CHAPTER 2. HEALTH EFFECTS

2.1 INTRODUCTION

The primary purpose of this chapter is to provide public health officials, physicians, toxicologists, and other interested individuals and groups with an overall perspective on the toxicology of cobalt. It contains descriptions and evaluations of toxicological studies and epidemiological investigations and provides conclusions, where possible, on the relevance of toxicity and toxicokinetic data to public health. When available, mechanisms of action are discussed along with the health effects data; toxicokinetic mechanistic data are discussed in Section 3.1.

A glossary and list of acronyms, abbreviations, and symbols can be found at the end of this profile.

To help public health professionals and others address the needs of persons living or working near hazardous waste sites, the information in this section is organized by health effect. These data are discussed in terms of route of exposure (inhalation, oral, and dermal) and three exposure periods: acute (≤14 days), intermediate (15–364 days), and chronic (≥365 days).

As discussed in Appendix B, a literature search was conducted to identify relevant studies examining health effect endpoints. Figure 2-1 provides an overview of the database of studies in humans or experimental animals included in this chapter of the profile. These studies evaluate the potential health effects associated with inhalation, oral, or dermal exposure to cobalt, but may not be inclusive of the entire body of literature. A systematic review of the scientific evidence of the health effects associated with exposure to cobalt was also conducted; the results of this review are presented in Appendix C. As discussed in *Prioritization of Human Data* in Appendix C, epidemiological studies included in the profile are focused on known environmental exposures to cobalt (e.g., occupational studies). There is a vast literature of general population studies that measure cobalt (and other trace essential elements) in the blood or urine and health outcomes. Studies of this nature without information on sources of environmental exposure above background levels are not included in this document. Additionally, studies evaluating kinetics and potential toxic effects associated with medical applications of cobalt (e.g., implantable medical devices, hydroxocobalamin for treatment of cyanide poisoning) are not included in the profile, as the profile is concerned with environmental exposures via inhalation, oral, or dermal routes.

Human and animal inhalation studies are presented in Table 2-1 and Figure 2-2, and human and animal oral studies are presented in Table 2-2 and Figure 2-3; limited dermal data were identified for cobalt and are presented in Table 2-3.

Levels of significant exposure (LSEs) for each route and duration are presented in tables and illustrated in figures. The points in the figures showing no-observed-adverse-effect levels (NOAELs) or lowest-observed-adverse-effect levels (LOAELs) reflect the actual doses (levels of exposure) used in the studies. Effects have been classified into "less serious LOAELs" or "serious LOAELs (SLOAELs)." "Serious" effects are those that evoke failure in a biological system and can lead to morbidity or mortality (e.g., acute respiratory distress or death). "Less serious" effects are those that are not expected to cause significant dysfunction or death, or those whose significance to the organism is not entirely clear. ATSDR acknowledges that a considerable amount of judgment may be required in establishing whether an endpoint should be classified as a NOAEL, "less serious" LOAEL, or "serious" LOAEL, and that in some cases, there will be insufficient data to decide whether the effect is indicative of significant dysfunction. However, the Agency has established guidelines and policies that are used to classify these endpoints. ATSDR believes that there is sufficient merit in this approach to warrant an attempt at distinguishing between "less serious" and "serious" effects. The distinction between "less serious" effects and "serious" effects is considered to be important because it helps the users of the profiles to identify levels of exposure at which major health effects start to appear. LOAELs or NOAELs should also help in determining whether or not the effects vary with dose and/or duration, and place into perspective the possible significance of these effects to human health.

A User's Guide has been provided at the end of this profile (see Appendix D). This guide should aid in the interpretation of the tables and figures for LSEs and MRLs.

This section provides information regarding potential adverse health effects of cobalt and cobalt compounds. In order to appropriately compare doses following exposures to various cobalt compounds, inhalation exposures are expressed throughout the profile as mg Co/m^3 and oral and dermal exposures are expressed as mg Co/kg/day, when possible. This allows for evaluation of relative toxicities and determination of whether certain chemical properties (e.g., solubility) impact toxicity. When available, information pertaining to relative toxicity of different cobalt compounds is discussed throughout Chapter 2.

Cobalt salts (e.g., cobalt sulfate, cobalt chloride) can exist in anhydrous and hydrated forms. Since hydration status does not impact the toxicity of cobalt salts, it is not discussed throughout Chapter 2 in the health

effects sections (e.g., both anhydrous and hydrated forms of cobalt sulfate are referred to as "cobalt sulfate"). However, hydration status impacts conversion into mg Co/m^3 or mg Co/kg/day; therefore, hydration status is provided in the LSE tables. In cases for which hydration status of a salt was not explicitly reported (e.g., study authors report test substance as "cobalt chloride" without clearly specifying "anhydrous cobalt chloride" or 'cobalt chloride hexahydrate"), dose conversions were based on the available information in the study. That is, the molecular weight of the salt (e.g., cobalt chloride) was used, without any assumptions regarding the degree of hydration. Test compounds, exactly as reported by study authors, are reported in the LSE tables. Hydration status is particularly important for inhalation studies with cobalt sulfate heptahydrate, which will convert to cobalt sulfate hexahydrate when air humidity levels are <70% (Redhammer et al. 2007). Behl et al. (2015) confirmed that predominant hydration species in inhalation chambers during the NTP (1991) studies of cobalt sulfate heptahydrate was actually cobalt sulfate hexahydrate. Therefore, the dose conversions listed in the LSE table for NTP (1991) are based on the molecular weight of cobalt hexahydrate. Additionally, based on the information provided by Behl et al. (2015) and information regarding relative humidity (Viegas 2024), the dose conversions for other inhalation studies of cobalt sulfate heptahydrate reported in the LSE tables were also based on the molecular weight of cobalt sulfate hexahydrate (e.g., Burzlaff et al. 2022a; Viegas et al. 2022a).

The health effects of cobalt and cobalt compounds have been evaluated in 75 human and 118 animal studies. As illustrated in Figure 2-1, most of the health effects data come from oral exposure studies in animals and inhalation (occupational) exposure studies in humans.

Human studies predominantly focused on respiratory endpoints following inhalation exposure and hematological and endocrine (thyroid) effects following oral exposure. Animal studies examined a comprehensive set of endpoints following inhalation and oral exposure. For inhalation studies, the most well-studied system in animals is also the respiratory system. For oral studies, the most well-studied effects (after survival) include body weight, reproductive endpoints, and hematological effects. Data from dermal exposure are very limited.

As outlined in Chapter 1, the most sensitive effects following exposure to cobalt appear to be respiratory effects following inhalation exposure and gastrointestinal, hematological, and thyroid effects following oral exposure. A systematic review was conducted on the available human and animal studies for these endpoints. The information in these studies indicate the following on the potential targets of cobalt toxicity:

- **Respiratory Endpoints.** Respiratory effects are a known health effect for humans following inhalation exposure to cobalt based on a high level of evidence in humans and animals. In humans, occupational exposure has been associated with increased respiratory symptoms, impaired lung function, and asthma. In animals, inhalation exposure is consistently associated with inflammatory changes throughout the upper and lower respiratory tract progressing to widespread lesions with increasing concentration and duration in multiple species. At high exposure levels, severe pulmonary damage was the cause of death in rodents.

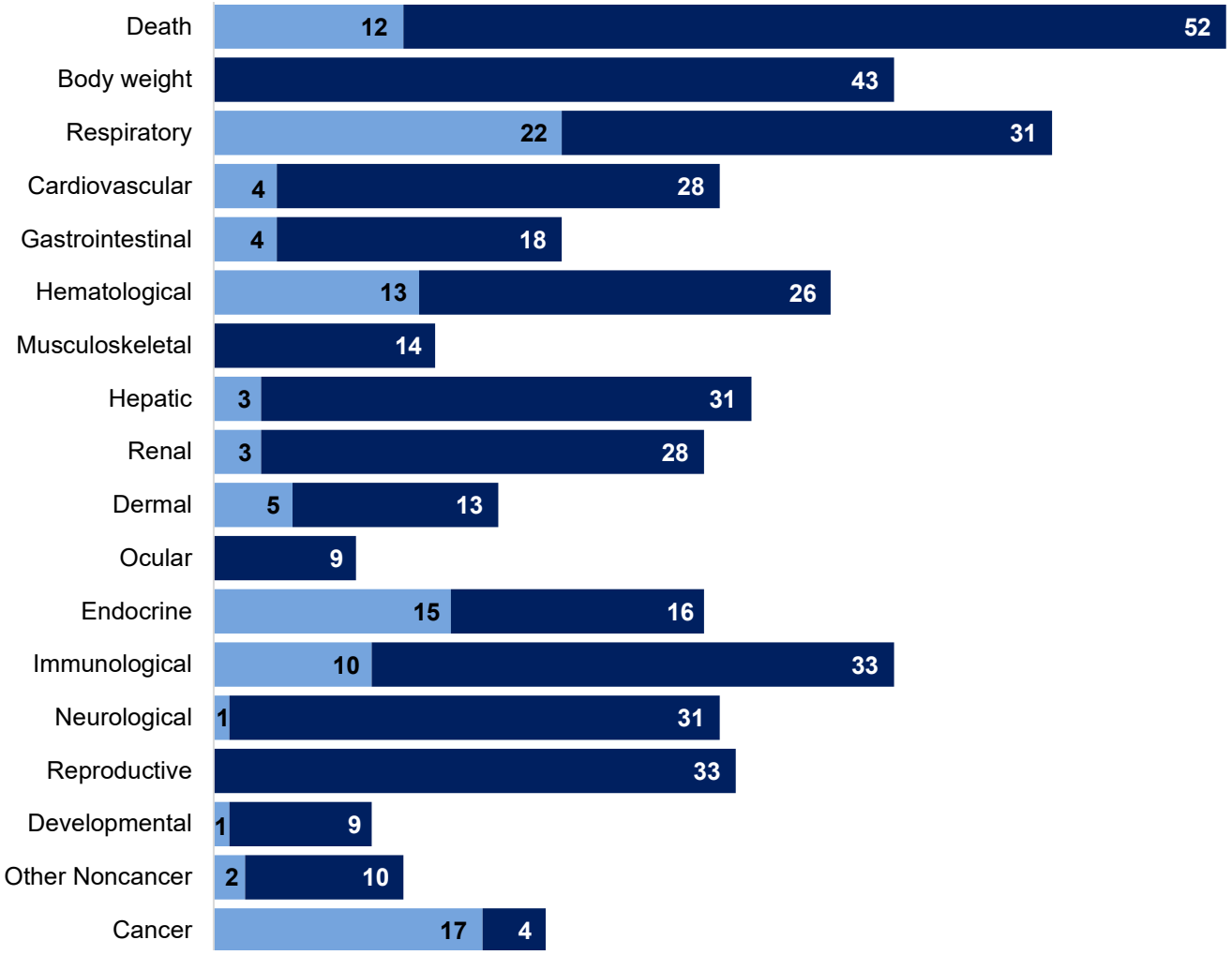
- **Gastrointestinal Endpoints.** Gastrointestinal effects are not classifiable as health effects for humans following oral exposure to cobalt due to inadequate data in humans and a low level of evidence in animals. Findings in humans are limited to complaints of gastrointestinal distress in some humans following oral exposure to cobalt potential treatment for anemia or hyperthyroidism. Animal data are limited to a few acute-duration oral studies in rats reporting damage to the small intestine and delays in gastric emptying; however, no histopathological changes were found throughout the gastrointestinal tract in intermediate-duration oral studies in rats.

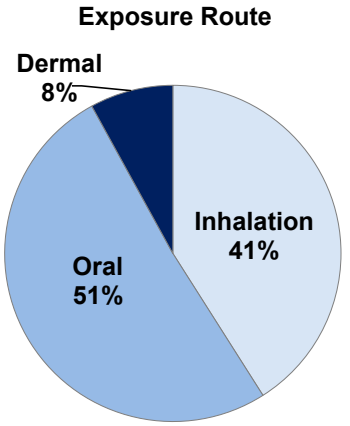
- **Hematological Endpoints.** Hematological effects are a presumed health effect for humans following oral exposure to cobalt based on a moderate level of evidence in humans and a high level of evidence in animals. One study reported an increase in red blood cell concentration (defined as polycythemia by the study authors) in healthy humans following acute- or intermediate-duration oral exposure to cobalt. Cobalt supplementation has also been shown to elevate red blood cell count when given to anemic patients. Polycythemia was not observed in healthy individuals in two additional studies with exposure to lower-dose cobalt exposure. Findings in humans are supported by numerous acute- and intermediate-duration rat studies that reported elevated erythrocytes, hematocrit, and/or hemoglobin following exposure to cobalt. Mechanistic data indicate that cobalt mimics hypoxic conditions, stimulating erythropoiesis.

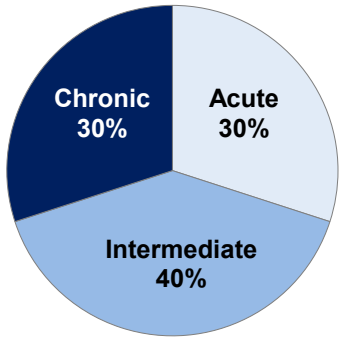
- **Thyroid Endpoints.** Thyroid effects are a suspected health effect for humans following oral exposure to cobalt based on a low level of evidence in humans and a moderate level of evidence in animals. There is limited evidence from case reports of goiter or impaired thyroid function in some patients taking cobalt as a treatment for anemia associated with sickle cell anemia, pregnancy, or chronic renal disease. Transient impairments in thyroid function were observed following acute- or intermediate-duration oral exposure to cobalt in some controlled human studies. Only a limited number of animal studies were identified, but they provide evidence of severe histopathological damage to the thyroid at high oral exposure doses in mice. A proposed mechanism of action is organic blocking of iodine by cobalt.

Figure 2-1. Overview of the Number of Studies Examining Cobalt Health Effects*

Most studies examined the potential lethal, body weight, respiratory, immunological, and hematological effects of cobalt
Fewer studies evaluated health effects in humans than animals (counts represent studies examining endpoint)







*Includes studies discussed in Chapter 2. A total of 193 studies (including those finding no effect) have examined toxicity; most studies examined multiple endpoints.

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
ACUTE EXPOSURE									
Kusaka et al. 1986a									**Cobalt Metal**
1	Human 15 M	6 hours	0, 0.038	CS, OF	Resp		0.038		Subjective complaints of respiratory irritation; decreased FVC
Burzlaff et al. 2022a									**Cobalt Tetraoxide**
2	Rat (Wistar) 5 M, 5 F	14 days 6 hours/day (N)	0, 9.86, 33.87, 160.90	LE, BW, BI, OW, GN, HP	Bd wt	160.9			
					Resp	9.86	33.87		Increased BALF levels of LDH and polymorphonuclear neutrophils
Palmes et al. 1959									**Cobalt Hydrocarbonyl**
3	Rat (Albino) 5–10 M	30 minutes (WB)	0, 7, 28, 47, 68, 78, 113, 191, 215, 222, 408	CS, LE	Death			165	LC_{50}
					Resp	68		78	Labored or disturbed respiration; severe pulmonary irritation
Exposure was to cobalt hydrocarbonyl plus oxide/carbonate decomposition products due to instability of test substance in oxygen.									
Palmes et al. 1959									**Cobalt Hydrocarbonyl**
4	Rat (Albino) 1–33 M	30 minutes (WB)	0, 7, 26, 83, 90, 106, 116, 137, 179, 236	GN, OW	Resp	26		83	Gross lung lesions (hemorrhage, edema, consolidation, congestion, pleuritis, bronchiectasis, emphysema, or atelectasis)
Exposure was to cobalt hydrocarbonyl plus oxide/carbonate decomposition products due to instability of test substance in oxygen.									
Viegas et al. 2022a, 2022b									**Cobalt Metal**
5	Rat (Sprague-Dawley) 3–5 M, 3–5 F	4 hours (N)	50, 500, 1,000, 5,000	LE, CS, HP	Death			50	100% mortality
Viegas et al. 2022a, 2022b									**Cobalt Hydroxide**
6	Rat (Sprague-Dawley) 3–5 M, 3–5 F	4 hours (N)	32, 320, 3,200	LE, CS, HP	Death			32	100% mortality

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Viegas et al. 2022a, 2022b									**Cobalt Carbonate**
7	Rat (Sprague-Dawley) 3–5 M, 3–5 F	4 hours (N)	32, 320, 3,200	LE, CS, HP	Death			3,200	LC_{50}
Viegas et al. 2022a, 2022b									**Cobalt Oxide**
8	Rat (Sprague-Dawley) 3–5 M, 3–5 F	4 hours (N)	39, 79, 390, 790, 3,900	LE, CS, HP	Death			47	LC_{50}
Viegas et al. 2022a, 2022b; Viegas 2024									**Cobalt Sulfate Heptahydrate**
9	Rat (Sprague-Dawley) 5 F	4 hours (WB)	0, 0.02, 0.07, 0.2, 2.2, 6.7	LE, CS, HP, BI	Resp	0.2[b]	2.2		Increased BALF neutrophils, decreased BALF cell viability
Test substance was likely converted to cobalt sulfate hexahydrate in the inhalation chamber due to relative humidity <70%.									
INTERMEDIATE EXPOSURE									
Bucher et al. 1990; NTP 1991, 2023									**Cobalt Sulfate Heptahydrate**
10	Rat (F344/N) 10 M, 10 F	13 weeks 5 days/week 6 hours + 12 minutes (T90 time) per day (WB)	0, 0.114, 0.376, 1.11, 3.78, 11.4	BC, BW, CS, GN, HE, HP, LE, OW, RX	Bd wt	11.4 F			
						3.78 M	11.4 M		14% decrease in final body weight
					Resp		0.114	3.78	LOAEL: Minimal-to-mild squamous metaplasia of the larynx SLOAEL: Lung fibrosis in females; laryngeal ulceration, necrosis, and polyps in both sexes
					Cardio	11.4			
					Gastro	11.4			
					Hemato	1.11 F	3.78 F		Polycythemia, decreased platelet count
						0.376 M	1.11 M		Polycythemia
					Musc/skel	11.4			
					Hepatic	11.4			

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
					Renal	11.4			
					Dermal	11.4			
					Endocr	1.11 F	3.78 F		Decreased serum T3
						3.78 M	11.4 M		Decreased serum TSH
					Immuno	11.4			
					Neuro	11.4			
					Repro	11.4			
					Other noncancer	11.4			
Exposure chamber analysis showed that under test conditions, the test substance converted to cobalt sulfate hexahydrate (Behl et al. 2015).									
Burzlaff et al. 2022a									**Cobalt Tetraoxide**
11	Rat (Wistar) 10 M, 10 F	28 days 6 hours/day (N)	0, 3.76, 15.05, 59.31	CS, FI, WI, BW, BC, BI, HE, UR, HP	Bd wt	59.31			
					Resp	3.76	15.05	59.31	LOAEL: Alveolar lipoproteinosis, increased LDH and polymorphonuclear neutrophils in BALF SLOAEL: Moderate interstitial fibrosis and interstitial inflammatory cell infiltration
					Cardio	59.31			
					Gastro	59.31			
					Hemato	59.31			
					Musc/skel	59.31			
					Hepatic	59.31			
					Renal	59.31			
					Endocr	59.31			
					Immuno	59.31			
					Neuro	59.31			
					Repro	59.31			

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Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Burzlaff et al. 2022a, 2022b									**Cobalt Sulfate Heptahydrate**
12	Rat (Wistar) 10 M, 10 F	28 days 6 hours/day (N)	0, 0.46	CS, WI, BW, BC, BI, HE, HP	Bd wt	0.46			
					Resp		0.46		Slight focal squamous metaplasia and inflammatory changes in larynx in both sexes; increased BALF LDH levels and polymorphonuclear neutrophils in males
Based on findings in other studies (Behl et al. 2015), the test substance likely converted to cobalt sulfate hexahydrate in the exposure chamber.									
NTP 1991, 2023									**Cobalt Sulfate Heptahydrate**
13	Rat (F344/N) 5 M, 5 F	16 days 5 days/week 6 hours + 12 minutes (T90 time) per day (WB)	0, 0.035, 0.19, 1.80, 19.0, 75.7	BW, CS, GN, HP, LE, OW	Death			75.7 F	5/5 died
								19 M	2/5 died
					Bd wt	1.8		19	Decrease in final body weight in males (47%) and females (23%)
					Resp	1.8		19	Lesions throughout the respiratory tract (inflammation, necrosis, hyperplasia, metaplasia, acanthosis, fibrosis, histiocytic infiltration)
					Cardio	75.7			
					Gastro	75.7			
					Musc/skel	75.7			
					Hepatic			75.7	Congestion and necrosis of liver (in rats that died)
					Renal	75.7			
					Dermal	75.7			
					Endocr	75.7			
					Immuno	1.8		19	Necrosis of thymus (in rats that died); decreased absolute and relative thymus weights
					Neuro	1.8		19	Congestion of vessels in brain (in rats that died); hypoactivity

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
					Repro	75.7 F			
						1.8 M		19 M	Testicular atrophy; decreased cellularity of the seminiferous tubules and atypical germinal epithelial cells in the epididymal ducts
Exposure chamber analysis showed that under test conditions, the test substance converted to cobalt sulfate hexahydrate (Behl et al. 2015).									
NTP 2014									**Cobalt Metal**
14	Rat (F344/N) 5 M, 5 F	16 days 5 days/week 6 hours + 12 minutes (T90 time) per day (WB)	0, 2.5, 5, 10, 20, 40	BW, CS, GN, HP, LE, OW, UR	Death			20	5/5 males and 3/5 females died
					Bd wt	5 F	10 F	20 F	LOAEL: 12% decrease in final body weight SLOAEL: 45% decrease in final body weight
						5 M		10 M	20% decrease in final body weight
					Resp		2.5	20	LOAEL: Minimal cytoplasmic vacuolization of bronchiolar epithelium; minimal-to-mild atrophy and necrosis of olfactory epithelium SLOAEL: Abnormal breathing; histiocytic infiltrates in the lungs; lung hemorrhage and acute inflammation in male; nasal respiratory epithelium necrosis in females
					Hepatic	2.5 F	5 F		Decreased absolute liver weight
							2.5 M		Decreased absolute and relative liver weight
					Renal	10 F	20 F		Increased urinary creatinine levels, decreased urine volume
						5 M	10 M		Increased urinary creatinine levels, decreased urine volume
					Immuno	10 F	20 F		Decreased relative thymus weight
						40 M			

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
					Neuro	20 F	40 F		Lethargy
						10 M	20 M		Lethargy
NTP 2014									**Cobalt Metal**
15	Rat (F344/N) 10 M, 10 F	14 weeks 5 days/week 6 hours + 12 minutes (T90 time) per day (WB)	0, 0.625, 1.25, 2.5, 5	BC, BW, CS, GN, HE, HP, LE, OW, RX, UR	Bd wt	5			
					Resp		0.625		Chronic active inflammation in lung, pulmonary alveolar proteinosis, increased relative lung weight
					Cardio	5			
					Gastro	5			
					Hemato	0.625 F	1.25 F		Increased hematocrit, hemoglobin, and red blood cell counts; decreased platelets
							0.625 M		Increased hemoglobin and red blood cell count
					Musc/skel	5			
					Hepatic	5			
					Renal	2.5 F	5 F		Increased relative kidney weight
						2.5 M	5 M		Increased serum creatinine
					Dermal	5			
					Ocular	5			
					Endocr	5			
					Immuno	5			
					Neuro	5			
					Repro	5 F			
						1.25 M	2.5 M		Decreased sperm motility, increased relative testes weight
					Other noncancer	5 F			
						0.625 M	1.25 M		Decreased serum glucose

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Palmes et al. 1959									**Cobalt Hydrocarbonyl**
16	Rat (Albino) 34–57 M	3 months 5 days/week 6 hours/day (WB)	0, 9	BW, CS, GN, HE, HP, LE	Bd wt	9			
					Hemato		9		Increased hemoglobin levels; decreased percent monocytes and increased percent basophils
Exposure was to cobalt hydrocarbonyl plus oxide/carbonate decomposition products due to instability of test substance in oxygen.									
Bucher et al. 1990; NTP 1991, 2023									**Cobalt Sulfate Heptahydrate**
17	Mouse (B6C3F1) 10 M, 10 F	13 weeks 5 days/week 6 hours + 12 minutes (T90 time) per day (WB)	0, 0.114, 0.376, 1.11, 3.78, 11.4	BC, BW, CS, GN, HE, HP, LE, OW, RX	Death			11.4 M	2/10 died
					Bd wt	3.78			
								11.4 F	22% decrease in final body weight
							11.4 M		14% decrease in final body weight
					Resp		0.114		Squamous metaplasia of the larynx in both sexes; histiocytic infiltrates in the lungs in males
					Cardio	11.4			
					Gastro	11.4			
					Hemato	3.78 F	11.4 F		Decreased platelets
						11.4 M			
					Musc/skel	11.4			
					Hepatic	11.4			
					Renal	11.4			
					Dermal	11.4			
					Endocr	11.4			
					Immuno	3.78	11.4		Lymphoid hyperplasia in mediastinal lymph nodes
					Neuro	11.4			
					Repro	3.78 F	11.4 F		Increased estrous cycle length
							1.11 M	11.4 M	SLOAEL: Testicular atrophy; 3-fold increase in percent abnormal sperm LOAEL: Decreased sperm motility

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
					Other noncancer	11.4			
Exposure chamber analysis showed that under test conditions, the test substance converted to cobalt sulfate hexahydrate (Behl et al. 2015).									
NTP 1991, 2023									**Cobalt Sulfate Heptahydrate**
18	Mouse (B6C3F1) 5 M, 5 F	16 days 5 days/week 6 hours + 12 minutes (T90 time) per day (WB)	0, 0.035, 0.19, 1.80, 19.0, 75.7	BW, CS, GN, HP, LE, OW	Death			19	4/5 male and 1/5 female mice died
					Bd wt	1.8		19	Decrease in final body weight in males (33%) and females (20%)
					Resp	0.19		1.8	Inflammation and necrosis of respiratory epithelium (larynx, trachea, bronchioles, nasal turbinates) and degeneration of olfactory epithelium
					Cardio	75.7			
					Gastro	75.7			
					Musc/skel	75.7			
					Hepatic	75.7 F			
						1.8 M		19 M	Necrosis of hepatocytes (in mice that died)
					Renal	75.7			
					Dermal	75.7			
					Endocr	75.7			
					Immuno	1.8		19	Lymphoid depletion and necrosis of thymus (in mice that died); decreased absolute and relative thymus weight
					Neuro	1.8		19	Congestion of vessels in brain (in mice that died); hypoactivity
					Repro	75.7			
Exposure chamber analysis showed that under test conditions, the test substance converted to cobalt sulfate hexahydrate (Behl et al. 2015).									

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
NTP 2014									**Cobalt Metal**
19	Mouse (B6C3F1) 5 M, 5 F	17 days 5 days/week 6 hours + 12 minutes (T90 time) per day (WB)	0, 2.5, 5, 10, 20, 40	BW, CS, GN, HP, LE, OW, UR	Death			40	3/5 males and 3/5 females died
					Bd wt	10 F	20 F	40 F	SLOAEL: 38% decrease in final body weight LOAEL: 16% decrease in final body weight
						20 M		40 M	27% decrease in final body weight
					Resp		2.5	20	LOAEL: Minimal-to-mild nasal lesions (atrophy of olfactory epithelium; vacuolization of respiratory epithelium); minimal cytoplasmic vacuolization of bronchiolar epithelium with histiocytic infiltrates in males SLOAEL: Pulmonary fibrosis, olfactory epithelial necrosis, respiratory epithelial metaplasia, alveolar/bronchiolar karyomegaly
					Hepatic		2.5		Decreased absolute and relative liver weight
					Renal	20			
					Endocr	40			
					Immuno	40			
					Neuro	5 F	10 F		Lethargy
						10 M	20 M		Lethargy
NTP 2014									**Cobalt Metal**
20	Mouse (B6C3F1)	14 weeks 5 days/week		BC, BW, CS, GN, HE, HP,	Bd wt	5	10		13–14% decrease in final body weight

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
	10 M, 9–10 F	6 hours + 12 minutes (T90 time) per day (WB)	0, 0.625, 1.25, 2.5, 5, 10	LE, OW, RX, UR	Resp		0.625	5	LOAEL: Squamous metaplasia of the larynx; cytoplasmic vacuolization of bronchiole epithelium and alveolar histiocytic cellular infiltration SLOAEL: Pulmonary hemorrhage
					Cardio	10			
					Gastro	10			
					Hemato	5	10		Increased red blood cell count in both sexes, increased hemoglobin in males
					Musc/skel	10			
					Hepatic	1.25 F	2.5 F		Decreased absolute and relative liver weight
						5 M	10 M		Decreased absolute and relative liver weight
					Renal	2.5	5		Decreased absolute and relative kidney weight
					Dermal	10			
					Ocular	10			
					Endocr	10			
					Immuno	10			
					Neuro	10			
					Repro	5 F	10 F		Prolonged estrous cycle
						1.25 M	2.5 M		Decreased percent motile sperm
					Other noncancer	10			

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Johansson et al. 1987									**Cobalt Metal**
21	Rabbit (NS) 8 M	17 weeks, 5 days/week, 6 hours/day (WB)	0, 0.4, 2.0	CS, HP, OF, OW	Resp		0.4	2	LOAEL: Moderate lung inflammation and accumulation of macrophages SLOAEL: Severe lung inflammation and accumulation of macrophages; lung edema (lower lobe)
Johansson et al. 1991									**Cobalt Chloride**
22	Rabbit (NS) 8 M	4 months, 5 days/week, 6 hours/day (WB)	0, 0.5	BI, CS, GN, HP	Resp	0.5			
Johansson et al. 1992									**Cobalt Chloride**
23	Rabbit (NS) 8 M	4 months, 5 days/week, 6 hours/day (WB)	0, 0.6	BI, CS, GN, HP	Resp		0.6		Inflammatory lesions in the lung; increased cellularity of BALF, with decreased percent macrophages and increased percent monocytes
Kerfoot 1974									**Cobalt Metal**
24	Pig 5 NS	3 months, 5 days/week, 6 hour/day	0, 0.115, 0.991	CS, GN, HE, HP, UR	Bd wt		0.115		16% decrease in final body weight
					Resp		0.115		Decreased lung compliance (a metric of mechanical ventilation)
					Cardio		0.115		Increased heart rate, EKG changes (decreased QRS amplitude)
					Hemato	0.991			
					Hepatic	0.991			
					Renal	0.991			
					Immuno	0.991			
Palmes et al. 1959									**Cobalt Hydrocarbonyl**
25		3 months	0, 9		Bd wt	9			

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
	Guinea pig (NS) 6–32 M	5 days/week 6 hours/day (WB)		BW, CS, GN, HE, HP, LE	Hemato		9		Increased hemoglobin levels; decreased percent lymphocytes and increased percent basophils
Exposure was to cobalt hydrocarbonyl plus oxide/carbonate decomposition products due to instability of test substance in oxygen.									
CHRONIC EXPOSURE									
Deng et al. 1991									**Cobalt Metal**
26	Human 362–1,370 B	21 years (occupational)	0, 0.0175	CS	Resp	0.0175			
Kusaka et al. 1986a									**Cobalt Metal**
27	Human 34–68 M, 8–16 F	3 years (occupational)	0, 0.126	CS, OF	Resp		0.126		Decreased FEV_1
Nemery et al. 1992									**Cobalt Metal**
28	Human 212 M, 41 F	Current employees; duration of employment not reported (occupational)	0.0004, 0.0053, 0.0151	CS, OF, UR	Resp	0.0053[c]	0.0151		Decreased FEV_1 and FVC: increased cough, wheezing, and upper airway irritation
Prescott et al. 1992									**Cobalt Aluminate**
29	Human 34–36 F	14.6 years (occupational)	0.05	BI, CS, EA, OW	Endocr	0.05			
Swennen et al. 1993									**Hard Metal**
30	Human 82 M	8 years (occupational)	0, 0.125	BC, CS, HE, UR	Resp		0.125		Increased self-reported dyspnea and wheezing in smoking workers, compared to unexposed smokers, without changes in lung function tests
					Hemato		0.125		Decreased red blood cell counts, hemoglobin, and hematocrit levels; increase in white blood cell count
					Endocr		0.125		Decreased T3 levels

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Behl et al. 2015; Bucher et al. 1999, 2022; NTP 1998, 2023									**Cobalt Sulfate Heptahydrate**
31	Rat (F344/N) 50 M, 50 F	105 weeks 5 days/week 6 hours + 12 minutes (T90 time) per day (WB)	0, 0.12, 0.39, 1.11	BW, CS, GN, HP, LE	Bd wt	1.11			
					Resp			0.12	Hyperplasia and metaplasia of upper and lower respiratory tract tissues; pulmonary fibrosis; inflammatory changes in lungs
					Cardio	1.11			
					Gastro	1.11			
					Musc/skel	1.11			
					Hepatic	1.11			
					Renal	1.11			
					Dermal	1.11			
					Endocr	1.11			
					Immuno	1.11			
					Neuro	1.11			
					Repro	1.11			
					Cancer			0.39 F	CEL: Alveolar/bronchiolar neoplasms
								0.39 M	CEL: Benign, complex, or malignant pheochromocytoma
Exposure chamber analysis showed that under test conditions, the test substance converted to cobalt sulfate hexahydrate (Behl et al. 2015).									
Behl et al. 2015; NTP 2014									**Cobalt Metal**
32	Rat (F344/N) 50 M, 50 F	105 weeks 5 days/week 6 hours + 12 minutes (T90 time) per day (WB)	0, 1.25, 2.5, 5	BW, CS, GN, HP, LE	Death			2.5 F	22% decrease in survival
					Bd wt	1.25	2.5	5	LOAEL: Final body weights decreased by 11% in males and 16% in females SLOAEL: Final body weights decreased by 29% in males and 30% in females

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
					Resp			1.25	Pulmonary lesions (alveolar epithelium hyperplasia and proteinosis, bronchiole epithelium hyperplasia) and nasal lesions (hyperplasia, metaplasia, necrosis, and atrophy of the olfactory epithelium and nasal turbinate)
					Cardio	5			
					Gastro	5			
					Musc/skel	5			
					Hepatic	2.5 F	5 F		Basophilic foci
							1.25 M		Basophilic foci
					Renal	5			
					Dermal	5			
					Ocular	5			
					Endocr		1.25 F		Adrenal medullary hyperplasia
						5 M			
					Immuno	5			
					Neuro	5			
					Repro	5 F			
						2.5 M		5 M	Testicular infarct (complete effacement of parenchyma due to necrosis)
					Cancer			1.25	CEL: Alveolar/bronchiolar carcinoma in both sexes; mononuclear cell leukemia in females and bilateral benign pheochromocytoma in males

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Behl et al. 2015; Bucher et al. 1999, 2022; NTP 1998, 2023									**Cobalt Sulfate Heptahydrate**
33	Mouse (B6C3F1) 50 M, 50 F	105 weeks 5 days/week 6 hours + 12 minutes (T90 time) per day (WB)	0, 0.11, 0.39, 1.14	BW, CS, GN, HP, LE	Bd wt	1.14			
					Resp		0.11		Minimal squamous metaplasia of the larynx
					Cardio	1.14			
					Gastro	1.14			
					Musc/skel	1.14			
					Hepatic	1.14			
					Renal	1.14			
					Dermal	1.14			
					Endocr	1.14			
					Immuno	1.14			
					Neuro	1.14			
					Repro	1.14			
					Cancer			0.39 F	CEL: Alveolar/bronchiolar adenoma or carcinoma
								1.14 M	CEL: Alveolar/bronchiolar adenoma or carcinoma

Exposure chamber analysis showed that under test conditions, the test substance converted to cobalt sulfate hexahydrate (Behl et al. 2015).

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Behl et al. 2015; NTP 2014									**Cobalt Metal**
34	Mouse (B6C3F1) 50 M, 50 F	105 weeks 5 days/week 6 hours + 12 minutes (T90 time) per day (WB)	0, 1.25, 2.5, 5	BW, CS, GN, HP, LE	Death			2.5 M	20% decrease in survival
					Bd wt	2.5		5	Final body weights decreased by 24% in males and 29% in females
					Resp			1.25	Hyperplasia and cytoplasmic vacuolization of alveolar/bronchiolar epithelium; atrophy, hyperplasia, and metaplasia of olfactory epithelium; cytoplasmic vacuolization and squamous metaplasia of nasal respiratory epithelium; nasal turbinate atrophy
					Cardio	5			
					Gastro	5			
					Musc/skel	5			
					Hepatic	5			
					Renal	5			
					Dermal	5			
					Ocular	5			
					Endocr	5			
					Immuno	5			
					Neuro	5			
					Repro	5 F			
						2.5 M	5 M		Minimal to mild germinal epithelium degradation
					Cancer			1.25	CEL: Alveolar/bronchiolar carcinoma

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Wehner et al. 1977									**Cobalt Oxide**
35	Hamster (ENG:ELA) 51 M	Lifetime, 5 days/week, 7 hours/day	0, 7.9	BW, CS, LE, OF	Bd wt	7.9			
					Resp			7.9	Lung Inflammation and emphysema

[a]The number corresponds to entries in Figure 2-2; differences in levels of health effects and cancer effects between male and females are not indicated in Figure 2-2. Where such differences exist, only the levels of effect for the most sensitive sex are presented.
[b]Used to derive an acute-duration inhalation MRL of 0.0003 mg Co/m^3; the NOAEL was converted into a HEC of 0.01 mg Co/m^3 using MPPD modeling (see Appendix A for calculations) and divided by an uncertainty factor of 30 (3 for extrapolation from animals to humans after dosimetric adjustment and 10 for human variability).
[c]Used to derive a chronic-duration inhalation MRL of 0.0001 mg Co/m^3; concentration adjusted for intermittent exposure and divided by an uncertainty factor of 10 (for human variability).

