elemental concentrations and pregnancy risk factors (53). Plasma and urine samples were obtained from a cohort of 18-week pregnant women ($N=128$, age >18 years) from a hospital in Adelaide, Australia. The pregnancy outcomes were divided into 4 groups: 13 pre-term births, 18 low placental weight, 10 small for gestational age (SGA) and 87 healthy infants (controls). ICP-MS was used to measure 27 plasma elements and 37 urinary elements, including molybdenum. Analysis was done by one-way ANOVA of individual elements, with Dunn's post hoc analysis for pairwise comparison. Mean concentration of plasma molybdenum in the group with low placental weight (0.6 µg/L) was significantly ($p<0.0001$) lower than the mean concentration in controls (0.89 µg/L). No differences were found in plasma and urine concentrations of molybdenum between the more conventional developmental outcomes preterm birth or SGA and control infants.

Tung et al. (2022) studied the associations between single and multiple placental metal concentrations (including molybdenum) and atypical neurobehaviour in newborns in a prospective cohort study including 192 mother-infant pairs (54). Due to the main aim of the cohort, over half of the infants were born small or large for gestational age. Placental samples were collected within 2 hours after delivery. Neurobehavioural performance was assessed with the NICU Network Neurobehavioral Scale (NNS), administered by certified psychometrists 24 – 72 hours after birth, and divided into 5 profiles. Profile 5 indicated the most atypical neurobehavioural performance. Multivariate logistic regression analysis with adjustment for infant sex, maternal age, maternal race, pre-pregnancy BMI, education, and smoking status during pregnancy resulted in an OR of approximately 2 (with a wide 95% CI including unity) for NNS profile 5 with each doubling of placental molybdenum concentration.

Case-control studies

Yan et al. (2017) conducted a case-control study to investigate if 'essential trace metals', including molybdenum, protect against the risk of a neural tube defect (55). The researchers identified 191 women with a pregnancy complicated by a neural tube defect in Shanxi and Hebei

Provinces, China in 2003-2007. These cases were matched with 261 women from the same birthing hospital who delivered healthy infants on residence (same county/city) and time since last menstrual period. Molybdenum and eight other metals were measured in hair segments grown just before and during early pregnancy. Associations, adjusted for multiple confounders, were found for below vs. above median molybdenum concentrations with anencephaly (OR 0.51, 0.28-0.94), spina bifida (0.54, 0.31-0.94), and all NTDs as a group (0.64, 0.42-0.98), with higher levels of molybdenum corresponding to lower risks. Dose-response analysis by quartiles of molybdenum concentrations showed decreasing trends in the risks with increasing molybdenum concentrations.

Associations between concentrations of 16 trace metals, including molybdenum, and infants small for gestational age (SGA) were investigated by **Deysenroth et al. (2018)** (56). The metals were measured in maternal toe nail clippings collected on average 2.8 months post-partum from participants in the Rhode Island Child Health Study (n=195). Logistic regression and weighted quantile sum regression were performed for molybdenum and panels of the 16 trace metals, respectively, adjusted for multiple confounders and corroborated by Bayesian kernel machine regression. No association was observed between molybdenum and SGA in the single metal model, and molybdenum did not contribute to the association between SGA and a metal mixture including As.

The associations between prenatal exposure to 22 metals including molybdenum and low birth weight (LBW) were investigated by **Hou et al. (2019)** in a nested case-control study in China (57) This study included 246 women with LBW children and 406 women with normal birth weight children from a prospective birth cohort study. Maternal serum samples collected during prenatal examination were analysed by ICP-MS. The median molybdenum serum concentrations were 1.18 µg/L in the cases and 1.07 µg/L in the control group. In single metal analysis, adjusted for multiple confounders, lower quartiles of serum molybdenum concentration were associated with a decreased risk of LBW compared to the highest quartile (P for trend = 0.018, OR 0.48 (0.30-0.77) for second quartile), but no dose-response relationship could be established in cubic splines. In multi-metal analysis, 15 metals including molybdenum were associated with LBW.

Cross-sectional studies

The cross-sectional study by **Ovayolu et al. (2020)** in Turkey was performed to determine the levels of 14 trace elements and heavy metals, including molybdenum, in amniotic fluid of pregnant women collected around 20 weeks of gestation, and the associations with neural tube defects (NTDs) (58). The study included 36 pregnant women whose foetuses were complicated with NTDs (case group) and 39 pregnant women with unaffected foetuses (control group), matched for body mass index and gestational weeks. The amniotic fluid levels of Mo were measured using ICP-MS and compared between the two groups. Lower mean concentrations of molybdenum were observed in the NTD group than in the control group: 1.11 µg/L (± 1.06) vs. 2.47 µg/L (±

1.92), respectively ($p < 0.001$). These results were not adjusted for potential confounders or co-exposure to other metals.

Pi et al. (2019) examined the associations between concentrations of 6 metals including molybdenum in placental tissues and orofacial clefts (OFCs) in offspring in a rural population in northern China with a high prevalence of OFCs (59). Cases and controls were recruited via a population-based birth defects surveillance program. The study included 103 newborns and terminated fetuses with OFCs (cases) and 206 newborns without congenital malformations (controls), matched on mother's residence, date of last menstrual period, and newborn sex. Placental tissue was sampled after delivery or pregnancy termination, mostly in the 2nd or 3rd trimester. Median (IQR) placental molybdenum concentrations were higher in cases than controls: 35.9 (31.7–41.8) vs. 32.1 (27.3–37.0) ng/g dry weight, respectively ($p < 0.001$). The unadjusted association between molybdenum concentrations above (≥ 33.6 ng/g) vs. below (< 33.6 ng/g) median concentrations and risk of OFCs (OR 2.20, 1.36–3.58) all but disappeared (OR 1.42, 0.78–2.59) by adjustment for multiple confounders, including gestational age, which may have resulted in overadjustment. The same may have happened to the increasing trend of OFC risk initially seen for tertiles of molybdenum concentration.

In a cross-sectional study from Italy, metal concentrations in maternal serum collected during the 2nd trimester of pregnancy were compared between cases and controls by **Troisi et al. (2019)** (60). Cases were 111 pregnant women carrying a foetus diagnosed with a malformations or chromosomal abnormality. Controls were 90 women with normally developed foetuses, recruited during second trimester routine anomaly scan. No differences in serum molybdenum concentrations were found between cases and controls, neither in a CNS group ($n=17$) including all CNS malformations with unknown aetiology nor in the 'other malformations' group ($n=94$) with unspecified malformations or chromosomal abnormalities. The analyses were not adjusted for potential confounders.

In a cross-sectional study repeated at three points in time, **Manduca et al. (2020)** evaluated whether potential exposure to molybdenum and other heavy metals from weaponry deposited in the environment or in wound tissue during military attacks, increased the risk of adverse birth outcomes and birth defects in Gaza, Palestine (61). Median levels of molybdenum in mothers' hair ($n=644$) was lower in 2018–2019 (0.02 ppm) as compared to both 2015 (0.06 ppm) and 2016 (0.06 ppm). No statistically significant ($p > 0.1$) associations between study period (2015, 2016 and 2018–2019) and incidence of preterm birth, low birth weight and birth defects were found. Analyses focused on exposure to war attacks and other questionnaire-assessed determinants of adverse birth outcomes other than metal concentrations in hair including molybdenum. Unfortunately, direct comparison of molybdenum concentration between adverse birth outcomes (preterm birth, low birth weight and birth defects) was not reliable because of small numbers. This analysis is therefore not informative for evidence of any relationship between molybdenum and the adverse birth outcomes under study.

Gomez Roig et al. (2021) studied associations between placental concentration of molybdenum (among other elements), foetal growth, and markers of placental function among 167 mother-infant pairs from Barcelona, Spain (62). Prenatal Doppler ultrasound examinations were done at the beginning of the third trimester of pregnancy. On the basis of estimated foetal weight, the group was divided into 71 small for gestational age fetuses (<10th percentile) and 96 normal growth fetuses (>10th percentile). Placental function was assessed by measuring the Pulsatility Index of the uterine artery, umbilical artery, and middle cerebral artery. After delivery, molybdenum was determined in a placental sample using ICP-OES. No statistically significant ($p>0.05$) differences in placental molybdenum concentrations were observed between small and normal fetuses or between normal or abnormal placental function parameters (>95th percentile vs. <5th percentile of the artery pulsatility index).