B= both males and females; BALF = bronchoalveolar lavage fluid; BC = serum (blood) chemistry; Bd wt or BW = body weight; BI = biochemical changes; Cardio = cardiovascular; CEL = cancer effect level; Co = cobalt; CS = clinical signs; EKG = electrocardiogram; Endocr = endocrine; F = female(s); FEV_1 = forced expiratory volume in 1 second; FVC = forced vital capacity; Gastro = gastrointestinal; GN = gross necropsy; HE = hematological; HEC = human equivalent concentration; Hemato = hematological; HP = histopathology; Immuno = immunological; LC_{50} = lethal concentration, 50% kill; LDH = lactate dehydrogenase; LE = lethality; LOAEL = lowest-observed-adverse-effect level; M = male(s); MPPD = multiple-path particle dosimetry; MRL = minimal risk level; Musc/skel = musculoskeletal; (N) = nose-only; Neuro = neurological; NOAEL = no-observed-adverse-effect level; NS = not specified; OF = organ function; OW = organ weight; polycythemia = author-reported term associated with increased hemoglobin or erythrocyte count; Repro = reproductive; Resp = respiratory; RX = reproductive function; SLOAEL = serious LOAEL; T3 = triiodothyronine; T90 = the time required for the inhalation chamber concentration to reach 90% of the target concentration; TSH = thyroid-stimulating hormone; UR = urinalysis; (WB) = whole-body; WI = water intake

Figure 2-2. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)
Acute (≤14 days)

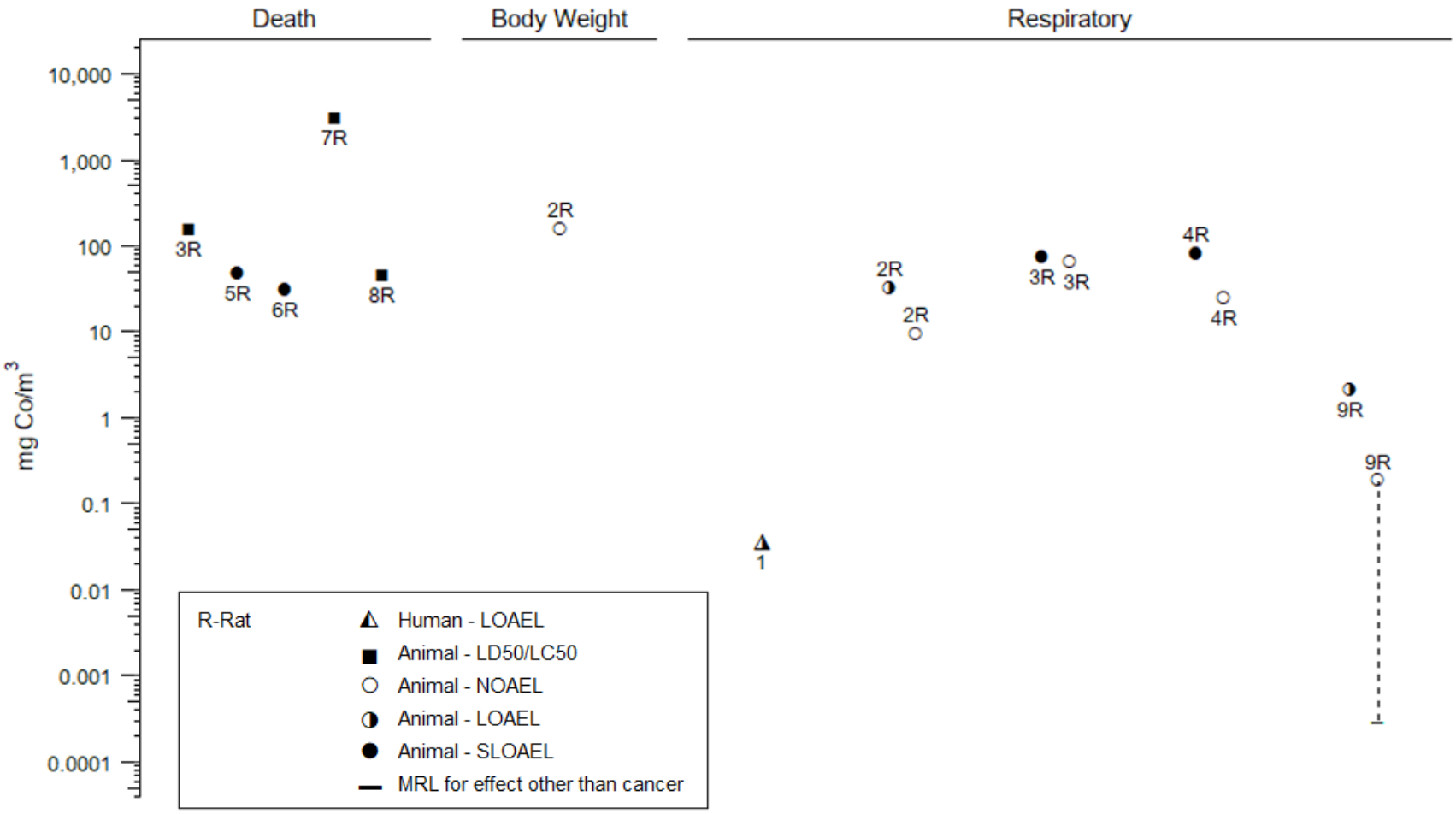


Figure 2-2. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)
Intermediate (15–364 days)

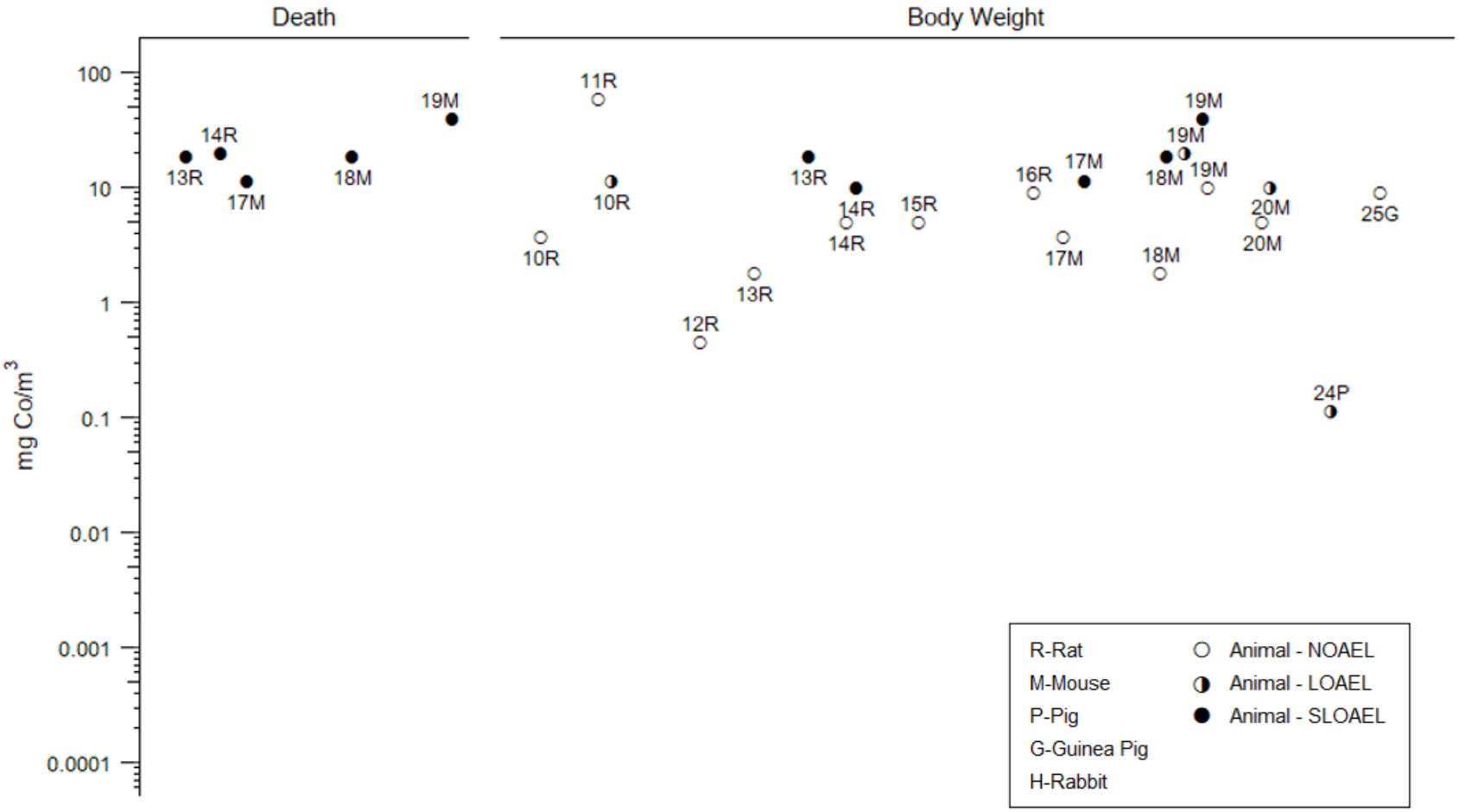


Figure 2-2. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)
Intermediate (15–364 days)

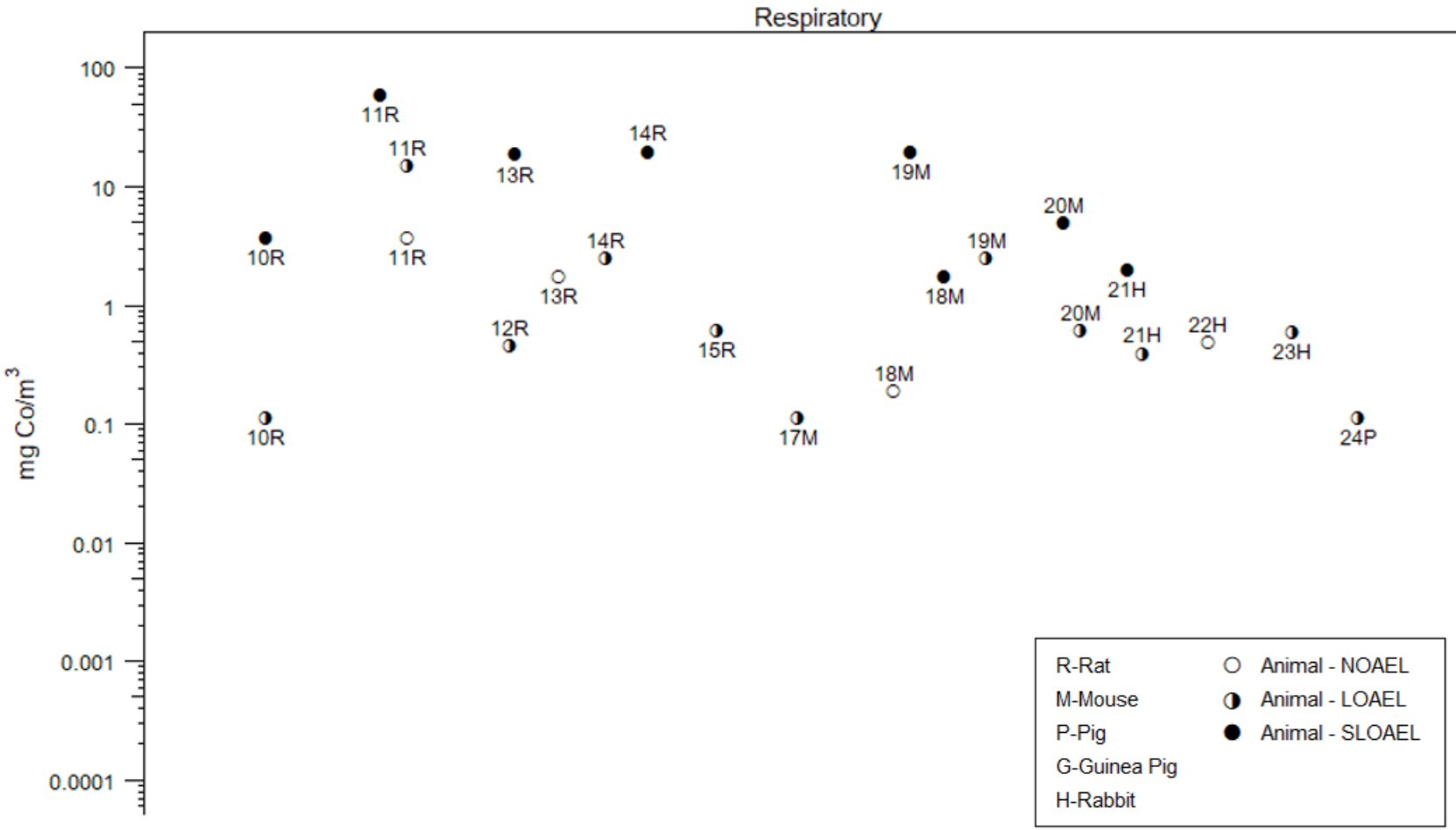


Figure 2-2. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)
Intermediate (15–364 days)

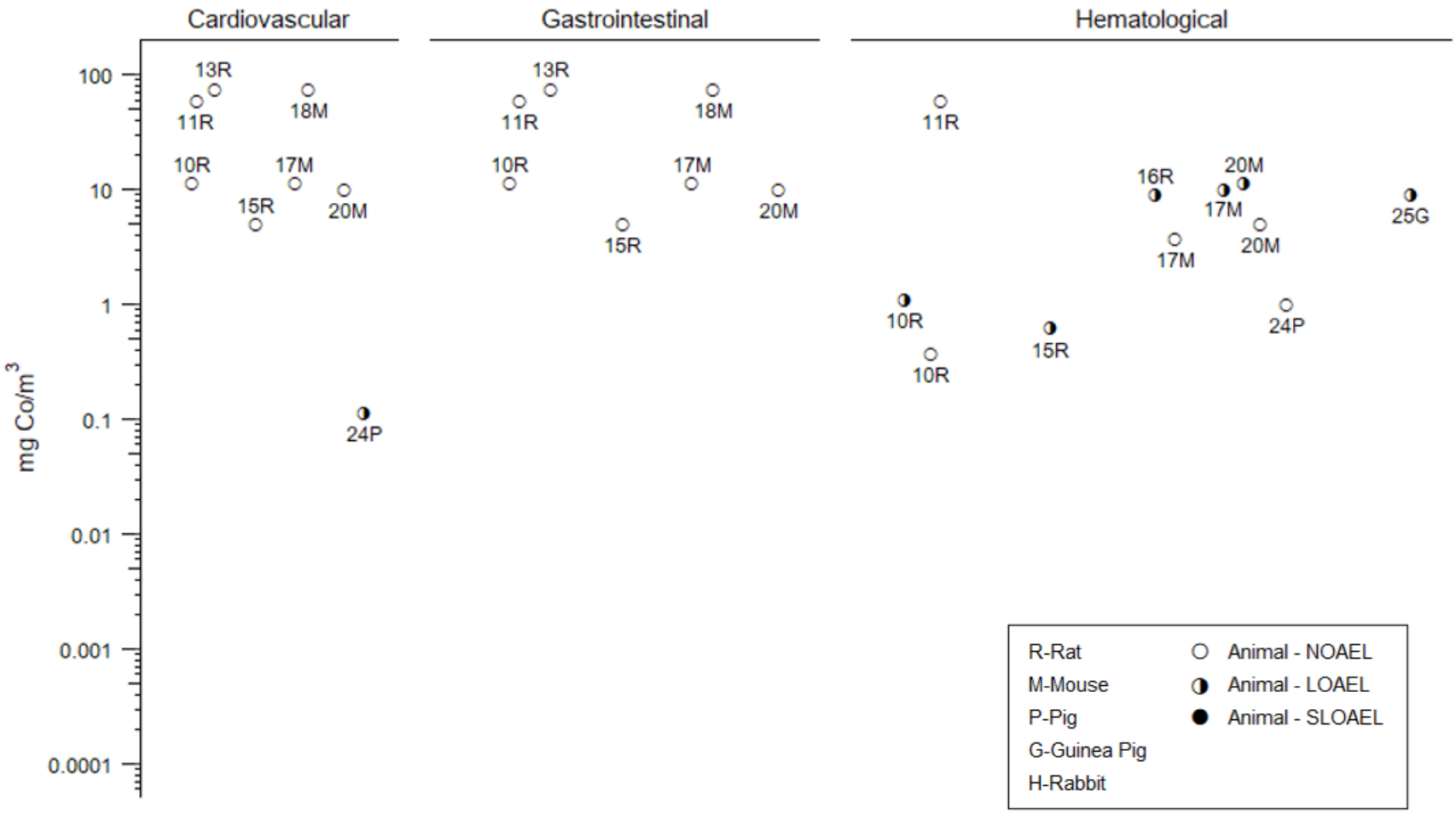


Figure 2-2. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)
Intermediate (15–364 days)

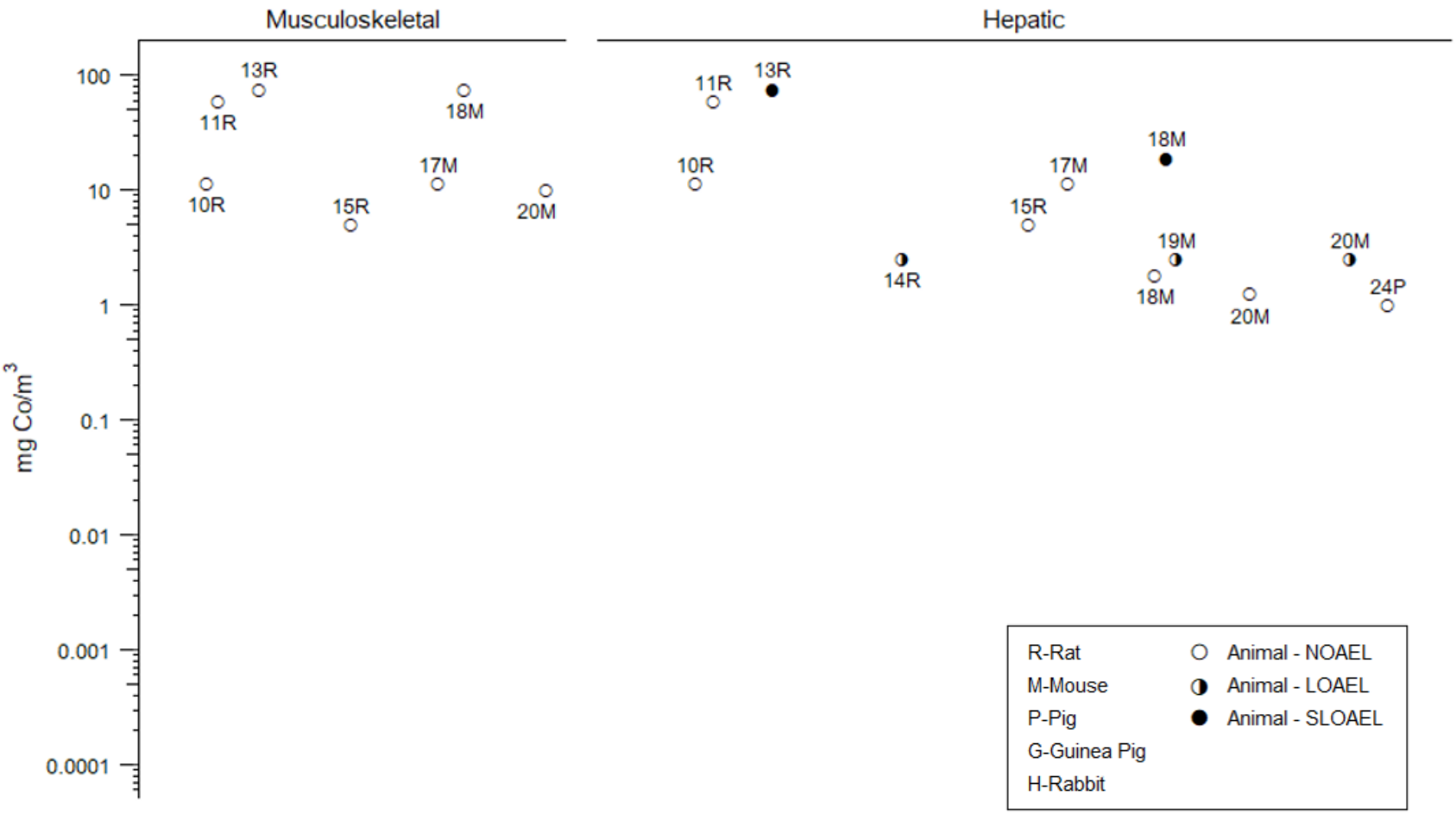


Figure 2-2. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)
Intermediate (15–364 days)

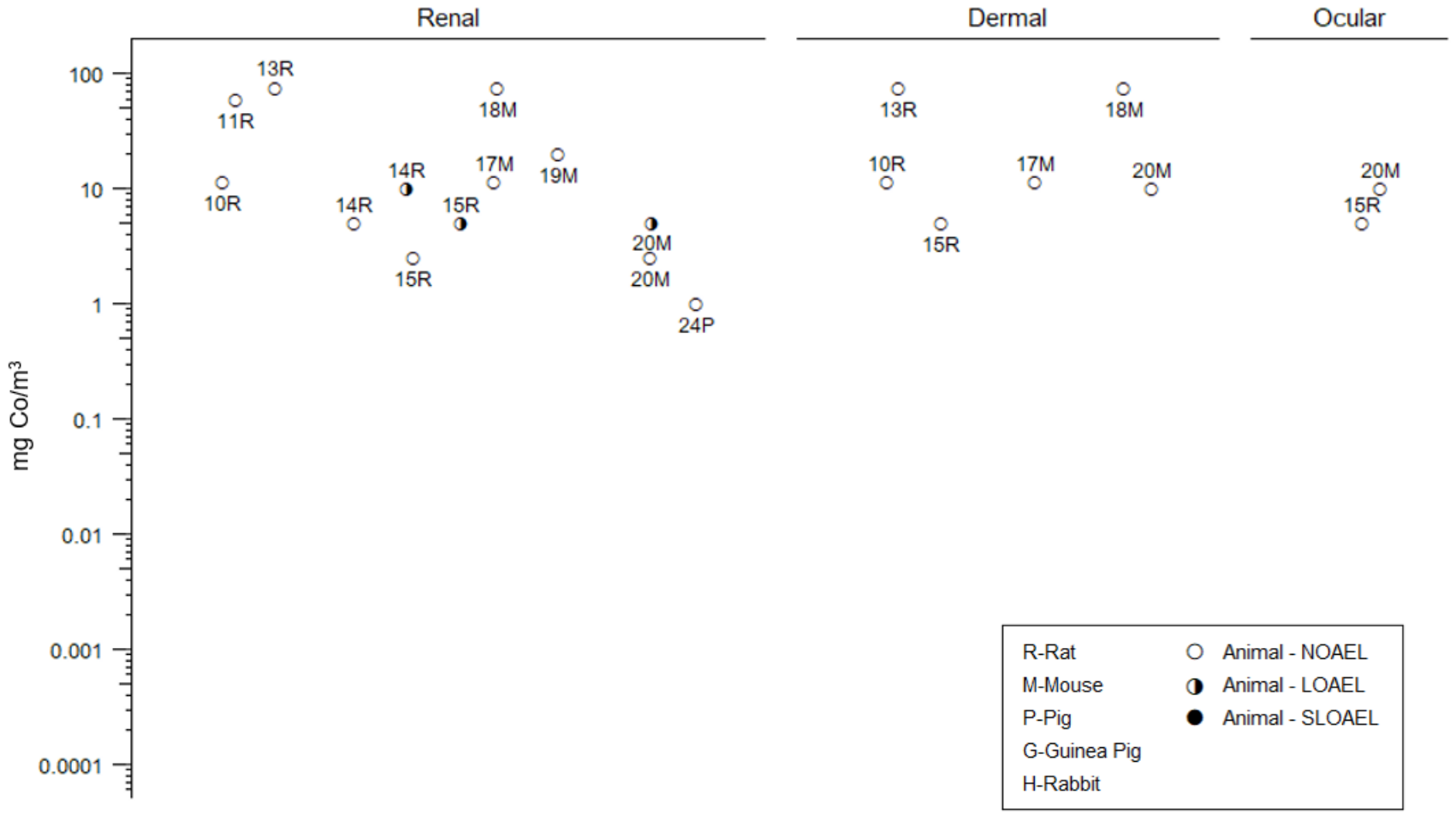


Figure 2-2. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)
Intermediate (15–364 days)

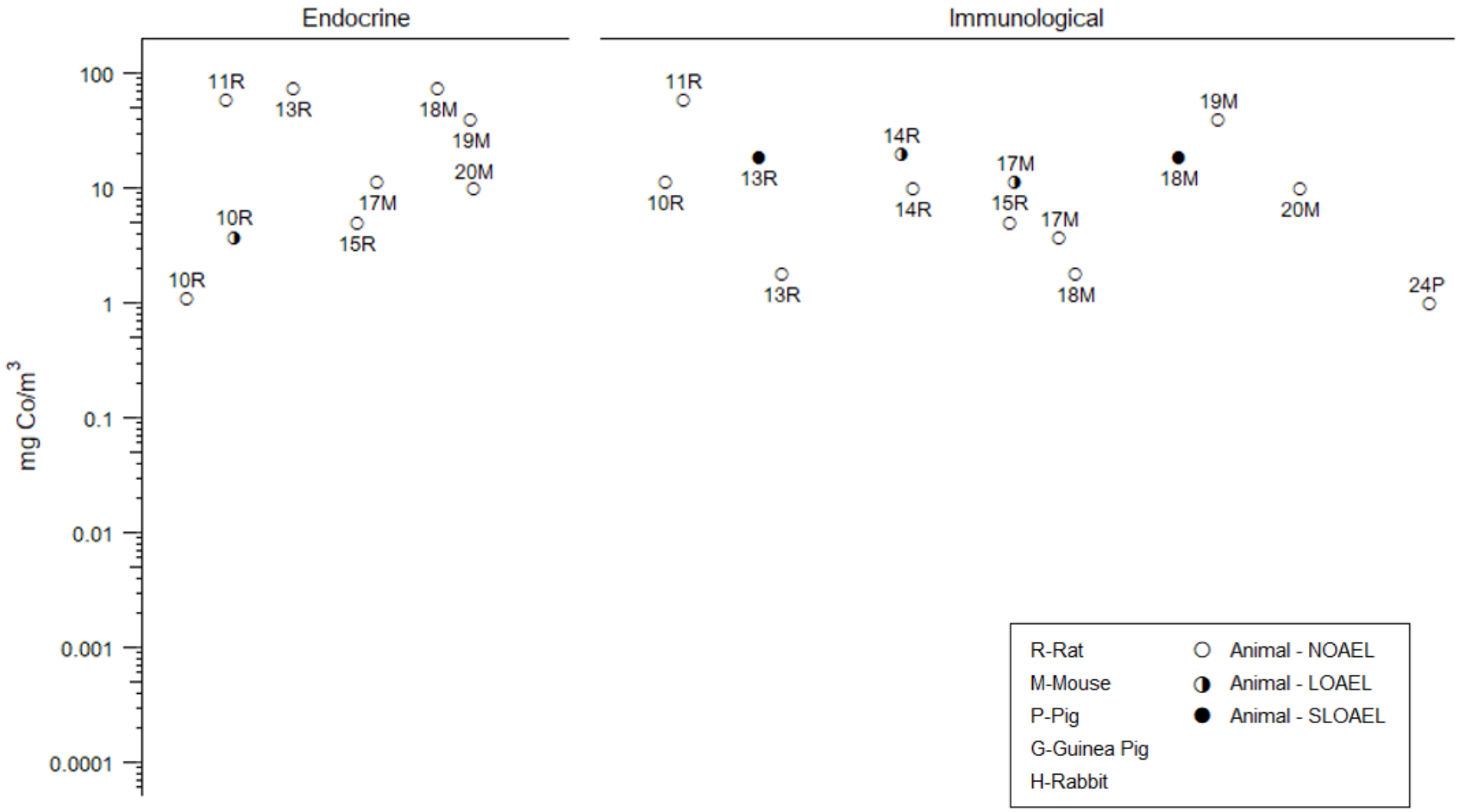


Figure 2-2. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)
Intermediate (15–364 days)

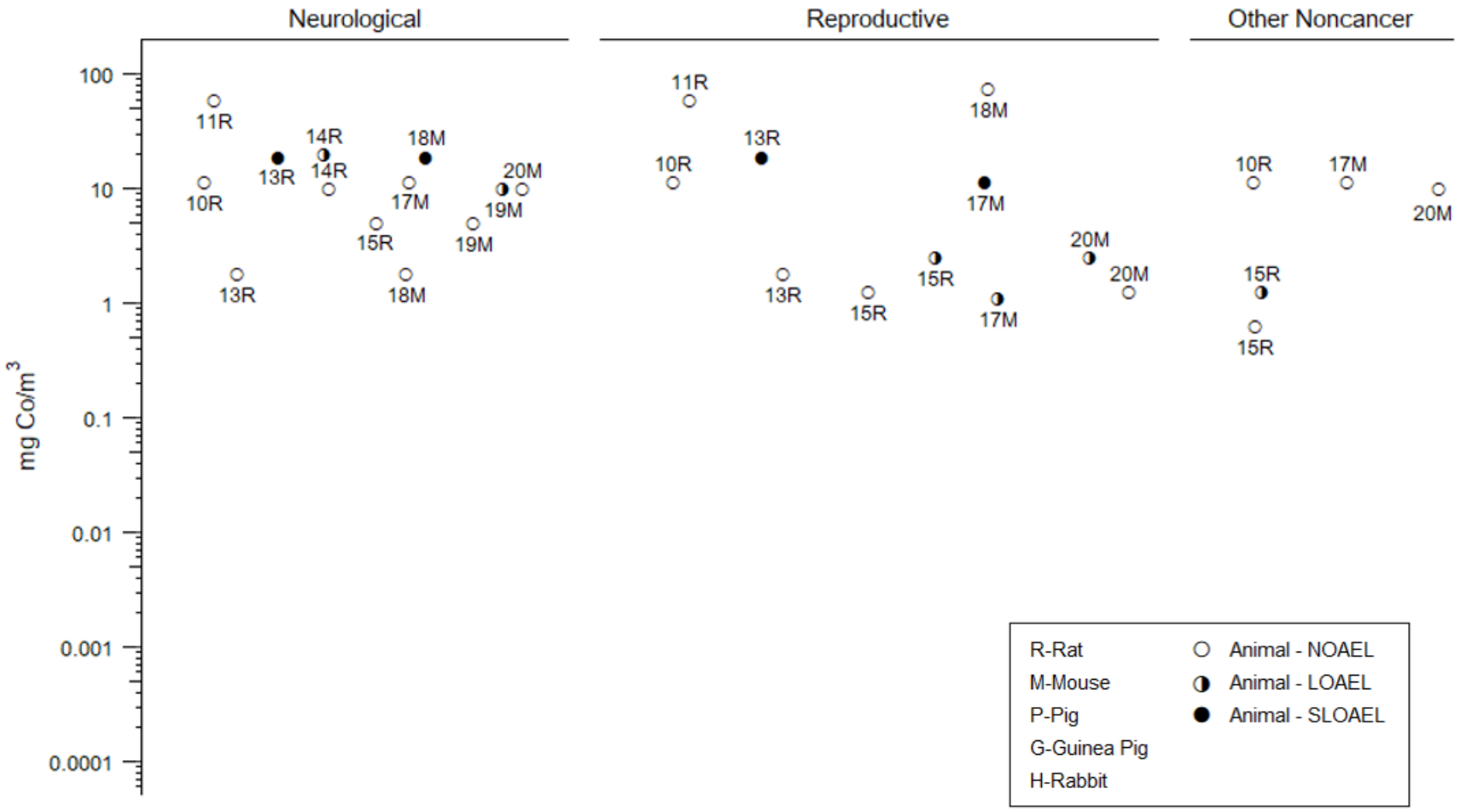


Figure 2-2. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)
Chronic (≥365 days)

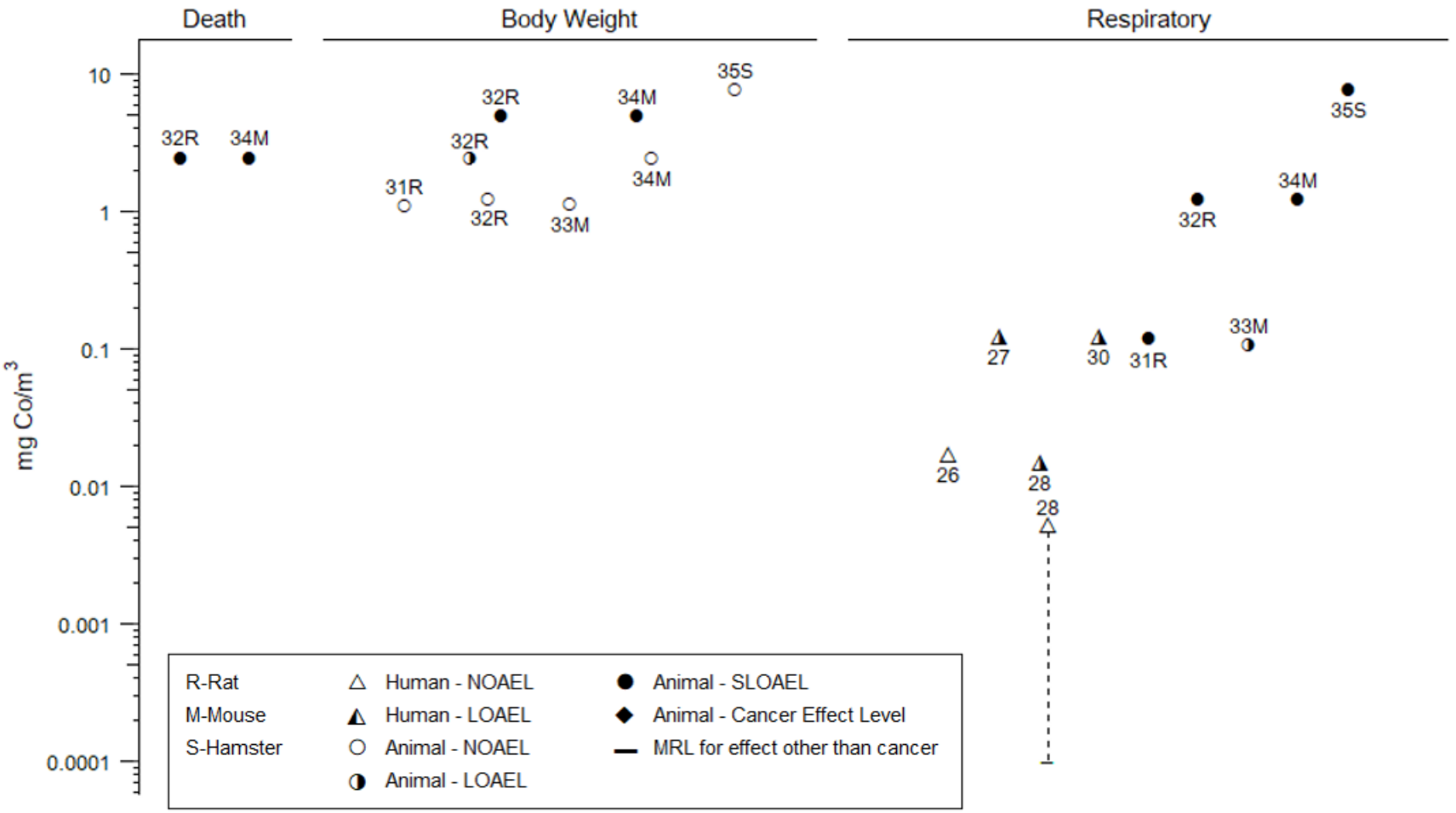


Figure 2-2. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)

Chronic (≥365 days)

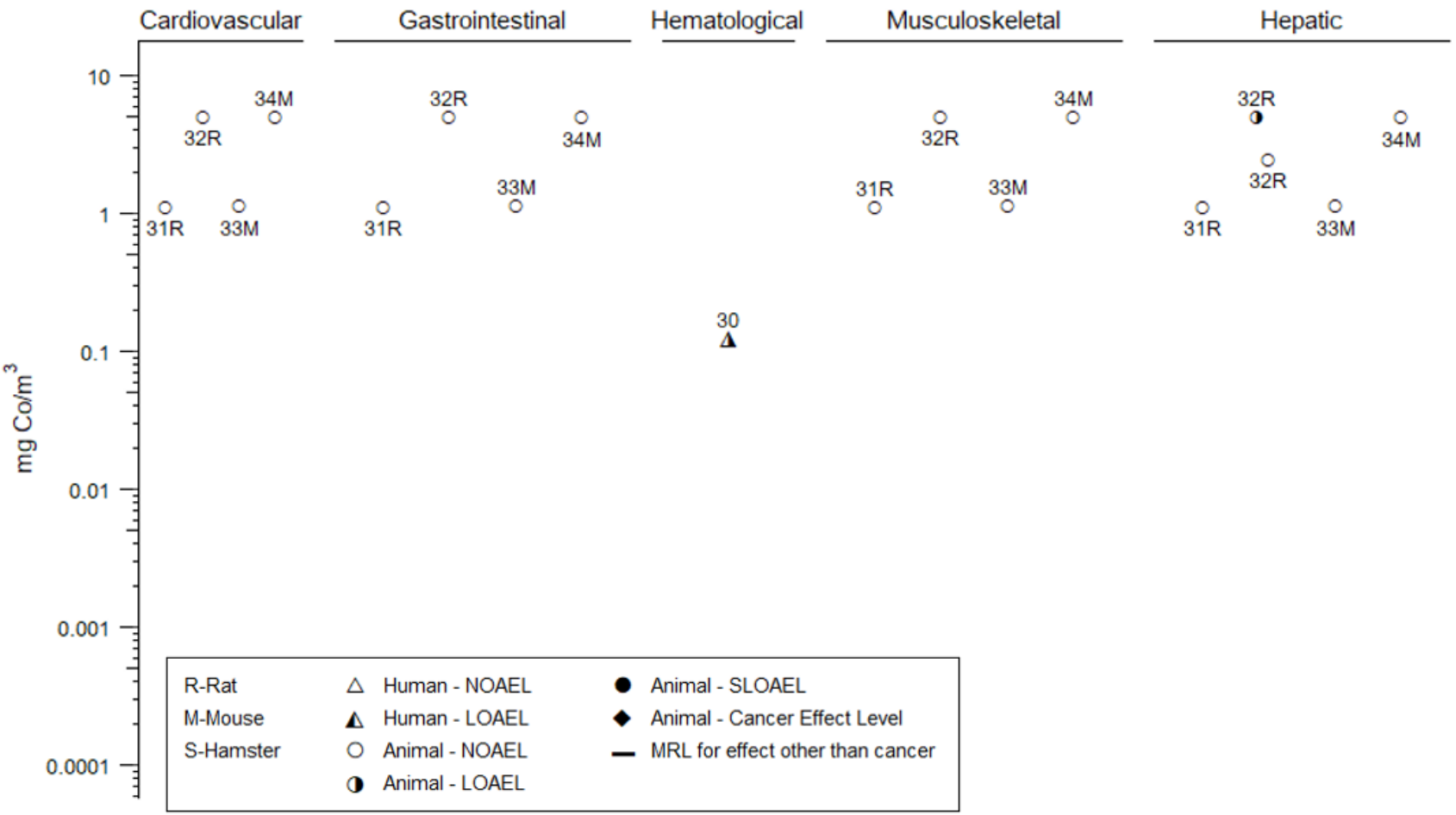


Figure 2-2. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)
Chronic (≥365 days)

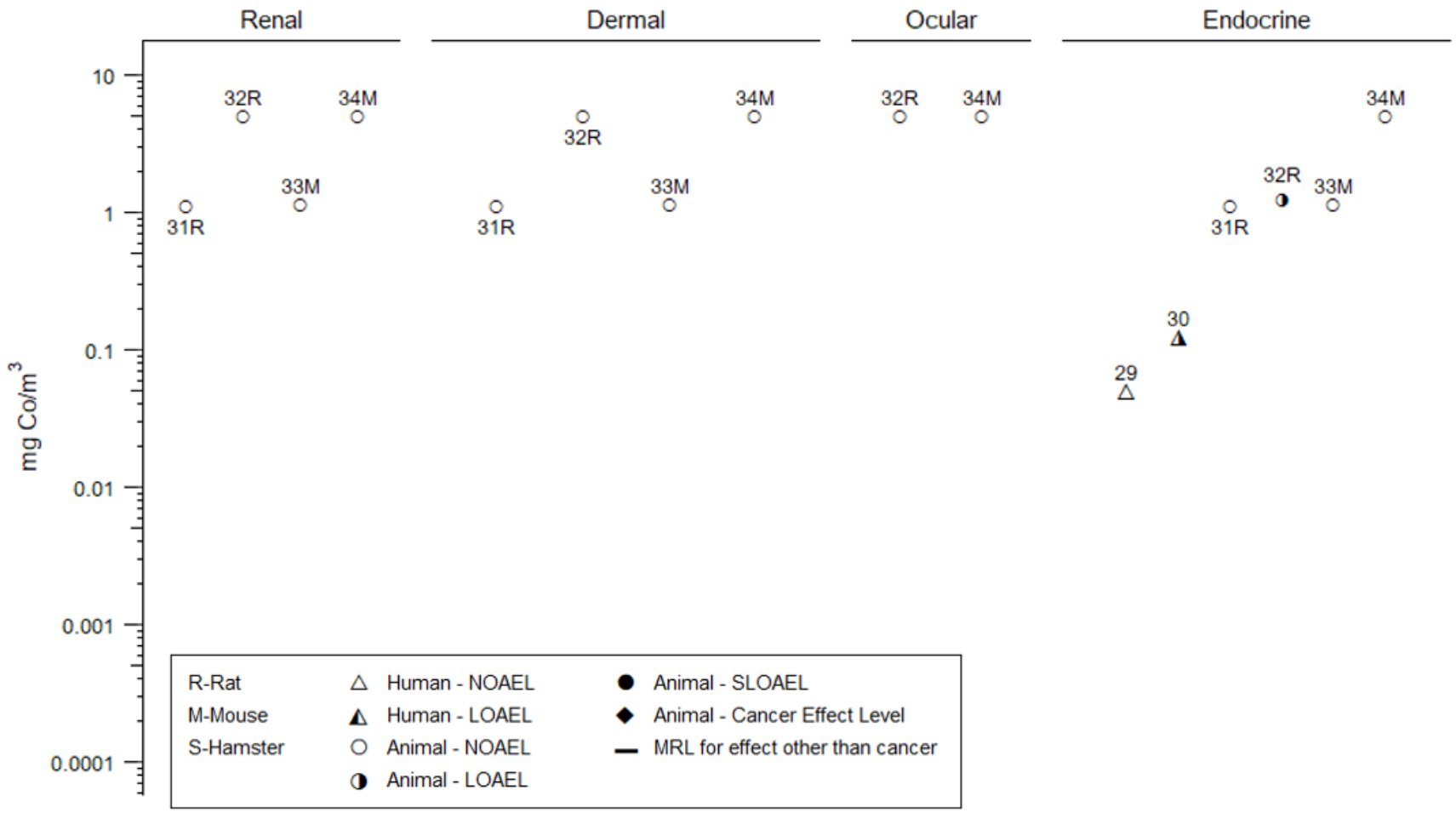
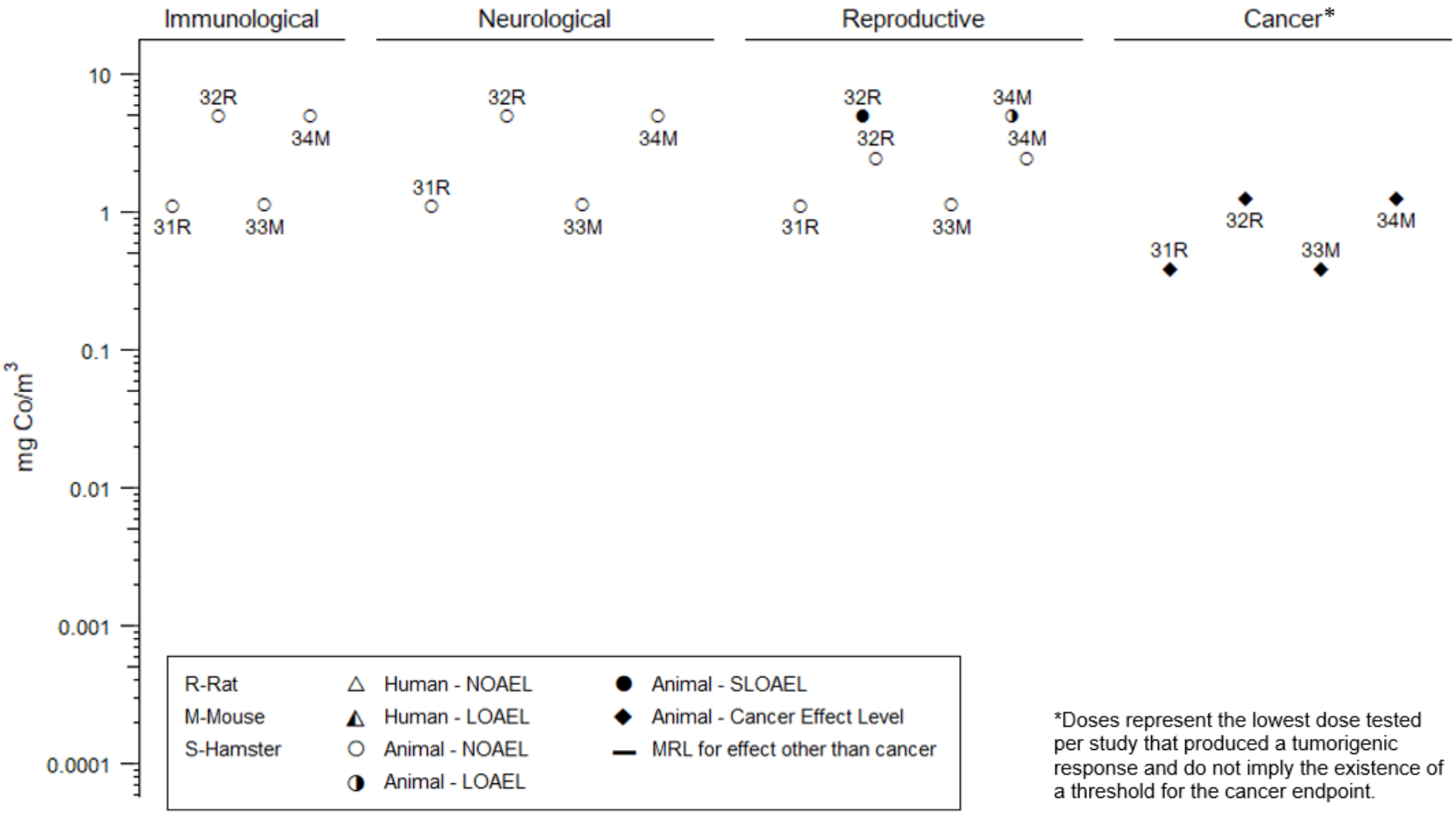


Figure 2-2. Levels of Significant Exposure to Cobalt – Inhalation (mg Co/m^3)
Chronic (≥365 days)



*Doses represent the lowest dose tested per study that produced a tumorigenic response and do not imply the existence of a threshold for the cancer endpoint.

Table 2-2. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
ACUTE EXPOSURE									
Davis and Fields 1958									**Cobalt Chloride**
1	Human 5 M	7–14 days (C)	0, 1	HE	Hemato		1[b]		Polycythemia (clinically high red blood cell levels)
Hoffmeister et al. 2018									**Cobalt (II)**
2	Human 8–16 M	7–14 days (C)	0, 0.03	HE	Hemato	0.03			
Paley et al. 1958									**Cobalt Chloride**
3	Human 3 M	10–14 days (C)	0, 0.54	BC, CS, OF	Gastro		0.54		Mild gastric distress
					Endocr		0.54		Impaired thyroid uptake of radioactive iodine-131
Roche and Layrisse 1956									**Cobalt Chloride**
4	Human 12 NS	2 weeks 7 days/week (C)	0, 1.0	BC, OF	Endocr		1		Impaired thyroid uptake of radioactive iodine-131
Ajibade et al. 2017									**Cobalt Chloride**
5	Rat (Wistar) 6 M	2 weeks 7 days/week (W)	0, 35	BC, BI, OF, HP	Cardio		35		Elevated systolic, diastolic, and mean arterial blood pressure; cellular infiltration and cardiac cell swelling
					Renal		35		Increased serum urea, inflammation of renal tissues
Akinrinde and Adebiyi 2019									**Cobalt (II) Chloride Hexahydrate**
6	Rat (Wistar) 12 M	7 days (GW)	0, 67.5	BC, BI, CS, NX	Immuno		67.5		Increased IL-1β and TNFα
					Neuro			67.5	Decreased motor strength, decreased activity and exploration in an open field; increased brain GFAP (reactive gliosis) and AChE activity

Table 2-2. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Akinrinde et al. 2016a									**Cobalt (II) Chloride Hexahydrate**
7	Rat (Wistar) 8 M	2 weeks, 7 days/week (W)	0, 10	BC, BI, GN, HP, OF	Cardio			10	Decreased systolic blood pressure; hemorrhagic lesions and congestion in the coronary blood vessels and infiltration of inflammatory cells in the myocardium and atrium
					Renal		10		Increased serum urea and creatinine; loss of normal renal cell morphology, loss of tubular and glomerular outlines, peritubular inflammatory cell infiltration
Akinrinde et al. 2016b									**Cobalt (II) Chloride Hexahydrate**
8	Rat (Wistar) 7 M	7 days (W)	0, 10	BI, HP, OF	Cardio			10	Decreased systolic, diastolic, and mean arterial blood pressure; Inflammation of the myocardium and areas of myocardial infarction
					Renal		10		Peritubular and perivascular inflammation; focal tubular necrosis
Akinrinde et al. 2016c									**Cobalt (II) Chloride Hexahydrate**
9	Rat (Wistar) 7 M	7 days (W)	0, 10	BC, BI, HP, OW	Gastro		10		Decreased relative small intestine weight; significant histopathological damage to intestine with depletion of absorptive epithelial cells
					Hepatic		10		Reduced relative liver weight; hepatocellular damage and areas of necrosis
					Immuno		10		Decreased serum TNFα and increased serum IL-1β
Awoyemi et al. 2017									**Cobalt (II) Chloride Hexahydrate**
10	Rat (Albino) 10 M	7 days (W)	0, 4.4, 8.9, 18	BC, BI, HP	Hepatic	4.4	8.9		Focal area of necrosis and congestion of vessels; mild infiltration by inflammatory cells

Table 2-2. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Corrier et al. 1985									**Cobalt (II) Chloride Hexahydrate**
11	Rat (Sprague-Dawley) 3 M	14 days daily (F)	0, 20	HP, RX	Repro	20			
Domingo and Llobet 1984									**Cobalt (II) Chloride Hexahydrate**
12	Rat (Sprague-Dawley) 20 M	Once (GW)	0, 161	BC, CS, HE, LE	Death			161	11/20 died
					Hemato		161		Increased hematocrit
					Hepatic	161			
					Renal		161		Increased serum urea
					Other noncancer		161		Decreased serum glucose
Domingo et al. 1985a									**Cobalt (II) Chloride Hexahydrate**
13	Rat (Sprague-Dawley) 20 M	Once (G)	0, 37, 61	LE	Death			37	10/20 died
Krasovskii and Fridlyand 1971									**Cobalt Chloride**
14	Rat (albino) NS	Once (GW)	Not reported	LE	Death			36	LD_{50}
Murdock 1959									**Cobalt Chloride**
15	Rat (NS) 10 M	Once (GW)	98, 122, 137, 150, 153, 191	LE	Death			144	LD_{50}
Oria et al. 2022									**Cobalt (II) Chloride Hexahydrate**
16	Rat (Wistar) 15 M	14 days (GW)	0, 9.9	BC, HP, NX	Immuno		9.9		Increased serum IL-1β and TNFα
					Neuro			9.9	Neurobehavioral effects (altered exploration, impaired memory, increased anxiety-like behaviors); ultrastructural changes in the hippocampus and amygdala; reactive gliosis

Table 2-2. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Oyagbemi et al. 2020									**Cobalt (II) Chloride Hexahydrate**
17	Rat (Wistar) 8 M	8 days (GW)	0, 37	BC, BI, OW, HP	Cardio			37	Histopathological changes to cardiac muscle (atrophy and patchy degeneration of myofibers, loss of striation); distension of interstitium
					Renal		37		Mild tubular atrophy, necrotizing nephritis, inflammatory cell infiltrate
Paternian and Domingo 1988									**Cobalt (II) Chloride Hexahydrate**
18	Rat (Sprague-Dawley) 20 F	10 days (GDs 6–15) (GW)	0, 6.2, 12.4, 24.8	BC, BW, DX, HE, OW	Bd wt			6.2	23% decrease in maternal body weight gain during gestation
					Hemato	12.4	24.8		Increased hematocrit, hemoglobin, and reticulocytes
					Develop	24.8			
Salami et al. 2023									**Cobalt Chloride**
19	Rat (Wistar) 5 M	8 days (G)	0, 11, 28, 68, 136	BW, OW, HP	Bd wt	28		68	Body weight loss (13% loss compared to starting weight; compared to 8% gain in controls)
					Gastro	28	68		Increased cryptal depth in small intestine, altered gut motility (increased gastric emptying time)
Shrivastava et al. 2008									**Cobalt (II) Chloride Hexahydrate**
20	Rat (Sprague-Dawley) 6 M	7 days (G)	0, 12.5	HE	Hemato		12.5		Increased hematocrit and hemoglobin levels
					Other noncancer		12.5		Elevated serum glucose levels
Shrivastava et al. 2010									**Cobalt (II) Chloride Hexahydrate**
21	Rat (Sprague-Dawley) 8 M	7 days (G)	0, 12.5	BC, BI, BW, FI, HE, HP, LE, NX, OP, OW, WI	Bd wt	12.5			
					Resp	12.5			
					Cardio	12.5			

Table 2-2. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
					Hemato		12.5		Increased red blood cell count, hematocrit, and hemoglobin; increased percent granulocytes and monocytes
					Hepatic	12.5			
					Renal	12.5			
					Ocular	12.5			
					Immuno	12.5			
					Neuro	12.5			
					Other noncancer		12.5		Elevated serum glucose levels
Singh and Junnarkar 1991									**Cobalt Chloride**
22	Rat (Wistar) 5 M, 5 F	Once (GW)	0, 7.8	CS, NX	Neuro		7.8		Moderate CNS depression (decreased spontaneous activity, muscle tone, touch response, respiratory rate, mild hypothermia; increased pentobarbitone-induced sleeping time)
Singh and Junnarkar 1991									**Cobalt Sulfate**
23	Rat (Wistar) 5 M, 5 F	Once (GW)	0, 35.2	CS, NX	Neuro		35.2		Mild CNS depression (decreased spontaneous activity, muscle tone, touch response, respiratory rate, mild hypothermia; increased pentobarbitone-induced sleeping time)
Singh and Junnarkar 1991									**Cobalt Chloride**
24	Rat (Wistar) 5 M, 5 F	Once (GW)	Not reported	LE	Death			77.6	LD_{50}
Singh and Junnarkar 1991									**Cobalt Sulfate**
25	Rat (Wistar) 5 M, 5 F	Once (GW)	Not reported	LE	Death			352	LD_{50}

Table 2-2. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Speijers et al. 1982									**Cobalt (II) Chloride Hexahydrate**
26	Rat (Wistar) 5 M, 5 F	Once (GW)	124, 149, 178, 214, 282	LE, CS	Death			190	LD_{50}
Speijers et al. 1982									**Cobalt Bromide**
27	Rat (Wistar) 5 M, 5 F	Once (GW)	53.9, 80.8, 121, 182, 272	LE, CS	Death			109	LD_{50}
Speijers et al. 1982									**Cobalt Sulfate Heptahydrate**
28	Rat (Wistar) 5 M, 5 F	Once (GW)	94.4, 142, 210, 315, 472	LE, CS	Death			161	LD_{50}
Speijers et al. 1982									**Cobalt fluoride**
29	Rat (Wistar) 5 M, 5 F	Once (G)	43, 68, 109, 176, 280	LE, CS, HP	Death			91	LD_{50}
Speijers et al. 1982									**Cobalt Nitrate Hexahydrate**
30	Rat (Wistar) 5 M, 5 F	Once (GW)	91.1, 137, 202, 304, 455	LE, CS	Death			140	LD_{50}
Speijers et al. 1982									**Cobalt Phosphate Octahydrate**
31	Rat (Wistar) 5 M, 5 F	Once (G)	104, 156, 234, 346, 519	LE, CS	Death			187	LD_{50}
Speijers et al. 1982									**Cobalt Oxide**
32	Rat (Wistar) 5 M, 5 F	Once (G)	157, 236, 354, 530, 794	LE, CS, HP	Death			159	LD_{50}
Speijers et al. 1982									**Cobalt Acetate Tetrahydrate**
33	Rat (Wistar) 5 M, 5 F	Once (GW)	59.1, 88.7, 132, 199, 298	LE, CS	Death			168	LD_{50}
Tanoğlu et al. 2022									**Cobalt (II) Chloride Hexahydrate**
34	Rat (Wistar) 6M	7 days (GW)	0, 37	BI, HP	Neuro			37	Moderate-to-severe sciatic nerve damage (degeneration of myelinated fibers; Schwann cell degeneration, perineurium disjunction)

Table 2-2. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Hassan et al. 2006									**Cobalt Chloride**
35	Mouse (NS) 3 M	5 days (NS)	0, 7.089, 14.18, 28.37	RX	Repro		7.089		Increased percent abnormal sperm
Krasovskii and Fridlyand 1971									**Cobalt Chloride**
36	Mouse (white) NS	Once (GW)	Not reported	LE	Death			36	LD50
Seidenberg et al. 1986									**Cobalt Chloride**
37	Mouse (ICR/SIM) 28 F	5 days (GDs 8–12) (GW)	0, 81	BW, CS, DX	Bd wt			81	32% decrease in maternal body weight gain
					Develop	81			
Singh and Junnarkar 1991									**Cobalt Sulfate**
38	Mouse (Swiss-Webster) 5 M	Once (GW)	0, 22.2	CS, NX	Neuro		22.2		Mild CNS excitation (elevated spontaneous activity and respiration)
Singh and Junnarkar 1991									**Cobalt Chloride**
39	Mouse (Swiss-Webster) 5 M	Once (GW)	0, 16	CS, NX	Neuro		16		Mild CNS depression (decreased spontaneous activity, touch response, muscle tone, and respiration)
Singh and Junnarkar 1991									**Cobalt Chloride**
40	Mouse (Swiss-Webster) 5 M	Once (GW)	Not reported	LE	Death			163	LD_{50}
Singh and Junnarkar 1991									**Cobalt Sulfate**
41	Mouse (Swiss-Webster) 5 M	Once (GW)	Not reported	LE	Death			222	LD_{50}

Table 2-2. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Krasovskii and Fridlyand 1971									**Cobalt Chloride**
42	Guinea pig (NS) NS	Once (GW)	Not reported	LE	Death			25	LD_{50}
INTERMEDIATE EXPOSURE									
Duckham and Lee 1976									**Cobalt Chloride**
43	Human 6 M, 6 F	12 weeks, 7 days/week (C)	0, 0.36	BC, CS, OF	Gastro		0.36 F		Nausea and constipation
Finley et al. 2013									**Cobalt Chloride**
44	Human 5 M, 5 F	31 days (IN)	0, 0.013	BC, CS, HE, IX	Hemato	0.013			
					Hepatic	0.013			
					Renal	0.013			
					Endocr	0.013			
					Immuno	0.013			
Hoffmeister et al. 2018									**Cobalt (II)**
45	Human 8–16 M	21 days (C)	0, 0.03	HE	Hemato	0.03			
Holly 1955									**Cobalt Chloride**
46	Human 20–55 F	13 weeks 7 days/week (C)	0, 0.57	BC, CS, DX, HE, UR	Gastro		0.57		Gastric intolerance
					Hepatic	0.57			
					Endocr	0.57			
					Develop	0.57			
Tvermoes et al. 2014									**Cobalt Chloride**
47	Human 5 M, 5 F	88–91 days daily (IN)	0, 0.013	BC, CS, HE, NX, OF	Cardio	0.013			
					Hemato	0.013			
					Hepatic	0.013			
					Renal	0.013			
					Endocr	0.013			
					Immuno	0.013			

Table 2-2. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Abdel-Daim et al. 2020									**Cobalt (II) Chloride Hexahydrate**
48	Rat (Sprague-Dawley) 10 M	4 weeks daily (W)	0, 8.9	BC, BI, BW, CS, LE, OW	Bd wt			8.9	192% decrease in body weight gain (11 g body weight loss compared to 12 g body weight gain in controls)
					Renal		8.9		Increased serum urea and creatinine
Bourg et al. 1985									**Cobalt Chloride**
49	Rat (Sprague-Dawley) 8 M	57 days daily (W)	0, 20.3	BW, NX, WI	Bd wt	20.3			
					Neuro		20.3		Enhanced avoidance retention in passive-avoidance testing (suggestive of decreased stress tolerance)
Chetty et al. 1979									**Cobalt Chloride**
50	Rat (Sprague-Dawley) 8–15 M	30 days daily (F)	0, 0.45, 2.2, 4.53, 8.99, 13.8	BI, BW, HE, IX, OW	Hemato	8.99	13.8		Decreased hemoglobin
					Immuno	2.2	4.53		Immune suppression (decreased immune response to sheep red blood cells)
Clyne et al. 1988									**Cobalt Sulfate Heptahydrate**
51	Rat (Sprague-Dawley) 5 M	8 weeks, 7 days/week (F)	0, 4.2	BW	Bd wt			4.2	33% decrease in body weight gain
Corrier et al. 1985									**Cobalt (II) Chloride Hexahydrate**
52	Rat (Sprague-Dawley) 3 M	98 days daily (F)	0, 20	HE, HP, RX	Hemato		20		Increased red blood cell count, hemoglobin level, and packed cell volume
					Repro			20	Marked degeneration and necrosis of germinal epithelium of seminiferous tubules; 43% decrease in spermatid reserve

Table 2-2. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Danzeisen et al. 2020a									**Cobalt (II) Chloride Hexahydrate**
53	Rat (Sprague-Dawley) 10 M, 10 F	90 days daily (G)	0, 0.74, 2.48, 7.44	BW, FI, HE, HP, LE, NX, OP, OW, RX, UR, WI	Bd wt	2.48	7.44		Decrease in body weight (11% in males at the end of exposure; 17% in males and 13% in females at the end of 4-week recovery period)
					Resp	7.44			
					Cardio	7.44			
					Gastro	7.44			
					Hemato	0.74[c]	2.48		Increased red blood cell count, hemoglobin, and hematocrit in males; erythroid hyperplasia in the bone marrow in both sexes; $BMDL_{1SD}$ for increased red blood cells in males=1.95 mg Co/kg/day[c]
					Musc/skel	7.44			
					Hepatic	7.44			
					Renal	7.44			
					Dermal	7.44			
					Ocular	7.44			
					Endocr	7.44			
					Immuno	7.44			
					Neuro	7.44			
					Repro	7.44			
Danzeisen et al. 2020a									**Cobalt Tetraoxide**
54	Rat (Sprague-Dawley) 10 M, 10 F	90 days daily (G)	0, 73.4, 220, 734	BW, FI, HE, HP, LE, NX, OP, OW, RX, UR, WI	Resp	734			
					Cardio	734			
					Gastro	734			
					Hemato	220 F	734 F		Increased red blood cell count, hemoglobin, and hematocrit
						73.4 M	220 M		Increased red blood cell count, hemoglobin, and hematocrit

Table 2-2. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
					Musc/skel	734			
					Hepatic	734			
					Renal	734			
					Dermal	734			
					Ocular	734			
					Endocr	734			
					Immuno	734			
					Neuro	734			
					Repro	734			
Danzeisen et al. 2020a									**Cobalt Sulfide**
55	Rat (Sprague-Dawley) 10 M, 10 F	One generation (2 weeks premating–PND 3) daily (G)	0, 64.8, 194, 648	BW, DX, FI, RX	Bd wt	648			
					Neuro	648			
					Repro	648			
					Develop	648			
Danzeisen et al. 2020a									**Cobalt Tetraoxide**
56	Rat (Sprague-Dawley) 10 M, 10 F	One generation (2 weeks premating–PND 3) daily (G)	0, 73.4, 220, 734	BW, DX, FI, RX	Bd wt	734			
					Neuro	734			
					Repro	734			
					Develop	220		734	Decreased pup weight at birth (18%) and on PND 4 (21%)
Domingo et al. 1984									**Cobalt (II) Chloride Hexahydrate**
57	Rat (Sprague-Dawley) 20 M	13 weeks 7 days/week (W)	0, 16.5	BC, CS, FI, GN, HE, HP, OW, UR, WI	Resp	16.5			
					Cardio	16.5			
					Gastro	16.5			
					Hemato		16.5		Increased hematocrit and hemoglobin
					Musc/skel	16.5			
					Hepatic	16.5			

Table 2-2. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
					Renal	16.5			
					Endocr	16.5			
					Immuno	16.5			
					Repro		16.5		Decreased relative testicular weight
Domingo et al. 1985b									**Cobalt Chloride**
58	Rat (Wistar) 15 F	28 days GD 14–PND 21 (G)	0, 5.4, 11, 22	DX	Develop			5.4	PNDs 1–21: Decreased pup weight in males (17-29%) and females (12–28%); decreased pup length in males (3–9%) and females (6%)
Garoui et al. 2011									**Cobalt Chloride**
59	Rat (Wistar) 6 F	28 days (GD 14–PND 14); daily (W)	0, 20	BC, BI, BW, DX, FI, GN, LE, OW, WI	Bd wt	20			
					Hepatic		20		Increased absolute liver weight; hepatic injury (infiltration of mononuclear cells and vascular congestion)
					Develop			20	Effects at PND 14: 40% decrease in body weight; hepatic damage (elevated plasma ALT, AST; infiltration of mononuclear cells and vascular congestion); decreased plasma glucose
					Other noncancer		20		Decreased plasma glucose levels; decreased maternal food intake
Garoui et al. 2012									**Cobalt Chloride**
60	Rat (Wistar) 6 F	28 days (GD 14–PND 14); daily (W)	0, 20	BI, BW, CS, DX, FI, GN, HP, LE, OW, UR, WI	Bd wt	20			
					Renal		20		Vascular congestion, reduction of glomerular space, and infiltration of leukocyte cells between tubules; increased plasma urea, decreased urinary creatinine and urea, and decreased relative kidney weight

Table 2-2. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
					Develop			20	Effects at PND 14: 40% decrease in body weight; renal damage (elevated plasma creatinine; decreased urinary creatinine and urea; vascular congestion and reduction of glomerular space)
					Other noncancer		20		Decreased maternal food intake
Garoui et al. 2013									**Cobalt Chloride**
61	Rat (Wistar) 6 F	28 days (GD 14–PND 14); daily (W)	0, 20	DX, LE	Develop			20	Findings at PND 14: Reduced AChE and BuChE levels in cerebrum and cerebellum; altered development of cerebellar architecture (poorly differentiated layers with frequent pyknotic cells)
Grice et al. 1969									**Cobalt Sulfate**
62	Rat (Wistar) 10 M	8 weeks 7 days/week (GW)	26	LE, HP	Death			26	50% mortality
					Cardio			26	Degenerative myocardial heart lesions
Haga et al. 1996									**Cobalt Sulfate Heptahydrate**
63	Rat (Sprague-Dawley) 10 M	24 weeks 7 days/week (F)	0, 8.4	BW, OF, OW	Bd wt			8.4	31% decrease in final body weight
					Cardio			8.4	Impaired left ventricular function
Haga et al. 1996									**Cobalt Sulfate Heptahydrate**
64	Rat (Sprague-Dawley) 8 M	16 weeks 7 days/week (F)	0, 8.4	BW, OF, OW	Bd wt			8.4	26% decrease in final body weight
					Cardio	8.4			
Holly 1955									**Cobalt Chloride**
65	Rat (Wistar) 3–8 M	4 months 7 days/week (G)	0, 18	HE, HP	Resp	18			
					Cardio	18			
					Gastro	18			

Table 2-2. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
					Hemato		18		Increased red blood cell count and hemoglobin levels
					Hepatic	18			
					Renal	18			
					Endocr	18			
					Immuno	18			
					Neuro	18			
Khalil et al. 2020									**Cobalt (II) Chloride Hexahydrate**
66	Rat (Sprague-Dawley) 8 M	4 weeks 7 days/week (W)	0, 68	BC, CS, LE	Hepatic		68		Increased serum AST, ALT, ALP, LDH, and total bilirubin
Krasovskii and Fridlyand 1971									**Cobalt Chloride**
67	Rat (NS) NS	7 months, 6 days/week (GW)	0, 0.05, 0.5, 2.5	HE, OF, IX, NX	Hepatic	2.5			
					Neuro	0.5	2.5		Learning impairment (altered operant behavior)
Mathur et al. 2011									**Cobalt (II) Chloride Hexahydrate**
68	Rat (Wistar) 8 M	60 days daily (GW)	0, 25	BC, BI, BW, HP, OW	Bd wt	25			
					Hepatic		25		Increased relative liver weight; increased AST and bilirubin; qualitatively reported changes in liver cells (altered morphology and atrophy)
Mohamed et al. 2019									**Anhydrous Cobalt Chloride**
69	Rat (Wistar) 10 M	60 days daily (NS)	0, 27	BI, CS, HP, LE	Death			27	4/10 rats died
					Neuro			27	Decreased neurotransmitter levels in the brain (serotonin, norepinephrine, dopamine, GABA); encephalopathy; increased GFAP (reactive gliosis)

Table 2-2. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Mollenhauer et al. 1985									**Cobalt Metal**
70	Rat (Sprague-Dawley) 3 M	98 days daily (F)	0, 20	HP	Repro		20		Thickening of basal lamina and collagen layer surrounding the seminiferous tubules and vessel walls; ultrastructural damage to testicular cells
Morvai et al. 1993									**Cobalt Chloride**
71	Rat (CFY) 8 M	3 weeks 7 days/week (G)	0, 22	BW, HP, OF, OW	Bd wt	22			
					Cardio			22	Decrease in cardiac output and arterial blood pressure; multifocal myocytolysis and myofibril degeneration
Murdock 1959									**Cobalt Chloride**
72	Rat (NS) 6–30 M	150 days 5 days/week (GW)	0, 10	HE	Hemato		10		Increased red blood cell count, hemoglobin, and hematocrit
Nation et al. 1983									**Cobalt Chloride**
73	Rat (Sprague-Dawley) 6 M	69 days daily (F)	0, 5, 20	CS, GN, OW, NX	Neuro	5	20		Impaired operant conditioning
					Repro	5		20	Testicular atrophy, decreased testicular weight
Pehrsson et al. 1991									**Cobalt Sulfate Heptahydrate**
74	Rat (Sprague-Dawley) 12 M	8 weeks 7 days/week (F)	0, 8.4	BW, OF	Bd wt			8.4	30% decrease in final body weight
					Cardio	8.4			
Saker et al. 1998									**Cobalt Chloride**
75	Rat (Sprague-Dawley) 6 M	12–16 days daily (W)	0, 18	BC, BI, BW, CS	Bd wt	18			
					Other noncancer	18			

Table 2-2. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Stanley et al. 1947									**Cobalt (II) Chloride Hexahydrate**
76	Rat (Sprague-Dawley) 4–6 NS	8 weeks 7 days/week (C)	0, 0.62, 2.5, 9.9	BW, CS, HE	Bd wt	9.9			
					Hemato	0.62	2.5		Increased red blood cell count and hemoglobin
Umar et al. 2016									**Cobalt Chloride**
77	Rat (Wistar) 5 B	28 days daily (NS)	0, 23	CS, NX	Neuro	23			
Anderson et al. 1992									**Cobalt (II) Chloride Hexahydrate**
78	Mouse (CD-1) 10 M	7–13 weeks 7 days/week (W)	0, 43.4	GN, HP, OW	Hepatic	43.4			
					Renal	43.4			
					Repro			43.4	Decreased absolute testicular weight at ≥9 weeks; damage to seminiferous tubules (mild at 9 weeks progressing to extensive Sertoli and germ cell loss at ≥11 weeks); testicular atrophy at ≥11 weeks
Anderson et al. 1993									**Cobalt (II) Chloride Hexahydrate**
79	Mouse (CD-1) 10 M	13 weeks 7 days/week (W)	0, 43.4	BW, HP, OW	Bd wt	43.4			
					Repro			43.4	Decreased absolute testes weight; seminiferous tubule damage and degeneration and hypercellularity of the interstitial areas
Elbetieha et al. 2008									**Cobalt (II) Chloride Hexahydrate**
80	Mouse (Swiss) 10 M	12 weeks 7 days/week (W)	0, 6.354, 11.62, 23.01	BW, CS, HP, LE, OW, RX, WI	Bd wt	23.01			
					Repro			6.354	Impaired male fertility (deceased viable pregnancies when mated to unexposed females), decreased epididymal sperm count

Table 2-2. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Gluhcheva et al. 2020									**Cobalt (II) Chloride Hexahydrate**
81	Mouse (ICR) 7–8 B	20–21 days; GD 19 or 20–PND 18 (W)	0, 19	DX	Develop			19	Effects on PND 18: 17% decrease in body weight; elevated red blood cell count; decreased relative spleen and kidney weights; histopathological changes in spleen, liver, and kidney
Huy et al. 2022									**Cobalt Chloride**
82	Mouse (B6C3F1) 5 F	17 days (GW)	0, 9, 18	BC, OW, IX	Immuno	18			
Legostaeva et al. 2013; Zaksas et al. 2013									**Cobalt (II) Chloride Hexahydrate**
83	Mouse (BALB/c) 8 M, 8 F	62-63 days; GD 19 or 20–PND 25 (via dam) + direct for 35 days postweaning (W)	0, 31	BC, BW, IX	Bd wt			31	33% decrease in body weight on PND 60
					Immuno		31		Decreased plasma IgG on PND 60
Pedigo and Vernon 1993									**Cobalt (II) Chloride Hexahydrate**
84	Mouse (B6C3F1) 10 M	10 weeks 7 days/week (W)	0, 58.9	BW, CS, LE, RX	Repro			58.9	Decreased fertility; decreased sperm concentration and motility
Pedigo et al. 1988									**Cobalt (II) Chloride Hexahydrate**
85	Mouse (CD-1) 5 M	7–13 weeks 7 days/week (W)	0, 58.9	HE, OW, RX, WI	Hemato	58.9			
					Repro			58.9	Decreased fertility at 13 weeks; decreased epididymal sperm concentration and percent motile sperm at ≥11 weeks; decreased absolute and relative testes weight at ≥9 weeks

Table 2-2. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Pedigo et al. 1988									**Cobalt (II) Chloride Hexahydrate**
86	Mouse (CD-1) 5 M	12 weeks 7 days/week (W)	0, 23.0, 42.0, 72.1	BC, BW, HE, OW, RX, WI	Bd wt	42	72.1		10% decrease in final body weight
					Hemato	72.1			
					Repro		23	72.1	LOAEL: Decreased relative testes weight, decreased epididymal sperm concentration, increased serum testosterone levels SLOAEL: Decreased fertility when mated to unexposed female, decreased sperm motility
Petrova et al. 2020									**Cobalt (II) Chloride Hexahydrate**
87	Mouse (ICR) NS F	27–28 days (2–3 days prior to parturition–PND 25) (W)	0, 75	DX	Develop			75	Decrease in body weight on PND 18 (24%) and PND 25 (15%)
Shrivastava et al. 1996									**Cobalt Chloride**
88	Mouse (Parkes) 6 F	45 days, daily (W)	0, 45	HP	Endocr			45	Degeneration and necrotic changes in thyroid epithelial cells; lymphocytic infiltration

Table 2-2. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
Mohiuddin et al. 1970									**Cobalt Sulfate**
89	Guinea pig (NS) 20 M	5 weeks, 7 days/week (F)	0, 20	BW, CS, GN, HP, LE, OF, OW	Death			20	4/20 died
					Bd wt	20			
					Cardio			20	Cardiomyopathy (pericardial effusion, pericarditis, vacuolar degeneration of the myocardium, thickened and edematous endocardium, mural thrombi; elevated heart weight), abnormal EKG

[a]The number corresponds to entries in Figure 2-3; differences in levels of health effects and cancer effects between male and females are not indicated in Figure 2-3. Where such differences exist, only the levels of effect for the most sensitive sex are presented.
[b]Used to derive an acute-duration oral MRL of 0.03 mg Co/kg/day; dose was divided by an uncertainty factor of 30 (10 for human variability, 3 for use of a minimal LOAEL).
[c]Used to derive an intermediate-duration oral MRL of 0.02 mg Co/kg/day; $BMDL_{1SD}$ of 1.95 mg Co/kg/day was divided by an uncertainty factor of 100 (10 for extrapolation from animals to humans and 10 for human variability).

AChE = acetylcholinesterase; ALP = alkaline phosphatase; ALT = alanine aminotransferase; AST = aspartate aminotransferase; B = both males and females; BC = serum (blood) chemistry; Bd wt or BW = body weight; BI = biochemical changes; BMDL = benchmark dose lower confidence limit; BuChE = butyrylcholinesterase; (C) = capsule; Cardio = cardiovascular; CNS = central nervous system; Co = cobalt; CS = clinical signs; Develop = developmental; DX = developmental toxicity; EKG = electrocardiogram; Endocr = endocrine; F = female(s); (F) = feed; FI = food intake; (G) = gavage, not specified; GABA = gamma-aminobutyric acid; Gastro = gastrointestinal; GD = gestational day; GFAP = glial fibrillary acidic protein; GN = gross necropsy; (GW) = gavage in water; HE = hematological; Hemato = hematological; HP = histopathology; IgG = immunoglobulin G; IL-1β = interleukin 1β; Immuno = immunological; IX = immune function; LD_{50} = median lethal dose; LDH = lactate dehydrogenase; LE = lethality; LOAEL = lowest-observed-adverse-effect level; M = male(s); MRL = minimal risk level; Musc/skel = musculoskeletal; Neuro = neurological; NOAEL = no-observed-adverse-effect level; NS = not specified; NX = neurological function; OF = organ function; OP = ophthalmology; OW = organ weight; PND = postnatal day; Repro = reproductive; Resp = respiratory; polycythemia = author-reported term associated with increased hemoglobin or erythrocyte count; RX = reproductive function; SD = standard deviation; SLOAEL = serious LOAEL; TNFα = tumor necrosis factor α; UR = urinalysis; (W) = water; WI = water intake

Figure 2-3. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)
Acute (≤14 days)

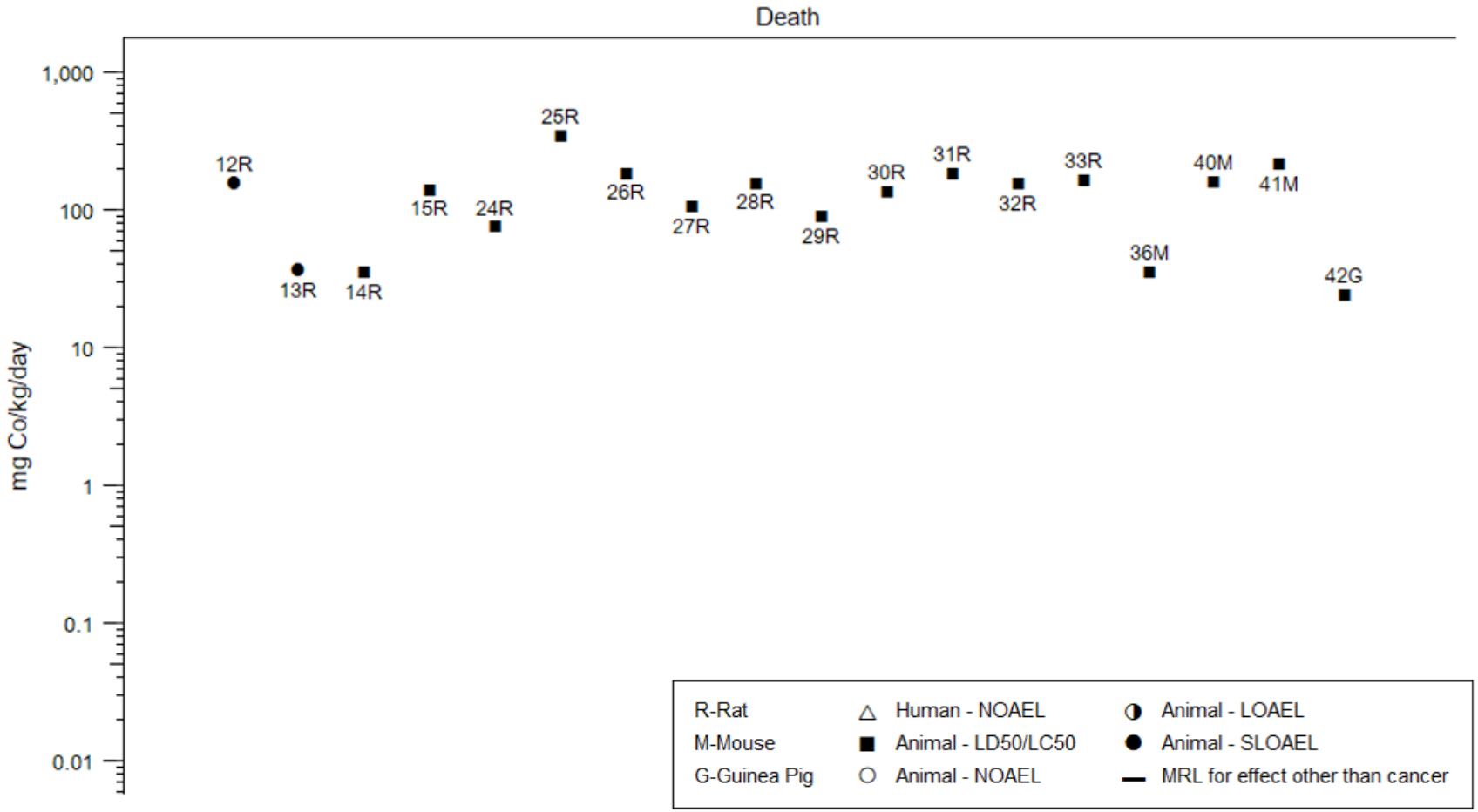


Figure 2-3. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)
Acute (≤14 days)

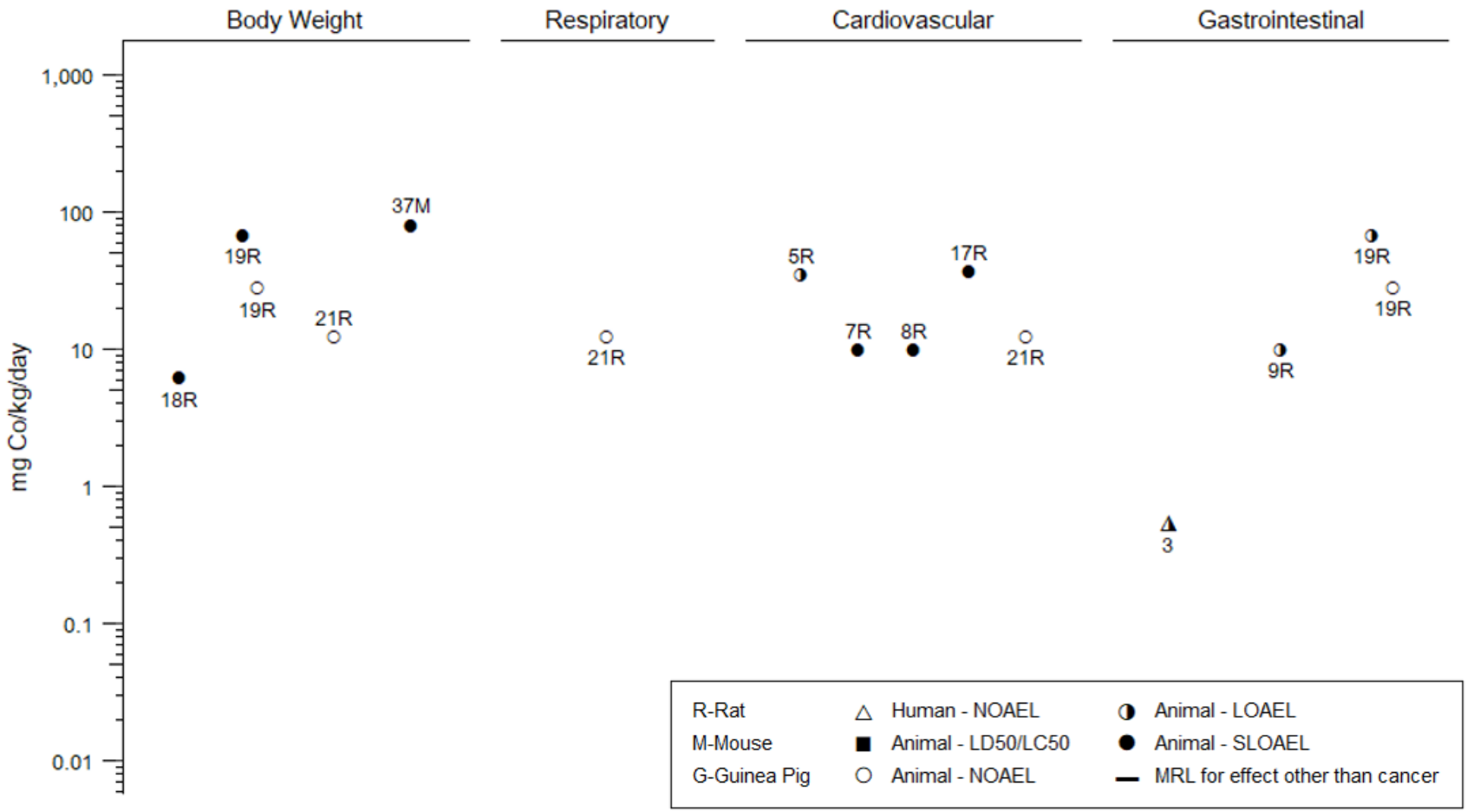


Figure 2-3. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)
Acute (≤14 days)

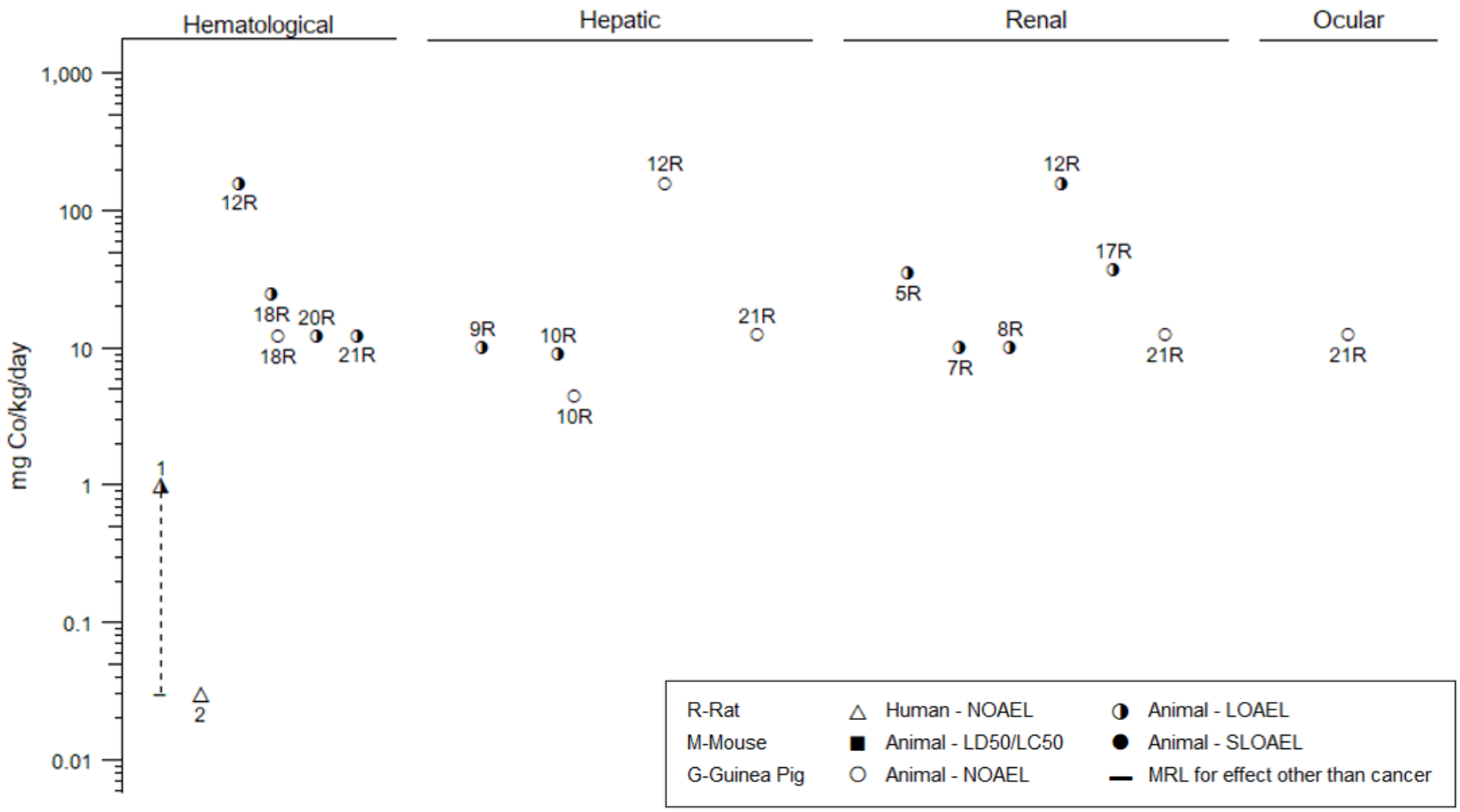


Figure 2-3. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)
Acute (≤14 days)