In a cross-sectional study in the Shanxi province of northern China, **Tian et al. (2021)** studied associations between serum metal concentrations, including molybdenum and neural tube defects (NTD) (63). Participants included 273 women with NTD-affected pregnancies (97 anencephaly, 127 spina bifida, 29 encephalocele, 20 NTD with other malformations) and 477 women with non-malformed fetuses or newborns who delivered at the same birthing hospital (controls), matched on last menstrual period (± 4 weeks). Fasting and non-fasting blood samples were collected during pregnancy, at termination of pregnancy, or at delivery (cases 15% ≥ 37 weeks; controls 91% ≥ 37 weeks). (67). Median (IQR) molybdenum concentrations were lower in cases than controls: 2.51 (1.43-3.07) vs. 2.66 (2.03-3.27) ng/ml ($p=0.001$). Statistical analysis using multilevel mixed-effects logistic regression, adjusted for multiple confounders, revealed that serum molybdenum concentrations were associated with a lower risk of NTDs with ORs (95% CI) for all NTD subtypes 0.48 (0.26-0.90) and 0.54 (0.29-1.00) for the 2nd and 3rd tertiles of molybdenum concentration, respectively, compared to the 1st tertile. Similar results but with wider confidence intervals were seen for anencephaly and spina bifida separately. In multi-metal analysis, the adjusted ORs for molybdenum were still inversely associated with risk of NTDs (0.87 (0.80-0.94) for IQR increase in molybdenum concentration with other metals set at 50th percentile).

Yin et al. (2020) studied the associations between placental metal concentrations and neural tube defects (NTDs) in the same rural source population (64). This study included 408 NTD cases with placental tissue available and 593 fetuses or newborns without structural malformations as controls from the same hospital and matched on mothers' province of residence and date of last menstrual period, including controls selected for OFC cases. Placental median (IQR) molybdenum concentrations were higher in cases than controls: 41.3 (32.8-51.2) vs. 32.8 (26.8-39.7) ng/g-dry weight, respectively ($p<0.001$). Molybdenum concentrations above the median of 35.7 ng/g were associated with a higher risk of NTDs: adjusted OR 3.73 (2.74-5.07), with similar results for anencephaly and spina bifida separately. Dose-response relationships were seen with increasing ORs for quartiles

of molybdenum concentration. molybdenum did not have any effect in the BKMR analysis in which all other metals were also included.

Yin et al. (2020) studied associations between concentrations of 6 trace elements, including molybdenum, in serum samples collected during pregnancy or after birth and orofacial clefts (OFCs) in China (65). The study included 130 women with fetuses or newborns with OFCs (cases) and 260 women with non-malformed fetuses or infants (controls), matched on province or city and first day of last menstrual period (± 4 weeks). The gestational weeks of blood sampling differed substantially between cases (57% ≥ 37 weeks) and controls (78% ≥ 37 weeks). Median (IQR) molybdenum concentrations were lower in cases than controls: 2.38 (1.76-2.94) vs. 2.82 (2.39–3.50) ng/ml ($p < 0.01$). The adjusted odds ratios for OFCs as a group, as well as for cleft lip with cleft palate and cleft lip only separately, decreased with increasing tertiles of molybdenum concentration (ORs ranging from 0.42 to 0.27, all 95% CI excluding unity; p for trend < 0.01). Molybdenum concentrations were also inversely associated with the risk of OFCs in multi-metal analyses including adjustment for multiple confounders.

In a cross-sectional study, **Zhao et al. (2021)** investigated associations between spot urinary molybdenum concentrations and ultrasound parameters of foetal growth at 22-28 weeks of gestation and offspring birth weight in 220 women in Hangzhou, China (66). Multivariable linear regression analyses were applied with adjustment for multiple potential confounders. Creatinine-adjusted molybdenum concentration was significantly associated with a 0.34 cm (95% CI 0.04 to 0.63) reduction in foetal abdominal circumference per unit increment in log-transformed urinary molybdenum, and potentially with a 18.2 g (95% CI -4.2 to 40.5) lower estimated foetal weight. No associations were observed between molybdenum concentration and biparietal diameter ($p = 0.68$), femur length ($p = 0.92$), head circumference ($p = 0.92$) and birth weight ($p = 0.90$).

8.2.3 *Other relevant information*

In addition to the *in vivo* animal studies and studies in humans, one *in vitro* study with zygotes from mice is available. Below, this additional study is described, and summarized in Table 38.

Table 38 Summary table of other studies relevant for development.

Reference	Species	Experimental period and design	Dose	Observations	Remarks
Bi et al., 2013 (68)	Mouse, Kunming and ICR strains	Zygotes were flushed from one outbred mouse strain (Kunming) and cultured in potassium simplex optimized medium (KSOM) containing 0, 5, 10, 20, 40, 80, 120, and 160 µg/ml of molybdenum for 5 days until the mid-blastocyst stage. In addition, normal blastocysts in morphology from various groups were used in embryo transfer experiments. Groups of 10 blastocysts were surgically transferred to each uterine horn on day 2.5 in pseudopregnant ICR recipient female mice.	0, 5, 10, 20, 40, 80, 120, and 160 µg/ml. Substance: Sodium molybdate dihydrate Purity: no data	<ul style="list-style-type: none"> • Significant decrease in the cleavage, blastocyst and birth rates, the average cell number, and increase in the proportion of degenerative blastocysts at 40 µg/ml and higher. • Inhibition of blastocysts development to birth at 120 µg/ml. • Overall developmental arrest (up to 16-cells) of embryos and their massive degeneration at 160 µg/ml. 	

In an in vitro study the effect of molybdenum on the development of mouse preimplantation embryos cultured in vitro was investigated. Zygotes were flushed from one outbred mouse strain (Kunming), and then were cultured in potassium simplex optimized medium (KSOM) containing 0, 5, 10, 20, 40, 80, 120, and 160 µg/ml of sodium molybdate hydrate for 5 days until the mid-blastocyst stage, at which time the quality of the embryos was evaluated.

The data show that the addition of ≤ 20 µg/ml molybdenum did not affect the blastocyst and birth rates. At 40 µg/ml and higher, the cleavage, blastocyst, birth rates and the average cell number were decreased and the proportion of degenerative blastocysts was increased. At 120 µg/ml, molybdenum inhibited the blastocysts development to birth. At 160 µg/ml, molybdenum caused overall developmental arrest (up to 16-cells) of embryos and their massive degeneration (Table 39) (68).

Table 39 Effects of sodium molybdate on the developmental capacity of mouse embryos cultured in vitro (68) .

Mo (µg/ml)	No. zygote	Cleavage rate, % (No. cleaved)	Arrest rate, % (No. 2-cells to morulas)	Blastocyst rate, % (No. blastocysts)		No. cells per blastocyst (n=10)	Birth rate, % (No. offsprings) (n=40)
				Normal	Degenerate		
Control	317	96.23 ± 1.33 (305)	4.23 ± 2.71 (13)	94.71 ± 2.37 (289)	0.98 ± 0.90 (3)	79.9 ± 4.04	57.50 ± 10.61 (23)
5	388	97.96 ± 1.07 (380)	2.85 ± 3.03 (11)	96.47 ± 2.68 (366)	0.78 ± 1.01 (3)	82.3 ± 4.59	62.50 ± 3.53 (25)
10	342	96.21 ± 3.47 (329)	4.27 ± 3.58 (14)	94.24 ± 3.07 (310)	1.53 ± 2.04 (5)	72.1 ± 4.64	52.50 ± 3.54 (21)
20	325	95.16 ± 2.20 (309)	6.38 ± 4.77 (20)	91.10 ± 3.77 (279)	3.23 ± 2.49 (10)	67.9 ± 5.22 ^a	47.50 ± 3.54 (19)
40	356	94.97 ± 1.80 (338)	49.92 ± 4.52 (168) ^a	44.05 ± 2.31 (147) ^a	6.59 ± 4.64 (23) ^a	63.1 ± 5.24 ^a	25.00 ± 7.07 (10) ^b
80	303	79.92 ± 5.64 (243) ^a	57.56 ± 6.21 (139) ^a	31.74 ± 4.13 (77) ^a	10.66 ± 4.34 (27) ^a	57.5 ± 4.03 ^a	27.50 ± 3.54 (11) ^b
120	418	67.60 ± 6.69 (283) ^a	65.62 ± 7.30 (184) ^a	19.51 ± 1.47 (56) ^a	14.64 ± 6.59 (43) ^a	44.7 ± 5.81 ^a	0(0) ^a
160	409	54.05 ± 7.22 (221) ^a	100.0 ± 0.00 (221) ^a	0 ^a	0 ^a	-	-

Mo: molybdenum. Data represent means ± SD; compared with the control group, ^a P < 0.01; ^b P < 0.05.

8.3 Adverse effects on or via lactation

In a multigenerational study, performed according to OECD TG 416, groups of 24 male and 24 female Sprague-Dawley rats were administered sodium molybdate dihydrate at 0, 5, 17, or 40 mg molybdenum (Mo)/kg bw/day in the drinking water or 40 mg Mo/kg bw/day in the diet over two generations to assess reproductive toxicity (18). This study is described in section 8.1.1 and 8.2.1. No adverse effects on the pups during lactation were observed. Viability index, lactation index, % male pups per litter, live litter size and pup weight were not affected by sodium molybdate. Overall, no adverse effects on reproductive function or development was observed at any dose level in either generation.