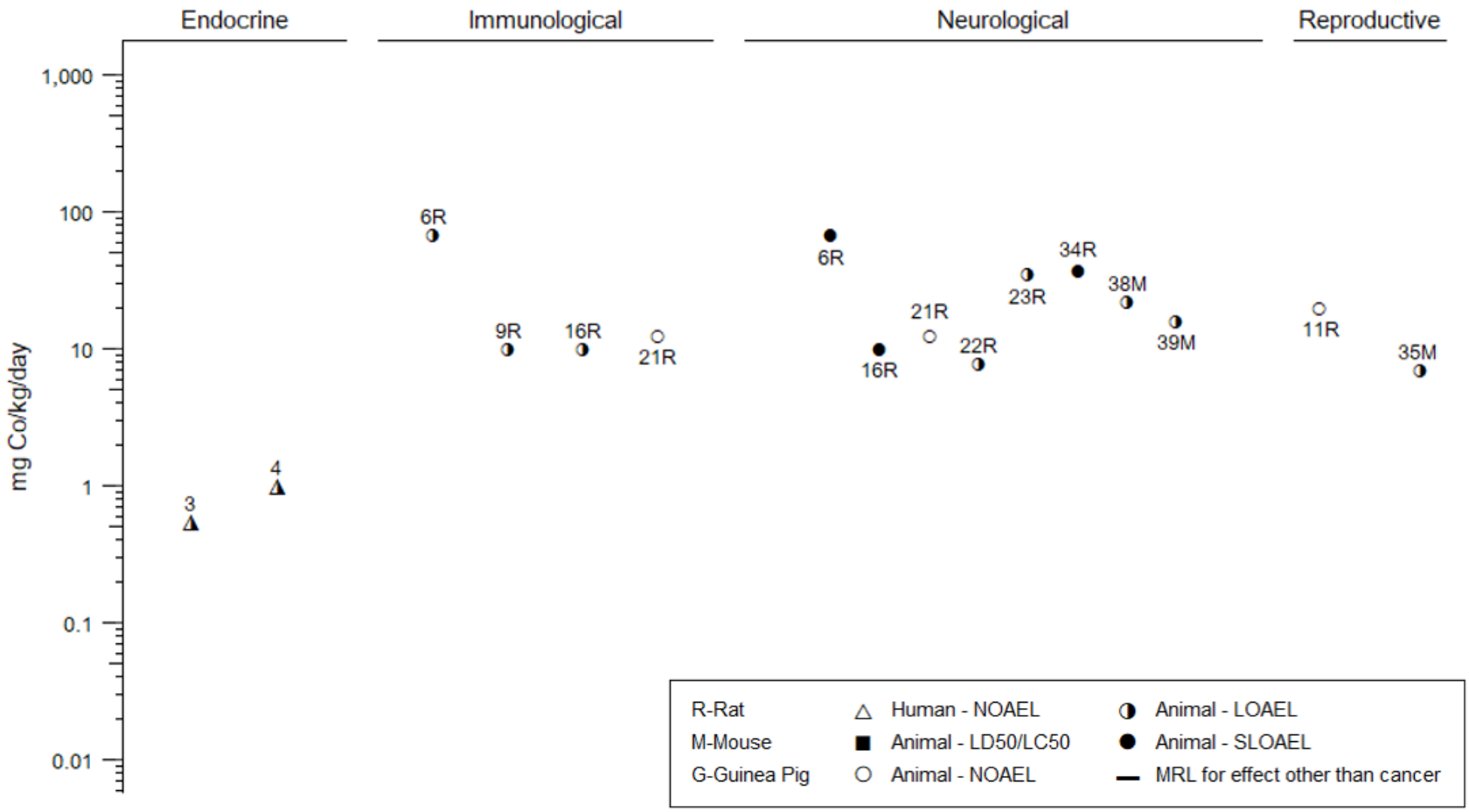


Figure 2-3. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)
Acute (≤14 days)

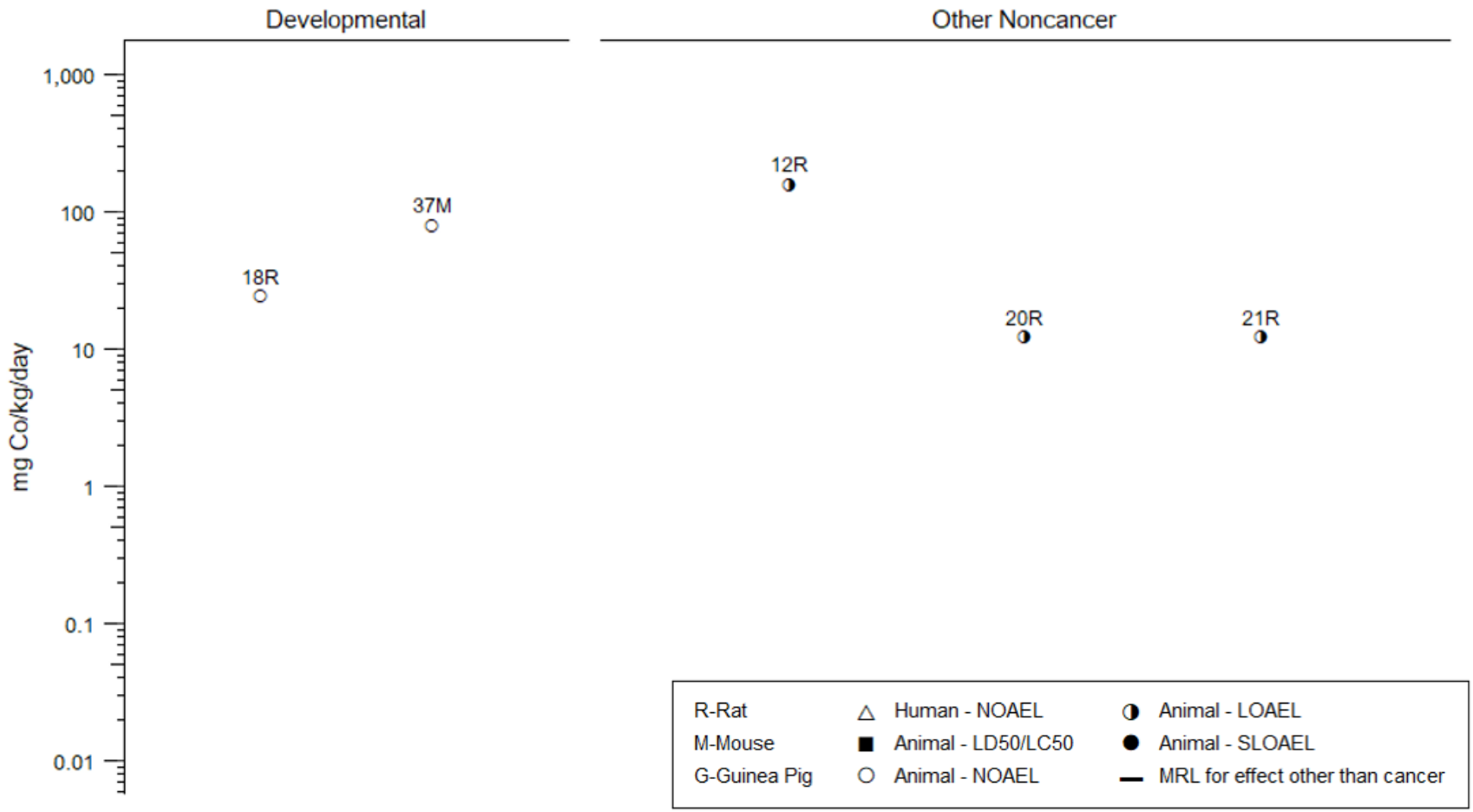


Figure 2-3. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)
Intermediate (15–364 days)

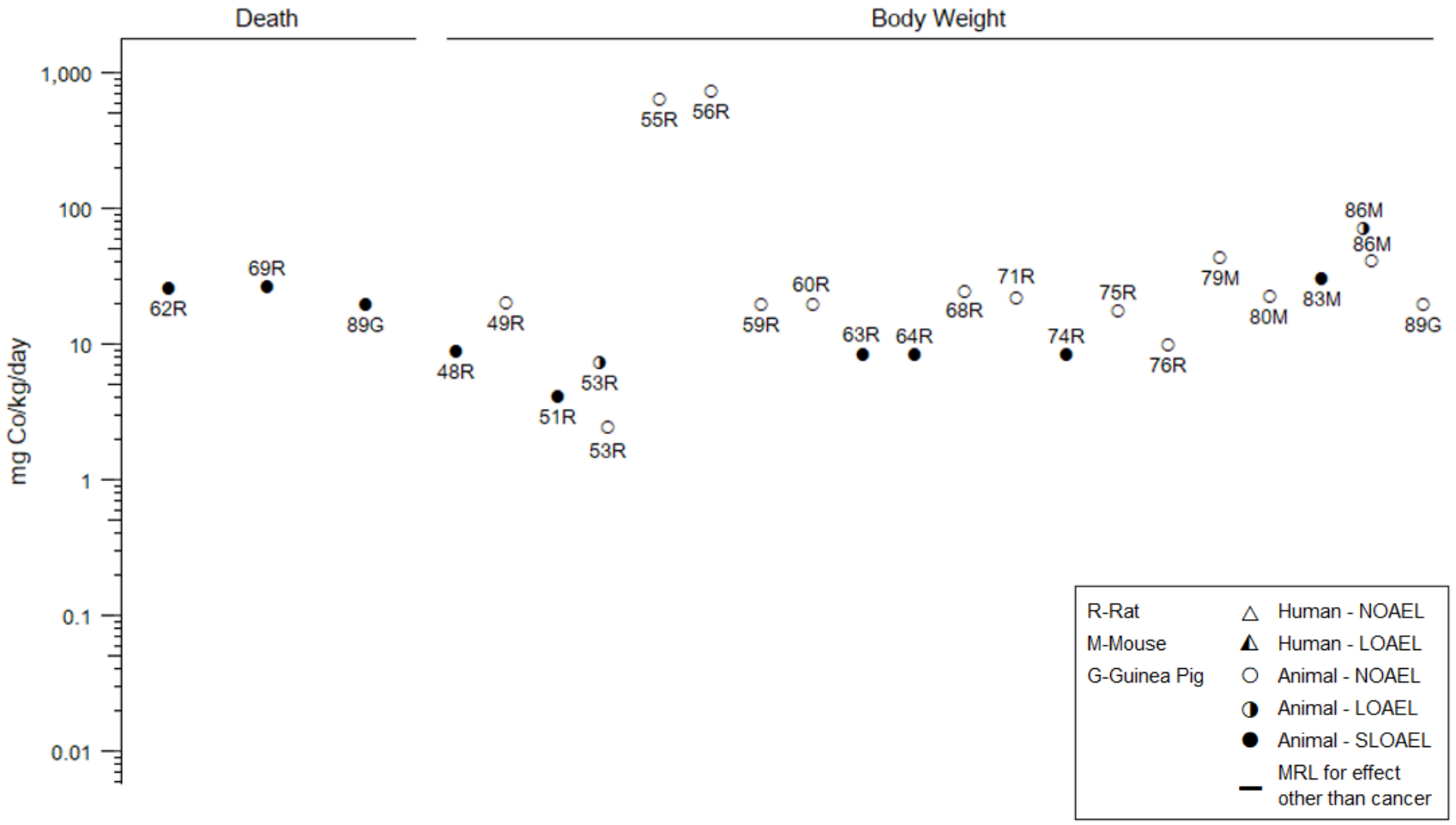


Figure 2-3. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)
Intermediate (15–364 days)

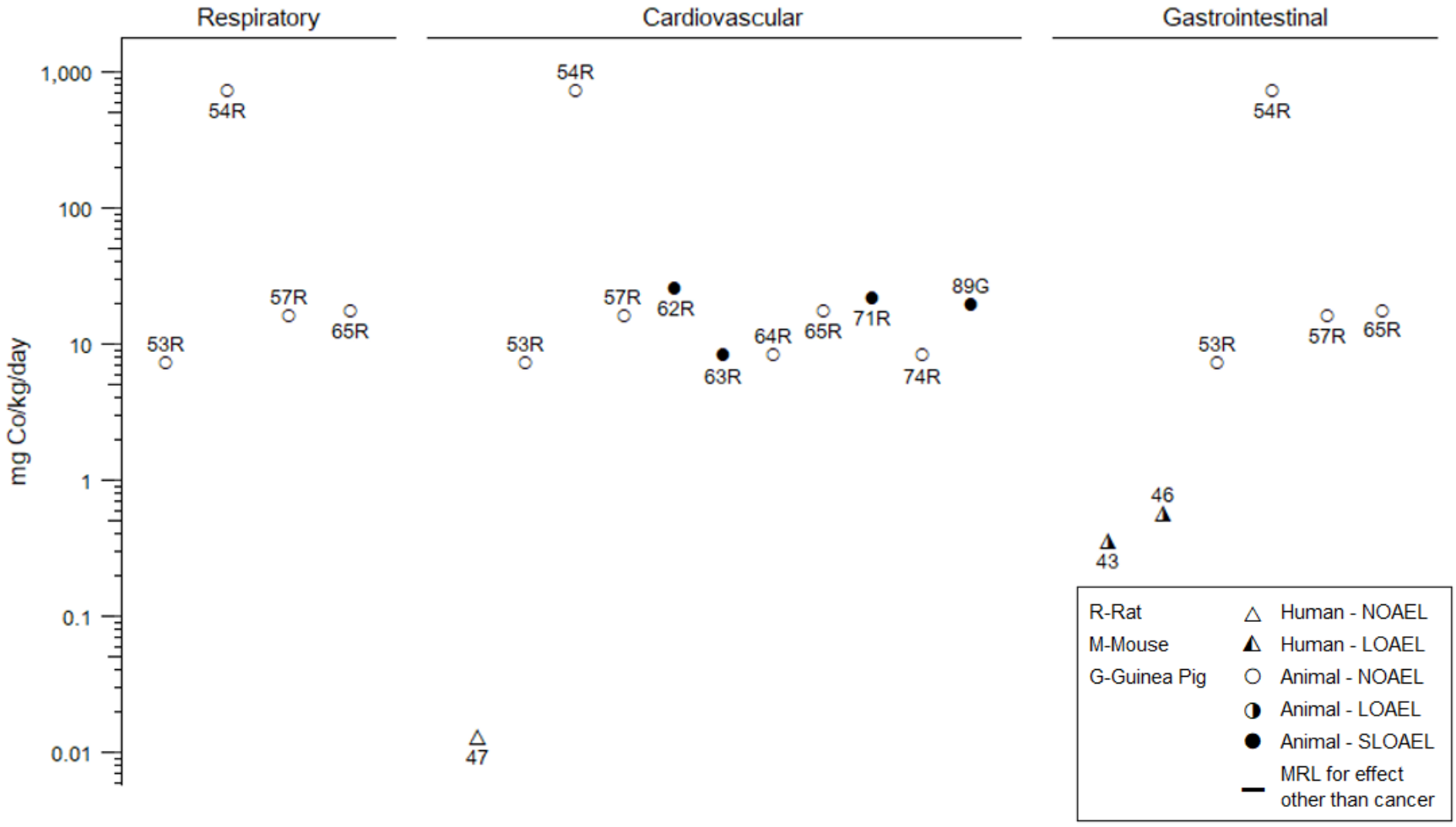


Figure 2-3. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)
Intermediate (15–364 days)

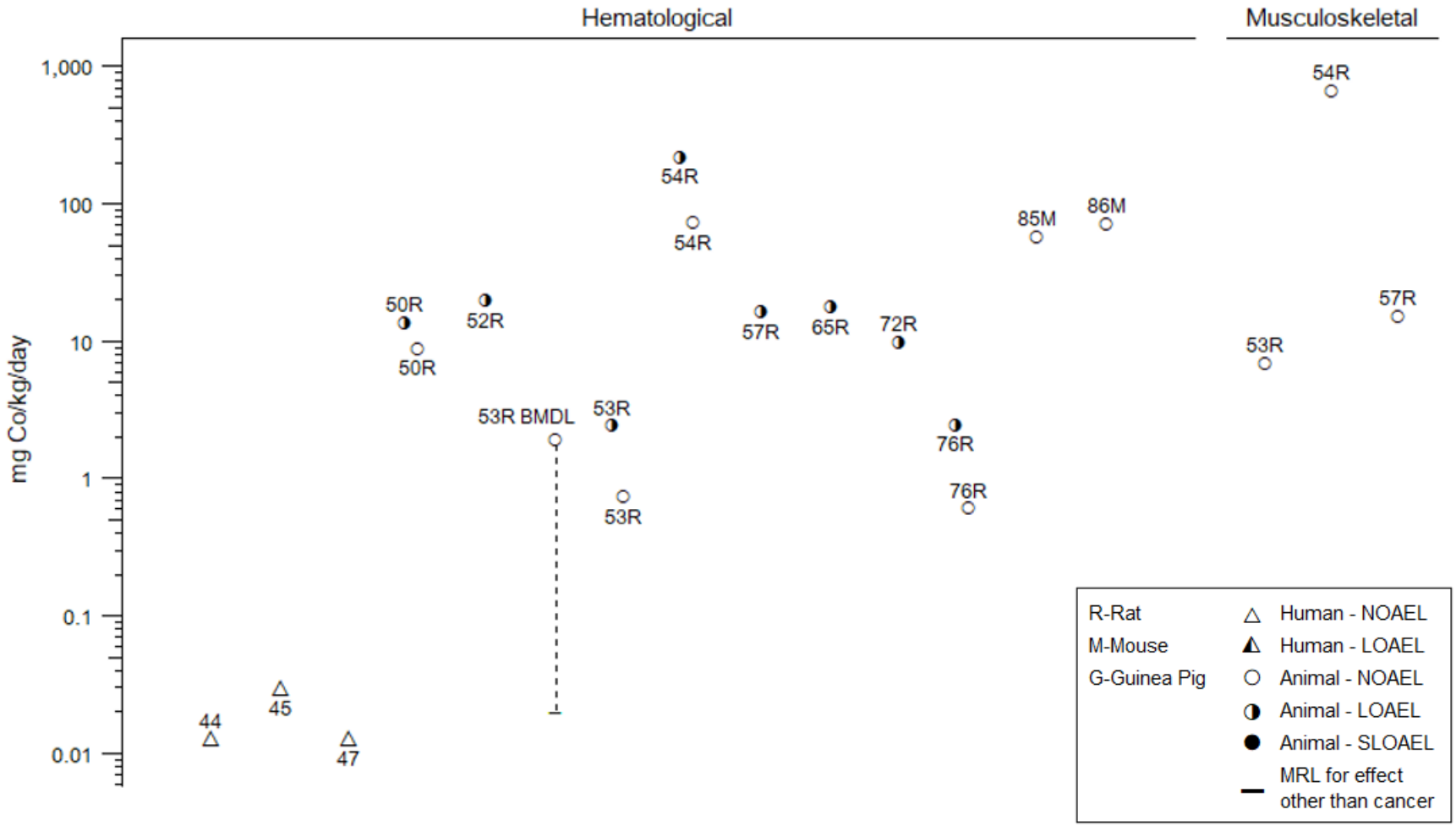


Figure 2-3. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)
Intermediate (15–364 days)

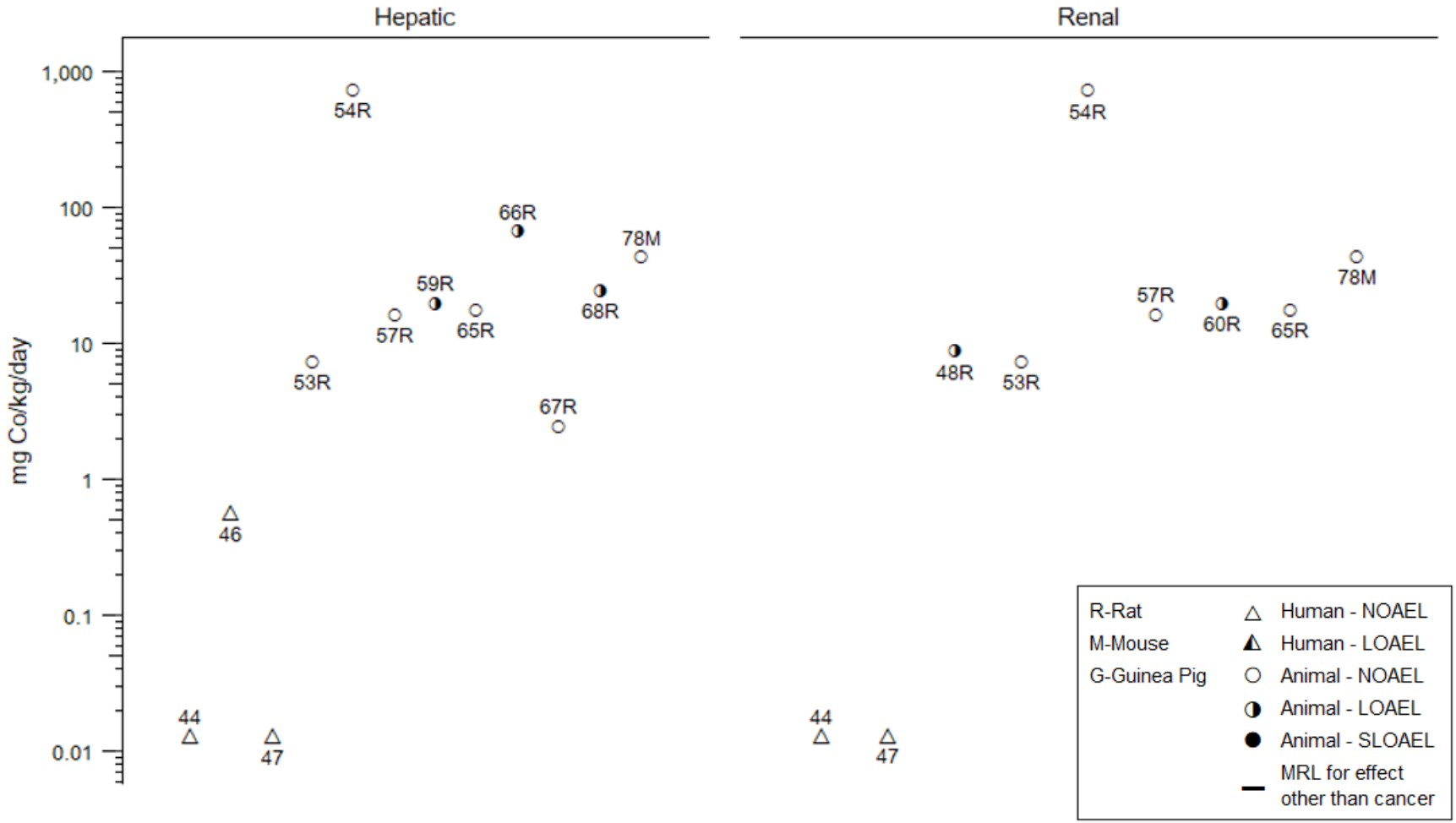


Figure 2-3. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)

Intermediate (15–364 days)

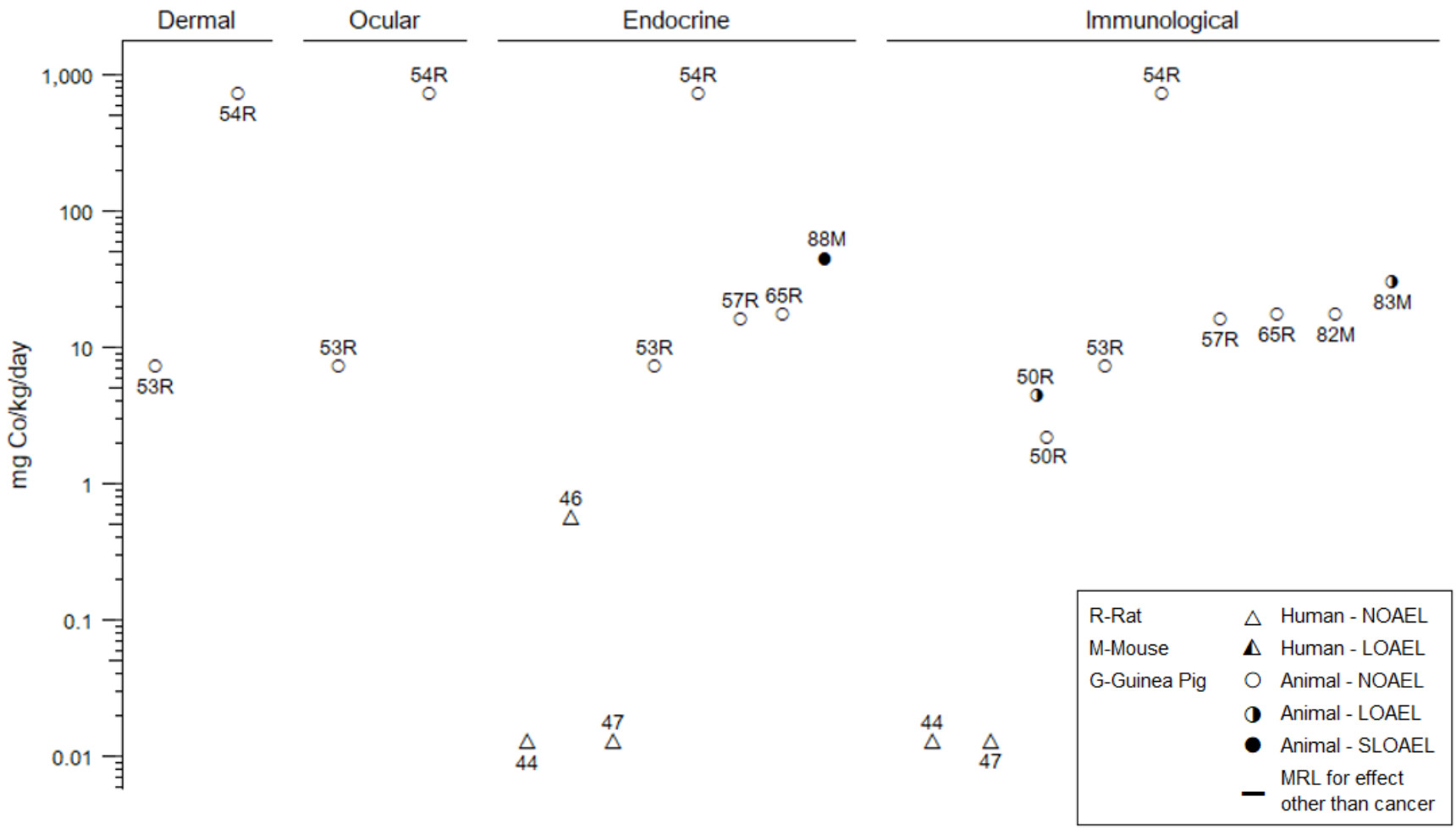


Figure 2-3. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)
Intermediate (15–364 days)

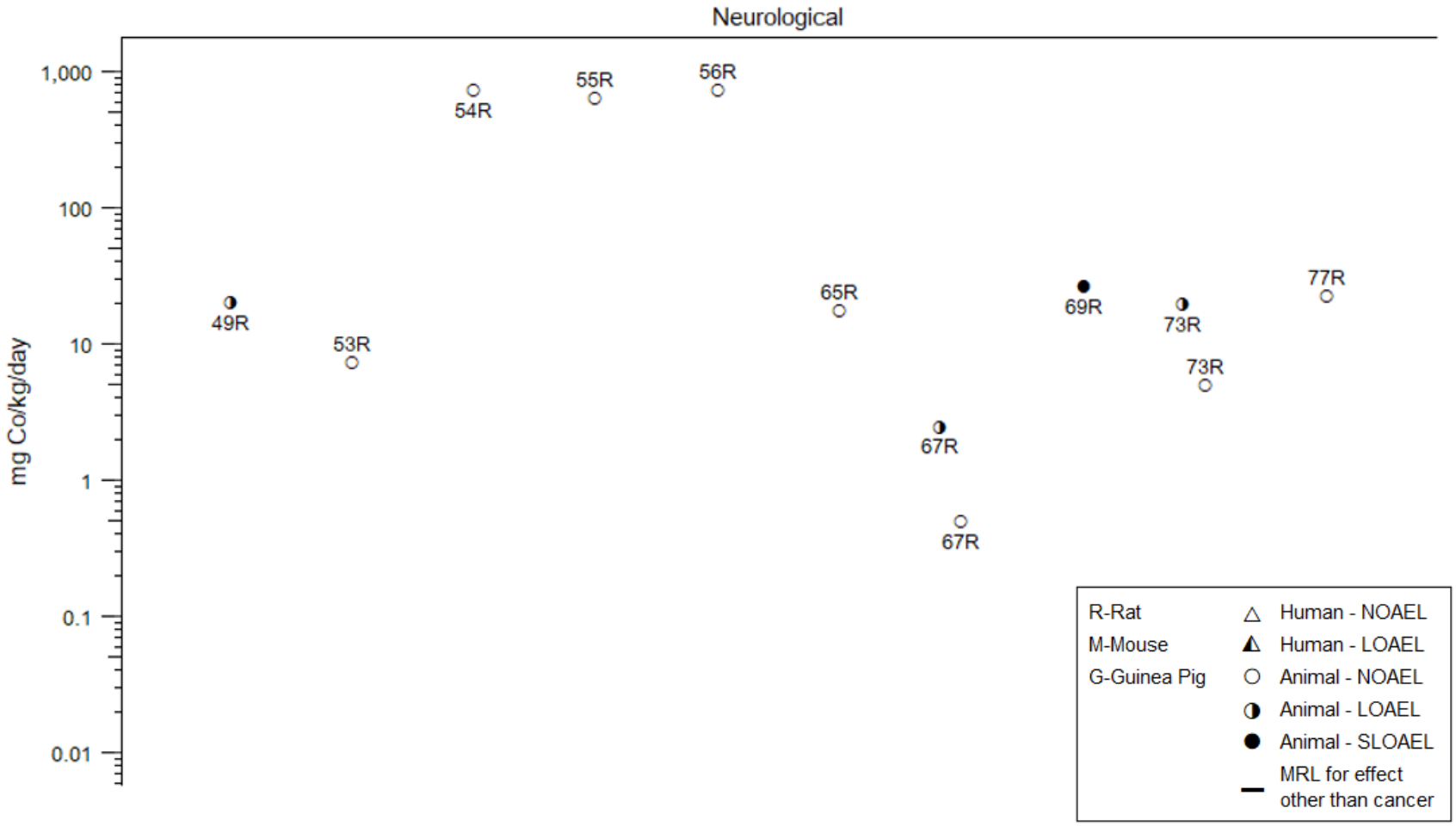


Figure 2-3. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)
Intermediate (15–364 days)

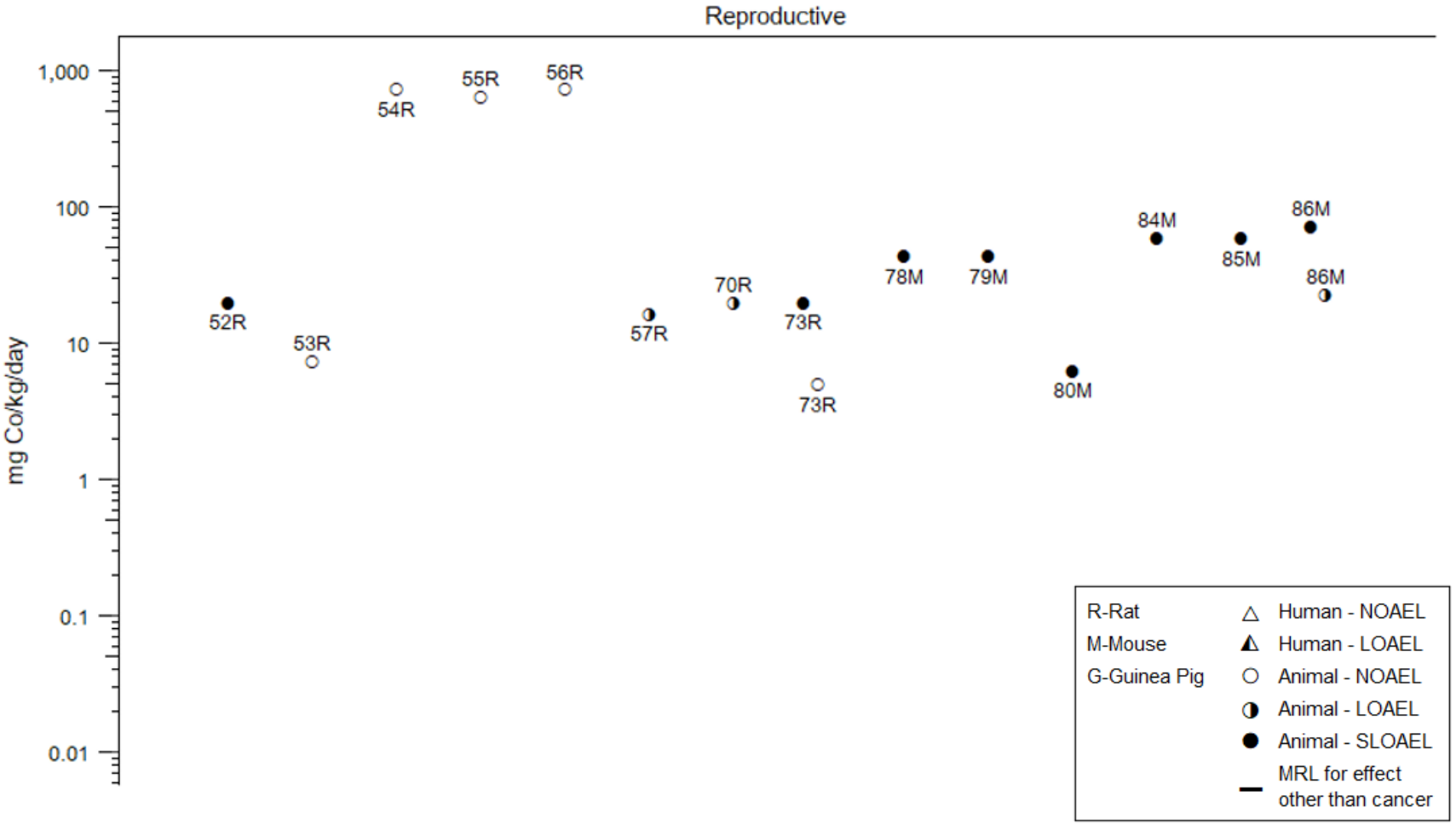


Figure 2-3. Levels of Significant Exposure to Cobalt – Oral (mg Co/kg/day)
Intermediate (15–364 days)

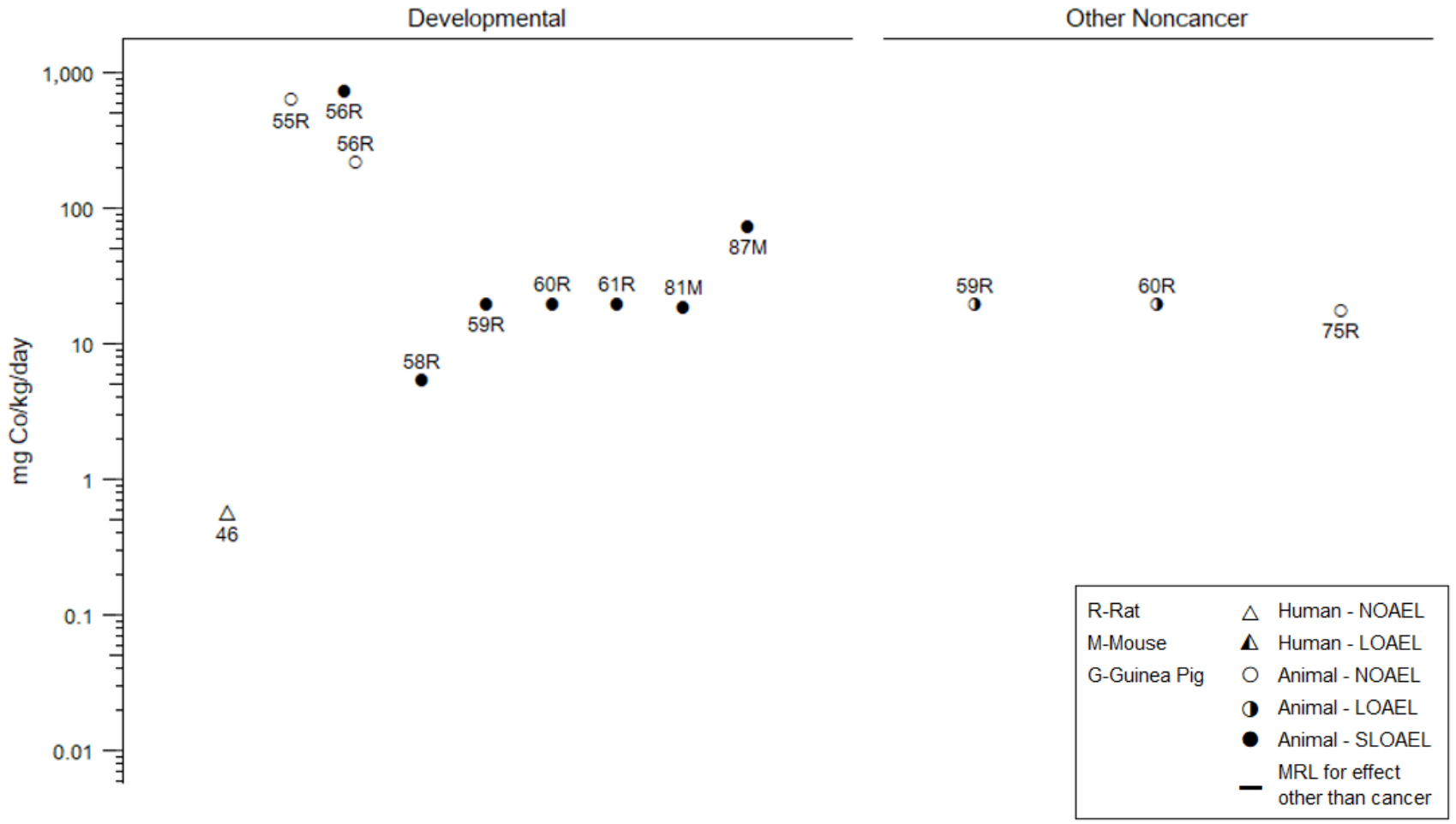


Table 2-3. Levels of Significant Exposure to Cobalt – Dermal

Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
ACUTE EXPOSURE								
Ikarashi et al. 1992a								**Cobalt Chloride**
Rat (Fischer-344) 3 F	3 days 1 time/day	0, 1.8, 3.6, 9.1, 18 mg Co/kg/day	LE, IX	Immuno	3.6 mg Co/kg/day	9.1 mg Co/kg/day		Increased proliferation of lymphatic cells (LLNA assay)
Bonefeld et al. 2015								**Cobalt Chloride**
Mouse (NS) NS	3 days (sensitization) + 2 days (challenge) 1 time/day	0, 4.5%	CS, IX	Immuno		4.5%		Skin sensitization (swelling, proliferation of lymphocytes) at challenge
Ikarashi et al. 1992a								**Cobalt Chloride**
Mouse (CBA/N) 3 F	3 days 1 time/day	0, 2.3, 4.5, 11, 23 mg Co/kg/day	LE, IX	Immuno	2.3 mg Co/kg/day	4.5 mg Co/kg/day		Increased proliferation of lymphatic cells (LLNA assay)
Ikarashi et al. 1992b								**Cobalt (II) Chloride Hexahydrate**
Mouse (BALB/c) 3 F	3 days 1 time/day	0, 2.5, 12, 25 mg Co/kg/day	LE, IX	Immuno	2.5 mg Co/kg/day	12 mg Co/kg/day		Increased proliferation of lymphatic cells (LLNA assay)
Mandervelt et al. 1997								**Cobalt (II) Chloride Hexahydrate**
Mouse (BALB/c) 3 F	3 days 1 time/day	0, 2.5, 6, 12 mg Co/kg/day	IX	Immuno	6 mg Co/kg/day	12 mg Co/kg/day		Increased proliferation of lymphatic cells (LLNA assay)
Camner et al. 1993								**Cobalt Chloride**
Guinea pig (Dunkin-Hartley) 16 F	24 hours	Sensitization: 0, 2.3% Challenge: 0.05, 0.2, 0.5%	IX	Immuno		2.3%		Skin sensitization
Ikarashi et al. 1992a								**Cobalt Chloride**
Guinea pig (Hartley) 3 F	3 days 1 time/day	0, 1.4, 2.8, 6.8, 14 mg Co/kg/day	LE, IX	Immuno	6.8 mg Co/kg/day	14 mg Co/kg/day		Increased proliferation of lymphatic cells (LLNA assay)

Table 2-3. Levels of Significant Exposure to Cobalt – Dermal

Species (strain) No./group	Exposure parameters	Doses	Parameters monitored	Endpoint	NOAEL	Less serious LOAEL	Serious LOAEL	Effects
INTERMEDIATE EXPOSURE								
NTP 1991, 2023								**Cobalt Sulfate Heptahydrate**
Rat (F344/N) 5 M, 5 F	16 days 5 days/week 6 hours + 12 minutes (T90 time) per day	0, 0.035, 0.19, 1.80, 19.0, 75.7 mg Co/m^3 in air	CS	Ocular	1.8 mg Co/m^3 in air	19 mg Co/m3 in air		Ocular irritation (chromodacryorrhea)
Exposure chamber analysis showed that under test conditions, the test substance converted to cobalt sulfate hexahydrate (Behl et al. 2015).								
NTP 1991, 2023								**Cobalt Sulfate Heptahydrate**
Mouse (B6C3F1) 5 M, 5 F	16 days 5 days/week 6 hours + 12 minutes (T90 time) per day	0, 0.035, 0.19, 1.80, 19.0, 75.7 mg Co/m^3 in air	CS	Ocular	1.8 mg Co/m^3 in air	19 mg Co/m^3 in air		Ocular irritation (chromodacryorrhea)
Exposure chamber analysis showed that under test conditions, the test substance converted to cobalt sulfate hexahydrate (Behl et al. 2015).								
Kincaid et al. 1954								**Dicobalt Octacarbonyl**
Guinea pig (NS) 3 NS	18 days 5 days/week 1 time/day	0, 51.7 mg Co/kg	CS, LE	Bd wt	51.7 mg Co/kg			
				Dermal		51.7 mg Co/kg		Skin lesions (scabs and denuded areas) at application site
CHRONIC EXPOSURE								
Swennen et al. 1993								**Hard Metal**
Human 82 M	8 years (occupational)	0, 0.125 mg Co/m^3 in air	CS	Dermal		0.125 mg Co/m^3 in air		Eczema and erythema

Bd wt = body weight; Co = cobalt; CS = clinical signs; F = female(s); Immuno = immunological; IX = immune function; LE = lethality; LLNA = local lymph node assay; LOAEL = lowest-observed-adverse-effect level; M = male(s); NOAEL = no-observed-adverse-effect level; NS = not specified; T90 = the time required for the inhalation chamber concentration to reach 90% of the target concentration

2.2 DEATH

The overall risk of death (from all causes) was not elevated in a cohort of 1,148 workers from an electrochemical plant producing sodium and cobalt in France; specific analysis of only cobalt production workers also showed no increased risk of death (Moulin et al. 1993; Mur et al. 1987). Many studies have evaluated the potential for increased mortality in hard metal workers exposed to cobalt (and other metals such as tungsten and tungsten carbide) from various countries around the world. The majority of them did not find any excess in total (all-cause) mortality in hard metal workers, compared to the general population, as determined by standard mortality ratios (SMRs) (Lasfargues et al. 1994; Marsh et al. 2017a; McElvenny et al. 2017; Morfeld et al. 2017; Moulin et al. 1998, 2000; Wallner et al. 2017; Wild et al. 2000). However, a large multinational cohort combining 32,354 hard metal workers from the United States, Austria, Germany, Sweden, and the United Kingdom exposed to long-term median levels of 0.006 mg Co/m^3 (as well as tungsten and nickel) observed an excess in total (all-cause) mortality compared to both national and regional rates (Marsh et al. 2017b). Specific causes of death that were elevated above national and/or regional rates included malignant neoplasms (buccal cavity and pharyngeal malignant neoplasms; bronchus, trachea, lung), nonmalignant respiratory diseases, emphysema, ischemic heart disease, and accidental deaths. However, due to concurrent exposure to other substances in hard metal, the contribution of cobalt exposure to these deaths is unclear.

A series of studies by Viegas et al. (2022a) demonstrate that acute-duration inhalation lethality in rats is dependent upon the administered compound. Cobalt hydroxide, cobalt metal powder, and cobalt oxide were the most toxic, with 100% mortality following 4-hour, nose-only exposures to 32, 50, and 79 mg Co/m^3, respectively. Other compounds included cobalt carbonate, with 50% mortality at 3,200 mg Co/m^3, and cobalt tetraoxide and cobalt sulfide, with 0% mortality at the limit test concentrations of 1,200 and 3,200 mg Co/m^3, respectively (Viegas et al. 2022a). Differences in toxicity were not attributable to lung disposition; rather, the study authors proposed that increased toxicity of cobalt hydroxide, metal powder, and oxide was attributable to their inflammatory "reactivity" in the lungs. Mortality was associated with inflammatory changes in the lung, including perivascular inflammatory edema, alveolar pulmonary edema, and pneumonia. Less toxic compounds (cobalt carbonate, tetraoxide, and sulfide) were generally associated only with mild perivascular inflammatory edema. Another study reported a 30-minute LC_{50} of 165 mg Co/m^3 for cobalt hydrocarbonyl (plus oxide/carbonate decomposition products) in albino rats (Palmes et al. 1959).

Increased mortality was observed in some intermediate-duration inhalation studies in rodents. Decreased survival was observed in F344/N rats and B6C3F1 mice exposed to cobalt metal at ≥20 and 40 mg Co/m^3, respectively, or cobalt sulfate at ≥19 mg Co/m^3 for 6 hours/day, 5 days/week for 16 or 17 days (NTP 1991, 2014). For cobalt sulfate, female rats were less susceptible, with no mortalities until 75.7 mg Co/m^3 (NTP 1991). Intermediate-duration exposure to 11.4 mg Co/m^3 for 13 weeks caused 20% mortality in male B6C3F1 mice but not female mice or F344/N rats of either sex (Bucher et al. 1990; NTP 1991). In other 13–14-week intermittent exposure inhalation studies, no exposure-related increases in mortality were observed in F344/N rats or B6C3F1 mice exposed to concentrations up to 5 mg Co/m^3 as cobalt metal (NTP 2014) or in albino rats or guinea pigs exposed to 9 mg Co/m^3 as cobalt hydrocarbonyl (plus oxide/carbonate decomposition products) (Palmes et al. 1959).

Increased mortality was observed in some chronic-duration studies in rodents. Chronic-duration exposure to cobalt metal for 105 weeks resulted in a 20–22% reduction in survival in male B6C3F1 mice and female F344/N rats exposed to ≥2.5 mg Co/m^3, compared to controls; survival in male rats or female mice was comparable to controls at concentrations up to 5 mg Co/m^3 (Behl et al. 2015; NTP 2014). Chronic-duration exposure for 105 weeks to cobalt sulfate did not have a significant effect on survival in F344/N rats or B6C3F1 mice at concentrations up to 1.11–1.14 mg Co/m^3 (Behl et al. 2015; Bucher et al. 1999; NTP 1998).

A 19-month-old male child who swallowed an unknown amount of a cobalt chloride solution died approximately 6.5 hours after ingestion, despite repeated induced vomiting, gastric lavage, and supportive therapy (Jacobziner and Raybin 1961).

In several studies, lethal cardiomyopathy was reported in people who consumed large quantities of beer containing cobalt sulfate (and ethyl alcohol) (Alexander 1969, 1972; Bonenfant et al. 1969; Morin et al. 1967, 1971; Sullivan et al. 1969). The deaths occurred during the early to mid-1960s, at which time, breweries in Canada, the United States, and Europe were adding cobalt to beer as a foam stabilizer (Alexander 1969, 1972; Bonenfant et al. 1969; Morin et al. 1967, 1971; Sullivan et al. 1969); this practice has been discontinued for decades. Deaths occurred following ingestion of beer containing 0.04–0.14 mg cobalt/kg/day for a period of years (approximately 8–30 pints of beer each day). "Acute mortality" (death within several days of admission) accounted for 18% of the deaths (Alexander 1972). Approximately 43% of the patients admitted to the hospital with cardiomyopathy died within several years of the initial hospital visit. It should be noted, however, that the cardiomyopathy may have also been due to the fact that the beer-drinkers had protein poor diets and may have had prior cardiac damage from alcohol abuse.

Therefore, the role of cobalt in cardiomyopathy, and subsequent death, is unclear. It is noted that treatment of both pregnant and nonpregnant anemic patients with doses of cobalt (0.6–1 mg/kg/day) that were much higher than the doses in the beer did not result in mortality (Davis and Fields 1958; Holly 1955).

Numerous acute-duration oral lethality studies have been conducted in animals. As seen with inhalation, oral LD_{50} values vary depending upon administered compound, from as low as 25 mg Co/kg for cobalt chloride to 352 mg Co/kg for cobalt sulfate (Singh and Junnarkar 1991). Reported LD_{50} values in rodents were 25–190 mg Co/kg for cobalt chloride, 91 mg Co/kg for cobalt fluoride, 109 mg Co/kg for cobalt bromide, 140 mg Co/kg for cobalt nitrate, 159 mg Co/kg/day for cobalt oxide, 161–352 mg Co/kg for cobalt sulfate, 168 mg Co/kg for cobalt acetate, and 187 mg Co/kg for cobalt phosphate (Krasovskii and Fridlyand 1971; Murdock 1959; Singh and Junnarkar 1991; Speijers et al. 1982). In other acute-duration oral studies, single oral exposures to 149 or 161 mg Co/kg as cobalt chloride caused death in 10/20 or 11/20 Sprague-Dawley rats, respectively (Domingo and Llobet 1984; Domingo et al. 1985a).

Oral intermediate-duration exposure to cobalt compounds in animals resulted in death in some studies. In an intermediate-duration exposure study, 4/10 Wistar rats died following oral exposure to 27 mg Co/kg/day as cobalt chloride for 60 days (Mohamed et al. 2019). Similarly, 5/10 Wistar rats died during an 8-week exposure to 26 mg Co/kg/day as cobalt sulfate (Grice et al. 1969). Following a 5-week exposure to 20 mg Co/kg/day as cobalt sulfate by gavage, 4 out of 10 guinea pigs (species not specified) died (Mohiuddin et al. 1970). No deaths were observed in Wistar rat dams exposed to at 20 mg Co/kg/day via gavage as cobalt chloride for 2 weeks during gestation and 2 weeks during lactation (Garoui et al. 2011, 2012, 2013). In Sprague-Dawley rats, no exposure-related mortalities were observed at doses up to 7.44 mg Co/kg/day as cobalt chloride or 734 mg Co/kg/day as cobalt tetraoxide for 90 days (Danzeisen et al. 2020a), 16.5 mg Co/kg/day as cobalt chloride in drinking water for 13 weeks (Domingo et al. 1984), or 68 mg Co/kg/day as cobalt chloride in drinking water for 4 weeks (Khalil et al. 2020). In Swiss mice, 1/10 and 2/10 mice died at 11.62 and 23.01 mg Co/kg/day as cobalt chloride in drinking water, respectively, during the 10^{th} week of a 12-week exposure; however, the cause of death was not discussed so it is unknown if these deaths were compound-related (Elbetieha et al. 2008). No deaths were observed in male B6C3F1 mice exposed to 58.9 mg Co/kg/day as cobalt chloride in drinking water for 10 weeks (Pedigo and Vernon 1993). Following a 5-week exposure to 20 mg Co/kg/day as cobalt sulfate by gavage, 4 out of 10 guinea pigs (species not specified) died (Mohiuddin et al. 1970).

No studies were identified regarding death in humans after dermal exposure to cobalt. In animals, no mortalities have been reported following dermal exposures to cobalt compounds. Acute-duration dermal exposure to cobalt chloride did not cause death in rats, mice, or guinea pigs at doses up to 18, 25, or 14 mg Co/kg/day, respectively (Ikarashi et al. 1992a, 1992b). Intermediate-duration dermal exposure once a day for 18 days to 51.7 mg Co/kg/day as dicobalt octacarbonyl did not cause death in guinea pigs (Kincaid et al. 1954).

Acute-duration exposure by subcutaneous injection of 45 mg Co/kg/day as dicobalt octacarbonyl did not cause death in guinea pigs (Kincaid et al. 1954). Acute-duration exposure to cobalt chloride at a dose of 12 mg Co/kg/day via intraperitoneal injection killed 13 Sprague-Dawley rats out of 20 in the treatment group (Domingo et al. 1985a). No Wistar rats died after a single subcutaneous injection of 7 mg Co/kg (Horiguchi et al. 2004). Domingo and Llobet (1984) showed that a single intraperitoneal injection of cobalt chloride at 12 mg Co/kg/day caused the death of 5 Sprague-Dawley rats in a treatment group of 20 (Domingo and Llobet 1984).

2.3 BODY WEIGHT

No studies in humans examined changes in body weight following inhalation, oral, or dermal exposure to cobalt.

Studies evaluating body weight in animals following acute-duration inhalation exposure are limited. No body weight effects were noted in Wistar rats exposed to cobalt tetraoxide for 6 hours/day at concentrations up to 160.90 mg Co/m^3 for 14 days (Burzlaff et al. 2022a).

Several studies in rats indicate that intermediate-duration inhalation exposure to cobalt results in exposure-related decreases in body weight, compared to control. In rats, no biologically significant decreases in body weight were observed following intermediate-duration inhalation exposure to cobalt metal or cobalt compounds at concentrations up to 9 mg Co/m^3 (Bucher et al. 1990; Burzlaff et al. 2022a, 2022b; NTP 1991, 2014; Palmes et al. 1959). Intermittent exposure to cobalt metal at 10 mg Co/m^3 for 16 days resulted in serious decreases in final body weight in male rats (20%) and less serious decreases in female rats (12%), compared to control (NTP 2014). Serious decreases in female body weight (45%) were observed following exposure to cobalt metal at 20 mg Co/m^3 for 16 days (NTP 2014). Male rats were also more sensitive to cobalt sulfate at a similar concentration of 11.4 mg Co/m^3 for 13 weeks, showing a 14% decrease in final body weight, compared to control; female rats did not show exposure-

related body weight changes at this concentration (Bucher et al. 1990; NTP 1991). Exposure to ≥19.0 mg Co/m^3 as cobalt sulfate for 16 days resulted in decreases in body weight ≥20% in female and male rats, compared to control rats (NTP 1991). No body weight effects were observed in Wistar rats exposed to cobalt tetraoxide for 6 hours/day at concentrations up to 59.31 mg Co/m^3 for 28 days (Burzlaff et al. 2022a).

Mice show similar body weight effects as rats following intermediate-duration inhalation exposure, with no exposure-related effects at concentrations up to 5 mg Co/m^3 (Bucher et al. 1990; NTP 1991, 2014). Intermittent exposure to cobalt metal at 10 mg Co/m^3 for 14 weeks resulted in a 13–14% decrease in final body weight in both male and female mice, compared to control (NTP 2014). Similarly, intermittent inhalation exposure to 11.4 mg Co/m^3 as cobalt sulfate for 13 weeks reduced the final body weights in male rats by 14% and in female rats by 22%, compared to control (Bucher et al. 1990; NTP 1991). Final body weight decreases >20% were observed in both sexes after exposure to 19 mg Co/m^3 as cobalt sulfate for 16 days, compared to control mice (Bucher et al. 1990; NTP 1991). Similarly, when compared to control, exposure to 20 mg Co/m^3 as cobalt metal for 17 days resulted in a 16% decrease in final body weight in female mice, and exposure to 40 mg Co/m^3 as cobalt metal for 17 days or 14 weeks resulted in >20% decrease in final body weight in both sexes (NTP 2014).

In other species, no exposure-related decreases in body weight were observed in guinea pigs exposed to 9 mg Co/m^3 as cobalt hydrocarbonyl (plus oxide/carbonate decomposition products) for 3 months, compared to control (Palmes et al. 1959). A 3-month exposure to cobalt metal for 5 days/week, 6 hours/day at 0.1 mg Co/m^3 resulted in a 16% decrease in final body weight in pigs, compared to control pigs (Kerfoot 1974).

Chronic-duration inhalation exposure to 5 mg Co/m^3 as cobalt metal for 105 weeks (5 days/week, 6 hours/day) caused a >20% decrease in final body weight in male and female rats and mice, compared to respective control groups (NTP 2014). In rats, the next lower concentration (2.5 mg Co/m^3) resulted in final body weight decreases of 11% in males and 16% in females, compared to controls (NTP 2014). No body weight effects were noted at 1.25 mg Co/m^3 in either species. No body weight effects were noted in rats or mice exposed to concentrations up to 1.11 or 1.14 Co/m^3, respectively, as cobalt sulfate for 105 weeks (Bucher et al. 1999; NTP 1998). Lifetime exposure (5 days/week, 7 hours/day) to 7.9 mg Co/m^3 as cobalt oxide did not result in decreased body weight gain in hamsters (Wehner et al. 1977).

Decreased body weight or body weight gain were observed in animals orally exposed to cobalt and its compounds. In acute-duration studies, exposure to oral doses up to 28 mg Co/kg/day as cobalt chloride were generally not associated with body weight effects in rats (Salami et al. 2023; Shrivastava et al. 2010). Acute doses ≥68 mg Co/kg/day as cobalt chloride resulted in body weight loss in exposed rats (>13% decrease compared to starting weights, compared to 8% body weight gains in control rats) (Salami et al. 2023). However, pregnant animals may be more susceptible to body weight effects. Maternal body weight gains were decreased by >20% in Sprague-Dawley rat dams exposed to doses ≥6.2 mg Co/kg/day via gavage as cobalt chloride on gestation days (GDs) 6–15 (Paternian and Domingo 1988) and in ICR mouse dams exposed to 81 mg Co/kg/day via gavage as cobalt chloride on GDs 8–12 (Seidenberg et al. 1986).

While several studies reported body weight effects in rats following intermediate-duration oral exposure to cobalt compounds, inconsistencies regarding dose-response across studies have been found for most studied compounds. Male Sprague-Dawley rats showed an 11% decrease in body weight at the end of a 90-day gavage exposure to 7.44 mg Co/kg/day as cobalt chloride, compared to control. At 4 weeks postexposure, body weight decreases were more pronounced in males at 17%, and females showed a 13% decrease, compared to control (Danzeisen et al. 2020a). No body weight effects were noted at ≤2.48 mg Co/kg/day for 90 days. Sprague-Dawley rats exposed to 8.9 mg Co/kg/day as cobalt chloride in drinking water for 4 weeks lost 7% of their initial body weight, compared to a body weight gain of 8% in controls (Abdel-Daim et al. 2020). One study reported a 45–61% reduction in body weight gain in Sprague-Dawley rats following dietary exposure 0.45–13.8 mg Co/kg/day as cobalt chloride for 30 days; however, there was a lack of a clear dose-response relationship and food intake data were not reported (Chetty et al. 1979). Additional studies in rats evaluating cobalt chloride did not report biologically significant decreases in body weight, including Sprague-Dawley rats exposed to drinking water doses up to 18 mg Co/kg/day for 16 days or 20.3 mg Co/kg/day for 57 days (Bourg et al. 1985; Saker et al. 1998), CFY rats exposed to a gavage dose of 22 mg Co/kg/day for 3 weeks (Morvai et al. 1993), Wistar rats exposed to a gavage dose of 25 mg Co/kg/day for 60 days (Mathur et al. 2011), pregnant Wistar rats exposed to a drinking water dose of 20 mg Co/kg/day for 28 days (2 weeks gestation plus 2 weeks lactation) (Garoui et al. 2011, 2012), or albino rats exposed to 9.9 mg Co/kg/day via gelatin capsule for 8 weeks (Stanley et al. 1947).

Sprague-Dawley rats showed >20% decreases in final body weights following dietary exposure to ≥4.2 mg Co/kg/day as cobalt sulfate for ≥8 weeks, compared to control (Clyne et al. 1988; Haga et al. 1996; Pehrsson et al. 1991). No body weight effects were noted in pregnant Sprague-Dawley rats

exposed to gavage doses up to 648 mg Co/kg/day as cobalt sulfide or 734 mg Co/kg/day as cobalt tetraoxide for 2 weeks premating through postnatal day (PND) 3 (Danzeisen et al. 2020a). Danzeisen et al. (2020a) also examined the effects of oral exposure to tricobalt tetraoxide at a dose of 734 mg Co/kg/day for 90 days in male and female Sprague-Dawley rats and reported "slight" reductions in male rat body weight and "marginal" effects in female body weight; however, quantitative data were not reported.

Data on body weight effects following intermediate-duration oral exposure in other species are limited. A 33% decrease in body weight was observed in BALB/c mice exposed to 31 mg Co/kg/day as cobalt chloride starting 2–3 days prior to birth and during lactation (via dam) and directly on PNDs 26–60 via drinking water (Legostaeva et al. 2013; Zaksas et al. 2013). In studies with adult mice, drinking water exposure to doses up to 43.4 mg Co/kg/day as cobalt chloride for 12–13 weeks did not result in body weight effects (Anderson et al. 1993; Elbetieha et al. 2008; Pedigo et al. 1988). Drinking water exposure to 72.1 mg Co/kg/day for 12 weeks was associated with a 10% decrease in body weight in male CD-1 mice (Pedigo et al. 1988). No body weight effects were observed in male guinea pigs exposed to dietary cobalt sulfate at 20 mg Co/kg/day for 5 weeks (Mohiuddin et al. 1970).

No body weight changes were observed after intermediate-duration dermal exposure to 51.7 mg Co/kg/day as dicobalt octacarbonyl in methyl ether ketone in guinea pigs (Kincaid et al. 1954).

A 24% weight loss was reported by week 6 of an 8-week study in albino rats exposed to 2.5 mg Co/kg/day as cobalt chloride via subcutaneous injection; no weight changes were observed at 0.6 mg Co/kg/day (Stanley et al. 1947).

2.4 RESPIRATORY

The respiratory tract is a sensitive target of toxicity following inhalation exposure to cobalt. Adverse effects observed in humans occupationally exposed to cobalt metal and cobalt compounds included altered spirometry and evidence of pulmonary irritation and dyspnea at higher exposure concentrations. In laboratory animal studies, inflammatory changes throughout the respiratory tract were consistently observed in multiple species following exposure for any duration, with necrosis progressing to hyperplasia and metaplasia with repeated exposure.

There are limited data regarding respiratory effects in humans following acute-duration exposure to cobalt. An acute 6-hour exposure to cobalt hard metal dust at an average concentration of 0.038 mg Co/m^3 resulted in decreased lung forced vital capacity (FVC) in 15 healthy male volunteers, compared to diurnal FVC changes during a comparable 6-hour period without exposure (Kusaka et al. 1986a). The study authors noted that the FVC changes did not correlate with cobalt concentration, although those data were not presented. No other measures of lung function were altered. The exposed volunteers also had subjective complaints of respiratory irritation. The same investigators did not observe any significant changes in lung function in a group of 42 metal shapers after a 7-hour work shift, compared to pre-shift lung function measures. Mean cobalt levels during the work shift were 0.085 mg Co/m^3 (mean exposure over the previous 3 years was 0.126 mg Co/m^3) and the mean employment duration for workers was 10 years and 4 months (Kusaka et al. 1986a).

Numerous occupational studies evaluated potential associations between long-term exposure to cobalt and adverse respiratory effects (Table 2-4). In general, factory workers evaluated in the studies discussed below were subjected to co-exposures with other metals, such as nickel and chromium, and irritant gases; therefore, the health effects observed might not be caused by cobalt alone. Additionally, some of the endpoints (such as lung function) may be confounded by the healthy worker effect or exposure to other chemicals outside working hours. More details on the quality and confidence in available epidemiological studies evaluating respiratory effects can be found in Appendix C.

Table 2-4. Results of Epidemiological Studies Evaluating Exposure to Cobalt and Respiratory Effects

Reference, study type, and population	Exposure concentration	Outcome evaluated	Result
Al-Abcha et al. 2021 Cohort; 498 cobalt exposed workers (Michigan, United States)	Current air concentration, mg Co/m^3 Low: ≤0.05 High: >0.05	Respiratory symptoms (daily or weekly chest tightness, shortness of breath, wheezing)	↔ (low versus high)
		Onset of asthma since employment	↔ (low versus high)
Andersson et al. 2020 Cohort with cross-sectional analysis; 72 workers (63 males, 9 females; mean age 42 years; mean	Current air concentration, mg Co/m^3 Median: 0.0009 Mean: 0.0017 8-hour TWA: 0.0034	Respiratory symptoms (asthmatic symptoms, nasal drip, cough without infection, cough with phlegm)	↔ (current 8-hour TWA, air; cumulative)
		Lung function FVC, FEV_1	↔ (current 8-hour TWA, air; cumulative)

Table 2-4. Results of Epidemiological Studies Evaluating Exposure to Cobalt and Respiratory Effects

Reference, study type, and population	Exposure concentration	Outcome evaluated	Result
employment 11 years) from the hard metal industry (Sweden)	Cumulative exposure, mg Co-year/m^3: T1: ≤0.01 T2: 0.02–0.05 T3: ≥0.06	Asthma diagnosis	↔ (cumulative)
Deng et al. 1991 Cross-sectional; 362 workers (75% males; mean age 45 years; mean employment 21 years), including 310 active workers and 52 retired workers from the sintered magnet industry, and 1,370 unexposed blue-collar workers (age range 20–59 years) (United States)	Current air concentration, mean: 0.0175 mg Co/m^3	Respiratory symptoms (chronic cough and bronchitis, dyspnea, wheezing with/without shortness of breath)	↔ (workers versus referents)
		Lung function FVC, FEV_1	↑ (workers versus referents)
		Abnormal findings on chest x-ray	↔ (workers versus referents)
Gennart and Lauwerys 1990 Retrospective cohort; 48 workers (14 females, 34 males; range of mean ages 28.7–33.8 years; mean employment 6 years) from the diamond-cobalt tool manufacturing industry and 23 unexposed workers (12 females, 11 males; mean age 32.5 years) (Belgium)	Current air concentration, geometric mean, mg Co/m^3: Mixing: 0.1355 Oven: 0.0152 Current urine cobalt levels, mean: Exposed nonsmokers: 40.7 µg/g creatinine Exposed smokers: 25.7 µg/g creatinine Unexposed controls, nonsmokers: 0.18 µg/g creatinine	Lung function FVC, FEV_1, PEF	↓ (nonsmokers: workers versus referents) ↓ (smokers: workers versus referents)
		FEV_1/FVC	↔ (nonsmokers: workers versus referents) ↔ (smokers: workers versus referents)
		MEF_{25} or MEF_{50}	↔ (nonsmokers: workers versus referents) ↔ (smokers: workers versus referents)
		MEF_{75}	↔ (nonsmokers: workers versus referents) ↓ (smokers: workers versus referents)
		Respiratory symptoms (cough, sputum, dyspnea)	↑ (workers versus referents)

Table 2-4. Results of Epidemiological Studies Evaluating Exposure to Cobalt and Respiratory Effects

Reference, study type, and population	Exposure concentration	Outcome evaluated	Result
Hamzah et al. 2014 Cross-sectional; 402 exposed male workers (mean age 36.8 years; mean employment 12.2 years) from the steel industry (Malaysia)	Current air concentration, range of 8-hour TWA means in different job categories: 0.01–0.19 mg Co/m^3	Respiratory symptoms (chronic cough and phlegm, chest tightness, shortness of breath)	↑
		Lung function	
		FEV_1	↓
		FVC	↓
		FEV_1/FVC	↔
Kusaka et al. 1986a Retrospective cohort; 42 workers (8 females, 34 males; mean age 42 years; employed for 3 years) from the hard metal shaping industry and 84 unexposed workers (16 females, 68 males; mean age 42 years) (Japan)	Current exposure, mean 7-hour exposure concentration (range): 0.085 (0.017–0.610) mg Co/m^3 Historical exposure, mean (range) over the past 3 years: 0.126 (0.006–0.610) mg Co/m^3	Lung function FVC, FEV_1, PEF, MMEF, Vmax	↔ (historical: workers versus referents)
		FEV_1/FVC[a]	↓ (historical: workers versus referents)
Kusaka et al. 1986b Retrospective cohort; 319 workers (sex and employment duration not reported; range of mean ages 31–61 years, employed 5–17 years) from the hard metal industry (Japan)	Measured air concentration during 3-year period, range of mean values: 0.003–1.292 mg Co/m^3	Occupational asthma	↑
		Abnormal chest radiograph (diffuse shadows of category 1 or greater)	↑
		Interstitial pneumonitis	↔

Table 2-4. Results of Epidemiological Studies Evaluating Exposure to Cobalt and Respiratory Effects

Reference, study type, and population	Exposure concentration	Outcome evaluated	Result
Linna et al. 2003 Retrospective cohort; 110 exposed male workers (mean age 50.3 years; mean employment 22.1 years) from the cobalt industry and 140 unexposed males (mean age 48.8 years; mean employment 24.7 years) (Finland)	Historical exposure levels, range: 0.01–0.1 mg Co/m^3 Cumulative exposure, median: 0.6 mg Co-year/m^3	Suspected asthma (general or work-related)	↑ (workers versus referents)
		Respiratory symptoms (phlegm, cough with wheezing, dyspnea with wheezing, breathlessness on exertion)	↑ (workers versus referents)
		Chest x-rays	↔ (workers versus referents)
		Lung function	
		FVC	↔ (workers versus referents)
		FEV_1	↓ (smokers: workers versus referents) ↔ (nonsmokers: workers versus referents)
		MEF_{50} or MEF_{25}	↓ (smokers: workers versus referents) ↔ (nonsmokers: workers versus referents)
		DLCO or DLCO/VA	↔ (workers versus referents)
Meyer-Bisch et al. 1989 Cross-sectional; 425 workers (351 men, 74 women) exposed to hard metal dusts and 88 unexposed workers (69 men, 19 women) from three hard metal plants (France)	Current air concentration, mean, mg Co/m^3: Finishing work area (“hard” carbide): 0.030–0.210 Powder, presses, and forming work areas (“soft” carbide): 0.030–0.272 Maintenance workers (men only): not reported	Lung function	
		Restrictive syndrome[b]	↔ (exposed versus unexposed)
		Obstructive syndrome[c]	↔ (exposed versus unexposed)
		Bronchial hyperreactivity[d]	↔ (men) ↑ (women; hard versus unexposed)
		Abnormal diffusing capacity (altered TCO_{ss})	↑ (men; soft versus unexposed) ↑ (women; hard versus unexposed)
		FuaCO	↓ (men; soft or hard versus unexposed) ↓ (women; soft or hard versus unexposed)

Table 2-4. Results of Epidemiological Studies Evaluating Exposure to Cobalt and Respiratory Effects

Reference, study type, and population	Exposure concentration	Outcome evaluated	Result
			↓ (nonsmokers; soft or hard versus unexposed
		FVC, FEV_1, FEV_1/FVC FEF	↔ (exposed versus unexposed; all or stratified by smoking status)
		Respiratory symptoms	
		Cough, sputum	↑ (men; soft versus unexposed) ↑ (women; hard versus unexposed)
		Dyspnea, bronchitis, rhinopharyngeal symptoms, asthma	↔ (exposed versus unexposed)
		Abnormal pulmonary radiograph	↑ (men; soft versus unexposed) ↔ (women)
Nemery et al. 1992 Cross-sectional; 194 workers (28 females,166 males, range of mean ages 25.4–32.8 years; employment duration not reported) from the diamond polishing industry and 59 unexposed workers (13 females, 46 males, range of mean ages 21.1–28.2 years) (Belgium)	Current air concentration, mean, mg Co/m^3: Low: 0.0053 High: 0.0151 Current urine cobalt level, mean, µg/g creatinine: Low: 7.0 High: 20.5	Respiratory symptoms	
		Upper airway irritation, cough, phlegm	↑ (high exposure versus referents) ↔ (low versus referents)
		Dyspnea, wheezing	↔ (workers versus referents)
		Lung function	
		FVC, FEV_1, MMEF, PEF	↓ (high exposure versus referents) ↔ (low versus referents)
		FEV_1/FVC	↔ (workers versus referents)
Rehfisch et al. 2012 Cohort; 582 workers (362 men, 220 women) from a hard metal plant; employed at least 1 year with at least two sets of spirometry data (Sweden)	Exposure categories (job-exposure matrix), mg Co/m^3: 0: Unexposed 1: <0.00099 2: >0.001–<0.049 3: >0.05	Change in lung function over time	
		FVC, FEV_1, FEV_1/FVC	↔ (smokers) ↔ (nonsmokers)

Table 2-4. Results of Epidemiological Studies Evaluating Exposure to Cobalt and Respiratory Effects

Reference, study type, and population	Exposure concentration	Outcome evaluated	Result
Roto 1980 Cross-sectional with nested case-control; 147 workers from a cobalt plant and 60 unexposed workers (duration of employment not reported); subjects included 21 clinically diagnosed cases of asthma and 55 randomly selected workers without asthma (Finland)	Historical concentrations, mg Co/m^3: Cobalt roasting building: Cobalt dust: 8–19 Soluble cobalt: 0.05–0.1 Cobalt packing area in leaching building: Cobalt dust: 0.01–0.1	Risk of asthma (case-control analysis)	↑ (exposed versus unexposed)
		Bronchial hyperreactivity[e] (cross-sectional analysis)	↔ (exposed versus unexposed)
Roto 1980 Cross-sectional; 224 male cobalt workers (mean age 33.6 years; mean of 7.3 years of employment) and 151 unexposed male referents (mean age 33.7 years) (Finland)	Historical concentrations, mg Co/m^3: Cobalt roasting building: Cobalt dust: 8–19 Soluble cobalt: 0.05–0.1 Cobalt packing area in leaching building: Cobalt dust: 0.01–0.1	Symptoms of chronic bronchitis (chronic cough, phlegm, wheezing)	↔ (workers versus referents, adjusted for smoking)
		Lung function FVC, FEV_1, FEV (%), Vmax	↔ (workers versus referents)
Swennen et al. 1993 Cross-sectional; 82 male workers (mean age 33 years; mean employment duration 8 years) from a cobalt refinery and 82 referents (mean age 38 years) (Belgium)	Current air concentration, mean: 0.125 mg Co/m^3 Current pre-shift urine cobalt level, median (µg/g creatinine): Monday: 22.9 Friday: 44.9 Current post-shift urine cobalt level, median (µg/g creatinine): Monday: 44.1 Friday: 72.4	Respiratory symptoms Dyspnea, wheezing (self-reported)	↑ (workers versus referents; smokers) ↔ (workers versus referents; nonsmokers)
		Cough or sputum in cold season, rhinitis, hay fever	↔ (workers versus referents)
		Lung function FVC, FEV_1, PEF, MEF_{50}, MEF_{75}	↔ (workers versus referents)

Table 2-4. Results of Epidemiological Studies Evaluating Exposure to Cobalt and Respiratory Effects

Reference, study type, and population	Exposure concentration	Outcome evaluated	Result
Verougstraete et al. 2004 Prospective cohort; 122 workers (mean age 43.7 years; mean employment duration 205.8 months) from a cobalt plant (Belgium)	Historical post-shift urine cobalt concentrations in workers from different job areas; µg/g creatinine[f] Job area Dry Wet Mixed 1992 275 20 35 1997 150 25 20 2001 65 15 10	Change in lung function (1988–2001) FVC FEV_1	↔ ↓ (smokers only)
Walters et al. 2012 Cross-sectional; 62 metal workers (toolmakers/grinders; 54 males, 8 females) involved in the manufacturing of precision engineering parts (mean age 39.5 years) (United Kingdom)	Current urine cobalt concentration, mean: 0.6 µg/g creatinine (0.6 µg/L) By diagnosis: Occupational asthma: 1.6 µg/L Non-occupational asthma: 0.4 µg/L Occupational rhinitis: 1.2 µg/L Asymptomatic: 0.4 µg/L	Occupational rhinitis	↔ (urinary cobalt)
		Occupational asthma	↑ (urinary cobalt)

[a]FEV_1/FVC reported as $FEV_{1\%}$ by the study authors.
[b]Restrictive syndrome is defined as a normal FEV_1/VC ratio with VC and TLC <80% of predicted values.
[c]Obstructive syndrome is defined as normal VC with FEV_1 or MMEF <80% of predicted values.
[d]Bronchial reactivity defined as a change of ≥10% in FEV_1 and/or ≥15% in $FEF_{25–75}$ after a challenge (exposure to acetylcholine at 100 mg/L via nebulizer for 3 minutes).
[e]Bronchial reactivity defined as decrease of ≥15% in FEV_1 after a challenge (exposure to 1% methacholine vapor via nebulizer; 1, 5, and 15 inhalations at intervals of 2 minutes).
[f]Estimated from Figure 1 in Verougstraete et al. (2004) using WebPlotDigitizer.

↑ = association; ↓ = inverse association; ↔ = no association; $FEF_{75/50/25}$ = forced expiratory flow at 75, 50, and 25% of the vital capacity, respectively; FEV_1 = forced expiratory volume in 1 second; FEV % = (FEV_1/FVC) x 100; FuaCO = alveolar capillary fractional uptake; FVC = forced vital capacity; DLCO = diffusing capacity of the lungs for carbon monoxide; DLCO/VA = specific diffusion capacity; $MEF_{75/50/25}$ = flow rate at 75, 50, and 25% of the vital capacity, respectively; MMEF = maximal mid expiratory flow; PEF = mean peak expiratory flow rate; T = tertile; TCO_{ss} = total steady-state carbon monoxide uptake; TLC = total lung capacity; TWA = time-weighted average; VA = alveolar volume; VC = vital capacity; Vmax = reduction in flow

Impaired lung function and increased complaints of respiratory symptoms were frequent findings in occupational cohorts of workers chronically exposed to cobalt. Decreased lung function, specifically a 2.7% decrease in forced expiratory volume in 1 second (FEV_1), was observed in 42 cobalt metal workers exposed to mean cobalt metal exposure levels of 0.126 mg Co/m^3 over the past 3 years, compared to unexposed referents (Kusaka et al. 1986a). Similarly, Belgian diamond-cobalt tool manufacturers

exposed to cobalt concentrations ranging from 0.0152 to 0.1355 mg Co/m^3 showed decreased FVC, FEV_1, and peak expiratory flow (PEF), compared to unexposed referents (Gennart and Lauwerys 1990). In workers who smoked only, decreased expiratory flow at 75% vital capacity was also observed compared to unexposed referents (nonsmokers). While cumulative exposure estimates were not determined, analysis showed that lung function parameters were negatively correlated with duration of employment. Findings were not correlated with current urinary cobalt levels. Verougstraete et al. (2004) also reported an association between cobalt exposure and a mild, but measurable, excess decline in FEV_1 over a 13-year follow-up period (1988–2001) in a cohort of workers from a Belgian cobalt production plant; however, this association was only observed in workers who smoked. No associations between cobalt exposure and decrements in FVC were observed. The highest airborne cobalt levels were measured in the early 1990s (1 mg Co/m^3) and declined thereafter (to an unspecified level). Mean urinary cobalt levels for different job areas ranged from approximately 35–275 µg/g creatinine in 1992, falling to approximately 10–65 µg/g creatinine in 2001 (Verougstraete et al. 2004). Impaired respiratory function was also reported in workers exposed to cobalt concentrations ranging from 0.01 to 0.1 mg Co/m^3, including decreased FVC, FEV_1, and maximal expiratory flow at 25 and 50% vital capacity (Linna et al. 2003). Reported respiratory symptoms in workers included phlegm, cough with wheezing, dyspnea with wheezing, and breathlessness on exertion. Calculated median cumulative cobalt exposure levels were 0.6 mg-year Co /m^3. No associations between cobalt exposure and changes in lung function over time, as measured by FVC, FEV_1, or FEV_1/FVC, were observed in workers employed in a Swedish hard metal plant for at least 1 year with at least two sets of spirometry data (Rehfisch et al. 2012). Mean exposure estimates were not reported; however, low-, medium-, and high-exposure groups (based on a job-exposure matrix) were defined as <0.00099, >0.001–<0.049, and >0.05 mg Co/m^3, respectively. However, a major limitation of this study was lack of a consistent latency between spirometry measurements and a limited number of measurements per study subject.

In a cross-sectional study from the diamond polishing industry, workers exposed to cobalt at a concentration of 0.0151 mg Co/m^3 showed decreases in several lung function parameters by approximately 5%, including FEV_1, FVC, maximal mid expiratory flow (MMEF), and PEF, compared to unexposed referents; workers exposed to a mean lower concentration of 0.0053 mg Co/m^3 did not show impaired lung function (Nemery et al. 1992). The exposed workers also exhibited increased incidence of cough (11/91), wheezing (4/91), and upper airway irritation (40/91). Among the workers subjected to work-related exposure, upper airway effects were seen in 30% of controls, 26% of low-exposure individuals, and 43% of high-exposure individuals. Work-related cough was not observed in the control subjects but was observed in 4% of low-exposure individuals and 12% of high-exposure individuals.

While the respiratory effects appeared at a greater rate in individuals who were exposed to higher concentrations of cobalt, the study collected, but did not report, the smoking status of this treatment group. There was no correlation between cobalt exposure and respiratory effects on an individual level within this group; correlations occurred only on a group level (low, high, and control). Therefore, smoking may have caused or contributed to the increase in cough in the 12% of individuals in the higher concentration exposure group. Personal and area air samples correlated well based on results of monitoring a set of individuals in each primary work area; correlations occurred on a group level (low, high, and control).

Additional cross-sectional studies reported associations between current estimates of cobalt exposure and lung function and/or respiratory symptoms. In Malaysian factory workers exposed to 8-hour cobalt concentrations ranging from 0.01 to 0.19 mg Co/m^3, exposure to cobalt was associated with significant increases in chronic phlegm and decreases in FVC and FEV_1 (Hamzah et al. 2014). In Belgian cobalt refinery workers exposed to mean concentrations of 0.125 mg Co/m^3, self-reported dyspnea and wheezing were increased in workers who smoked compared to unexposed referents who smoked; this effect was not observed in nonsmokers (Swennen et al. 1993). Lung function parameters in all workers were comparable to referents in this cohort. No changes in FVC, FEV_1, or end respiratory flow (FEF) were observed in French hard metal workers exposed to cobalt dust concentrations of 0.030–0.272 mg Co/m^3 (Meyer-Bisch et al. 1989). However, decreased alveolar capillary fractional uptake of carbon monoxide and altered diffusing capacity (i.e., decreased steady-state carbon monoxide uptake) were observed along with increased incidence of self-reported cough and sputum in exposed workers, compared to unexposed workers. Women workers from the finishing work area (exposed to "hard" carbides) also showed increased bronchial reactivity, while male workers from the powder, presses, and forming work areas (exposed to "soft" carbides) showed increased incidence of abnormal pulmonary radiographs (Meyer-Bisch et el. 1989). In other cross-sectional studies, no associations between occupational exposure to cobalt and prevalence of respiratory symptoms, abnormal chest x-ray, or impaired lung function were reported at mean air concentrations ranging from 0.0017 to 19 mg Co/m^3 (Andersson et al. 2020; Deng et al. 1991; Roto 1980) or urinary concentrations of 0.6 µg/g creatinine (Walters et al. 2012). In some cases, measurements of lung function were improved in cobalt-exposed workers, compared to unexposed referents from blue-collar industries (Deng et al. 1991).

Findings of adverse lung effects from cohort and cross-sectional studies of cobalt-exposed workers are supported from case reports of lung disease and damage in cobalt-exposed workers. Demedts et al. (1984) reported five cases of "cobalt lung" in diamond polishers. Unlike hard metal workers, mineralogic

analysis of lung tissue in these patients found that cobalt was not alloyed to carbides of hard metals, therefore concluding that cobalt was the only toxic agent. Cases of "cobalt lung" presented with various respiratory complaints (rhinitis, cough, chest tightness, dyspnea), impaired lung function (restrictive deficits), fibrosing alveolitis, mononuclear cell infiltrate, interalveolar desquamation, multinucleated giant cells, and/or centrilobular fibrosis. Cessation of exposure relieved subjective complaints and partially improved lung function. Cobalt exposure levels were not measured; however, the study authors noted that in some cases, workshops did not have adequate ventilation.

Evidence for an association between occupational cobalt exposure and increased risk of asthma is mixed. In retrospective cohorts, increased prevalence of asthma in workers was observed at exposure levels of cobalt ranging from 0.003 to 1.292 mg Co/m^3; findings were associated with abnormal chest radiographs in some cases (Kusaka et al. 1986b; Linna et al. 2003). Walters et al. (2012) found that current urinary cobalt concentrations were significantly higher in a cross-sectional study of workers with probable or definite occupational asthma, compared to asymptomatic workers. However, no association was observed between estimated cumulative exposure and the prevalence of asthma diagnosis in a cohort of 72 hard metal workers employed for an average of 11 years (Andersson et al. 2020).