Molybdenum is present in human breast milk, with mean concentrations ranging between 0.02 to 24 µg/L (3). Molybdenum concentrations in human milk appear to be highest during the first few days of breastfeeding, and decrease during the course of lactation (69). Wappelhorst et al. (2002) compared the element intake of nursing mothers and element content of human milk (70). The mean intake of molybdenum was 132 ± 60 µg/day (2.07 µg/kg/day) and the mean milk concentration of the same group of women was 0.72 µg/L (0.27-1.62).

In a human study, breast milk concentrations of 32 metals and elements were determined in early lactation (days 14-21) in a random sample of first time Swedish mothers (n = 60), collected in 2002-2009. The elements were measured using ICP-MS. The mean molybdenum concentrations in breast milk was 3.5 µg/L (± 2.7); the median concentration was 2.4 µg/L (range: 0.8 – 12 µg/L). The results were compared to concentrations found in Swedish women in a WHO study from 1989. The median molybdenum concentration in milk in the WHO study was 0.40 µg/L. In this WHO study, samples were taken at about three months after the birth of the baby, instead of 2-3 weeks after birth, which could explain the difference (71).

In a Japanese study, breast milk trace element concentrations were determined at 1 and 3 months postpartum using ICP-MS. Samples were collected from 79 Japanese healthy mothers who gave birth to a single infant after 37 weeks of gestation. The mean molybdenum concentration in breast milk was 1.0 and 1.1 µg/dL at 1 and 3 months postpartum respectively. The median concentration was 0.7 µg/dL for both 1 and 3 months postpartum (72).

Trace elements, including molybdenum, were determined in human breast milk from Jordanian mothers (n=76) and measured by ICP-MS. The mean molybdenum concentration was 32.6 µg/L (± 28) and the median concentration was 25.0 µg/L, with an overall range of 2.70-236 µg/L (73).

Concentrations of 35 minerals, essential and toxic trace elements were quantified in human milk samples obtained from full-term and preterm mothers in Santiago de Compostela, Spain (n=170). Samples were obtained from full-term (colostrum, intermediate and mature human milk) and preterm (mature human milk) mothers. ICP-MS analyses

showed mean concentrations of 1.88 ± 1.2 , 1.22 ± 1.98 and 0.96 ± 1.16 $\mu\text{g/L}$ in full-term (colostrum, intermediate and mature) milk. The concentration in preterm (mature) milk was 0.70 ± 1.17 $\mu\text{g/L}$ (74).

Intra- and inter-day variation of elements in breast milk in 11 women at 12 sampling points over three days by ICP-MS. The median concentration of molybdenum was 1.5 ng/g, with a minimum of 0.18 and maximum of 12 ng/g (75).

Human milk samples were collected pre- and post- every feed in a 24-hour period from 11 mothers and analysed for elements by ICP-MS. Pre-feed concentrations of molybdenum in milk were 0.48 ± 0.28 $\mu\text{g/L}$; post-feed concentrations were 1.20 ± 0.28 $\mu\text{g/L}$ (76).

9 References

1. Health Council of the Netherlands. Molybdenum and molybdenum compounds. Health-based recommendation occupational exposure limit. . The Hague: Health Council of the Netherlands. Dutch Expert Committee on Occupational Standards; 2013.
2. REGULATION (EC) No 1272/2008 OF THE EUROPEAN PARLIAMENT AND OF THE COUNCIL of 16 December 2008 on classification, labelling and packaging of substances and mixtures, amending and repealing Directives 67/548/EEC and 1999/45/EC, and amending Regulation (EC) No 1907/2006., (2008).
3. ATSDR. Toxicological profile for molybdenum. Agency for Toxic Substances and Disease Registry; 2020.
4. CRC. Handbook of Chemistry and Physics. 96th edition ed.
5. ECHA. ECHA's dissemination website [Available from: <https://echa.europa.eu/nl/information-on-chemicals/registered-substances>].
6. NIOSH. Method No. 7302: Elements by ICP (Microwave digestion). NIOSH Manual of Analytical Methods (NMAM) National Institute for Occupational Safety and Health; 2014.
7. NIOSH. Method No. 7304: Elements by ICP Microwave digestion. NIOSH Manual of Analytical Methods (NMAM) National Institute for Occupational Safety and Health; 2014.
8. NIOSH. Method No. 7306: Elements by Cellulosic Internal Capsule Sampler. NIOSH Manual of Analytical Methods (NMAM) National Institute for Occupational Safety and Health; 2015.
9. Kim SS, Meeker JD, Carroll R, Zhao S, Mourgas MJ, Richards MJ, et al. Urinary trace metals individually and in mixtures in association with preterm birth. *Environ Int.* 2018;121(Pt 1):582-90.
10. NIOSH. Method No. 8200: Elements in Tissues. NIOSH Manual of Analytical Methods (NMAM) National Institute for Occupational Safety and Health; 2018.
11. DFG. List of MAK and BAT values 2019. Deutsche Forschungsgemeinschaft; 2019, Report 55.
12. Hays SM, Macey K, Poddalgoda D, Lu M, Nong A, Aylward LL. Biomonitoring Equivalents for molybdenum. *Regul Toxicol Pharmacol.* 2016;77:223-9.
13. HSE. EH40/2005 Workplace exposure limits. Health and Safety Executive; 2020, 4th edition.
14. Su CK, Yang CH, Lin CH, Sun YC. In-vivo evaluation of the permeability of the blood-brain barrier to arsenicals, molybdate, and methylmercury by use of online microdialysis-packed minicolumn-inductively coupled plasma mass spectrometry. *Anal Bioanal Chem.* 2014;406(1):239-47.
15. Murray FJ, Sullivan FM, Tiwary AK, Carey S. 90-Day subchronic toxicity study of sodium molybdate dihydrate in rats. *Regul Toxicol Pharmacol.* 2014;70(3):579-88.
16. Tokalioglu S, Clough R, Foulkes M, Worsfold P. Bioaccessibility of Cr, Cu, Fe, Mg, Mn, Mo, Se and Zn from nutritional supplements by the unified BARGE method. *Food Chem.* 2014;150:321-7.

17. ECHA. Registration dossier Disodium molybdate: European Chemicals Agency; 2021 [updated 19 July 2021. Available from: <https://echa.europa.eu/nl/registration-dossier/-/registered-dossier/15826>.
18. Murray FJ, Sullivan FM, Hubbard SA, Hoberman AM, Carey S. A two-generation reproductive toxicity study of sodium molybdate dihydrate administered in drinking water or diet to Sprague-Dawley rats. *Reprod Toxicol*. 2019;84:75-92.
19. Khorami H, Eidi A, Mortazavi P, Modaresi M. Effect of sodium molybdate on cadmium-related testicular damage in adult male Wistar rats. *Journal of Trace Elements in Medicine and Biology*. 2020;62.
20. Zhai XW, Zhang YL, Qi Q, Bai Y, Chen XL, Jin LJ, et al. Effects of molybdenum on sperm quality and testis oxidative stress. *Syst Biol Reprod Med*. 2013;59(5):251-5.
21. Zhang YL, Liu FJ, Chen XL, Zhang ZQ, Shu RZ, Yu XL, et al. Dual effects of molybdenum on mouse oocyte quality and ovarian oxidative stress. *Syst Biol Reprod Med*. 2013;59(6):312-8.
22. Wang HW, Zhou BH, Zhang S, Guo HW, Zhang JL, Zhao J, Tian EJ. Reproductive toxicity in male mice after exposure to high molybdenum and low copper concentrations. *Toxicol Ind Health*. 2016;32(9):1598-606.
23. Fungwe TV, Buddingh F, Demick DS, Lox CD, Yang MT, Yang SP. The role of dietary molybdenum on estrous activity, fertility, reproduction and molybdenum and copper enzyme activities of female rats. *Nutrition Research*. 1990;10(5):515-24.
24. Meeker JD, Rossano MG, Protas B, Padmanahban V, Diamond MP, Puscheck E, et al. Environmental exposure to metals and male reproductive hormones: circulating testosterone is inversely associated with blood molybdenum. *Fertil Steril*. 2010;93(1):130-40.
25. Syrkasheva AG, Frankevich VE, Dolgushina NV. Elemental composition of blood of infertile patients participating in assisted reproduction programs. *Bulletin of Russian State Medical University*. 2021(1):45-50.
26. Ashrap P, Meeker JD, Sánchez BN, Basu N, Tamayo-Ortiz M, Solano-González M, et al. In utero and peripubertal metals exposure in relation to reproductive hormones and sexual maturation and progression among boys in Mexico City. *Environmental Health: A Global Access Science Source*. 2020;19(1).
27. Guzikowski W, Szykowska MI, Motak-Pochrzęst H, Pawlaczyk A, Sypniewski S. Trace elements in seminal plasma of men from infertile couples. *Arch Med Sci*. 2015;11(3):591-8.
28. Lewis RC, Meeker JD. Biomarkers of exposure to molybdenum and other metals in relation to testosterone among men from the United States National Health and Nutrition Examination Survey 2011-2012. *Fertil Steril*. 2015;103(1):172-8.
29. Skalnaya MG, Serebryansky EP, Yurasov VV, Tinkov AA, Demidov VA, Skalny AV. Association between semen quality and level of 20 essential and toxic metals in ejaculate. *Trace Elements and Electrolytes*. 2015;32(3):126-32.