Sauni et al. (2010) conducted a review of cases of occupational asthma in cobalt plant workers in Finland from 1967 to 2003, where the mean air concentrations of cobalt in different departments ranged from 0.03 to 0.15 mg/m^3. Until 1987, cobalt was being produced from pyrite ore concentrate, which led to co-exposures with irritant gases like sulfur dioxide and ammonia (known respiratory irritants) (Andersson et al. 2006; ATSDR 1998; Huber and Loving 1991). Starting in 1987, and in subsequent years, cobalt was instead predominantly produced using byproducts of the metallurgic industry as raw material, which eliminated the co-exposure to the irritant gases. After this switch in production method, the incidence of asthma decreased to only 1 case between 1987 and 2003 compared to 21 cases between 1967 and 1987 (Sauni et al. 2010). Therefore, it is likely that the health effects observed in Sauni et al. (2010) were due to the co-exposure to sulfur dioxide and ammonia and not cobalt alone. In a small case-control study of 21 cases of asthma and 55 controls, the risk of asthma was increased in subjects with a work history of intermittent or regular employment in a cobalt plant with exposure to cobalt dust ranging from 0.01 to 19 mg Co/m^3 (depending on work area), compared to unexposed subjects (Roto 1980). Similar to Sauni et al. (2010), findings in this study are also confounded by co-exposure to irritant gases (ammonia, hydrogen sulfide, sulfur dioxide).

Al-Abcha et al. (2021) reported a case-series of 35 work-related cases of asthma between 1988 and 2017 in the state of Michigan that were attributed to cobalt exposure, prompting industrial hygiene surveys of 21 workplaces and interviews of 498 exposed coworkers of asthma cases. Most workers were involved in the hard metal industry. Of these workplaces, six had exposure levels above the Michigan permissible exposure limit (PEL) of 0.05 mg Co/m^3 and five had cobalt air levels above the federal PEL of 0.1 mg Co/m^3. Respiratory symptoms, including daily or weekly chest tightness, shortness of breath, wheezing or new onset of asthma since employment began, were reported in 11% of coworkers; however, reported symptoms were not associated with cobalt exposure levels (Al-Abcha et al. 2021). Other potential occupational exposures in these cases where not addressed. More convincing evidence for cobalt-induced asthma comes from review of 14 cases of occupational asthma in workers involved in grinding and polishing automotive engine valves containing cobalt between 1996 and 2005 (Walters et al. 2014). Nine of the cases had confirmed sensitization to cobalt chloride. A limited number of cases of occupational asthma associated with cobalt exposure have also been reported in diamond polishers, in which co-exposure to other substances in hard metal does not occur (Gheysens et al. 1985).

Animal studies have consistently observed respiratory effects following inhalation exposure in multiple species, consistent with findings in human studies. Findings show both dose- and duration-dependency, and toxicity differed between administered compounds.

As discussed in Section 2.2 (Death), acute lethality following exposure to high concentrations of cobalt or cobalt concentrations is often due to, and/or associated with, severe pulmonary effects (Palmes et al. 1959; Viegas et al. 2022a). Compounds showing increased "inflammatory" reactivity in the lungs, such as cobalt hydroxide, metal powder, and oxide, were more acutely toxic; however, concentrations associated with inflammatory changes for these compounds were not reported (Viegas et al. 2022a). Following a 30-minute exposure to cobalt hydrocarbonyl (plus oxide/carbonate decomposition products), severe pulmonary irritation was listed as the cause of death for rats that died at concentrations ≥78 mg Co/m^3 (Palmes et al. 1959). Rats that survived exhibited labored breathing and disturbed respiration. Concentration-dependent increases in the incidence of pulmonary edema and gross lung damage (hemorrhage, edema, consolidation, congestion, pleuritis, bronchiectasis, emphysema, or atelectasis) were observed at ≥83 mg Co/m^3 (Palmes et al. 1959).

At sublethal concentrations, evidence of inflammatory reactivity in the lungs following acute-duration exposure was detected as increased neutrophil numbers in bronchoalveolar lavage fluid (BALF) collected from rats 1 day after being exposed to concentrations ≥2.2 mg Co/m^3 as cobalt sulfate for 4 hours (Viegas

et al. 2022a, 2022b). BALF cell viability was also decreased at 4–16 hours after exposure to ≥2.1 mg Co/m^3. No histopathological lesions were noted immediately postexposure, but squamous cell metaplasia of the epiglottis in the larynx was observed in almost all rats exposed to 6.7 mg Co/m^3 and evaluated 16 days postexposure (Viegas et al. 2022a). Increased BALF neutrophils, as well as lactate dehydrogenase (LDH) levels, were also observed in rats intermittently exposed to cobalt tetraoxide at concentrations ≥33.87 mg Co/m^3 for 14 days (Burzlaff et al. 2022a). Observed inflammatory changes in the lungs of rats exposed to poorly soluble cobalt tetraoxide resembled those associated with exposure to inert dust. No histopathological changes were noted at concentrations up to 160.90 mg Co/m^3.

Widespread respiratory damage was consistently observed in rats and mice following intermittent intermediate- or chronic-duration inhalation exposure to cobalt sulfate; the larynx was the most sensitive location in the respiratory tract. Focal squamous metaplasia with inflammatory changes in the larynx, along with elevated BALF neutrophils and LDH levels, were observed in Wistar rats exposed to 0.46 mg Co/m^3 for 28 days (Burzlaff et al. 2022a, 2022b). Various histopathological lesions were observed throughout the respiratory tract epithelium (nasal turbinates, larynx, trachea, and bronchioles) in F344/N rats exposed to 19 mg Co/m^3 and B6C3F1 mice exposed to concentrations ≥1.9 mg Co/m^3 for 16 days, including inflammation, necrosis, hyperplasia, metaplasia, acanthosis, fibrosis, histiocytic infiltration, and/or degeneration (NTP 1991). Increased lung weight was observed in the mice at 19 mg Co/m^3. With longer exposure, respiratory tract lesions were observed at all tested concentrations (≥0.1 mg Co/m^3), including squamous metaplasia of the larynx in rats exposed for 13 or 104 weeks, histiocytic infiltrates in the lung in male mice exposed for 13 weeks, and hyperplasia and metaplasia of various upper and lower respiratory tract tissues, pulmonary fibrosis, and inflammatory changes in lungs in mice exposed for 104 weeks (Behl et al. 2015; Bucher et al. 1990, 1999; NTP 1991, 1998). In rats and mice exposed for 13 weeks and rats exposed for 104 weeks, findings became widespread throughout the respiratory system at higher concentrations, consistent with the 16-day studies and the findings at the low concentration in the chronic-duration mouse study (e.g., elevated lung weights, histiocytic infiltrates of the lung, chronic inflammation of the larynx and lung, alveolar epithelium hyperplasia, bronchiolar epithelium regeneration, pulmonary fibrosis, olfactory epithelium degeneration, and respiratory epithelium squamous hyperplasia/metaplasia in the nose).

As seen in studies with cobalt sulfate, intermediate- and chronic-duration exposure to cobalt metal consistently produced adverse respiratory effects in multiple species. In rats and mice, mild lesions in the respiratory tract were observed after exposure for 16–17 days at all tested concentrations (≥2.5 mg Co/m^3) in both rats and mice (NTP 2014). Findings included minimal cytoplasmic vacuolization of bronchiolar

epithelium and minimal-to-mild nasal lesions (necrosis and/or atrophy of olfactory epithelium; vacuolization of respiratory epithelium) in both species with histiocytic infiltrates in the lungs of male mice. Elevated absolute and/or relative lung weights were observed in female rats exposed to ≥10 mg Co/m^3 for 16 days and male and female mice exposed to ≥5 mg Co/m^3 for 16 days (NTP 2014). At ≥20 mg Co/m^3, rats showed abnormal breathing with lung hemorrhage in males and nasal respiratory epithelium necrosis in females. Mice exposed to ≥20 mg Co/m^3 showed pulmonary fibrosis, olfactory epithelial necrosis, respiratory epithelial metaplasia, and alveolar/bronchiolar karyomegaly. Lesions were observed in both rats and mice following exposure for 14 weeks at all tested concentrations (≥0.625 mg Co/m^3), including chronic active inflammation in lung, pulmonary alveolar proteinosis, and increased relative lung weight in rats and squamous metaplasia of the larynx, cytoplasmic vacuolization of bronchiole epithelium, and alveolar histiocytic cellular infiltration in mice (NTP 2014). Pulmonary hemorrhage was observed in mice exposed to ≥5 mg Co/m^3 for 14 weeks (NTP 2014). Similarly, lesions were observed in both rats and mice following exposure for 105 weeks to ≥1.25 mg Co/m^3 (Behl et al. 2015; NTP 2014). Rats showed lesions in the lungs (hyperplasia and cytoplasmic vacuolization of alveolar/bronchiolar epithelium), nasal turbinate (atrophy), nasal olfactory epithelium (atrophy, hyperplasia, and metaplasia), and nasal respiratory epithelium (cytoplasmic vacuolization and squamous metaplasia). Mice similarly showed lesions of the lungs (alveolar epithelium hyperplasia and proteinosis, bronchiole epithelium hyperplasia) and nose (hyperplasia, metaplasia, necrosis, and atrophy of the olfactory epithelium and nasal turbinate). In mice, elevated lung weights were only observed at higher concentrations, ≥2.5 mg Co/m^3 in males and ≥5 mg Co/m^3 in females (NTP 2014). In other species, a 3-month exposure to 0.1 mg Co/m^3 in pigs decreased respiratory compliance, a metric of mechanical ventilation (Kerfoot 1974). A 17-week intermittent exposure in male rabbits to 0.4 mg Co/m^3 caused inflammation in lungs and accumulation of macrophages; at 2 mg Co/m^3, severe inflammation and edema of the lower lobes of the lung were noted (Johansson et al. 1987).

Findings following repeated inhalation exposure to other forms of cobalt are limited. Inflammatory lesions in the lung and increased cellularity BALF, with decreased percent macrophages and increased percent monocytes, were observed in male rabbits after intermittent exposure to 0.6 mg Co/m^3 as cobalt chloride for 4 months (Johansson et al. 1992). No adverse lung effects were observed in male rabbits similarly exposed to 0.5 mg Co/m^3 as cobalt chloride for 4 months (Johansson et al. 1991). Increased LDH and neutrophil count in BALF and alveolar lipoproteinosis were observed in Wistar rats intermittently exposed to cobalt tetraoxide at concentrations ≥15.05 mg Co/m^3 for 28 days (Burzlaff et al. 2022a). Moderate interstitial fibrosis and interstitial inflammatory cell infiltration were also observed at 59.31 mg Co/m^3 (Burzlaff et al. 2022a). Lifetime intermittent exposure to cobalt oxide at 7.9 mg Co/m^3

caused lung inflammation and emphysema in male ENG:ELA hamsters (Wehner et al. 1977). The "most prominent" finding in the lungs in male albino rats and guinea pigs (unspecified strain) exposed to 9 mg Co/m^3 as cobalt hydrocarbonyl (plus oxide/carbonate decomposition products) intermittently for 3 months was a transient increase in particle-laden macrophages (Palmes et al. 1959), indicative of active clearance of inhaled particles. Palmes et al. (1959) also reported transient evidence of moderate interstitial and peribronchial fibrosis, mild emphysema, and moderate peribronchial lymphoid hyperplasia in exposed animals; however, it is unclear if these findings were observed in both species and at what incidence; therefore, a NOAEL/LOAEL determination could not be made for lung effects from this study.

No studies examined respiratory toxicity in humans following oral exposure to cobalt. In oral studies in rats, no changes in lung weight or histology were observed following exposure to cobalt chloride at a dose of 12.5 mg Co/kg/day for 7 day (Shrivastava et al. 2010), cobalt chloride at doses up to 7.44 mg Co/kg/day for 90 days (Danzeisen et al. 2020a), cobalt chloride at doses up to 18 mg Co/kg/day for 4 months (Holly 1955) or 16.5 mg/kg/day for 13 weeks (Domingo et al. 1984), or cobalt tetraoxide at doses up to 734 mg Co/kg/day for 90 days (Danzeisen et al. 2020a).

No studies were identified that examined respiratory effects in humans or animals following dermal exposure to cobalt.

2.5 CARDIOVASCULAR

A few studies examined cardiovascular effects in humans after occupational inhalation exposure to cobalt (Table 2-5). These studies provided limited and contradictory evidence of cardiovascular toxicity following inhalation exposure to cobalt. Using Doppler analysis, echocardiogram, and electrocardiogram (ECG), Lantin et al. (2013) found no association between urinary cobalt levels or estimated cumulative cobalt exposures and dilated cardiomyopathy (i.e., increased left ventricular volume) in a study of 256 male cobalt refinery workers in Belgium exposed to 0.001–0.108 mg Co/m^3. In fact, the echocardiogram showed an association between increased urinary cobalt and decreased left ventricle volume. The only other observed association was a positive association between urinary cobalt and heart rate (Lantin et al. 2013). In a study of Finnish factory workers and a 6-year follow-up, no differences in ECG findings, heart rate, or blood pressure were associated with historical exposure to concentrations ranging from 0.01 to 1.0 mg Co/m^3 (Linna et al. 2004, 2020). At the initial evaluation, exposed workers did show significant changes in left ventricular relaxation and filling on the echocardiogram, indicating altered diastole (Linna et al. 2004). However, no abnormalities were observed in the echocardiogram in

workers at the 6-year follow-up (Linna et al. 2020). Additionally, at follow-up, prevalences of heart disease, hypertension, and stroke were similar in exposed and unexposed workers (Linna et al. 2020).

Table 2-5. Results of Epidemiological Studies Evaluating Exposure to Cobalt and Cardiovascular Effects

Reference, study type, and population	Exposure concentration	Outcome evaluated	Result
Lantin et al. 2013 Cohort with cross-sectional analysis; 256 male workers (median age 46 years; median employment 12.42 years), including 237 active workers and 19 retired workers from the cobalt industry (Belgium)	Measured air concentration (2007), range: 0.001–0.108 mg Co/m^3 Current urine cobalt levels, median (µg/g creatinine): Day of ECG: 3.90 Day of ECHO: 3.95 Cumulative exposure (IEI), median (µg/g creatinine x years): ECG: 106.72 ECHO: 107.25	Doppler	↔ (current, cumulative)
		ECG	
		Heart rate	↑ (current urine)
		ECHO	
		LVIDd	↓ (current urine)
		LVIDs	↓ (current urine)
		LV mass	↓ (current urine)
Linna et al. 2004 Cohort; 203 exposed male workers (median age 45 years; median employment 20 years) from the cobalt industry and 94 unexposed males (median age 44 years; median employment 25 years) (Finland)	Historical exposure levels, range: 0.01–1.0 mg Co/m^3 Cumulative exposure, median: 0.18 mg Co-year/m^3	Abnormal ECHO[a]	↑ (workers versus referents)
		ECG	↔ (workers versus referents)
		Blood pressure	↔ (workers versus referents)
		Heart rate	↔ (workers versus referents)

Table 2-5. Results of Epidemiological Studies Evaluating Exposure to Cobalt and Cardiovascular Effects

Reference, study type, and population	Exposure concentration	Outcome evaluated	Result
Linna et al. 2020 Cohort; 93 exposed male workers (median age 56 years; median employment 31 years) from the cobalt industry and 49 unexposed males (median age 56 years; median employment 32 years) (Finland) *Follow-up to Linna et al. (2004)*	Historical exposure levels, range: 0.01–1.0 mg Co/m^3 Cumulative exposure, median: 0.55 mg Co-years	ECHO	↔ (workers versus referents)
		ECG	↔ (workers versus referents)
		Blood pressure	↔ (workers versus referents)
		Heart rate	↔ (workers versus referents)
		Prevalence of heart disease, hypertension, and stroke	↔ (workers versus referents)

[a]Increased left ventricular isovolumetric relaxation time and deceleration time of the velocity of the early rapid filling wave.

↑ = association; ↓ = inverse association; ↔ = no association; ECG = electrocardiogram; ECHO = echocardiogram; IEI = integrated exposure index; LV = left ventricle; LVIDd = left ventricular diameter at diastole; LVIDs = left ventricular diameter at systole

There is very limited evidence from animal studies suggesting altered cardiac function following inhalation exposure to cobalt. Intermittent exposure to cobalt metal at 0.1 mg Co/m^3 for 3 months in pigs caused a 14% increase in heart rate, a 38% decrease in QRS amplitude, and ECG abnormalities that may reflect ventricular impairment (Kerfoot 1974). In rodents, no exposure-related changes in heart weight or histology were observed in F344/N rats or B6C3F1 mice following exposure to cobalt sulfate at concentrations up to 75.7 mg Co/m^3 for 16 days, 11.4 mg Co/m^3 for 13 weeks, or 1.11–1.14 mg Co/m^3 for 105 weeks (Behl et al. 2015; Bucher et al. 1990, 1999; NTP 1991, 1998) or in F344/N rats or B6C3F1 mice following exposure to cobalt metal at concentrations up to 5 or 10 mg Co/m^3 for 14 weeks, respectively, or 5 mg Co/m^3 for 105 weeks (Behl et al. 2015; NTP 2014). No exposure-related histopathological changes in the heart were observed in Wistar rats intermittently exposed to cobalt tetraoxide at concentrations up to 59.31 mg Co/m^3 for 28 days (Burzlaff et al. 2022a).

As discussed in Section 2.2 (Death), an increase in lethal cardiomyopathy was observed in the mid-1960s in people who heavily and routinely consumed beer containing ethyl alcohol with cobalt sulfate added as a foam stabilizer (Alexander 1969, 1972; Bonenfant et al. 1969; Kesteloot et al. 1968; Morin et al. 1967, 1971; Sullivan et al. 1969). So-called "beer-cobalt cardiomyopathy" was observed in drinkers who

consumed large volumes of beer daily (approximately 8–30 pints of beer each day), resulting in an average daily cobalt intake of 0.04–0.14 mg Co/kg/day (Alexander 1969, 1972; Morin et al. 1971, n=28). The cardiomyopathy was characterized by sinus tachycardia, left ventricular failure, cardiogenic shock, diminished myocardial compliance, absence of a myocardial response to exercise or catecholamine, enlarged heart, pericardial effusion, and extensive intracellular changes (changes in the myofibers, mitochondria, glycogen, and lipids). The beer-cobalt cardiomyopathy appeared to be similar to alcoholic cardiomyopathy, but the onset of beer-cobalt cardiomyopathy was very abrupt, suggesting that cobalt may have an etiological contribution. However, due to confounding by potential cardiac damage from alcohol abuse combined with concurrent protein-poor diets, the role of cobalt in cardiomyopathy is unclear. No exposure-related changes were found in echocardiograms of 10 volunteers given low-dose cobalt supplements (mean dose of 0.013 mg Co/kg/day) for 3 months (Tvermoes et al. 2014).

Animal studies indicate that acute-duration oral exposure to cobalt can alter cardiovascular function and cause structural heart damage in rats; however, observed effects were sometimes inconsistent between studies. Elevated systolic (45%), diastolic (60%), and mean arterial (50%) blood pressure were observed in Wistar rats exposed to 35 mg Co/kg/day as cobalt chloride in drinking water for 2 weeks (Ajibade et al. 2017). In contrast, decreased systolic (17%), diastolic (24%), and mean arterial (21%) blood pressure were observed in Wistar rats exposed to 10 mg Co/kg/day as cobalt chloride in drinking water for 7 days (Akinrinde et al. 2016b). Wistar rats similarly exposed to 10 mg Co/kg/day for 2 weeks showed a 12% decrease in systolic blood pressure; no changes were observed in diastolic or mean arterial blood pressure or ECG measurements (Akinrinde et al. 2016a). All three studies qualitatively reported histopathological changes in cardiac tissue, including inflammatory changes (cellular infiltration, cardiac cell swelling) and/or areas of myocardial infarction with damage to coronary blood vessels. However, no changes in heart weight or histology were observed in Sprague-Dawley rats exposed to 12.5 mg Co/kg/day for 7 days as cobalt chloride via gavage (Shrivastava et al. 2010). At higher doses (37 mg Co/kg/day as cobalt chloride), histopathological changes to cardiac muscle (atrophy and patchy degeneration of myofibers, loss of striation) as well as distension of interstitium were observed in Wistar rats exposed via gavage for 8 days (Oyagbemi et al. 2020). While there were no control groups included, Wistar rats exposed to a single gavage dose ≥176.6 mg Co/kg as cobalt fluoride or ≥795 mg Co/kg as cobalt oxide in an acute lethality study showed a proliferation of interstitial tissue, swollen muscle fibers, and focal degeneration in the cardiac tissues (Speijers et al. 1982).

Some intermediate-duration studies in rodents also reported altered cardiovascular function and/or structural heart damage; however, findings varied across administered compounds and, in some cases,

were inconsistent across studies evaluating the same compound. Most adverse effects were observed in rodents exposed to cobalt sulfate. Exposure of Sprague-Dawley rats to 8.4 mg Co/kg/day as cobalt sulfate for 24 weeks via the diet resulted in impaired left ventricular systolic and diastolic functions (Haga et al. 1996). Dietary exposure to 8.4 mg Co/kg/day as cobalt sulfate for shorter durations (8 or 16 weeks) did not impair ventricular function or cardiac hemodynamics (Haga et al. 1996; Pehrsson et al. 1991). Daily exposures for 2 months to 26 mg Co/kg/day as cobalt sulfate via gavage resulted in degenerative heart lesions in Wistar rats, including degeneration and swelling in myocardial cells, decreased myofibrils, and ultrastructural mitochondrial damage (Grice et al. 1969). An oral exposure to 20 mg Co/kg/day for 5 weeks as cobalt sulfate in guinea pigs resulted in a 32% increase in relative heart weight, along with pericardial effusion in 45% of the animals and combined endocardial, myocardial, and pericardial lesions in 75% of the samples examined microscopically. Lesions observed included pericarditis, vacuolar degeneration of the myocardium, thickened and edematous endocardium, and mural thrombi. Exposure also caused an increase in relative bradycardia, decrease in QRS voltage, and a significant increase in abnormal ECG findings (Mohiuddin et al. 1970).

A 3-week exposure to 22 mg Co/kg/day as cobalt chloride in male CFY rats resulted in cardiac damage, presenting as multifocal myocytolysis with myofibril degeneration, as well as decreased cardiac output and arterial blood pressure (Morvai et al. 1993). However, no histopathological changes in the heart were observed in Wistar rats exposed to 18 mg Co/kg/day as cobalt chloride via gavage for 4 months (Holly 1955). In Sprague-Dawley rats, no pathological changes were observed in the heart following drinking water exposure to 16.5 mg Co/kg/day as cobalt chloride for 3 months (Domingo et al. 1984) or gavage exposure to doses up to 7.44 mg Co/kg/day as cobalt chloride or 734 mg Co/kg/day as cobalt tetraoxide for 90 days (Danzeisen et al. 2020a).

No studies were identified regarding cardiovascular toxicity in humans or animals after dermal exposure to cobalt.

2.6 GASTROINTESTINAL

No studies were identified that examined gastrointestinal effects in humans after inhalation exposure to cobalt. In laboratory animals, no exposure-related histopathological changes were observed in the esophagus, stomach, duodenum, ileum, jejunum, cecum, colon, or rectum of F344/N rats or B6C3F1 mice following intermittent inhalation exposure to cobalt sulfate at concentrations up to 75.7 mg Co/m^3 for 16 days, 11.4 mg Co/m^3 for 13 weeks, or 1.11–1.14 mg Co/m^3 for 105 weeks (Behl et al. 2015; Bucher et

al. 1990, 1999; NTP 1991, 1998). Similarly, no exposure-related histopathological changes in the gastrointestinal tract were observed in F344/N rats or B6C3F1 mice following exposure to cobalt metal at concentrations up to 5 or 10 mg Co/m^3 for 14 weeks, respectively, or 5 mg Co/m^3 for 105 weeks (Behl et al. 2015; NTP 2014). No exposure-related histopathological changes in the esophagus or stomach were observed in Wistar rats intermittently exposed to cobalt tetraoxide at concentrations up to 59.31 mg Co/m^3 for 28 days (Burzlaff et al. 2022a).

In studies evaluating cobalt chloride supplements in humans as a potential treatment for anemia or hyperthyroidism, doses of 0.36–0.57 mg Co/kg/day resulted in gastric intolerance in some patients, including nausea, vomiting, and constipation (Duckham and Lee 1976; Holly 1955; Paley et al. 1958). Some patients stopped treatment due to severity of effects.

There is limited evidence that acute-duration oral exposure to cobalt may result in altered gastrointestinal function and/or damage in animals. Decreased relative small intestine weight as well as qualitatively reported histopathological damage to the intestine, including significant depletion of absorptive epithelial cells, were observed in Wistar rats exposed to 10 mg Co/kg/day as cobalt chloride via drinking water for 7 days (Akinrinde et al. 2016c). Salami et al. (2023) found that exposure to ≥68 mg Co/kg/day as cobalt chloride via drinking water for 8 days increased cryptal depth in the small intestine and altered gut motility in Wistar rats, resulting in increased gastric emptying time. Acute-duration oral exposure to cobalt chloride at 37 mg Co/kg/day also altered the overall composition of the gut microbiota in Sprague-Dawley rats; changes were predominantly a relative increase in bacteria from the Verrucomicrobia phylum (Richardson et al. 2018). However, no specific genus within the phylum differed significantly from control. Of the 42 tested genera, only *Allobaculum* from the phylum *Erysipelotrichaceae* was increased significantly. However, the biological adversity of altered composition of the gut microbiota is unknown.

In intermediate-duration oral studies, no histopathological changes in the gastrointestinal system were observed in Sprague-Dawley rats exposed to 16.5 mg Co/kg/day as cobalt chloride in drinking water for 3 months (Domingo et al. 1984), in Sprague-Dawley rats exposed to gavage doses up to 7.44 mg Co/kg/day as cobalt chloride or 734 mg Co/kg/day as cobalt tetraoxide for 90 days (Danzeisen et al. 2020a), or in Wistar rats exposed to 18 mg Co/kg/day via gavage for 4 months (Holly 1955).

No studies were identified regarding gastrointestinal effects in humans or animals after dermal exposure to cobalt.

2.7 HEMATOLOGICAL

Occupational studies evaluating hematological effects in humans from inhalation exposure to cobalt have reported mixed findings (Table 2-6). In a cross-sectional study, mild decreases in red blood cells, hemoglobin, and hematocrit levels and increased total white blood cell counts were observed in 82 refinery workers exposed to 0.125 mg Co/m^3, compared to unexposed referents (Swennen et al. 1993). Other cross-sectional studies in cobalt production or hard metal industries did not find consistent associations between measures of cobalt exposure and red or white blood cell parameters (Hedbrant et al. 2022; Lantin et al. 2011). Andersson et al. (2021) reported some associations between cobalt exposure and alterations in coagulation parameters in a cross-sectional study of 72 workers from the hard metal industry in Sweden. The study authors proposed that observed alterations may be a risk for cardiovascular disease. Positive associations were observed between respirable cobalt dust levels and two of five measures of coagulation (coagulation factor VIII and von Willebrand factor); these factors were not associated with cobalt levels in the blood or urine. A slight inverse association was observed between urinary cobalt levels and serum fibrinogen levels; no associations were observed between fibrinogen levels and cobalt dust levels or cobalt blood levels. No associations were observed between measures of cobalt exposure and other biomarkers of coagulation (plasminogen activator inhibitor-1, D-dimer).

Table 2-6. Results of Epidemiological Studies Evaluating Exposure to Cobalt and Hematological Effects

Reference, study type, and population	Exposure concentration	Outcome evaluated	Result
Andersson et al. 2021 Cross-sectional; 72 workers (9 females, 63 males; mean age 42 years; mean employment 11 years) from the hard metal industry (Sweden)	Current 8-hour TWA, median: 0.0023 mg Co/m^3 Stationary cobalt measurements: Inhalable cobalt, mg Co/m^3: Low: ≤0.0001500 Mid: 0.0001501–0.0004700 High: ≥0.0004701 Total dust cobalt, mg Co/m^3: Low: ≤0.0001200 Mid: 0.0001201–0.0004100 High: ≥0.0004101 Respirable dust cobalt, mg Co/m^3: Low: ≤0.0000594	Coagulation factor VIII	↑ (high versus low stationary: inhalable, total, respirable) ↔ (blood, urine)
		von Willebrand factor	↑ (high versus low stationary: respirable) ↔ (blood, urine)
		Fibrinogen	↓ (high versus low, urine) ↔ (air, blood)
		Plasminogen activator inhibitor-1	↔ (air, blood, urine)

Table 2-6. Results of Epidemiological Studies Evaluating Exposure to Cobalt and Hematological Effects

Reference, study type, and population	Exposure concentration	Outcome evaluated	Result
	Mid: 0.0000595–0.0000665 High: ≥0.0000666 Current cobalt concentrations, Blood, nmol/L: Low: ≤5.20 Mid: 5.21–8.00 High: ≥8.21 Urine, nmol/L: Low: ≤25.56 Mid: 25.57–53.67 High: ≥53.68 Cumulative exposure, range: 0.21–≤0.0870 mg Co-year/m^3	D-dimer	↔ (air, blood, urine)
Hedbrant et al. 2022 Cross-sectional; 72 workers (9 females, 63 males; mean age 42.3 years; mean employment 10.4 years) from the hard metal industry (Sweden)	Current 8-hour TWA, mean: 0.0034 mg Co/m^3; respirator adjusted: 0.0017 mg Co/m^3 Current cobalt concentrations, mean: AM blood: 6.5 nmol/L PM blood: 7.1 nmol/L AM urine: 34 nmol/L PM urine: 44 nmol/L	Total WBC	↔ (air, blood, urine)
		Neutrophils	↔ (air, blood, urine)
		Lymphocytes	↑ (AM urine) ↔ (PM urine, air, blood)
		Monocytes	↔ (air, blood, urine)
		Eosinophils	↔ (air, blood, urine)
Lantin et al. 2011 Cross-sectional; 249 male workers (median age 46 years; median employment 12.27 years), including 230 active workers and 19 retired workers (~5 years of retirement) from a cobalt production department (Belgium)	Measured air concentration (2007), range: 0.001–0.108 mg Co/m^3 Current cobalt concentrations, median: Blood: 0.10 µg/100 mL Urine: 3.90 µg/g creatinine Cumulative exposure, median: 106.09 µg/g creatinine x years	Hemoglobin	↔ (current air, blood, urine; cumulative)
		Hematocrit	↔ (current air, blood, urine; cumulative)
		WBC count	↔ (current blood, urine; cumulative)

Table 2-6. Results of Epidemiological Studies Evaluating Exposure to Cobalt and Hematological Effects

Reference, study type, and population	Exposure concentration	Outcome evaluated	Result
Swennen et al. 1993 Cross-sectional; 82 male workers (mean age 33 years; mean employment duration 8 years) from a cobalt refinery and 82 referents (mean age 38 years) (Belgium)	Current air concentration, mean: 0.125 mg Co/m^3 Current pre-shift urine cobalt level, median (µg/g creatinine): Monday: 22.9 Friday: 44.9 Current post-shift urine cobalt level, median (µg/g creatinine): Monday: 44.1 Friday: 72.4	RBC count	↓ (workers versus referents)
		Hemoglobin	↓ (workers versus referents)
		Hematocrit	↓ (workers versus referents)
		WBC count	↑ (workers versus referents)
		Platelets	↔ (workers versus referents)

↑ = association; ↓ = inverse association; ↔ = no association; RBC = red blood cell; TWA = time weighted average; WBC = white blood cell

In rodents, most inhalation studies reported increases in erythrocyte (red blood cell) count, hemoglobin, and/or hematocrit following intermediate-duration exposure. Mild increases in hemoglobin levels were observed in albino rats (10%) and guinea pigs of an unspecified strain (5%) following intermittent exposure to 9 mg Co/m^3 as cobalt hydrocarbonyl (plus oxide/carbonate decomposition products) for 3 months; no other red cell parameters were measured (Palmes et al. 1959). Polycythemia (as defined by the study authors) was reported in male and female F344/N rats exposed to ≥1.11 and ≥3.78 mg Co/m^3, respectively, as cobalt sulfate for 13 weeks; hematological changes included 4–32% increases in erythrocyte count, hemoglobin, and hematocrit levels (Bucher et al. 1990; NTP 1991). Female rats also showed increased reticulocytes at 11.4 mg Co/m^3. In B6C3F1 mice similarly exposed to cobalt sulfate, no consistent, exposure-related changes in red blood cell parameters were observed at concentrations up to 11.4 mg Co/m^3 (Bucher et al. 1990; NTP 1991). Increased erythrocyte parameters (hematocrit, hemoglobin, and/or erythrocyte count) were also observed following a 14-week exposure to cobalt metal at concentrations ≥0.625 mg Co/m^3 in male F344/N rats, ≥1.25 mg Co/m^3 in female F344/N rats, and 10 mg Co/m^3 in male and female B6C3F1 mice (NTP 2014). Reticulocytes were also elevated in male and female rats at 5 and ≥2.5 mg Co/m^3, respectively. In a study with cobalt tetraoxide, no exposure-related hematological effects were observed in Wistar rats following intermittent exposure to concentrations up to 59.31 mg Co/m^3 for 28 days (Burzlaff et al. 2022a). No hematological effects were seen in pigs after a 3-month exposure to cobalt metal (Kerfoot 1974).

Additional hematological effects were occasionally noted in rodents following intermediate-duration inhalation. In white cell differentials, the percent of monocytes was decreased and the percent of basophils was increased in albino rats exposed to 9 mg Co/m^3 as cobalt hydrocarbonyl (plus oxide/ carbonate decomposition products) for 3 months (Palmes et al. 1959). In similarly exposed guinea pigs, the percent of lymphocytes was decreased and the percent of basophils was increased at 9 mg Co/m^3 (Palmes et al. 1959). Following intermittent exposure to cobalt sulfate for 13 weeks, decreased platelets were observed in male and female F344/N rats at ≥3.78 mg Co/m^3 and in female B6C3F1 mice at 11.4 mg Co/m^3 (NTP 1991). Similarly, following intermittent exposure to cobalt metal for 14 weeks, platelets were decreased in male and female F344/N rats at ≥1.25 mg Co/m^3 and in male B6C3F1 mice at 10 mg Co/m^3 (NTP 2014).

Due to observed effects on the hematological system, cobalt chloride has been evaluated as a potential treatment for anemia. To evaluate the effects, several studies have been conducted in small groups of both healthy and anemic individuals. Acute-duration oral exposure to 1 mg Co/kg/day for 7–14 days induced an average 14% increase in red blood cells in five healthy male volunteers, compared to pre-exposure levels (Davis and Fields 1958). In all five volunteers, red blood cells were outside the clinically normal range at the end of exposure, indicating polycythemia. Red blood cell counts returned to baseline levels (within medical norms) for all individuals 4–9 days after cessation of cobalt administration. This study also evaluated the effect of exposure to 0.8 mg Co/kg/day for 15 days in a single volunteer; no biologically relevant changes in red blood cell counts were observed, while another volunteer (one from the acute-duration study) exposed to 1 mg Co/kg/day for 15 days showed an 18% increase over pre-exposure values (Davis and Fields 1958). When the dose for the single volunteer at 0.8 mg Co/kg/day was increased to 1 mg Co/kg/day for the next 7 days, red blood cell counts increased by 5%. These data suggest that the no-adverse-effect level may be around 0.8 mg Co/kg/day for intermediate-duration oral studies; however, with only a single subject per dose group, this study is of insufficient study design to make that determination. Other volunteer studies did not find clinically adverse hematological effects following exposure to low-dose cobalt chloride supplements for 7–21 days at mean intakes of 0.03 mg Co/kg/day (Hoffmeister et al. 2018) or 31–91 days at mean intakes of 0.013 mg Co/kg/day (Finley et al. 2013; Tvermoes et al. 2014).

Additional studies conducted in anemic patients have evaluated potential therapeutic effects of cobalt. In a controlled human study designed to evaluate the potential for cobalt chloride supplementation to reverse pregnancy-related anemia, no changes in hematological parameters were observed in 20 pregnant women exposed to 0.57 mg Co/kg/day for at least 90 days prior to delivery, compared to 55 unexposed controls

(Holly 1955). However, in anephric patients (with non-functioning kidneys) with anemia, cobalt chloride supplementation for ≥12 weeks to ≥0.16 mg Co/kg/day has been shown to increase hemoglobin levels, eliminating the need for transfusions in some patients (Duckham and Lee 1976; Taylor et al. 1977). Hematological endpoints from these studies are not included in the LSE table since NOAEL/LOAEL determinations could not be made (i.e., elevation of red blood cell parameters in anemic patients would be therapeutic); however, these studies were useful for evaluating potential adverse side effects in other systems (e.g., gastrointestinal, hepatic, etc.).

Acute-duration oral exposure to cobalt has also led to hematological effects in rats. An 8% increase in hematocrit levels was observed in Sprague-Dawley rats following a single gavage exposure to 161 mg Co/kg as cobalt chloride (Domingo and Llobet 1984). Elevated red blood cells (44%), hematocrit (7–25%), and hemoglobin (11–20%) were also increased in Sprague-Dawley rats exposed to cobalt chloride for 7 days at 12.5 mg Co/kg/day via gavage (Shrivastava et al. 2008, 2010). Differential lymphocyte counts also showed increased percentages of granulocytes and monocytes (Shrivastava et al. 2010). In pregnant rats, increased hematocrit (11%), hemoglobin (14%), and reticulocytes (100%) were observed following exposure to 24.8 mg CO/kg/day as cobalt chloride via gavage on GDs 6–15 (Paternain and Domingo 1988).

Intermediate-duration oral exposure to cobalt also consistently induced hematological effects in rats, predominantly related to polycythemia. The most sensitive studies were by Stanley et al. (1947) and Danzeisen et al. (2020a), which reported dose-related increases in red cell parameters in Sprague-Dawley rats following exposure to cobalt chloride at doses of ≥2.5 mg Co/kg/day via capsule for 8 weeks or ≥2.48 mg Co/kg/day via gavage for 90 days, respectively. Stanley et al. (1947) reported increases in erythrocyte numbers and hemoglobin levels starting in week 4 of exposure. By 8 weeks of exposure, respective increases in erythrocytes and hemoglobin levels, compared to control, were 15 and 23% at 2.5 mg Co/kg/day and 26 and 34% at 9.9 mg Co/kg/day; values at 0.6 mg Co/kg/day were comparable to control (Stanley et al. 1947). In the study by Danzeisen et al. (2020a), male rats showed no alterations in hematological parameters at 0.74 mg Co/kg/day; however, at a dose of 2.48 mg Co/kg/day, there were 11, 9, and 10% increases in hemoglobin, erythrocytes, and hematocrit, respectively. Parameters were further altered at 7.44 mg Co/kg/day, showing respective increases of 26, 20, and 24%. While the male rats were more sensitive and showed changes in hematological parameters at lower doses, female rats showed increases of 13 and 10% in hemoglobin and erythrocytes, respectively, at 7.44 mg Co/kg/day (Danzeisen et al. 2020a). In both sexes, erythroid hyperplasia in the bone marrow was observed at ≥2.48 mg Co/kg/day. A satellite group of animals exposed to 7.44 mg Co/kg/day for 90 days followed by a 28-day

recovery period demonstrated reversibility of hematological and bone marrow effects in both male and female rats (Danzeisen et al. 2020a).

Danzeisen et al. (2020a) also examined effects of cobalt tetraoxide on hematological parameters and found that a daily oral dose of 220 mg Co/kg/day increased hemoglobin, erythrocytes, and hematocrit by 10, 10, and 9%, respectively, in male rats; a mild 6% increase in hemoglobin level was observed in female rats. At the highest dose of 734 mg Co/kg/day, males and female rats showed an increase in hemoglobin (25% males and 16% females), erythrocytes (23% males and 13% females), and hematocrit (24% males and 14% females) (Danzeisen et al. 2020a). Bone marrow hyperplasia was not observed in rats following exposure to cobalt tetraoxide.

In other intermediate-duration studies in rats, similar findings were observed. Increase in red blood cells and hemoglobin were observed in Wistar rats exposed to cobalt chloride at 18 Co/kg/day via gavage for 4 months (Holly 1955). Sprague-Dawley rats exposed to 16.5–20 mg Co/kg/day as cobalt chloride for 13–14 weeks in food or drinking water showed increases in red blood cells (41%), hemoglobin (28–31%), hematocrit (29%), and/or packed cell volume (56%) (Corrier et al. 1985; Domingo et al. 1984). Increased red blood cell count, hemoglobin, and hematocrit were also observed in rats (strain not specified) exposed to 10 mg Co/kg/day as cobalt chloride 5 days/week via gavage for 150 days (Murdock 1959). In contrast, a 30-day exposure to 13.8 mg Co/kg/day as cobalt chloride caused a 20% decrease in hemoglobin in male Sprague-Dawley rats (Chetty et al. 1979).

Another study briefly noted “mild and transient polycythemia” in rats exposed to 0.5 mg Co/kg/day as cobalt chloride via gavage for 7 months, with increased hemoglobin and red blood cell levels at 2.5 mg Co/kg/day (Krasovskii and Fridlyand 1971). However, due to the qualitative nature of the reporting, the magnitude (and therefore the biological significance) of the findings cannot be determined. Therefore, a NOAEL/LOAEL determination cannot be made for this study.

In mice, no exposure-related changes in hematocrit were observed in male CD-1 mice exposed to cobalt chloride in drinking water at doses of 58.9 mg Co/kg/day for 7–13 weeks or doses up to 72.1 mg Co/kg/day for 12 weeks (Pedigo et al. 1988). Minimal changes in the levels of blood proteins (transferrin, several haptoglobulins, and ceruloplasmin) were noted in male Swiss mice following 4, 24, and 48 hours of treatment with 140 mg Co/kg as cobalt chloride in the drinking water; however, findings are difficult to interpret due to concurrent decreases in water intake resulting in dehydration (Bryan and Bright 1973). When mice were similarly exposed for 3 or 13 weeks, these mild changes in serum protein

levels were no longer observed (Bryan and Bright 1973). Due to minimal effects of unclear adversity, along with limited endpoints evaluated, this study was not included in the LSE table.

No studies were identified regarding hematological effects in humans or animals after dermal exposure to cobalt.