30. Zeng Q, Feng W, Zhou B, Wang YX, He XS, Yang P, et al. Urinary metal concentrations in relation to semen quality: a cross-sectional study in China. *Environ Sci Technol*. 2015;49(8):5052-9.
31. Wang YX, Sun Y, Huang Z, Wang P, Feng W, Li J, et al. Associations of urinary metal levels with serum hormones, spermatozoa apoptosis and sperm DNA damage in a Chinese population. *Environ Int*. 2016;94:177-88.
32. Zhou Y, Fu XM, He DL, Zou XM, Wu CQ, Guo WZ, Feng W. Evaluation of urinary metal concentrations and sperm DNA damage in infertile men from an infertility clinic. *Environmental Toxicology and Pharmacology*. 2016;45:68-73.
33. Branch FM, Perry MJ, Chen Z, Louis GMB. Metal(loid)s and human semen quality: The LIFE Study. *Reproductive Toxicology*. 2021;106:94-102.
34. Buck Louis GM, Schisterman EF, Sweeney AM, Wilcosky TC, Gore-Langton RE, Lynch CD, et al. Designing prospective cohort studies for assessing reproductive and developmental toxicity during sensitive windows of human reproduction and development--the LIFE Study. *Paediatr Perinat Epidemiol*. 2011;25(5):413-24.
35. Bommarito PA, Kim SS, Meeker JD, Fry RC, Cantonwine DE, McElrath TF, Ferguson KK. Urinary trace metals, maternal circulating angiogenic biomarkers, and preeclampsia: a single-contaminant and mixture-based approach. *Environ Health*. 2019;18(1):63.
36. Wang P, Ma W, Zhou Y, Zhao Y, Shi H, Yang Q, Zhang Y. Circulating metal concentrations, inflammatory cytokines and gestational weight gain: Shanghai MCPC cohort. *Ecotoxicology and Environmental Safety*. 2020;199.
37. Mandiá N, Bermejo-Barrera P, Herbello P, López-Suárez O, Fraga JM, Fernández-Pérez C, Couce ML. Human milk concentrations of minerals, essential and toxic trace elements and association with selective medical, social, demographic and environmental factors. *Nutrients*. 2021;13(6).
38. Elongi Moyene JP, Scheers H, Tandu-Umba B, Haufroid V, Buassa-Bu-Tsumbu B, Verdonck F, et al. Preeclampsia and toxic metals: A case-control study in Kinshasa, DR Congo. *Environmental Health: A Global Access Science Source*. 2016;15(1).
39. Li D, Jiang T, Wang X, Yin T, Shen L, Zhang Z, et al. Serum Essential Trace Element Status in Women and the Risk of Endometrial Diseases: a Case-Control Study : Serum Essential Trace Element Status in Women and the Risk of Endometrial Diseases: a Case-Control Study. *Biological trace element research*. 2022.
40. Kolesarova A, Capcarova M, Sirotkin AV, Medvedova M, Kalafova A, Filipejova T, Kovacic J. In vitro assessment of molybdenum-induced secretory activity, proliferation and apoptosis of porcine ovarian granulosa cells. *J Environ Sci Health A Tox Hazard Subst Environ Eng*. 2011;46(2):170-5.
41. Roychoudhury S, Detvanova L, Sirotkin AV, Toman R, Kolesarova A. In vitro changes in secretion activity of rat ovarian fragments induced by molybdenum. *Physiol Res*. 2014;63(6):807-9.

42. Jay Murray F, Tyl RW, Sullivan FM, Tiwary AK, Carey S. Developmental toxicity study of sodium molybdate dihydrate administered in the diet to Sprague Dawley rats. *Reprod Toxicol.* 2014;49:202-8.
43. ECHA. Developmental toxicity study: European Chemicals Agency; 2021 [updated 19 July 2021. Available from: <https://echa.europa.eu/nl/registration-dossier/-/registered-dossier/15826/7/9/3/?documentUUID=cc2281be-c909-4aef-ae87-db7531dd6f86>.
44. Mohamed HRH, El-Atawy RH, Ghoneim AM, El-Ghor AA. Induction of fetal abnormalities and genotoxicity by molybdenum nanoparticles in pregnant female mice and fetuses. *Environmental science and pollution research international.* 2020.
45. Shirai S, Suzuki Y, Yoshinaga J, Mizumoto Y. Maternal exposure to low-level heavy metals during pregnancy and birth size. *Journal of Environmental Science and Health - Part A Toxic/Hazardous Substances and Environmental Engineering.* 2010;45(11):1468-74.
46. Vázquez-Salas RA, López-Carrillo L, Menezes-Filho JA, Rothenberg SJ, Cebrián ME, Schnaas L, et al. Prenatal molybdenum exposure and infant neurodevelopment in Mexican children. *Nutr Neurosci.* 2014;17(2):72-80.
47. Bloom MS, Buck Louis GM, Sundaram R, Maisog JM, Steuerwald AJ, Parsons PJ. Birth outcomes and background exposures to select elements, the Longitudinal Investigation of Fertility and the Environment (LIFE). *Environ Res.* 2015;138:118-29.
48. Bloom MS, Buck Louis GM, Sundaram R, Maisog JM, Steuerwald AJ, Parsons PJ. Corrigendum to Birth outcomes and background exposures to select elements, the Longitudinal Investigation of Fertility and the Environment (LIFE) [*Environ. Res.*, 138, (2015), 118-129] DOI: 10.1016/j.envres.2015.01.008. *Environmental Research.* 2015;142:760-1.
49. Howe CG, Claus Henn B, Eckel SP, Farzan SF, Grubbs BH, Chavez TA, et al. Prenatal Metal Mixtures and Birth Weight for Gestational Age in a Predominately Lower-Income Hispanic Pregnancy Cohort in Los Angeles. *Environ Health Perspect.* 2020;128(11):117001.
50. Kim SS, Meeker JD, Aung MT, Yu Y, Mukherjee B, Cantonwine DE, et al. Urinary trace metals in association with fetal ultrasound measures during pregnancy. *Environmental Epidemiology.* 2020;4(2).
51. Howe CG, Claus Henn B, Farzan SF, Habre R, Eckel SP, Grubbs BH, et al. Prenatal metal mixtures and fetal size in mid-pregnancy in the MADRES study. *Environ Res.* 2021;196:110388.
52. Karakis I, Landau D, Gat R, Shemesh N, Tirosh O, Yitshak-Sade M, et al. Maternal metal concentration during gestation and pediatric morbidity in children: an exploratory analysis. *Environmental Health and Preventive Medicine.* 2021;26(1).
53. McKeating DR, Clifton VL, Hurst CP, Fisher JJ, Bennett WW, Perkins AV. Elemental Metabolomics for Prediction of Term Gestational Outcomes Utilising 18-Week Maternal Plasma and Urine Samples. *Biological Trace Element Research.* 2021;199(1):26-40.