Acute-duration exposure to cobalt chloride by 10 subcutaneous injections in a controlled exposure human study (9-day gap between two blocks of five consecutive injections) of 18 mg Co/kg/day increased erythropoietin (Taylor et al. 1977). In a human case study of cobalt exposure of unknown origin, Jefferson et al. (2002) found a correlation between serum cobalt and excessive erythrocytosis ($p=0.002$) and packed-cell volume ($r=0.4$, $p=0.01$). Domingo and Llobet (1984) showed that single intraperitoneal injections of cobalt chloride at a dose of 12 mg Co/kg caused a 10% increase in hematocrit levels in Sprague-Dawley rats (Domingo and Llobet 1984). Wistar rats were exposed to a single dose of cobalt chloride by a subcutaneous injection (7 mg Co/kg), which resulted in an approximately 17% increase in excretion of methemoglobin within 3 hours of exposure (Horiguchi et al. 2004). A subcutaneous injection study in Sprague-Dawley rats showed a 29% increase in erythrocyte number and a 38–39% increase in hemoglobin concentration following administration of cobalt chloride at doses of 0.6 or 2.5 mg Co/kg/day for 8 weeks (Stanley et al. 1947). Elevated levels were first observed after 4 weeks of exposure. At the highest dose, 9.9 mg Co/kg/day, slight increases of 8–11% were observed in erythrocyte number and hemoglobin by 2 weeks of exposure; however, all animals died prior to the 4-week analysis (Stanley et al. 1947).

2.8 MUSCULOSKELETAL

No studies were identified regarding toxicity of cobalt on musculoskeletal effects in humans after inhalation, oral, or dermal exposure to cobalt.

No exposure-related histopathological changes were observed in the skeletal bone of F344/N rats or B6C3F1 mice following intermittent inhalation exposure to cobalt sulfate at concentrations up to 75.7 mg Co/m^3 for 16 days, 11.4 mg Co/m^3 for 13 weeks, or 1.11–1.14 mg Co/m^3 for 105 weeks (Behl et al. 2015; Bucher et al. 1990, 1999; NTP 1991, 1998). Similarly, no exposure-related histopathological changes in the musculoskeletal system were observed in F344/N rats or B6C3F1 mice following exposure to cobalt metal at concentrations up to 5 or 10 mg Co/m^3 for 14 weeks, respectively, or 5 mg Co/m^3 for 105 weeks (Behl et al. 2015; NTP 2014). No exposure-related histopathological changes in the femur were observed

in Wistar rats intermittently exposed to cobalt tetraoxide at concentrations up to 59.31 mg Co/m^3 for 28 days (Burzlaff et al. 2022a).

No morphological changes were found in the skeletal muscle of Sprague-Dawley rats exposed to drinking water doses of 16.5 mg Co/kg/day as cobalt chloride for 3 months (Domingo et al. 1984) or gavage doses up to 7.44 mg Co/kg/day as cobalt chloride or 734 mg Co/kg/day as cobalt tetraoxide for 90 days (Danzeisen et al. 2020a).

No studies were identified regarding musculoskeletal effects in animals after dermal exposure to cobalt.

2.9 HEPATIC

No studies were identified regarding hepatic effects in humans after inhalation exposure to cobalt.

The liver is not a consistent or sensitive target of cobalt toxicity following inhalation exposure. Decreased absolute and/or relative liver weights were observed following intermittent exposure to cobalt metal at concentrations ≥2.5 mg Co/m^3 in male F344/N rats and male and female B6C3F1 mice and ≥5 mg Co/m^3 in female F344/N rats for 16–17 days (NTP 2014). However, no exposure-related changes in liver weight were observed in rats or mice of either sex exposed to concentrations up to 5 mg Co/m^3 for 14 or 105 weeks (Behl et al. 2015; NTP 2014). Decreased absolute and relative liver weights were observed in mice exposed to 10 mg Co/m^3 for 14 weeks; rats were not evaluated at this exposure concentration for this duration (NTP 2014). No histopathological lesions were observed at concentrations up to 40 mg Co/m^3 for 16–17 days or 5 mg Co/m^3 (rats) or 10 mg Co/m^3 (mice) for 14 weeks (NTP 2014). After 105 weeks, basophilic foci were observed in male and female rats exposed to 1.25 and 5 mg Co/m^3, respectively; no histopathological liver lesions were observed in similarly exposed mice at concentrations up to 5 mg Co/m^3 (Behl et al. 2015; NTP 2014). No exposure-related changes in serum alkaline phosphatase (ALP), alanine aminotransferase (ALT) or sorbitol dehydrogenase (SDH) were observed in rats or mice at concentrations up to 5 or 10 mg Co/m^3, respectively, for 14 weeks; hepatic clinical chemistry was not evaluated at additional timepoints (NTP 2014). In a similar set of experiments with cobalt sulfate, liver effects were only observed in F344/N rats and B6C3F1 mice that died following exposure to 75 and ≥19 mg Co/m^3, respectively, for up to 16 days; findings at autopsy included congestion and necrosis of the liver (NTP 1991). No changes in liver weight or histology were observed in rats or mice exposed to cobalt sulfate at concentrations up to 11.4 mg Co/m^3 for 13 weeks or 1.11–1.14 mg Co/m^3 for 105 weeks (Behl et al. 2015; Bucher et al. 1990, 1999; NTP 1991, 1998). No

exposure-related changes in hepatic clinical chemistry parameters or liver weight or histology were observed in Wistar rats intermittently exposed to concentrations up to 59.31 mg Co/m^3 as cobalt tetraoxide for 28 days (Burzlaff et al. 2022a). No histological effects on the liver were found in pigs (strain not specified) exposed ≤1.0 mg Co/m^3 as cobalt metal dust intermittently for 3 months (Kerfoot 1974).

In a controlled human study designed to evaluate the potential for cobalt chloride supplementation to reverse pregnancy-related anemia, no adverse changes in serum or urinary markers of liver function were noted in 20 pregnant women exposed to 0.57 mg Co/kg/day for at least 90 days prior to delivery, compared to 55 unexposed controls (Holly 1955). No exposure-related changes were found in hepatic serum clinical chemistry parameters of 10 volunteers given low-dose cobalt chloride supplements (mean dose of 0.013 mg Co/kg/day) for up to 91 days (Finley et al. 2013; Tvermoes et al. 2014).

In acute-duration oral exposure studies in rats, decreased relative liver weight and histopathological alterations were observed at doses ≥8.9 mg Co/kg/day as cobalt chloride in drinking water for 7 days (Akinrinde et al. 2016c; Awoyemi et al. 2017). Observed lesions included evidence of hepatocellular damage, focal necrosis, vascular congestion, and mild infiltration of inflammatory cells; lesions were not observed at 8.2 mg Co/kg/day. While there were no controls included in this study, hyperemia of the liver and cytoplasmic changes in hepatocytes (clumpy cytoplasm located along the cell membrane) were found in Wistar rats exposed once to doses ≥68 mg Co/kg as cobalt fluoride or ≥157 mg Co/kg as cobalt oxide (Speijers et al. 1982). However, no changes in liver weight or histology were observed in Sprague-Dawley rats exposed to 12.5 mg Co/kg/day for 7 days as cobalt chloride via gavage (Shrivastava et al. 2010). No biologically relevant changes in hepatic clinical chemistry were observed in rats following a single oral exposure to 161 mg Co/kg as cobalt chloride (Domingo and Llobet 1984) or a 7-day oral exposure up to 18 mg Co/kg as cobalt chloride (Akinrinde et al. 2016c; Awoyemi et al. 2017; Shrivastava et al. 2010).

Hepatic effects were noted in some intermediate-duration oral exposure studies following exposure to cobalt. A 13-week exposure to 16.5 mg Co/kg/day as cobalt chloride in the drinking water did not result in adverse changes in serum ALT, ALP, or aspartate aminotransferase (AST) in rats (Domingo et al. 1984). Garoui et al. (2011) demonstrated that daily exposure to 20 mg Co/kg/day as cobalt chloride via drinking water for 2 weeks during gestation and 2 weeks during lactation resulted in a 10% decrease in absolute liver weight with evidence of liver injury (infiltration of mononuclear cells and vascular congestion) in maternal Wistar rats. Wistar rats exposed to 25 mg Co/kg/day as cobalt chloride for

60 days showed an increase in relative liver weight by 13%, along with degradation and alteration in the morphology and atrophy of liver cells; alterations in liver biochemistry included a 126% increase in AST and a 122% increase in bilirubin (Mathur et al. 2011). A 4-week exposure to 68 mg Co/kg/day in drinking water as cobalt chloride increased LDH by 3.6-fold; increased ALP, AST, and ALT by 1.7-, 4.5-, and 1.7-fold, respectively; and increased total bilirubin levels by 1.9-fold in Sprague-Dawley rats (Khalil et al. 2020).

In other intermediate-duration oral studies, no hepatic effects were observed. No changes in hepatic function, as measured by bromsulphthalein challenge, were observed in rats exposed to oral doses up to 2.5 mg Co/kg/day for 7 months (Krasovskii and Fridlyand 1971). No histopathological changes in the liver were observed in Sprague-Dawley rats exposed to 16.5 mg Co/kg/day as cobalt chloride in drinking water for 3 months (Domingo et al. 1984), in Sprague-Dawley rats exposed to gavage doses up to 7.44 mg Co/kg/day as cobalt chloride or 734 mg Co/kg/day as cobalt tetraoxide for 90 days (Danzeisen et al. 2020a), or in Wistar rats exposed to 18 mg Co/kg/day via gavage for 4 months (Holly 1955).

No studies were identified regarding hepatic effects in humans or animals after dermal exposure to cobalt.

2.10 RENAL

In a prospective cohort that followed 100 male welders over the course of 2 years, urinary cobalt levels were associated with elevated serum creatinine levels (Wu et al. 2023a). No additional renal endpoints were evaluated in this cohort, and no additional studies evaluating renal endpoints in humans following inhalation exposure to cobalt were identified.

The kidney is not a consistent or sensitive target of cobalt toxicity following inhalation exposure. No exposure-related renal lesions were observed in F344/N rats or B6C3F1 mice following exposure to cobalt metal at concentrations up to 5 and 10 mg Co/m^3 for 14 weeks, respectively, or 5 mg Co/m^3 for 105 weeks (Behl et al. 2015; NTP 2014). However, elevated urinary creatinine levels and decreased urine volume were observed in male and female rats following exposure to concentrations ≥10 and 20 mg Co/m^3 for 16 days, respectively (NTP 2014). After exposure to 5 mg Co/m^3 for 14 weeks, findings included increased serum creatinine in male rats, increased relative kidney weight in female rats, and decreased absolute and relative kidney weights in male and female mice (NTP 2014). Kidney weight, urinalysis, and serum clinical chemistry were not evaluated at 105 weeks. No exposure-related changes in kidney weight or histology were observed in F344/N rats or B6C3F1 mice following intermittent

inhalation exposure to cobalt sulfate at concentrations up to 75.7 mg Co/m^3 for 16 days, 11.4 mg Co/m^3 for 13 weeks, or 1.11–1.14 mg Co/m^3 for 105 weeks (Behl et al. 2015; Bucher et al. 1990, 1999; NTP 1991, 1998). No exposure-related changes were observed in serum creatinine levels in the 13-week studies (not evaluated at other timepoints) (NTP 1991). No exposure-related changes in renal clinical chemistry parameters or kidney weight or histology were observed in Wistar rats intermittently exposed to concentrations up to 59.31 mg Co/m^3 as cobalt tetraoxide for 28 days (Burzlaff et al. 2022a). No histological effects on the kidneys were found in pigs exposed ≤1.0 mg Co/m^3 as cobalt metal for 3 months (Kerfoot 1974).

Changes in renal clinical chemistry and histology have been reported following exposure to high acute oral doses of cobalt compounds. Domingo and Llobet (1984) showed that a single gavage exposure to 161 mg Co/kg as cobalt chloride caused a 68% increase in serum urea in Sprague-Dawley rats, which suggests alterations in renal function; however, no additional renal endpoints were evaluated in this study (Domingo and Llobet 1984). Wistar rats exposed to ≥10 mg Co/kg/day as cobalt chloride via drinking water for 2 weeks showed an increase in serum creatinine and/or serum urea levels (Ajibade et al. 2017; Akinrinde et al. 2016a). Both studies qualitatively reported histopathological changes in renal tissue including inflammatory changes (cellular infiltration) and renal tubular degeneration. Inflammation in the peritubular and perivascular areas, along with focal tubular necrosis, were also observed in Wistar rats exposed to 10 mg Co/kg/day as cobalt chloride in drinking water for 7 days (Akinrinde et al. 2016b). Mild tubular atrophy, necrotizing nephritis, and inflammatory cell infiltrate were observed in Wistar rats exposed to 37 mg Co/kg/day via gavage as cobalt chloride for 8 days (Oyagbemi et al. 2020). While there were no control animals in this study, renal injury, evidenced by swelling and degeneration of the proximal tubules, was observed in Wistar rats exposed once to doses ≥42 mg Co/kg as cobalt fluoride (Speijers et al. 1982). However, no changes in serum clinical chemistry or kidney weight or histology were observed in Sprague-Dawley rats exposed to 12.5 mg Co/kg/day for 7 days as cobalt chloride via gavage (Shrivastava et al. 2010).

No exposure-related changes were found in renal serum clinical chemistry parameters of 10 volunteers given low-dose cobalt chloride supplements (mean dose of 0.013 mg Co/kg/day) for up to 91 days (Finley et al. 2013; Tvermoes et al. 2014). No additional studies evaluating renal endpoints in humans following oral exposure to cobalt were identified.

There is limited evidence for renal effects in rats following intermediate-duration oral exposure to cobalt compounds. Abdel-Daim et al. (2020) showed increased serum urea (105%) and creatinine (137%) in

Sprague-Dawley rats exposed to 8.9 mg Co/kg/day in drinking water as cobalt chloride for 4 weeks; no change in relative kidney weight was observed. Garoui et al. (2012) observed that daily exposure to 20 mg Co/kg/day in drinking water as cobalt chloride for 2 weeks during gestation and 2 weeks during lactation caused vascular congestion, reduction of glomerular space, and infiltration of leukocyte cells between tubules in the kidneys of Wistar rat dams. Additional renal findings included a 35% increase in plasma urea, a 34% decrease in urinary creatinine, 38% decrease in urinary urea, and a slight 8% reduction in relative kidney weight, compared to controls (Garoui et al. 2012).

No exposure-related renal changes were observed in other intermediate-duration oral studies in rodents. After a 13-week exposure to 16.5 mg Co/kg/day as cobalt chloride in the drinking water, no adverse changes in renal clinical chemistry or morphological changes in the kidney were observed in Sprague-Dawley rats; the observed decrease in urine volume was attributable to decreased water intake (Domingo et al. 1984). No changes in kidney weight or histology were observed in Sprague-Dawley rats exposed to gavage doses up to 7.44 mg Co/kg/day as cobalt chloride or 734 mg Co/kg/day as cobalt tetraoxide for 90 days (Danzeisen et al. 2020a) or in Wistar rats exposed to 18 mg Co/kg/day via gavage for 4 months (Holly 1955). No histopathological changes in the kidney were observed in CD-1 mice exposed to 43.4 mg Co/kg/day in drinking water as cobalt chloride for 7–13 weeks (Anderson et al. 1992).

No studies were identified regarding renal effects in humans or animals after dermal exposure to cobalt.

Domingo and Llobet (1984) showed that a single intraperitoneal injection of cobalt chloride at a dose of 12 mg Co/kg did not alter renal function in Sprague-Dawley rats (Domingo and Llobet 1984). Wistar rats were exposed to a single dose of cobalt chloride by a subcutaneous injection (7 mg Co/kg). This acute exposure resulted in an approximately 10-fold transient increase in excretion of methemoglobin within 3 hours from the renal tissues in Wistar rats (Horiguchi et al. 2004). Singh and Junnarkar (1991) examined the effects of intraperitoneal and intravenous injections on both Wistar rats and Swiss-Webster mice and observed that it increased urine volume at various doses of cobalt chloride and cobalt sulfate (Singh and Junnarkar 1991). An intermediate-duration exposure to 1.6 mg Co/kg/day as subcutaneous injections of cobalt nitrate for 4 weeks caused glomerular-tubular nephrosis with degenerative changes and was toxic to the renal tubule cells in albino rats (Hanafy and Soltan 2004).

2.11 DERMAL

Studies in humans evaluating the potential association between exposure to cobalt and dermal effects are limited to two occupational cohort studies and several case reports of dermal exposure. In a cohort of metal factory workers (n=71) exposed to air cobalt concentrations ranging from 0.0001 to 0.019 mg/m^3, there was a high prevalence of self-reported dry skin (42%) and eczema (6–7%) (Wahlqvist et al. 2020). Similarly, increased numbers of skin lesions (eczema, erythema) were identified in a cross-sectional study of 82 workers from a cobalt refinery with mean airborne cobalt concentrations of 0.125 mg/m^3 (range 0.001–7.7 mg/m^3) (Swennen et al. 1993). In both occupational studies, it is likely that any association between cobalt exposure and dermal conditions is due to direct contact with cobalt particles in the air. Dermal exposure to cobalt has also been associated with eczema and contact dermatitis in several case reports (Alinaghi et al. 2020; Krecisz et al. 2009; Laing et al. 2005). Four cases of eczema of the hands, feet, and/or limbs were associated with exposure to objects ranging from 0.01 to >10% cobalt by weight (Alinaghi et al. 2020). Clothing dye containing 0.32 mg/kg cobalt caused pruritic rash in a 20-year-old female (Krecisz et al. 2009). In another case study, exposure to blue paint containing cobalt caused eczema, hives, swelling, and anaphylactic reaction (Laing et al. 2005).

No exposure-related histopathological changes to the skin were observed in F344/N rats or B6C3F1 mice following intermittent inhalation exposure to cobalt sulfate at concentrations up to 75.7 mg Co/m^3 for 16 days, 11.4 mg Co/m^3 for 13 weeks, or 1.11–1.14 mg Co/m^3 for 105 weeks (Behl et al. 2015; Bucher et al. 1990, 1999; NTP 1991, 1998). Similarly, no exposure-related histopathological changes in the gastrointestinal tract were observed in F344/N rats or B6C3F1 mice following exposure to cobalt metal at concentrations up to 5 or 10 mg Co/m^3 for 14 weeks, respectively, or 5 mg Co/m^3 for 105 weeks (Behl et al. 2015; NTP 2014).

No histopathological changes to the skin were observed in Sprague-Dawley rats exposed to gavage doses up to 7.44 mg Co/kg/day as cobalt chloride or 734 mg Co/kg/day as cobalt tetraoxide for 90 days (Danzeisen et al. 2020a).

In an intermediate-duration dermal exposure study where guinea pigs (strain not specified) were exposed to 2.4% cobalt for 18 days, scabs and denuded areas were formed around the area where dicobalt octacarbonyl was applied (Kincaid et al. 1954).

Ulcerations were observed at the site after a single subcutaneous injection of 45 mg Co/kg as dicobalt octacarbonyl in guinea pigs (strain not specified) (Kincaid et al. 1954).

2.12 OCULAR

No studies examined ocular effects in humans following inhalation, oral, or dermal exposure to cobalt.

Ocular irritation (chromodacryorrhea) was observed in F344/N rats and B6C3F1 mice during and after 6-hour exposures to cobalt sulfate at concentrations of ≥19 mg Co/m^3/day; clinical signs worsened over the 16-day exposure period (NTP 1991). This finding is likely due to direct ocular exposure to cobalt particles in the air, rather than a systemic effect of inhalation exposure.

No histological lesions were reported in the eyes of F344/N rats intermittently exposed to cobalt metal at concentrations up to 5 mg Co/m^3 for 14 or 105 weeks (Behl et al. 2015; NTP 2014). Similarly, no histological lesions were reported in the eyes of B6C3F1 mice intermittently exposed to cobalt metal at concentrations up to 10 mg Co/m^3 for 14 weeks or 5 mg Co/m^3 for 105 weeks (Behl et al. 2015; NTP 2014).

No ophthalmological changes were observed in Sprague-Dawley rats exposed to cobalt chloride via gavage at 12.5 mg Co/kg/day for 7 days (Shrivastava et al. 2010). No ophthalmological or histopathological changes were observed in Sprague-Dawley rats exposed to gavage doses up to 7.44 mg Co/kg/day as cobalt chloride or 734 mg Co/kg/day as cobalt tetraoxide for 90 days (Danzeisen et al. 2020a).

2.13 ENDOCRINE

Thyroid. Limited data from occupational studies do not provide consistent evidence of alterations in serum thyroid hormone levels following inhalation exposure to cobalt or cobalt compounds (Table 2-7). In a cohort of 249 cobalt refinery workers in Belgium, Lantin et al. (2011) found no association between current blood or urine levels or estimated cumulative cobalt exposures and serum thyroxine (free or total T4), triiodothyronine (free or total T3), or thyroid stimulating hormone (TSH). Measured air concentrations ranged from 0.001 to 0.108 mg/m^3. In contrast, Swennen et al. (1993) reported a significant reduction in serum T3 in a cohort of 82 workers from Belgium occupationally exposed to cobalt oxides, cobalt salts, and cobalt metal at a mean air concentration of 0.125 mg/m^3, compared to

unexposed referents. No changes were observed in serum T4 or TSH levels. A study of Danish porcelain workers found alterations in serum thyroid hormone levels in workers utilizing semi-soluble cobalt dye (cobalt-zinc silicate), but not in workers utilizing insoluble cobalt dye (cobalt aluminate), despite measured air cobalt levels of 0.05 mg Co/m^3 (Prescott et al. 1992). Compared to unexposed referents, workers exposed to cobalt-zinc silicate showed elevated levels of serum total and free T4 but no change in serum T3 or TSH levels. It is unclear if findings in workers exposed to cobalt-zinc silicate are attributable to increased solubility of this form of cobalt, as these workers showed elevated urinary levels of cobalt and workers exposed to cobalt aluminate did not, or due to co-exposure with zinc.

Table 2-7. Results of Epidemiological Studies Evaluating Exposure to Cobalt and Thyroid Effects

Reference, study type, and population	Exposure concentration	Outcome evaluated	Result
Lantin et al. 2011 Cohort; 249 male workers (median age 46 years; median employment 12.27 years), including 230 active workers and 19 retired workers (~5 years of retirement) from a cobalt production department (Belgium)	Measured air concentration (2007), range: 0.001–0.108 mg Co/m^3 Current cobalt concentrations, median: Blood: 0.10 µg/100 mL Urine: 3.90 µg/g creatinine Cumulative cobalt exposure (IEI), median: 106.09 µg/g creatinine x years	TSH	↔ (current blood, urine; cumulative)
		FT4	↔ (current blood, urine; cumulative)
		T4	↔ (current blood, urine; cumulative)
		FT3	↔ (current blood, urine; cumulative)
		T3	↔ (current blood, urine; cumulative)
Prescott et al. 1992 Cross-sectional; 35 female plate painters from Factory 1 exposed to cobalt aluminate (mean age 41.4 years, mean length of employment 14.6 years), 25 female plate painters from Factory 2 exposed to cobalt-zinc silicate (mean age 42.9 years, mean length of employment 16.2 years) and 48 referents (mean age 41.3 years) (Denmark)	Measured air concentrations (1987–1988) in both factories: 0.05 mg Co/m^3 Current urinary cobalt concentrations, mean: Factory 1: 0.20 µg/mmol Factory 2: 1.17 µg/mmol Referent: 0.13 µg/mmol	TSH	↔ (Factory 1 or 2 versus referents)
		FT4	↔ (Factory 1 versus referents) ↑ (Factory 2 versus referents)
		T4	↔ (Factory 1 versus referents) ↑ (Factory 2 versus referents)
		FT3	↔ (Factory 1 or 2 versus referents)
		T3	↔ (Factory 1 or 2 versus referents)

Table 2-7. Results of Epidemiological Studies Evaluating Exposure to Cobalt and Thyroid Effects

Reference, study type, and population	Exposure concentration	Outcome evaluated	Result
Swennen et al. 1993 Cross-sectional; 82 male workers (mean age 33 years; mean employment duration 8 years) from a cobalt refinery and 82 referents (mean age 38 years) (Belgium)	Current air concentration, mean: 0.125 mg Co/m^3 Current pre-shift urine cobalt level, median (µg/g creatinine): Monday: 22.9 Friday: 44.9 Current post-shift urine cobalt level, median (µg/g creatinine): Monday: 44.1 Friday: 72.4	TSH	↔ (workers versus referents)
		T4	↔ (workers versus referents)
		T3	↓ (workers versus referents)

↑ = association; ↓ = inverse association; ↔ = no association; FT3 = free triiodothyronine; FT4 = free thyroxine; IEI = integrated exposure index; T3 = triiodothyronine; T4 = thyroxine; TSH = thyroid stimulating hormone

Data pertaining to thyroid function in animals following inhalation exposure to cobalt are limited. Exposure-related changes in serum thyroid hormone levels observed in F344/N rats following intermittent exposure to cobalt sulfate for 13 weeks included decreased serum T3 in females at ≥3.78 mg Co/m^3 and decreased TSH in males at 11.4 mg Co/m^3; no exposure-related changes in serum total or free T4 were observed in either sex (Bucher et al. 1990; NTP 1991). No other inhalation studies identified measured serum thyroid hormone levels following cobalt exposure.

No exposure-related changes in thyroid gland weight and/or histology were observed in F344/N rats or B6C3F1 mice following exposure to cobalt sulfate at concentrations up to 75.7 mg Co/m^3 for 16 days, 11.4 mg Co/m^3 for 13 weeks, or 1.11–1.14 mg Co/m^3 for 105 weeks (Behl et al. 2015; Bucher et al. 1990, 1999; NTP 1991, 1998) or in F344/N rats or B6C3F1 mice following exposure to cobalt metal at concentrations up to 5 or 10 mg Co/m^3 for 14 weeks, respectively, or 5 mg Co/m^3 for 105 weeks (Behl et al. 2015; NTP 2014). No exposure-related histopathological changes in the thyroid were observed in Wistar rats intermittently exposed to cobalt tetraoxide at concentrations up to 59.31 mg Co/m^3 for 28 days (Burzlaff et al. 2022a).

Several patients, including children, have developed goiter (enlargement of the thyroid gland) and/or hypothyroidism following cobalt therapy for anemia associated with sickle-cell anemia, pregnancy, or chronic renal disease (Chamberlain 1961; Duckham and Lee 1976; Gross et al. 1955; Kriss et al. 1955;

Little and Sunico 1958; Washburn and Kaplan 1964). Due to case reports of altered thyroid function, a study by Roche and Layrisse (1956) examined iodine-131 uptake in 12 euthyroid (normal thyroid) patients who were orally administered approximately 1 mg Co/kg/day (assuming a body weight of 70 kg) as cobalt chloride for 2 weeks. After 1 week, reduced uptake of 48-hour radioactive iodine-131 by the thyroid was observed. By 2 weeks, uptake of iodine-131 was almost zero (Roche and Layrisse 1956). Iodine uptake returned to baseline following cessation of treatment. Similar findings were observed in three euthyroid males exposed to 0.54 mg Co/kg/day for 10–14 days (Paley et al. 1958). Decreased iodine-131 uptake was more pronounced in a hyperthyroid patient similarly exposed to 0.54 mg Co/kg/day for 21 days (Paley et al. 1958). No other clinical details, such as thyroid hormone levels, were provided for the human subjects in these studies. Paley et al. (1958) proposed that the decreased uptake was likely a result of cobalt blocking the organic binding of iodine-131.

In another controlled trial, no exposure-related changes in serum thyroid hormone levels were observed in 9/10 volunteers given low doses of a cobalt chloride dietary supplement (0.013 mg Co/kg/day) for up to 91 days (Tvermoes et al. 2014). One volunteer showed TSH levels elevated above clinically normal ranges at the end of exposure, coupled with serum T4 levels at the lower end of clinically normal ranges (Tvermoes et al. 2014). No changes were observed in serum thyroid hormone levels in 10 volunteers similarly exposed to a cobalt chloride supplement at 0.013 mg Co/kg/day for 31 days (Finley et al. 2013). In a controlled human study designed to evaluate the potential for cobalt chloride supplementation to reverse pregnancy-related anemia, no evidence of thyroid enlargement was observed in 20 pregnant women exposed to 0.57 mg Co/kg/day for at least 90 days prior to delivery, compared to 55 unexposed controls (Holly 1955). No additional measures of thyroid function (e.g., serum hormone levels) were monitored in this study.

In a 90-day repeat-dose gavage study, no histopathological changes were observed in the thyroid glands of Sprague-Dawley rats exposed to doses up to 7.44 mg Co/kg/day as cobalt chloride or 734 mg Co/kg/day as cobalt tetraoxide (Danzeisen et al. 2020a). While not explicitly stated, this study was conducted according to Organisation for Economic Co-operation and Development (OECD) 408 guidelines (OECD 2018), which require evaluation of serum T4, T3, and TSH; the study authors indicated that “only those endpoints that were affected by the treatment are reported,” suggesting that these parameters were not altered by treatment. However, it is not entirely clear based upon study reporting. No other identified animal studies evaluated serum thyroid hormone levels following oral exposure to cobalt. However, female Parkes mice exposed to 45 mg Co/kg-day as cobalt chloride in the drinking water for 15–45 days showed histopathological changes to the thyroid gland, including

degeneration and necrotic changes in thyroid epithelial cells and lymphocytic infiltration; findings increased in severity in a duration-dependent manner (Shrivastava et al. 1996). No morphological changes were seen in the thyroid gland of Wistar rats treated with 18 mg Co/kg/day for 4 months (Holly 1955).

No studies were identified regarding endocrine effects in humans or animals after dermal exposure to cobalt.

Other Endocrine Endpoints. Data for non-thyroid endocrine endpoints are limited. In a year-long prospective cohort from Taiwan of 69 shipyard workers exposed to particulate matter and metal fumes, urinary cobalt levels were not associated with urinary cortisol levels (Lai et al. 2021).

In intermediate-duration inhalation studies in animals, no exposure-related histopathological changes were observed in the adrenal gland, pancreas, parathyroid gland, or pituitary gland in F344/N rats or B6C3F1 mice following exposure to cobalt sulfate at concentrations up to 75.7 mg Co/m^3 for 16 days or 11.4 mg Co/m^3 for 13 weeks (Bucher et al. 1990, 1999; NTP 1991) or cobalt metal at concentrations up to 5 or 10 mg Co/m^3 for 14 weeks, respectively (NTP 2014). Similarly, no histopathological changes were observed in the adrenal gland in Sprague-Dawley rats exposed to gavage doses up to 7.44 mg Co/kg/day as cobalt chloride or 734 mg Co/kg/day as cobalt tertraoxide for 90 days (Danzeisen et al. 2020a).

There is limited evidence for non-neoplastic effects in the adrenal gland following chronic-duration inhalation exposure to cobalt. An increased incidence of medullary hyperplasia in the adrenal gland was seen in female F344/N rats after exposure to cobalt metal at 1.25 mg Co/m^3 for 105 weeks; this was not observed in similarly exposed male F344/N rats or B6C3F1 mice at concentrations up to 5 mg Co/m^3 (Behl et al. 2015; NTP 2014). No exposure-related, non-neoplastic lesions were observed in other endocrine organs (pancreas, parathyroid gland, pituitary gland) in rats or mice at concentrations up to 5 mg Co/m^3 as cobalt metal (Behl et al. 2015; NTP 2014). In studies with cobalt sulfate, no histopathological lesions were observed in the adrenal gland, pancreas, parathyroid gland, or pituitary gland in F344/N rats or B6C3F1 mice exposed to concentrations up to 1.11–1.14 mg Co/m^3 for 105 weeks (Bucher et al. 1999; NTP 1998).

No histopathological changes to the adrenal gland or pancreas were observed in Sprague-Dawley rats exposed to 16.5 mg Co/kg/day as cobalt chloride via drinking water for 30 days (Domingo et al. 1984). No histopathological changes were observed in the adrenal gland, pancreas, parathyroid gland, or

pituitary gland in Sprague-Dawley rats exposed to gavage doses up to 7.44 mg Co/kg/day as cobalt chloride or 734 mg Co/kg/day as cobalt tetraoxide for 90 days (Danzeisen et al. 2020a). No morphological changes were seen in the adrenal gland, pancreas, or pituitary gland of Wistar rats treated with 18 mg Co/kg/day for 4 months (Holly 1955).

Fifteen-day intermediate-duration exposure of 30 mg Co/kg/day as cobalt chloride by intraperitoneal injections in guinea pigs (strain not specified) altered hormones in the pancreas and had cytotoxic effects on the alpha cells in the pancreas (Beskid 1963). Single intravenous doses of 25–40 mg Co/kg as cobalt chloride in female rabbits (strain not specified) also caused cytotoxicity in the alpha cells in the pancreas (Goldner et al. 1952). Another acute-duration study that exposed pigmented guinea pigs to cobalt chloride parenterally by a single intravenous dose corroborated the results described in the previous studies by showing damages to alpha cells in pancreatic islets (Hakanson et al. 1974). A similar study in dogs (strain not specified) also showed damage to alpha cells in the pancreatic islets after an acute-duration intravenous exposure to cobalt chloride (Lazarus et al. 1953). Acute-duration exposure to cobalt nitrate salts subcutaneously was detrimental to the alpha cells in the pancreas in guinea pigs (strain not specified) (Van Campenhout 1955).

2.14 IMMUNOLOGICAL

As discussed in Section 2.4 (Respiratory), occupational exposure to cobalt has been associated with increased risk of asthma in some cohorts (Kusaka et al. 1986b; Linna et al. 2003; Roto 1980; Walters et al. 2012). Shirakawa et al. (1988, 1989) showed an increase in immunoglobin E (IgE) antibodies specific to cobalt in several cases of occupational asthma associated with hard metal exposure. Similarly, a case-study in a diamond polisher diagnosed with occupational asthma suggests that cobalt-mediated asthma may be IgE-mediated, and that sensitization of lymphocytes could play a crucial role in disease development (Krakowiak et al. 2005). Several studies have examined potential associations between occupational exposure to cobalt and markers of inflammation (e.g., inflammatory cytokines, C-reactive protein, exhaled nitric oxide), but none have observed adverse associations (Table 2-8). In studies of workers in cobalt production or hard metal industries, no positive associations were observed between estimates of current or cumulative exposure to cobalt and markers of inflammation (Andersson et al. 2020, 2021; Hedbrant et al. 2022; Lantin et al. 2011). In a year-long prospective cohort of 69 shipyard workers exposed to particulate matter and metal fumes, urinary cobalt levels were not associated with urinary levels of inflammatory cytokines or exhaled nitric oxide levels (Lai et al. 2021).

Table 2-8. Results of Epidemiological Studies Evaluating Exposure to Cobalt and Immunological Effects

Reference, study type, and population	Exposure concentration	Outcome evaluated	Result
Andersson et al. 2020 Cohort; 72 workers (9 females, 63 males; mean age 42 years; mean employment 11 years) from the hard metal industry (Sweden)	Current air concentration, mg Co/m^3 Median: 0.0009 Mean: 0.0017 8-hour TWA: 0.0034 Cumulative exposure, range: ≤0.02–>0.07 mg-year/m^3	FENO	↔ (cumulative)
Andersson et al. 2021 Cohort; 72 workers (9 females, 63 males; mean age 42 years; mean employment 11 years) from the hard metal industry (Sweden)	Current 8-hour TWA, median: 0.0023 mg Co/m^3 Stationary cobalt measurements, median, mg Co/m^3: Inhalable cobalt: 0.00042 Total cobalt: 0.00078 Respirable cobalt: 0.000062 Current cobalt concentrations, range of medians: Blood: 5.9–6.5 nmol/L Urine: 33–35 nmol/L Cumulative exposure, range: 0.21–≤0.0870 mg Co-year/m^3	Cytokines (TNF, IL-6, IL-8, IL-10)	↔ (current air, blood, urine; cumulative)
		CRP	↔ (current air, blood, urine; cumulative)
Hedbrant et al. 2022 Cross-sectional; 72 workers (9 females, 63 males; mean age 42.3 years; mean employment 10.4 years) from the hard metal industry (Sweden)	Current 8-hour TWA, mean: 0.0034 mg Co/m^3; respirator adjusted: 0.0017 mg Co/m^3 Current cobalt concentrations, mean: AM blood: 6.5 nmol/L PM blood: 7.1 nmol/L AM urine: 34 nmol/L PM urine: 44 nmol/L	IL-18	↓ (AM, PM blood)
		IL-1β	↔ (current, blood, urine)
		IL-1Ra	↔ (current, blood, urine)
Lai et al. 2021 Prospective cohort; shipyard workers compromised of: 49 welders (92% male, mean age 44 years; employment duration not reported) and 20 male office workers (mean age 51 years; employment duration not reported) (Taiwan)	Urinary cobalt concentration (post-shift), mean (μg/g creatinine): Initial: 0.00007 1 year later: 0.00009	FENO	↔ (urinary cobalt)
		IL-6	↔ (urinary cobalt)

Table 2-8. Results of Epidemiological Studies Evaluating Exposure to Cobalt and Immunological Effects

Reference, study type, and population	Exposure concentration	Outcome evaluated	Result
Lantin et al. 2011 Cohort; 249 male workers (median age 46 years; median employment 12.27 years), including 230 active workers and 19 retired workers (~5 years of retirement) from a cobalt production department (Belgium)	Measured air concentration (2007), range: 0.001—0.108 mg Co/m^3 Current cobalt concentrations, median: Blood: 0.10 µg/100 ml Urine: 3.90 µg/g creatinine Cumulative exposure (IEI), median: 106.09 µg/g creatinine x years	Cytokines (TNF, IL-6, IL-8, IL-10)	↔ (current air, blood, urine; cumulative)
		CRP	↔ (current air, blood, urine; cumulative)

↑ = association; ↓ = inverse association; ↔ = no association; CRP = C-reactive protein; FENO = fraction of nitric oxide in exhaled air; IEI = integrated exposure index; IL = interleukin; TNF = tumor necrosis factor; TWA = time weighted average

No studies evaluating immunological function in animals following inhalation exposure to cobalt were identified; however, several studies evaluated organ weight and/or histology of immune organs.

There is limited evidence that exposure to high concentrations can damage the thymus and/or lymph nodes after intermediate-duration exposure; however, findings differed between studies and administered compound. In 16-day studies, lymphoid depletion and/or necrosis of the thymus accompanied by decreased absolute and relative thymus weights were observed in F344/N rats and B6C3F1 mice intermittently exposed to concentrations ≥19 mg Co/m^3 as cobalt sulfate (NTP 1991). In contrast, exposure to ≥20 mg Co/m^3 as cobalt metal for 16 days only resulted in decreased thymus weight in female, but not male, F344/N rats (NTP 2014). No exposure-related changes in thymus weight were observed in male or female B6C3F1 mice exposed to cobalt metal at concentrations up to 40 mg Co/m^3 for 17 days (NTP 2014). No changes in other immune organs (spleen, lymph nodes, or bone marrow) were observed in either species at cobalt sulfate concentrations up to 75.7 mg Co/m^3 or cobalt metal concentrations up to 40 mg Co/m^3 (NTP 1991, 2014). In 13-week studies, hyperplasia in the mediastinal lymph nodes was observed in B6C3F1 mice exposed to 11.4 mg Co/m^3 as cobalt sulfate (Bucher et al. 1990; NTP 1991). However, no histopathological changes in the spleen, thymus, lymph nodes, or bone marrow were seen in F344/N rats exposed to cobalt sulfate at concentrations up to 11.4 mg Co/m^3 for 13 weeks (Bucher et al. 1990; NTP 1991) or in F344/N rats or B6C3F1 mice exposed to cobalt metal at concentrations up to 5 or 10 mg Co/m^3 for 14 weeks, respectively (Behl et al. 2015; NTP 2014). No

exposure-related histopathological changes in the thymus, spleen, or bone marrow were observed in Wistar rats intermittently exposed to cobalt tetraoxide at concentrations up to 59.31 mg Co/m^3 for 28 days (Burzlaff et al. 2022a). No pathological changes in immune organs were seen in pigs after a 3-month intermittent exposure to cobalt metal at 0.1 mg Co/m^3 (Kerfoot 1974).

In chronic-duration inhalation studies, no non-neoplastic histopathological changes were observed in immune system organs in F344/N rats or B6C3F1 mice exposed at concentrations up to 1.11–1.14 mg Co/m^3 as cobalt sulfate or 5 mg Co/m^3 as cobalt metal (Behl et al. 2015; Bucher et al. 1999; NTP 1998, 2014).

No evidence of cobalt sensitization was observed in 10 volunteers after exposure to low-dose cobalt chloride supplements (mean dose of 0.013 mg Co/kg/day) for 31 or 91 days, as assessed by an *in vitro* lymphocyte transformation test (Finley et al. 2013; Tvermoes et al. 2014). No additional studies evaluating immunological endpoints in humans following oral exposure to cobalt were identified.

The function of the immune system was evaluated in a few oral studies in animals. Decreased immune responses were observed in male Sprague-Dawley rats exposed to dietary cobalt chloride for 30 days, including decreased plaque formation in response to sheep red blood cells at ≥4.53 mg Co/kg/day and decreased hemagglutinin antibody response at 8.99 mg Co/kg/day (Chetty et al. 1979). Decreased relative thymus weights were also reported at ≥4.53 mg Co/kg/day; however, findings are difficult to interpret due to concomitant decreases in body weight gain (absolute organ weight data not reported). A 1.5-fold decrease of total immunoglobulin G (IgG) was observed in BALB/c mice exposed to 31 mg Co/kg/day as cobalt chloride starting 2–3 days prior to birth and during lactation (via dam) and directly on PNDs 26–60 via drinking water (Legostaeva et al. 2013). Together, these studies suggest that cobalt may suppress the immune system's ability to fight infection. In contrast, the ability to fight infection was not suppressed in mice following exposure to ≥9 mg Co/kg/day for 17 days via gavage as cobalt chloride (Huy et al. 2022). In fact, Huy et al. (2022) reported an enhanced immune response to *Brucella abortus* infection in mice following exposure to ≥9 mg Co/kg/day for 17 days via gavage as cobalt chloride.

Increased levels of proinflammatory cytokines have been reported following acute- and intermediate-duration oral exposure to cobalt in some studies. A 1-week oral exposure to cobalt chloride via gavage caused an approximate 4-fold increase in interleukin 1β (1L-1β) and an approximate 80% increase in tumor necrosis factor-alpha (TNF-α) at 67.5 mg Co/kg/day in Wistar rats (Akinrinde and Adebiyi 2019). At a lower dose, serum 1L-1β and TNF-α were increased by approximately 3- and 2-fold, respectively, in

Wistar rats exposed to cobalt chloride at 9.9 mg Co/kg/day for 14 days via gavage (Oria et al. 2022). However, in a 7-day oral exposure study by Akinrinde et al. (2016c), Wistar rats exposed to 19 mg Co/kg/day as cobalt chloride showed an increase IL-1β by about 50% but a decrease in TNF-α by about 33%. In an intermediate-duration exposure, TNF-α was elevated >10-fold in Sprague-Dawley rats exposed to 16.24 mg Co/kg/day as cobalt chloride (Abdel-Daim et al. 2020). Other inflammatory markers (C-reactive protein, nitric oxide, myeloperoxidase) were also elevated.