54. Tung PW, Burt A, Karagas M, Jackson BP, Punshon T, Lester B, Marsit CJ. Prenatal exposure to metal mixtures and newborn neurobehavior in the Rhode Island Child Health Study. *Environmental Epidemiology*. 2022;6(1):E194.
55. Yan L, Wang B, Li Z, Liu Y, Huo W, Wang J, et al. Association of essential trace metals in maternal hair with the risk of neural tube defects in offspring. *Birth Defects Res*. 2017;109(3):234-43.
56. Deyssenroth MA, Gennings C, Liu SH, Peng S, Hao K, Lambertini L, et al. Intrauterine multi-metal exposure is associated with reduced fetal growth through modulation of the placental gene network. *Environ Int*. 2018;120:373-81.
57. Hou Q, Huang L, Ge X, Yang A, Luo X, Huang S, et al. Associations between multiple serum metal exposures and low birth weight infants in Chinese pregnant women: A nested case-control study. *Chemosphere*. 2019;231:225-32.
58. Ovayolu A, Ovayolu G, Karaman E, Yuce T, Ozek MA, Turksoy VA. Amniotic fluid levels of selected trace elements and heavy metals in pregnancies complicated with neural tube defects. *Congenit Anom (Kyoto)*. 2020;60(5):136-41.
59. Pi X, Wei Y, Li Z, Jin L, Liu J, Zhang Y, et al. Higher concentration of selenium in placental tissues is associated with reduced risk for orofacial clefts. *Clin Nutr*. 2019;38(5):2442-8.
60. Troisi J, Giugliano L, Sarno L, Landolfi A, Richards S, Symes S, et al. Serum metallome in pregnant women and the relationship with congenital malformations of the central nervous system: A case-control study. *BMC Pregnancy and Childbirth*. 2019;19(1).
61. Manduca P, Al Baraquni N, Parodi S. Long term risks to neonatal health from exposure to war-9 years long survey of reproductive health and contamination by weapon-delivered heavy metals in Gaza, Palestine. *International Journal of Environmental Research and Public Health*. 2020;17(7).
62. Gómez-Roig MD, Mazarico E, Cuadras D, Muniesa M, Pascal R, Ferrer P, et al. Placental chemical elements concentration in small fetuses and its relationship with Doppler markers of placental function. *Nutrients*. 2021;110:1-8.
63. Tian T, Yin S, Jin L, Liu J, Wang C, Wei J, et al. Single and mixed effects of metallic elements in maternal serum during pregnancy on risk for fetal neural tube defects: A Bayesian kernel regression approach. *Environmental Pollution*. 2021;285.
64. Yin S, Wang C, Wei J, Wang D, Jin L, Liu J, et al. Essential trace elements in placental tissue and risk for fetal neural tube defects. *Environ Int*. 2020;139:105688.
65. Yin S, Wang C, Wei J, Jin L, Liu J, Wang L, et al. Selected essential trace elements in maternal serum and risk for fetal orofacial clefts. *Science of the Total Environment*. 2020;712.
66. Zhao H, Wu W, Zhang X, Zhu Q, Tang J, He H, et al. Associations between molybdenum exposure and ultrasound measures of fetal growth parameters: A pilot study. *Chemosphere*. 2021;269.
67. Wei J, Wang C, Yin S, Pi X, Jin L, Li Z, et al. Concentrations of rare earth elements in maternal serum during pregnancy and risk for fetal neural tube defects. *Environ Int*. 2020;137:105542.

68. Bi CM, Zhang YL, Liu FJ, Zhou TZ, Yang ZJ, Gao SY, et al. The effect of molybdenum on the in vitro development of mouse preimplantation embryos. *Syst Biol Reprod Med*. 2013;59(2):69-73.
69. EFSA NDA Panel. Scientific Opinion on Dietary Reference Values for molybdenum. *EFSA Journal*. 2013;11(8):35.
70. Wappelhorst O, Kuhn I, Heidenreich H, Markert B. Transfer of selected elements from food into human milk. *Nutrition*. 2002;18(4):316-22.
71. Bjorklund KL, Vahter M, Palm B, Grander M, Lignell S, Berglund M. Metals and trace element concentrations in breast milk of first time healthy mothers: a biological monitoring study. *Environ Health*. 2012;11:92.
72. Motoyama K, Isojima T, Sato Y, Aihara A, Asakura H, Hiraike H, et al. Trace element levels in mature breast milk of recent lactating Japanese women. *Pediatrics international : official journal of the Japan Pediatric Society*. 2020.
73. Tahboub YR, Massadeh AM, Al-sheyab NA, El shrafat D, Nsserat IA. Levels of Trace Elements in Human Breast Milk in Jordan: a Comparison with Infant Formula Milk Powder. *Biological Trace Element Research*. 2021.
74. Mandia N, Bermejo-Barrera P, Herbello P, Lopez-Suarez O, Fraga JM, Fernandez-Perez C, Couce ML. Human Milk Concentrations of Minerals, Essential and Toxic Trace Elements and Association with Selective Medical, Social, Demographic and Environmental Factors. *Nutrients*. 2021;13(6).
75. Iwai K, Iwai-Shimada M, Asato K, Nakai K, Kobayashi Y, Nakayama SF, Tatsuta N. Intra- and Inter-Day Element Variability in Human Breast Milk: Pilot Study. *Toxics*. 2022;10(3).
76. Bilston-John SH, Narayanan A, Tat Lai C, Rea A, Joseph J, Geddes DT. Daily and within-feed variation of macro- and trace-element concentrations in human milk and implications for sampling. *Food Chem*. 2021;363:130179.

10 Annex A Selection of molybdenum and molybdenum compounds.

First, an overall list with molybdenum and molybdenum compounds was compiled. This list was based on data from a report of The Dutch Expert Committee on Occupational Standards (2013), the ATSDR report (2020), REACH registrations and the Handbook of chemistry and physics.

For each compound, information on reproduction toxicity, water solubility and the existence of a REACH registration dossier was collected. Reproduction toxicity data were collected based on the literature search and from the registration dossiers when available.

Second, criteria were set to be used for grouping of molybdenum compounds. Grouping and read-across should be done with caution, especially for inorganic substances including metals and metal compounds. Guidances such as ECHA's Read-Across Assessment Framework (RAAF)²³ may be interpreted differently to come to justified and scientifically sound grouping.

According to the RAAF, solubility is the main parameter for grouping of metals and metal compounds.

A selection of molybdenum compounds was made based on the following criteria:

1. Availability of reproduction toxicity data. Data were available for four compounds, which are molybdenum, sodium molybdate, ammonium molybdate and molybdenum trioxide. These compounds were used as the starting point.
2. Water solubility. The solubility of molybdenum compounds determines (partly) the bioavailability of the compounds, and thus also the potential of inducing toxicity. Therefore, only compounds for which water solubility data are available and that have a similar solubility as one of the selected compounds mentioned above under 1), were considered. Compounds with a solubility of >10 g/L were considered soluble; compounds with a solubility of 0.001 – 10 g/L were considered slightly soluble.

Applying these criteria, this resulted in the selection of the following three groups of compounds:

1. Molybdenum
2. Ammonium molybdate(VI), sodium molybdate, ammonium paramolybdate, diammonium dimolybdate, ammonium tetrathio molybdate and potassium (soluble compounds)
3. Molybdenum trioxide, tetraammonium hexamolybdate and molybdenum sulfide (MoS₂), roasted (slightly soluble compounds)

An overview of all molybdenum compounds considered is presented in Table A1, indicating the selected molybdenum compounds for grouping.

²³ https://echa.europa.eu/documents/10162/13628/raaf_en.pdf/614e5d61-891d-4154-8a47-87efebd1851a

Next to the mono-constituent molybdenum substances, more complex structures containing molybdenum exist, such as reaction products, UVCBs and mixtures. These compounds cannot be used for grouping and read-across with the mono-constituent molybdenum substances and were therefore excluded from further selection. However, reproduction toxicity data from those complex structures may still be of interest for interpretation of the overall data. Reproduction toxicity data of those structures were obtained from the ECHA database and included in Annex B.

Table A1 List of all molybdenum compounds considered for this overview.

Chemical name	Synonyms	Chemical formula	CAS number	REACH registration dossier	Reproduction toxicity data	Solubility in water
<i>Selected molybdenum compounds</i>						
Molybdenum	Metallic molybdenum	Mo	7439-98-7	+	+	Insoluble
Ammonium orthomolybdate	Ammonium molybdate (VI), diammonium molybdate	(NH ₄) ₂ MoO ₄	13106-76-8	+	+	Soluble
Sodium molybdate	Sodium molybdate (VI), disodium molybdate	Na ₂ MoO ₄	7631-95-0; 10102-40-6 (dihydrate)	+	+	Soluble
Ammonium paramolybdate	Ammonium molybdate (VI) tetrahydrate; hexaammonium heptamolybdate	(NH ₄) ₆ Mo ₇ O ₂₄ (NH ₄) ₆ Mo ₇ O ₂₄ · 4H ₂ O	12027-67-7 12054-85-2; (tetrahydrate)	+	-	Soluble
Diammonium dimolybdate	Ammonium dimolybdate	(NH ₄) ₂ Mo ₂ O ₇	27546-07-2	+	-	Soluble
Dipotassium tetraoxomolybdate	Potassium molybdate	K ₂ MoO ₄	13446-49-6	+	-	Soluble
Ammonium tetrathiomolybdate	Tiomolibdate diammonium; ammonium molybdenum sulfide; ammonium tetrathiomolybdate; thiomolybdic acid, diammonium salt;	(NH ₄) ₂ MoS ₄	15060-55-6	-		Soluble
Molybdenum trioxide	Molybdenum (VI) oxide, molybdenum peroxide	MoO ₃	1313-27-5	+	-	Slightly soluble
Tetraammonium hexamolybdate		(NH ₄) ₄ Mo ₈ O ₂₆	12411-64-2	+	-	Slightly soluble
Molybdenum sulfide (MoS ₂), roasted		MoS ₂	86089-09-0	+	-	Slightly soluble
Molybdic acid (→ see Mo trioxide)		H ₂ MoO ₄	7782-91-4	+	-	Slightly soluble

Chemical name	Synonyms	Chemical formula	CAS number	REACH registration dossier	Reproduction toxicity data	Solubility in water
<i>Compounds considered but not selected for grouping</i>						
Molybdenite	Molybdenum (IV) sulfide, molybdenum disulfide	MoS ₂	1317-33-5	-		Insoluble
Molybdenum chloride	Molybdenum (V) chloride, Molybdenum pentachloride	MoCl ₅	10241-05-1	-		Insoluble
Calcium molybdate	Calcium molybdate (VI), Powellite	CaMoO ₄	7789-82-4	+	-	Insoluble
Molybdenum dioxide	Dioxomolybdenum, Molybdenum(IV)oxide	MoO ₂	18868-43-4	+	-	Insoluble
Molybdenum disilicide		MoSi ₂	12136-78-6	+	+ ²⁴	Insoluble
Molybdenum sulfide		H ₂ MoS	12612-50-9	+	-	Insoluble
Dimolybdenum carbide		CMo ₂	12069-89-5	+	-	Insoluble
Molybdophosphoric acid		H ₃ P(Mo ₃ O ₁₀) ₄	51429-74-4	-	-	-
Molybdenum boride		Mo ₂ B	12006-99-4	-	-	-
Molybdenum boride		Mo ₂ B ₅	12007-97-5	-	-	-
Molybdenum carbide		MoC	12011-97-1	-	-	-
Molybdenum carbonyl		Mo(CO) ₆	13939-06-5	-	-	Insoluble
Molybdenum nitride		MoN	12033-19-1	-	-	-
Molybdenum nitride		Mo ₂ N	12033-31-7	-	-	-
Molybdenum phosphide		MoP	12163-69-8	-	-	-
Molybdenum(II) bromide		MoBr ₂	13446-56-5	-	-	Insoluble
Molybdenum(II) chloride		MoCl ₂	13478-17-6	-	-	Insoluble
Molybdenum(II) iodide		MoI ₂	14055-74-4	-	-	Insoluble
Molybdenum(III) bromide		MoBr ₃	13446-57-6	-	-	Insoluble
Molybdenum(III) chloride		MoCl ₃	13478-18-7	-	-	Insoluble

²⁴ The registration dossier on molybdenum disilicide (EC number 235-231-8; CAS number 12136-78-6) contains a reproduction / developmental toxicity screening test (OECD 421). According to the summary, no treatment-related effects were observed. However, no details were provided at all. Since this is the only insoluble molybdenum compound with reproduction toxicity data and no details were available for this study, this compound was not included in the selection of molybdenum compounds.

Chemical name	Synonyms	Chemical formula	CAS number	REACH registration dossier	Reproduction toxicity data	Solubility in water
Molybdenum(III) fluoride		MoF ₃	20193-58-2	-	-	Insoluble
Molybdenum(III) iodide		MoI ₃	14055-75-5	-	-	Insoluble
Molybdenum(III) oxide		Mo ₂ O ₃	1313-29-7	-	-	Insoluble
Molybdenum(IV) bromide		MoBr ₄	13520-59-7	-	-	Reacts with H ₂ O
Molybdenum(IV) chloride		MoCl ₄	13320-71-3	-	-	Reacts with H ₂ O
Molybdenum(IV) fluoride		MoF ₄	23412-45-5	-	-	Reacts with H ₂ O
Molybdenum(IV) iodide		MoI ₄	14055-76-6	-	-	Insoluble
Molybdenum(IV) selenide		MoSe ₂	12058-18-3	-	-	-
Molybdenum(IV) telluride		MoTe ₂	12058-20-7	-	-	-
Molybdenum(V) fluoride		MoF ₅	13819-84-6	-	-	Reacts with H ₂ O
Molybdenum(V) oxytrichloride		MoOCl ₃	13814-74-9	-	-	Reacts with H ₂ O
Molybdenum(VI) dioxydichloride		MoO ₂ Cl ₂	13637-68-8	-	-	Reacts with H ₂ O
Molybdenum(VI) dioxydifluoride		MoO ₂ F ₂	13824-57-2	-	-	-
Molybdenum(VI) fluoride		MoF ₆	7783-77-9	-	-	Reacts with H ₂ O
Molybdenum(VI) metaphosphate		Mo(PO ₃) ₆	133863-98-6	-	-	Insoluble
Molybdenum(VI) oxytetrachloride		MoOCl ₄	13814-75-0	-	-	-
Molybdenum(VI) oxytetrafluoride		MoOF ₄	14459-59-7	-	-	-
Molybdenum(VI) sulfide		MoS ₃	12033-29-3	-	-	Insoluble
Ammonium molybdophosphate		(NH ₄) ₃ PO ₄ ·12MoO ₃	12026-66-3	-	-	Slightly soluble

11 Annex B Reproduction toxicity data of additional compounds containing molybdenum

The REACH registration database was searched for compounds containing molybdenum, such as reaction products and substances with unknown or variable composition (UVCBs). Reproduction toxicity data for those additional compounds are presented in Table B1.

Table B1 Animal studies on sexual function and fertility and development with compounds containing molybdenum.

Reference	Species	Experimental period and design	Dose and route	General toxicity	Effects on reproductive organs or reproduction	Remarks
Study report, 2018 ²⁵	Wistar rats, males and females. N=12/sex/dose	OECD Guideline 422 (combined repeated dose toxicity study with the reproduction / developmental toxicity screening test)	0, 100, 300 and 1000 mg/kg bw/day, by oral gavage. Test item: Xanthylum, 9-[2-(ethoxycarbonyl)phenyl]-3,6-bis(ethylamino)-2,7-dimethyl-, molybdatesilicate (trade name: Lumière Pink S.M. 8135N) (CAS# 63022-06-0; EC# 263-793-4) Purity: not specified	Kidney toxicity was observed at doses of 100 mg/kg bw/day and above. Thymus of both sexes at 1000 mg/kg/day: minimal involution/atrophy or minimally increased cortical apoptosis. Cortical hypertrophy was also observed in the affected females. Adrenal glands of females at 1000 mg/kg/day: bilateral, minimal to slight diffuse cortical hypertrophy.	No effects on reproduction (estrous cycle, pre-coital interval, mating performance, fertility and gestation length or in the offspring on litter size, survival, sex ratio, clinical signs, body weights, ano-genital distances or macropathology). Mean ano-genital distance in male offspring was higher in all treatment groups compared with the controls, but no differences were seen between the three treatment groups (no data provided). The differences were statistically significant. Despite that, the study authors considered it not dose related. NOAEL was 1000 mg/kg bw/day.	OECD guideline study. Only summary available. Only quantitative data available.

²⁵ <https://echa.europa.eu/nl/registration-dossier/-/registered-dossier/20458/7/9/2>

Reference	Species	Experimental period and design	Dose and route	General toxicity	Effects on reproductive organs or reproduction	Remarks
Study report, 2018 ²⁶	Sprague-Dawley rats, males and females. N=12/sex/dose	OECD 422 (Combined Repeated Dose Toxicity Study with the Reproduction / Developmental Toxicity Screening Test)	0, 100, 300 and 700 mg/kg bw/day, by oral gavage. Test item: UVCB containing 6 constituents. 1. Molybdenum, bis[O,O-bis(2-ethylhexyl)phosphorodithioato-.kappa.S, .kappa.S']dioxodi-.mu.-thioxodi-, (Mo-Mo) (EC # 615-708-0; CAS # 72030-25-2); 2. Molybdenum, bis[O,O-bis(2-ethylhexyl)phosphorodithioato-.kappa.S, .kappa.S']-.mu.-oxodioxo-.mu.-thioxodi- (CAS # 153128-45-1). 3. Molybdenum, bis[O,O-bis(2-ethylhexyl)	Mean body weights of the high dose males group were consistently lower than that of control animals from study day 8 to 29 (-23.5% to -39.9%). Minimal increases in ALT activity in animals administered 300 or 700 mg/kg/day and in gamma glutamyl transpeptidase (GGT) activities in animals administered 700 mg/kg/day. Minimal increases in GGT activities in females at PND 13, administered 700 mg/kg/day. Test item-related microscopic changes were observed in the thyroid at doses \geq 100 mg/kg/day in males and at doses \geq 300 mg/kg/day in females, in the liver in both sexes at doses \geq 300 mg/kg/day and the stomach in males at doses \geq 300 mg/kg/day and females at a dose level of 700 mg/kg/day.	No reproductive or developmental toxicity. <i>Reproductive toxicity</i> No reproductive toxicity observed at all dose levels, up to 700 mg/kg/day. In the males at doses up to 700 mg/kg/day, there were no differences in reproductive organ weights or reproductive parameters evaluated. In females at doses up to 700 mg/kg/day, there were no differences in oestrus cycles, pregnancy, or conception rates. <i>Developmental toxicity</i> The test substance did not induce developmental toxicity at all dose levels evaluated. The litter size, pup weight and observations on the morphology of the pups were performed from birth to PND 13 and there was no indication that the test substance affected development.	GLP study. Only summary available. Mainly quantitative data. Test item was not specified.

²⁶ <https://echa.europa.eu/nl/registration-dossier/-/registered-dossier/25978/7/9/2>

Reference	Species	Experimental period and design	Dose and route	General toxicity	Effects on reproductive organs or reproduction	Remarks
			<p>phosphorodithioato- .kappa.S, (N.kappa.S']oxodi- .mu.-thioxothioxodi- (no CAS/EC number) 4. Unspecified 2- ethylhexyl thiophosphate esters (no CAS/EC number) 5. Unspecified 2- ethylhexyl phosphate esters (no CAS/EC number) 6. Unspecified 2- ethylhexyl dithiophosphate esters (no CAS/EC number) Purity: assumed 100%</p>			
Study report, 2018 ²⁷	Crl:CD(SD) rats, males and females. N=10/sex/dose	OECD 422 (Combined Repeated Dose Toxicity Study with the Reproduction / Developmental	0, 100, 330 and 1000 mg/kg bw/day, by oral gavage. Test item: Molybdenum, bis	No general toxicity was observed.	No reproduction or developmental toxicity. <i>Reproduction toxicity</i> Estrous cycles, pre-coital interval, fertility, mating performance, gestation length and index were	GLP study. Only summary available.

²⁷ <https://echa.europa.eu/nl/registration-dossier/-/registered-dossier/23767/7/9/2>

Reference	Species	Experimental period and design	Dose and route	General toxicity	Effects on reproductive organs or reproduction	Remarks
		Toxicity Screening Test)	(dibutylcarbamodithioato), CAS 68412-26-0 Purity: UVCB substance		unaffected by treatment. There was no effect of treatment on the number of implantations or litter size. <i>Developmental toxicity</i> There was no effect of treatment on the number of implantations, litter size, the growth of the offspring, anogenital distance or nipple counts.	
Study report, 2019 ²⁸	Wistar rats, males and females. N=10/sex/dose	OECD 422 (Combined Repeated Dose Toxicity Study with the Reproduction / Developmental Toxicity Screening Test)	0, 100, 300 and 1000 mg/kg bw/day, by oral gavage. Test item: Molybdenum, bis(N,N-dibutylcarbamodithioato-κS,κS')dioxodi-μ-thioxodi-, stereoisomer (EC # 825-571-0; CAS # 60428-79-7). Purity: no data	All males and females at 300 and 1000 mg/kg showed pale faeces starting at Treatment Day 27. In 1000 mg/kg males, the faeces appeared normal from Day 26 of the recovery period onwards. In 1000 mg/kg females, faeces were normal from Day 5 of the recovery period.	No reproduction or developmental toxicity. No effects observed on oestrus cycle, sperm measures, reproductive performance or litter observations (number and sex of pups, stillbirths, live births, postnatal mortality, gross anomalies, weight gain, physical or behavioural abnormalities, anogenital distance (AGD), pup weight on the day of AGD, presence of nipples/areolae in male pups).	GLP study. Only summary available. Mainly quantitative data available. Test item was not specified.
Study report 2010 ²⁹	Crj:CD(SD) rats, males and females.	OECD 421 (Reproduction / Developmental	100, 300, and 1000 mg/kg bw/day	Clinical signs at 1000 mg/kg bw/day:	No reproduction or developmental toxicity. <i>Reproduction toxicity</i>	GLP study. Only summary

²⁸ <https://echa.europa.eu/nl/registration-dossier/-/registered-dossier/28090/7/9/2>

²⁹ <https://echa.europa.eu/registration-dossier/-/registered-dossier/12908/7/9/2>

Reference	Species	Experimental period and design	Dose and route	General toxicity	Effects on reproductive organs or reproduction	Remarks
	N = 12/sex/group	Toxicity Screening Test)	Test item: long chain alkyl thio carbamide metal complex (EC# 457-320-2) Purity: no data	<p>- intermittent tremors, repetitive movement of mouth and jaws, excessive pawing and/or licking of the cage floor and/or walls (females only, beginning on study day 13), and/or head twitch, in males and females. Seen at 1-3 hours following dose administration, beginning on study day 12, continuing sporadically throughout the remainder of treatment.</p> <p>- salivation, throughout the treatment period.</p> <p>Effects on organ weight, at 1000 mg/kg bw/day:</p> <p>- Females had a higher mean liver weight when compared with the control group. 1 of those females had yellow areas on the liver, as observed by histopathology</p>	<p>There were no test substance-related effects on male and female reproductive performance (mating, fertility, copulation, and conception indices and pre-coital intervals), mean gestation lengths, or the process of parturition at any dosage level. In reproductive tissues, no test substance-related changes in organ weights, gross necropsy observations, or histologic changes were detected. In addition, the mean numbers of unaccounted-for sites, former implantation sites, and corpora lutea were similar between the control and test substance-treated groups.</p> <p><i>Developmental toxicity</i> There were no effects on the numbers of pups born, live litter size, percentage of males at birth, pup survival, or on mean male and female pup body weights and body weight gains during PND 1-4. No test substance-related clinical findings were noted for pups at any dosage level; no test substance-related macroscopic</p>	available, and mainly quantitative data.

Reference	Species	Experimental period and design	Dose and route	General toxicity	Effects on reproductive organs or reproduction	Remarks
					findings were observed in pups that were found dead.	
Study report, 2018 ³⁰	Crl:CD(SD) rats, males and females. N = 10/sex/group	OECD 422 (Combined Repeated Dose Toxicity Study with the Reproduction / Developmental Toxicity Screening Test)	0, 100, 330 and 1000 mg/kg bw/day, by oral gavage. Test item: multi-constituent substance, with four constituents. 1. Bis(C11-14-branched and linear alkyl)dithiocarbamic acid 2. Molybdenum, bis(C11-14 branched and linear alkyl) carbamodithioato-.kappa.S,.kappa.S') oxodi-thioxothioxodi- 3. Molybdenum, bis(C11-14 branched and linear alkyl) carbamodithioato-.kappa.S,.kappa.S') dioxodi-thioxodi-	No treatment-related general toxicity was observed. Some effects observed were considered not related to treatment.	No reproduction or developmental toxicity. <i>Reproduction toxicity</i> The reproductive endpoints assessed which were unaffected by treatment included oestrus cycles, pre-coital interval, gestation length, mating performance, fertility, litter size, offspring survival or offspring sex ratio. <i>Developmental toxicity</i> Offspring clinical condition, body weight, body weight gain, anogenital distance and external development were also unaffected by treatment.	GLP study. Only summary available. Mainly quantitatively data. Study is part of registration dossier of Molybdenum, N,N-bis(C11-14-branched and linear alkyl)carbamodithioate oxothioxo complexes (EC number 948-019-1).

³⁰ <https://echa.europa.eu/nl/registration-dossier/-/registered-dossier/27562/7/9/2>

Reference	Species	Experimental period and design	Dose and route	General toxicity	Effects on reproductive organs or reproduction	Remarks
			4. Molybdenum, bis(N,N-di-C11-14 branched and linear alkyl) carbamodithioato-.kappa.S,.kappa.S')-oxodioxo-thioxodi- Purity: >90% Control: vehicle (arachis oil)			
Study report, 2006 ³¹	CrI:CD (SD) rats, males and females. N(F ₀) = 28/sex/group N(F ₁) = 24/sex/group	OECD 416 (Two-Generation Reproduction Toxicity Study)	0, 250, 500 and 1000 mg/kg bw/day (3000, 6000 and 12000 ppm), by diet. Test item: reaction product of cocoalkyldiethanolamides and cocoalkylmonoglycerides and molybdenumtrioxide (1.75-2.2: 0.75-1.0:0.1-1.1) (Trade name: Molyvan 855) (EC number 430-380-7; CAS number 445409-27-8)	General toxicity was observed at 500 and 1000 mg/kg bw/day. Effects on body weight and weight changes, food consumption, organ weights and histopathological findings (kidneys, mesenteric lymph nodes, thymus, and bone marrow) were observed.	No treatment-related effects were observed on the reproductive function and performance of the F ₀ and F ₁ generation. Effects in ovaries: fine cytoplasmic vacuolation of interstitial cells was observed with lower incidence among animals treated with 12000 ppm (P <0.001) or 6000 ppm (P <0.05) of the test material. The significance of this change is uncertain, according to the study authors. A greater incidence of higher grades of severity of yellow pigment accumulation was also observed for females treated with 12000 ppm (P <0.01). The pigment was demonstrated to be	GLP study. Only summary available, and only quantitative data.

³¹ <https://echa.europa.eu/nl/registration-dossier/-/registered-dossier/31478/7/9/2>

Reference	Species	Experimental period and design	Dose and route	General toxicity	Effects on reproductive organs or reproduction	Remarks
			Purity: no data		<p>haemosiderin by Perl's staining technique.</p> <p>F0 adult females, high dose:</p> <ul style="list-style-type: none"> - Reduced live litter size at birth - Reduced live birth index - Reduction corpora lutea <p>F1 adult females, high dose:</p> <ul style="list-style-type: none"> - Reduction corpora lutea - Lower number of implantations - Reduced litter size at birth - However, no dose related response for corpora lutea and the deterioration in the physical condition of the animals <p>F1 offspring, high dose:</p> <ul style="list-style-type: none"> - increase in age at completion of sexual maturation - reduction in bodyweight at sexual maturation - reduction in absolute brain, spleen and thymus weight with the effect on brain 	

Reference	Species	Experimental period and design	Dose and route	General toxicity	Effects on reproductive organs or reproduction	Remarks
					weight extending to the mid dose group. F2 offspring: - reduction in absolute brain, spleen and thymus weight with the effect on brain weight extending to the mid dose group.	
Study report, 2018 ³²	Wistar rat, males and females. N = 12/sex/group	OECD 422 (Combined Repeated Dose Toxicity Study with the Reproduction / Developmental Toxicity Screening Test)	0, 100, 300 and 1000 mg/kg bw/day, by oral gavage. Test item: Benzenamine, N,N-dimethyl-, oxidized, molybdatetungstate phosphates, EC number 309-916-8) (trade name: Pigment Violet 3 PTM) Purity: preparation containing ≥80% UVCB	No treatment-related general toxicity was observed.	No effects observed at reproductive function or reproductive organs. No effects observed on development.	GLP study. Only summary available, and only quantitative data.
Study report, 2016 ³³	Wistar rat, males and females.	OECD 422 (Combined Repeated Dose	0, 25, 100 and 400 mg/kg bw/day, by oral gavage.	Clinical signs of discomfort/toxicity (hunched posture, piloerection) were	<i>Reproduction toxicity</i>	GLP study. Only summary

³² <https://echa.europa.eu/nl/registration-dossier/-/registered-dossier/24634/7/9/1>

³³ <https://echa.europa.eu/nl/registration-dossier/-/registered-dossier/27857/7/9/1>

Reference	Species	Experimental period and design	Dose and route	General toxicity	Effects on reproductive organs or reproduction	Remarks
	N = 10/sex/group	Toxicity Study with the Reproduction / Developmental Toxicity Screening Test)	Test item: Bis[O,O-bis(2-ethylhexyl)dithiophosphorato-S,S']dioxodi- μ -thioxodimolybdenum (EC number 273-381-6). Purity: no data	observed in parental animals of both sexes at the highest dose level of 400 mg/kg bw/day. In addition, several effects were observed at study termination (28 days for males and 51-62 days for females having offspring) that all were reversible after a 28 day recovery period without dosing.	No effects observed on reproduction or reproductive organs. <i>Developmental toxicity</i> One pup of the control group, two pups at 100 mg/kg (from two litters) and four pups at 400 mg/kg (from four litters) went missing (presumed cannibalized) on PND 2, 3 or 4. This post-natal loss was regarded by the authors of the study as unrelated to treatment due to the incidental occurrence (within the range considered normal for pups of this age). In female pups at PND 14-16, a trend was seen towards an increase in total T4 with increasing dose levels. The relative differences from controls were 4, 10 and 18% at 25, 100 and 400 mg/kg, respectively (statistical significance was not achieved). Mean values were within historical control; half of the individual values (at 100 and 400 mg/kg bw/day) exceeded the historical range.	available, and only quantitative data.

Reference	Species	Experimental period and design	Dose and route	General toxicity	Effects on reproductive organs or reproduction	Remarks
					TSH levels in female PND 14-16 pups were decreased at 400 mg/kg (0.85x of control), although not reaching statistical significance.	
Study report, 2018 ³⁴	Wistar rat, males and females. N = 12/sex/group	OECD 422 (Combined Repeated Dose Toxicity Study with the Reproduction / Developmental Toxicity Screening Test)	0, 100, 300 and 1000 mg/kg bw/day, by oral gavage. Test item: Ethanaminium, N-[4-[[4-(diethylamino)phenyl][4-(ethylamino)-1-naphthalenyl]methylene]-2,5-cyclohexadien-1-ylidene]-N-ethyl-, molybdatetungstate phosphate (EC number 215-410-7) Purity: Preparation containing ≥90% UVCB.	Treatment-related general toxicity is observed. - Mortality at the mid and high dose group (1 animal/group) - Changes in organ weight, not specified. - Kidney: minimal, bilateral, diffuse tubular basophilia in the renal cortices at high dose, and in one animal at mid dose. - Adrenal glands: high incidence of bilateral diffuse cortical hypertrophy in females at all dose levels. - Thymus: a minimal decrease in cortical cellularity in a male at high dose level; the incidence and severity of decreased cortical cellularity were higher in females in the mid and high dose group. - Spleen: minimally to slightly decreased marginal zone cellularity in one male and one female in the high dose group.	<i>Reproduction toxicity</i> No effects were observed on reproductive function. <i>Developmental toxicity</i> Effects on development were observed. - Slight reduction in gestation index in the mid and high dose group. - Body weights in male and female offspring in all dose groups tended to be lower than those recorded in the control group. Mean values at 100 and 300 mg/kg/day are similar; the differences with respect to the control are greater at 1000 mg/kg/day and increase with the days of lactation. - Slightly lower offspring survival index in the mid and high dose group.	GLP study. Only summary available, and only quantitative data.

³⁴ <https://echa.europa.eu/nl/registration-dossier/-/registered-dossier/27465/7/9/1>

Reference	Species	Experimental period and design	Dose and route	General toxicity	Effects on reproductive organs or reproduction	Remarks
Study report 2013 ³⁵	Crl:CD(SD) rats, males and females. Number of animals per sex and per group: no data	OECD 421 (Reproduction / Developmental Toxicity Screening Test)	0, 100, 300 and 1000 mg/kg bw/day, by oral gavage. Test item: S-900 (Reaction mass of Di- μ -thio-[[bis(2-ethylhexyl)carbamato-S,S']oxo molybdenum(V)], Di- μ -thio-[[{(2-ethylhexyl)carbamato-S,S'}{(branched dtridecyl)carbamato-S,S'}oxo molybdenum(V)] and Di- μ -thio-[[bis(branched dtridecyl)carbamato-S,S']oxo molybdenum(V)]; EC number 441-570-4) Purity: 98.7%	Overall body weight gain in females receiving 300 or 1000 mg/kg/day was low during the two week pre-mating period, compared with control animals.	<i>Reproduction toxicity</i> No adverse effects on reproduction observed. <i>Developmental toxicity</i> Clinical signs recorded for the offspring identified occasional pups with findings, but the type of findings and incidence were typical and showed no relationship to parental treatment. The predominant macropathological finding among the small number of decedent offspring on the study was no milk in stomach. This is a common finding and, in the absence of an effect upon offspring survival, is considered to be of no toxicological significance by the authors of this study.	GLP study. Only summary available, and only quantitative data.
Study report, 2019 ³⁶	Wistar rats, males and females. N = 10 males/dose	OECD 422 (Combined Repeated Dose Toxicity Study with the Reproduction /	0, 75, 150 and 300 mg/kg bw/day, by oral gavage. Test item: BiLUBE 4131	No treatment-related general toxicity was observed.	<i>Reproduction toxicity</i> No treatment-related effects on reproduction. It is noted that a slight, but not statistically significant decrease in	GLP study. Only summary available, and only

³⁵ <https://echa.europa.eu/nl/registration-dossier/-/registered-dossier/20044/7/9/1>

³⁶ <https://echa.europa.eu/nl/registration-dossier/-/registered-dossier/20183/7/9/1>

Reference	Species	Experimental period and design	Dose and route	General toxicity	Effects on reproductive organs or reproduction	Remarks
	group, and 15 females/dose group	Developmental Toxicity Screening Test)	Purity: no data		mating efficiency in all dose groups was observed (80% efficiency, compared to 100% in control group). <i>Developmental toxicity</i> No developmental toxicity was observed.	quantitative data. Study is part of the registration dossier of Alkanoic acid and polyoxomolybdate borate alkanoate clusters.

Published by:

**National Institute for Public Health
and the Environment, RIVM**

P.O. Box 1 | 3720 BA Bilthoven

The Netherlands

www.rivm.nl/en

December 2023

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