No additional oral studies in animals were identified that evaluated immune function (or markers of immune function); findings in studies evaluating only the weight and/or histology of immune organs were unremarkable. No changes in spleen weight or histology were observed in Sprague-Dawley rats exposed to 12.5 mg Co/kg/day for 7 days as cobalt chloride via gavage (Shrivastava et al. 2010). No changes in spleen histology were observed in Sprague-Dawley rats after a 13-week exposure to 16.5 mg Co/kg/day as cobalt chloride in the drinking water (Domingo et al. 1984) or Wistar rats after a 4-month exposure to 18 mg Co/kg/day as cobalt chloride via gavage (Holly 1955). No histopathological changes in the spleen, thymus, or lymph nodes were observed in Sprague-Dawley rats exposed to gavage doses up to 7.44 mg Co/kg/day as cobalt chloride or 734 mg Co/kg/day as cobalt tetraoxide for 90 days (Danzeisen et al. 2020a). Findings in bone marrow were restricted to erythroid hyperplasia in rats exposed to ≥2.48 mg Co/kg/day as cobalt chloride, as discussed in Section 2.7 (Hematological).

Cobalt is a skin sensitizer. A meta-analysis of contact allergy in the general population found that cobalt was the third most prevalent hapten (2.7%) in individuals with positive patch tests, behind nickel (11.4%) and fragrance mix (3.5%); however, in most cases, the source of cobalt sensitization exposure was unknown (Alinaghi et al. 2019). Skin sensitization has been confirmed in patch-testing in guinea pigs following initial exposure to 5% cobalt chloride (2.3% cobalt) (Camner et al. 1993). In the local lymph node assay (LLNA), a 3-day dermal exposure resulted in increased proliferation of lymphocytes in rats, mice, and guinea pigs exposed to doses ≥9.1, ≥4.5, and 14 mg Co/kg/day as cobalt chloride, respectively (Ikarashi et al. 1992a). Additional LLNA studies in mice have confirmed skin sensitization at ≥12 mg Co/kg/day as cobalt chloride (Ikarashi et al. 1992b; Mandervelt et al. 1997). Bonefeld et al. (2015) also reported skin sensitization in mice following exposure to 10% cobalt chloride (4.5% cobalt), which resulted in increased ear swelling and proliferation of both B and T lymphocytes when challenged with re-exposure 3 weeks later, compared with unsensitized mice (Bonefeld et al. 2015).

2.15 NEUROLOGICAL

Studies examining neurotoxicity in humans following inhalation exposure to cobalt are limited to a couple of occupational studies evaluating neurotransmitter levels in workers exposed to hard metals. In a year-long prospective cohort from Taiwan of 69 shipyard workers exposed to particulate matter and metal fumes, urinary cobalt levels were not associated with urinary serotonin levels (Lai et al. 2021). In a cross-sectional study of 186 Chinese welders, urinary cobalt levels were not associated with serum dopamine levels (Wu et al. 2023b).

Available intermediate-duration inhalation studies in rodents indicate that exposure to high levels of cobalt caused central nervous system depression, while histopathological damage was only observed at concentrations associated with lethality. A 16- or 17-day intermittent exposure to 20 and 40 mg Co/m^3 as cobalt metal caused lethargy in male and female F344/N rats, respectively, and in male and female B6C3F1 mice at 20 and 10 mg Co/m^3, respectively (NTP 2014). Hypoactivity was also observed in rats and mice intermittently exposed to cobalt sulfate at concentrations ≥19 mg Co/m^3 for 16 days; rodents that died at those exposure concentrations showed congestion in the vessels of the brain (NTP 1991). No exposure-related clinical signs or changes in brain weight or histology were seen in F344/N rats or B6C3F1 mice exposed to concentrations up to 11.4 mg Co/m^3 as cobalt sulfate for 13 weeks (Bucher et al. 1990; NTP 1991). Similarly, no exposure-related changes in clinical signs or changes in brain weight or histology were observed in F344/N rats or B6C3F1 mice following exposure to cobalt metal at concentrations up to 5 or 10 mg Co/m^3 for 14 weeks, respectively (NTP 2014).

No neurological effects have been reported at the lower concentrations evaluated in chronic-duration inhalation studies in rodents. No clinical signs of toxicity or histopathological changes in the brain, spinal cord, or peripheral nerves were observed in F344/N rats and B6C3F1 mice following exposure to cobalt sulfate at concentrations up to 1.11–1.14 mg Co/m^3 or at cobalt metal concentrations up to 5 mg Co/m^3 for 105 weeks (Behl et al. 2015; Bucher et al. 1999; NTP 1998, 2014).

No studies were identified that examined neurotoxicity in humans following oral exposure to cobalt.

Neurological effects were reported frequently in animals following acute-duration oral exposure. Mild central nervous system depression was observed in Wistar rats or Swiss-Webster mice exposed once to cobalt sulfide at 32.2 or 22.2 mg Co/kg, respectively, and Swiss-Webster mice exposed once to cobalt chloride at 16 mg Co/kg via gavage (Singh and Junnarkar 1991). Observed effects included decreased

spontaneous activity, touch response, muscle tone, and respiration; mild hypothermia; and increased pentobarbitone-induced sleeping time. Similar effects that were classified as more severe (moderate in nature) were observed in Wistar rats exposed once to cobalt chloride at 7.8 mg Co/kg via gavage (Singh and Junnarkar 1991). No changes in functional observation battery, cage-side observations, or brain weight or histology were observed in Sprague-Dawley rats exposed to 12.5 mg Co/kg/day for 7 days as cobalt chloride via gavage (Shrivastava et al. 2010). However, neurobehavioral impairments, including decreased motor strength in the hanging wire tasks and decreased motor activity and exploration in an open field, were observed in male Wistar rats following exposure to 67.5 mg Co/kg/day via gavage as cobalt chloride for 7 days (Akinrinde and Adebiyi 2019). No learning or memory impairments were noted in the Morris water maze. Neurobehavioral alterations were associated with a 2-fold increase in glial fibrillary acidic protein (GFAP) positive astrocytes, a marker of reactive gliosis, and a 60% increase in acetylcholinesterase (AChE) activity (Akinrinde and Adebiyi 2019). Altered exploration in the Y-maze test, impaired memory in the novel object recognition test, and increased anxiety-like behaviors in the elevated plus maze were also observed in Wistar rats exposed to cobalt chloride at 9.9 mg Co/kg/day for 14 days via gavage (Oria et al. 2022). Neurobehavioral alterations were accompanied by reactive gliosis in the hippocampus (increased GFAP-positive astrocytes) and ultrastructural changes in the hippocampus and amygdala. Moderate-to-severe sciatic nerve damage (degeneration of myelinated fibers; Schwann cell degeneration, perineurium disjunction) was observed in Wistar rats exposed to 37 mg Co/kg/day as cobalt chloride via gavage for 7 days (Tanoğlu et al. 2022).

Some intermediate-duration rodent studies have reported altered performance in operant conditioning tasks following oral exposure to cobalt. For example, Sprague-Dawley rats exposed to 20.3 mg Co/kg/day as cobalt chloride for 57 days in water showed enhanced avoidance retention in passive-avoidance testing, as determined by increased latency to step down on an electrified grid after the initial operant training acquisition phase (Bourg et al. 1985). This finding is suggestive of decreased stress tolerance. Findings were not attributed to enhance nociception in exposed animals, as no differences in analgesic tolerance were observed. Impaired operant conditioning in food-reward based paradigms has also been observed in rats exposed to cobalt chloride at gavage doses of 2.5 mg Co/kg/day for 4–7 months (Krasovskii and Fridlyand 1971) or dietary doses of 20 mg Co/kg/day for 69 days (Nation et al. 1983). Krasovskii and Fridlyand (1971) reported time-dependent increases in conditioned latent reflexes and loss of memory retention from 4 to 7 months. Findings reported by Nation et al. (1983) were milder, showing a reduced rate of lever pressing than controls, but no change in overall number of food reinforcements received (Nation et al. 1983). In contrast to Bourg et al. (1985), no evidence of decreased stress tolerance was observed in the conditioned suppression (shock) task in rats exposed to dietary doses up to 20 mg

Co/kg/day via diet for 69 days (Nation et al. 1983). It is noted that findings from food-based operant conditioning need to be interpreted with caution, as they may be due to conditioned taste aversion rather than impaired cognitive function (Wellman et al. 1984).

Findings in other intermediate-duration studies are mixed. Increased GFAP-positive brain regions, encephalopathy, and decreased levels of several neurotransmitter levels, including serotonin, norepinephrine, dopamine, and gamma-aminobutyric acid (GABA), were observed in Wistar rats orally exposed to 27 mg Co/kg/day as cobalt chloride for 60 days (Mohamed et al. 2019). In a series of studies by Danzeisen et al. (2020a), no clinical signs of neurotoxicity or exposure-related changes in neurological screening or functional neurological testing were observed in Sprague-Dawley rats following intermediate-duration exposure to gavage doses up to 7.44 mg Co/kg/day as cobalt chloride, 648 mg Co/kg/day as cobalt sulfide, or 734 mg Co/kg/day as cobalt tetroxide. Additionally, no changes in brain weight or histology were observed in Sprague-Dawley rats exposed to gavage doses up to 7.44 mg Co/kg/day as cobalt chloride or 734 mg Co/kg/day as cobalt tetraoxide for 90 days (Danzeisen et al. 2020a). No morphological changes were seen in the brains of Wistar rats treated with 18 mg Co/kg/day for 4 months (Holly 1955). No changes in pain threshold or responses were observed in Wistar rats following oral exposure to 23 mg Co/kg/day as cobalt chloride for 28 days (Umar et al. 2016).

No studies were identified regarding neurological effects in humans or animals after dermal exposure to cobalt.

Acute-duration exposure to 6 mg Co/kg/day as cobalt chloride via intraperitoneal injections resulted in a 25% decrease in response latency in Balb/c mice (Alexa et al. 2015). In another study, rats (strain not specified) were exposed by intraperitoneal administration of cobalt sulfate at 114 mg Co/kg/day for 5 consecutive days resulting in a decrease in avoidance response (Inozemtsev et al. 2008). Balb/c mice showed a decrease in auditory brainstem response thresholds after an intraperitoneal injection of 22.7 mg Co/kg/day once as cobalt chloride. This effect indicates that cobalt is potentially ototoxic (Lee et al. 2016). A single intraperitoneal injection of 25 mg/kg/day did not alter brain serotonin levels in Swiss albino mice but did cause hypothermia (Burke et al. 1978).

Altered neurotransmission has been reported *ex vivo* in rat tissues following exposure to 6.4 mg Co/kg/day in drinking water as cobalt nitrate for 30 days (Mutafova-Yambolieva et al. 1994; Vassilev et al. 1993). Vassilev et al. (1993) evaluated cumulative concentration-effect curves for carbachol (a cholinergic agonist) ileum and trachea isolated from control or treated rats. Increased sensitivity and

decreased maximal response to carbachol was observed in isolated ileum of treated rats. Mutafova-Yambolieva et al. (1994) evaluated peripheral sympathetic neurotransmission in isolated vas deferens tissue, a classic model for evaluation of pre- and postjunctional mechanisms of peripheral sympathetic neurotransmission due to its dense sympathetic innervation. Specifically, *in vivo* exposure to cobalt nitrate resulted in increased *ex vivo* adrenoceptor- and purinoceptor-mediated contractile responses of the vas deferens. Due to challenges associated with interpreting the adversity of findings from *ex vivo* studies, these studies are not included in the LSE table. Additionally, these findings may represent changes in the examined organ tissue (e.g., respiratory, gastrointestinal, reproductive tissues) rather than (or in addition to) changes in the neurological system.

2.16 REPRODUCTIVE

No studies were identified that examined reproductive toxicity in humans following inhalation, oral, or dermal exposure to cobalt.

Intermediate-duration inhalation exposure to cobalt produced reproductive effects in some studies; the male rodent appears to be more susceptible than the female. A series of intermediate-duration studies evaluated reproductive endpoints in F344/N rats and B6C3F1 mice following intermittent exposure to either cobalt metal dust or cobalt sulfate for 16–17 days or 13–14 weeks (Bucher et al. 1990; NTP 1991, 2013). Male and female reproductive organ weight were examined at both timepoints, histology was examined at both timepoints for cobalt sulfate but only at 14 weeks for cobalt metal, and estrous cyclicity and sperm morphology and motility evaluated at 13–14 weeks. In the shorter-duration studies (16 days), testicular atrophy, along with a decrease in number of cells in the seminiferous tubules and atypical germinal epithelial cells in the epididymal ducts, were observed in male rats exposed to ≥19.0 mg Co/m^3 for 16 days (NTP 1991). However, no exposure-related changes in organ weight or histology were observed in female rats or male or female mice exposed to concentrations up to 75.7 mg Co/m^3 as cobalt sulfate (NTP 1991). Similarly, no exposure-related changes in organ weights were observed in rats or mice exposed to concentrations up to 40 mg/m^3 as cobalt metal; however, due to the lack of histological examination at this timepoint, a NOAEL could not be confidently established at this exposure level (NTP 2014). In the longer intermediate-duration studies (13–14 weeks), effects noted included decreased percent motile sperm at cobalt metal concentrations ≥2.5 mg Co/m^3 in male rats and mice and at cobalt sulfate concentrations ≥1.11 mg Co/m^3 in male mice (NTP 1991, 2014). Additional effects observed in exposed male rodents included increased relative testes weights in male rats at ≥2.5 mg Co/m^3 as cobalt metal, decreased absolute testes weight in male mice at ≥5 mg Co/m^3 as cobalt metal, and a 3-fold

increase in percent abnormal sperm with testicular atrophy in male mice at 11.4 mg Co/m^3 as cobalt sulfate. In contrast, male rats similarly exposed to cobalt sulfate at concentrations up to 11.4 mg/m^3 for 13 weeks did not show any changes in sperm parameters or reproductive organ weight or histology (NTP 1991). In female rodents, prolonged estrous cycle length was observed in mice exposed to cobalt metal at 10 mg Co/m^3 for 14 weeks or cobalt sulfate at 11.4 mg Co/m^3 for 13 weeks; this effect was not observed in similarly exposed female rats (NTP 1991, 2014). No changes in female reproductive organ weight or histology were observed in either rats or mice at cobalt sulfate concentrations up to 11.4 mg Co/m^3 or cobalt metal concentrations up to 5 or 10 mg Co/m^3, respectively. In another intermediate-duration inhalation study, no exposure-related histopathological changes in the testes, ovaries, or uterus were observed in Wistar rats intermittently exposed to cobalt tetraoxide at concentrations up to 59.31 mg Co/m^3 for 28 days (Burzlaff et al. 2022a).

Reproductive endpoints evaluated in chronic-duration studies were restricted to histological examination of reproductive organs. As observed in intermediate-duration studies, findings indicate that the male reproductive system is more sensitive to cobalt toxicity than the female reproductive system. Histopathological changes in the testes were observed in male F344/N rats and B6C3F1 mice intermittently exposed to 5 mg Co/m^3 as cobalt metal for 105 weeks (Behl et al. 2015; NTP 2014). In rats, findings were severe, classified as testicular infarcts (complete effacement of parenchyma due to necrosis). Findings in mice were less severe, including minimal-to-mild degeneration of the testicular epithelium. No histopathological changes were observed in female reproductive organs in rats or mice at cobalt metal concentrations up to 5 mg Co/m^3 (Behl et al. 2015; NTP 2014). No histopathological changes in male or female reproductive organs were observed in F344/N rats or B6C3F1 mice following exposure to cobalt sulfate at concentrations up to 1.11 or 1.14 mg Co/m^3, respectively, for 105 weeks (Bucher et al. 1999; NTP 1998).

Acute-duration oral studies evaluating reproductive effects in animals are limited. The percent of abnormal sperm was increased in a dose-related manner in male Swiss mice orally exposed to doses ranging from 7.089 to 28.37 mg Co/kg/day as cobalt chloride (4.74 to 9.86% abnormal), compared to 2.10% abnormal in controls (Hassan et al. 2006). No exposure-related changes in sperm parameters or male reproductive organ histology were observed in male Sprague-Dawley rats exposed to cobalt chloride at dietary doses up to 20 mg Co/kg/day for 14 days (Corrier et al. 1985).

Several intermediate-duration studies have reported male reproductive effects following oral exposure to cobalt. The lowest dose associated with impaired fertility was reported in male Swiss mice exposed to

cobalt chloride at ≥6.354 mg Co/kg/day via drinking water for 12 weeks, resulting in a decreased sperm count and reduced number of viable pregnancies when mated to unexposed females (Elbetieha et al. 2008). Additional effects noted at ≥11.62 mg Co/kg/day included a decreased percentage of pregnant females, decreased absolute and relative testes weights, increased absolute and relative seminal vesicles weights, decreased testicular sperm count and daily sperm count/testis, and histopathological changes in the testes (necrosis, hypertrophy of Leydig cells, degeneration of spermatogonial cells).

A series of intermediate-duration drinking water studies in CD-1 mice showed duration-dependent testicular effects following exposure to 43.4–58.9 mg Co/kg/day as cobalt chloride for 7–13 weeks (Anderson et al. 1992; Pedigo et al. 1988). Exposure for 7 weeks was not associated with adverse effects on testes weight, histology, sperm count or motility, or fertility. At 9 weeks, decreased absolute (29%) and relative (25%) testes weights were observed, along with morphological changes in the seminiferous tubules, including vacuolation of Sertoli cells and spermatid, reduced thickness of germinal epithelium, and sloughing of germ cells. Severity of effects increased at 11–13 weeks, with decreased absolute (62%) and relative (58%) testes weights, decreased epididymal sperm concentration (85%), decreased motile sperm (71–83%), and severe degeneration of seminiferous tubules characterized by extensive Sertoli and germ cell loss. After 13 weeks, additional findings not observed at earlier timepoints included grossly apparent testicular atrophy and decreased fertility, as measured by a 78% decrease in fertilized ova when mated to unexposed female mice. Testicular weight, histopathological findings, and decreased fertility after the 13-week exposure persisted following a 20-week recovery period (Anderson et al. 1992; Pedigo et al. 1988). Findings at 13 weeks were confirmed in a follow-up study by Anderson et al. (1993) utilizing the same protocol, with a 61% decrease in testicular weight, seminiferous tubule damage and degeneration, and hypercellularity of the interstitial areas. Pedigo et al. (1988) also showed that reproductive effects in male mice were dose-dependent following exposure to cobalt chloride via drinking water for 12 weeks. At ≥23.0 mg Co/kg/day, adverse effects included decreased absolute (33–74%) and relative (29–70%) testes weights, decreased epididymal sperm concentration (34–92%), and increased serum testosterone levels (5–7-fold). Additional effects observed at the highest dose (72.1 mg Co/kg/day) included decreased percent of motile sperm (58%) and decreased fertility, as measured by a 74% decrease in fertilized ova when mated to unexposed female mice. Similar decreases in fertility (when mated to unexposed females), sperm concentration, and sperm motility have also been observed in male B6C3F1 mice exposed to 58.9 mg Co/kg/day as cobalt chloride in drinking water for 10 weeks (Pedigo and Vernon 1993). In this study, fertilization rates no longer differed from control 6 weeks postexposure.

For other cobalt compounds, no exposure-related impairments in fertility, mating, or reproductive indices or sperm parameters were observed in male and female Sprague-Dawley rats exposed to gavage doses up to 648 mg Co/kg/day as cobalt sulfide or 734 mg Co/kg/day as cobalt tetroxide for 2 weeks prior to mating through PND 3 (Danzeisen et al. 2020a).

Mollenhauer et al. (1985) demonstrated that exposure to dietary cobalt metal at 20 mg Co/kg/day for 98 days caused deterioration of cell architecture and a decrease in testicular volume in Sprague-Dawley rats. This damage included thickening of basal lamina and basement membranes, increased packing of red blood cells in veins and arteries, change in sperm morphology, and degeneration in sperm mitochondria. Testicular atrophy and marked decrease in testicular weight (58%) were observed in Sprague-Dawley rats exposed dietary doses of 20 mg Co/kg/day as cobalt chloride for 69 days (Nation et al. 1983). Evidence of testicular damage, including marked degeneration and necrosis of germinal epithelium in 27–90% of seminiferous tubules, a 43% drop in spermatid reserves, and atrophy/marked decreases in testicular weight, have been reported in Sprague-Dawley rats exposed to 16.5–20 mg Co/kg/day as cobalt chloride for 13–14 weeks in food or drinking water (Corrier et al. 1985; Domingo et al. 1984). No changes in serum hormone levels (testosterone, progesterone, estradiol) or testes or prostate weight or histology were observed in male Sprague-Dawley rats exposed to gavage doses up to 7.44 mg Co/kg/day as cobalt chloride or 734 mg Co/kg/day as cobalt tetraoxide for 90 days (Danzeisen et al. 2020a).

Data on female reproductive toxicity following oral exposure to cobalt in animals are limited. No changes in estrous cyclicity, serum hormone levels (testosterone, progesterone, estradiol), or female reproductive organ weight or histology were observed in female rats exposed to gavage doses up to 7.44 mg Co/kg/day as cobalt chloride or 734 mg Co/kg/day as cobalt tetraoxide for 90 days (Danzeisen et al. 2020a).

No studies were identified regarding reproductive effects in animals after dermal exposure to cobalt.

Acute-duration exposure by intraperitoneal injections to cobalt chloride resulted in structural and functional alterations of the testes in Syrian hamsters (Lukac et al. 2007).

2.17 DEVELOPMENTAL

No studies were identified that examined developmental toxicity in humans or animals following inhalation exposure to cobalt.

In a controlled human study designed to evaluate the potential for cobalt chloride supplementation to reverse pregnancy-related anemia, all pregnancies were reportedly "normal" with no evidence of "enlargement" of infants for 20 pregnant women exposed to 0.57 mg Co/kg/day for at least 90 days prior to delivery, compared to 55 unexposed controls (Holly 1955). No further details on pregnancy or infant outcomes were provided.

Acute-duration exposure to cobalt during gestation did not produce developmental effects in rats, even at doses associated with maternal toxicity. Maternal exposure to 81 mg Co/kg/day via gavage as cobalt chloride on GDs 8–12 had no effects on fetal growth, mortality, or incidence of external malformations (Seidenberg et al. 1986). Similarly, no effects on fetal growth, survival, or external or internal malformations or variations were found following maternal exposure to doses up to 24.8 mg Co/kg/day as cobalt chloride during GDs 6–15 in Sprague-Dawley rats (Paternian and Domingo 1988). In both studies, maternal toxicity (>20% decreases in body weight gains) were observed.

In contrast to gestation-only studies, gestation plus postnatal exposure studies reported impaired pup development and survival. Impaired pre- and postnatal growth was observed in Wistar rat pups following maternal gavage exposure to cobalt chloride at doses ≥5.4 mg Co/kg/day from GD 14 through PND 21 (Domingo et al. 1985b). Findings included decreased pup weights >10% on PNDs 1, 4, and 21, as well as mild decreases (<10%) in body length. Higher maternal doses of 22 mg Co/kg/day were associated with decreased postnatal survival, primarily between PNDs 1 and 4 (Domingo et al. 1985b). No gross malformations were observed, and no dose-related changes in hematology, clinical chemistry, or organ weights were reported in pups surviving until PND 21. In a one-generation study in Sprague-Dawley rats, pup weight was decreased by up to 18% on PND 1 and by up to 21% on PND 4 following F0 exposure to 734 mg Co/kg/day as cobalt tetraoxide from 2 weeks premating through PND 3 (Danzeisen et al. 2020a). Pup viability at PND 4 was also decreased at this dose (87.6%) compared to controls (100%); however, this finding was attributed by the study authors to the unexplained total loss of one litter (quantitative data were not provided). There were no exposure-related external abnormalities. In Sprague-Dawley rats similarly exposed to cobalt sulfide, no exposure-related changes in pup viability,

growth, or malformations were observed at gavage doses up to 648 mg Co/kg/day (Danzeisen et al. 2020a).

Gluhcheva et al. (2020) reported various systemic effects in PND 18 mouse pups in a study designed to evaluate potential effects of perinatal exposure to cobalt chloride following maternal exposure to 19 mg Co/kg/day via drinking from GD 19 or 20 through PND 18. Pups showed a 17% decrease in body weight on PND 18, along with reduced relative spleen and kidney weights; relative liver weights were comparable to controls. Histological examinations showed various abnormalities in these organs, including reduced red pulp in the spleen; leukocyte infiltration, mesangial cellularity, and reduced capsular space in the kidney; and leukocyte infiltration, binucleated hepatocytes, abundant Kupffer cells, and apoptotic bodies in the liver. Hematological analysis showed a 17% increase in red blood cell count; hematocrit and hemoglobin levels were comparable to controls. Similar studies have shown serious body weight effects (40% decrease) accompanied by liver, kidney, and brain damage in PND 14 rat pups following maternal exposure to 20 mg Co/kg/day as cobalt chloride from GD 14 to PND 14 (Garoui et al. 2011, 2012, 2013). Observed liver effects included increased plasma levels of ALT and AST of approximately 2-fold and infiltration of mononuclear cells and vascular congestion in the liver (Garoui et al. 2011). Absolute liver weights were also significantly decreased but findings were confounded by significant body weight decreases in pups and lack of relative liver weight data reporting. Observed kidney effects included increased plasma creatinine (31%), decreased urinary creatinine (29%) and urea (47%), reduced relative kidney weight (4%), and vascular congestion with reduction of glomerular space (Garoui et al. 2012). Neurological findings included altered development of cerebellar architecture, including poorly differentiated layers marked by frequent pyknotic cells, fewer overall cells, and an overly developed external granular layer. Pups also showed decreases in the levels of AChE and butyrylcholinesterase (BuChE) in the cerebrum by 33 and 36%, respectively, and in the cerebellum by 33% and 47%, respectively (Garoui et al. 2013).

No studies were identified regarding developmental effects in humans or animals after dermal exposure to cobalt.

2.18 OTHER NONCANCER

A limited number of studies have evaluated endpoints relevant to metabolic syndrome in workers exposed to cobalt. In a prospective cohort that followed 100 male welders over the course of 2 years, urinary cobalt levels were not associated with body mass index, fasting blood glucose, or serum lipids, and an

inverse (non-adverse) association was observed between urinary cobalt levels and serum adiponectin levels (Wu et al. 2023a). In a cross-sectional study of 769 ferro-manganese refinery workers, current cobalt plasma levels were inversely associated with fasting blood glucose levels; however, once adjusted for multiple metal exposure, the association was no longer significant (Ge et al. 2021).

Studies in rodents indicate that exposure to cobalt may impact glucose homeostasis. Decreased serum glucose was observed in male rats exposed to cobalt metal at concentration ≥1.25 mg Co/m^3 for 14 weeks; this finding was not observed in similarly exposed female rats at concentrations up to 5 mg Co/m^3 or male or female mice at concentrations up to 10 mg Co/m^3 (NTP 2014). No exposure-related changes in serum glucose were observed in rats or mice exposed to cobalt sulfate at concentrations up to 11.4 mg Co/m^3 for 13 weeks (Bucher et al. 1990; NTP 1991). Following oral exposure, decreased serum glucose was also reported in rats exposed once to 161 mg Co/kg as cobalt chloride via gavage (Domingo and Llobet 1984). Maternal rats exposed to 20 mg Co/kg/day as cobalt chloride in drinking water from GD 14 to PND 14 also showed decreased plasma glucose; however, this may have been secondary to decreased maternal food intake (Garoui et al. 2012). A dose of 18 mg Co/kg/day as cobalt chloride in drinking water for 12–16 days also lowered blood glucose levels in diabetic rat models; however, no exposure-related changes were observed in similarly exposed non-diabetic rats (Saker et al. 1998). In contrast, elevated serum glucose levels were observed in male rats exposed to 12.5 mg Co/kg/day as cobalt chloride via gavage for 7 days (Shrivastava et al. 2008, 2010).

Several oral studies reported decreased food and/or water intake in rodents; in most cases, findings are likely attributable to palatability issues and are not considered evidence of toxicity. Acute-duration exposure of 45 mg Co/kg/day as cobalt chloride in food for 3 consecutive days decreased food consumption in Sprague-Dawley rats (Wellman et al. 1984). Intermediate-duration exposure of 16.5 mg Co/kg/day in drinking water as cobalt chloride for 13 weeks decreased water intake, resulting in decreased urine output, in Sprague-Dawley rats (Domingo et al. 1984). Decreased water intake was also reported in rats exposed to 20.3 mg Co/kg/day as cobalt chloride in drinking water for 57 days, 72.1 mg Co/kg/day as cobalt chloride in drinking water for 12 weeks, or 58.9 mg Co/kg/day as cobalt chloride in drinking water for 13 weeks (Bourg et al. 1985; Pedigo et al. 1988). Decreases in both water and food intake were observed in maternal Wistar rats exposed to 20 mg Co/kg/day in drinking water as cobalt chloride for 2 weeks during gestation plus 2 weeks during lactation (Garoui et al. 2011, 2012).

No studies were identified regarding other noncancer effects in animals after dermal exposure to cobalt for any duration.

An acute-duration (10 days) subcutaneous exposure in ICR mice to 0.59 mg Co/kg/day as cobalt chloride resulted in increased adipocyte messenger ribonucleic acid (mRNA) by nearly 100% and adiponectin levels by 42% (Kawakami et al. 2012). These effects were directly related with decreases in white adipose tissue weight and size, which were potentially a direct result of cobalt toxicity (Kawakami et al. 2012). The relevance of these effects to human health are currently unknown as they have not been studied in humans.

2.19 CANCER

IARC classified cobalt metal (without tungsten carbide or other metal alloys) and soluble cobalt (II) salts (cobalt chloride, cobalt sulfate) as probably carcinogenic to humans and cobalt (II) oxide as possibly carcinogenic to humans (IARC 2023). Metal mixtures containing cobalt, including cobalt metal with tungsten carbide and weapons-grade tungsten (with nickel and cobalt) are classified as probably and possibly carcinogenic to humans, respectively (IARC 2006, 2023). IARC (2023) determined that cobalt (II, III) oxide (cobalt tetraoxide), cobalt (II) sulfide, and other cobalt (II) compounds are not classifiable as to their carcinogenicity to humans. NTP determined that cobalt and cobalt compounds that release cobalt ions *in vivo* are reasonably anticipated to be human carcinogens (NTP 2021). The EPA Integrated Risk Information System (IRIS) program is currently conducting an inhalation cancer risk assessment for cobalt and cobalt compounds and has released the protocol for the assessment along with its dose-response methodology (EPA 2022a).

Several retrospective cohort studies have evaluated cancer risk in workers exposed to cobalt (Table 2-9). Exposure to cobalt, tungsten, and nickel and cancer mortality risk was evaluated in an international cohort of hard metal production workers (Marsh et al. 2017b). Workers (32,534) from 3 companies, 17 sites among 5 countries, including the United States, Austria, Germany, Sweden, and the United Kingdom, were evaluated. Information on deaths was obtained from various national datasets, and phone interviews were completed for participants when possible. These interviews provided information on demographic and lifestyle factors. Kennedy et al. (2017) described the job class plus exposure matrix that was used and reported the estimated cobalt, nickel, and tungsten exposures. Employee history was obtained from occupational records. Among just the U.S. cohort in this study, which included eight sites, there was no increased lung cancer mortality risk or trends in standardized mortality ratios (SMRs) from long-term exposure to cobalt or from the other metals studied (Marsh et al. 2017a). No sex-related differences in SMRs were observed. While two plants observed excess lung cancer mortality, SMRs did not differ from

the general population. The study authors stated that the lung cancer risks were higher in females than in males in Germany, the United States, and Sweden likely due to lifestyle and behavioral factors, such as increased smoking, and not from occupational exposure (Marsh et al. 2017a). When pooling data from all international cohorts, there was a slight excess in all cancer and lung cancer mortality; however, there was no evidence of an exposure-response relationship for lung cancer (Marsh et al. 2017b). Additionally, there was no indication that occupation duration or cumulative exposure to cobalt impacted lung cancer mortality risk.

Table 2-9. Results of Epidemiological Studies Evaluating Exposure to Cobalt and Cancer

Reference, study type, and population	Exposure concentration	Outcome evaluated	Result
Lasfargues et al. 1994 Retrospective cohort; 709 workers from the hard metal industry; employed for at least 1 year from 1956 to 1989 (France)	Job-exposure matrix (historic air concentrations and urinary cobalt levels) Unexposed Low: Air: <0.01 mg Co/m^3 Urine: 0.01–0.02 µmol/L Medium: Air: 0.015–0.04 mg Co/m^3 Urine: 0.01–0.10 µmol/L High: Air: >0.05 mg Co/m^3 Urine: 0.02–0.28 µmol/L	Cancer deaths All malignant neoplasms Buccal cavity, pharynx, larynx Esophagus Larynx Leukemia Lung	 ↑ (high) ↔ ↔ ↔ ↔ ↑ (medium, current smokers) ↑ (high, current smokers)
Marsh et al. 2017a Retrospective cohort; 7,304 workers from the hard metal industry (United States)	Average intensity of exposure during a 56-year period, median: 0.006 mg Co/m^3 Cumulative exposure, median: 0.020 mg Co-year/m^3	Lung cancer deaths	↔
Marsh et al. 2017b Retrospective cohort; 32,354 workers from the hard metal industry (Austria, Germany, Sweden, United Kingdom, United States)	Average intensity of exposure during a 62-year period, median: 0.006 mg Co/m^3 Cumulative exposure, median: 0.020 mg Co-year/m^3	Lung cancer deaths	↔

Table 2-9. Results of Epidemiological Studies Evaluating Exposure to Cobalt and Cancer

Reference, study type, and population	Exposure concentration	Outcome evaluated	Result
McElvenny et al. 2017 Retrospective cohort; 1,538 workers from the hard metal industry (United Kingdom)	Historical air concentration, median, mg Co/m^3: 1980s: 0.0105 1994–2003: 0.01 2004–2004: 0.0071	All cancer deaths	↔
Morfeld et al. 2017 Retrospective cohort; 6,865 workers from the hard metal industry (Germany)	Long-term average concentration, median: 0.04 mg Co/m^3 Cumulative exposure, estimated median (range): 0.16–0.23 mg Co-year/m^3	Lung cancer mortality risk	↔
Moulin et al. 1993; Mur et al. 1987 Retrospective cohort 1,148 workers from an electrochemical plant producing cobalt and sodium (France)	Exposure levels not reported	Lung cancer deaths	↑ (1950–1980) ↔ (1950–1988)
		Buccal cavity, pharynx, larynx cancer deaths	↔ (1950–1980) ↔ (1950–1988)
		Brain cancer deaths	↑ (1950–1988)
		Other cancer deaths (gastrointestinal, pancreas, bladder, prostate, blood, bone)	↔ (1950–1988)
Moulin et al. 1998 Retrospective cohort; 7,459 workers from the hard metal industry (France)	Measured air levels in workshops, range of geometric means: 0.01825–1.6534 mg Co/m^3	Cancer deaths All cancer sites	↔
		Buccal cavity, pharynx	↔
		Larynx	↔
		Esophagus	↔
		Lung	↔
		Pleura	↔
		Bladder	↔
Moulin et al. 2000 Retrospective cohort; 4,897 workers from the hard metal industry (France)	Job-exposure matrix (not reported)	Cancer deaths All cancer sites	↔
		Buccal cavity, pharynx	↔
		Larynx	↔
		Esophagus	↔
		Lung	↔

Table 2-9. Results of Epidemiological Studies Evaluating Exposure to Cobalt and Cancer

Reference, study type, and population	Exposure concentration	Outcome evaluated	Result
		Pleura	↔
		Bladder	↔
		Leukemia	↔
Sauni et al. 2017 Retrospective cohort; 955 workers from a cobalt production plant industry (Finland)	Historical air concentration, range of means in mg Co/m^3 (in different departments): 1968–1976: 0.017–0.082 1977–1986: 0.02–0.10 1987–1999: 0.019–0.065 2000–2003: 0.017–0.065 2004–2014: 0.018–0.075	All cancer incidence	↔
		Lung cancer incidence	↔
Svartengren et al. 2017 Retrospective cohort; 3,713 workers from the hard metal industry (Sweden)	Historical air concentration 1970–2012, range: 0.0001–2.8 mg Co/m^3 Quartiles: Q1: ≤0.001 Q2: 0.002–0.0038 Q3: 0.0039–0.0088 Q4: ≥0.0089	Lung cancer incidence	↑ (Q4)
		Other cancers (lip, larynx, pleura, gastrointestinal, pancreas, prostate, bladder, skin, blood)	↔
Tüchsen et al. 1996 Retrospective cohort; 874 women occupationally exposed to cobalt-aluminate spinel dye (Factory 1) or cobalt silicate dye (Factory 2) in porcelain factories (Denmark)	Historical air concentration, range: 0.01–1.5 mg Co/m^3	Cancer incidence All cancers	↔
		Lung cancer	↑ (all workers) ↔ (Factory 1) ↑ (Factory 2)
		Cervix uteri	↑ (all workers)
		Other cancers (gastrointestinal, pancreatic, breast, ovary, kidney, bladder, skin, brain, blood)	↔
Wallner et al. 2017 Retrospective cohort; 1,965 workers from the hard metal industry (Austria)	Historical air concentration, average: 0.05 mg Co/m^3 Cumulative exposure, average: 0.52 mg Co-year/m^3	All cancer deaths	↔
		Lung cancer deaths	↔

Table 2-9. Results of Epidemiological Studies Evaluating Exposure to Cobalt and Cancer

Reference, study type, and population	Exposure concentration	Outcome evaluated	Result
Westberg et al. 2017a, 2017b Retrospective cohort; 16,999 workers from the hard metal industry (Sweden)	Historical air concentration (1970–2012), median: 0.01 mg Co/m^3	Lung cancer deaths	↔
Wild et al. 2000 Retrospective cohort; 2,216 men, 644 women working in hard metal industry (France)	Job exposure matrix based on potential exposure to hard metal dust (concentrations not measured)	Cancer deaths	
		All cancer sites	↔
		Lung cancer	↑ (men, ever smokers) ↔ (men, nonsmokers) ↔ (women)
		Other cancers (oral cavity or pharynx, larynx, esophagus, stomach, intestine, rectum, liver, pancreas, breast, pleura, bladder, brain, Hodgkin's disease and lymphoma, leukemia)	↔

↑ = association; ↔ = no association; Q = quartile

A retrospective mortality study in workers from a French electrochemical plant producing cobalt and sodium found a small increase in the number of lung cancer deaths between the years of 1950 and 1980 (Mur et al. 1987); however, when the cohort was re-assessed 8 years later, the association was no longer apparent (Moulin et al. 1993). Deaths associated with brain cancer in this cohort was slightly increased from 1950 to 1988 (Moulin et al. 1993). No other cancer deaths were increased in this worker cohort, compared to the general population (SMRs). An increased incidence of lung cancer was also reported in workers in the highest quartile of cobalt exposure (≥0.0089 mg/m^3) from a Swedish cohort of hard metal workers (Svartengren et al. 2017). However, after various analyses looking at exposure intensity, duration, and lag, no clear association with cobalt was observed. In both the French and Swedish cohorts, the study authors noted that the lack of control for smoking status was a major confounding factor. When smoking status was specifically evaluated in a French cohort of hard metal workers, increased lung cancer deaths were observed only in workers exposed to ≥0.015 mg Co/m^3 who were current smokers, compared

to the general population (Lasfargues et al. 1994). Lung cancer death rates were not increased in workers who formerly or never smoked. Consistent with this study, an evaluation of 14 French hard metal workshops found an increased rate of lung cancer deaths in male workers exposed to cobalt, compared to the general population, but only if they were "ever smokers" (Wild et al. 2000). The incidence of lung cancer deaths was not elevated in male workers who never smoked or female workers. Across the various departments, male workers with increased risk of lung cancer death were employed in the hard metal production before sintering and maintenance departments. However, workers were exposed to various other compounds, including asbestos, polycyclic aromatic hydrocarbons, silica, nickel compounds, and chromium compounds (Wild et al. 2000).

Other studies evaluating cobalt production facilities or hard metal factories in the United Kingdom and European countries did not find significant exposure-response relationships between cancer incidence or SMRs and occupational exposure to cobalt (McElvenny et al. 2017; Moulin et al. 1998, 2000; Morfeld et al. 2017; Sauni et al. 2017; Wallner et al. 2017; Westberg et al. 2017a). A small retrospective cohort study in Danish female porcelain workers found an increased incidence in lung cancer in workers using cobalt silicate dye, but not in workers using cobalt aluminate dye (Tüchsen et al. 1996). As reported by Prescott et al. (1992), the silicate dye (cobalt-zinc silicate) is semi-soluble, while the cobalt aluminate is insoluble, which may account for the differential finding. Additionally, the contribution of the zinc and silica elements of the semi-soluble dye are unknown. Tüchsen et al. (1996) indicated that findings need to be confirmed in a larger sample size.

No associations were observed between occupational exposure to cobalt and overall cancer risk in a meta-analysis of nine studies (Marsh et al. 2017a; McElvenny et al. 2017; Morfeld et al. 2017; Moulin et al. 1993, 1998, 2000; Sauni et al. 2017; Svartengren et al. 2017; Tüchsen et al. 1996) conducted by Zhang et al. (2021). Similarly, a systematic review and meta-analysis conducted by Holy et al. (2022) determined at there is "insufficient to conclude that there exists an increased risk" of developing any specific type of cancer from exposure to cobalt via occupational exposure, exposure from implanted medical devices (total joint replacements), or a combination thereof.

Chronic-duration inhalation exposure to cobalt produces exposure-related lung tumors in rats and mice, pheochromocytomas in the adrenal glands of rats, and hematopoietic cancers in rats. Chronic intermittent exposure to cobalt sulfate for 105 weeks caused an increased incidence in alveolar/bronchiolar adenoma or carcinoma in F344/N rats and B6C3F1 mice at ≥0.39 mg Co/m^3 in females and 1.11–1.14 mg Co/m^3 in males (Behl et al. 2015; Bucher et al. 1999; NTP 1998). Benign, complex, or malignant

pheochromocytoma were also observed in the adrenal glands in males at 0.39 mg Co/m^3 and in females at 1.11 mg Co/m^3. Increased incidence in alveolar/bronchiolar carcinoma was also observed in male and female F344/N rats and B6C3F1 mice exposed to concentrations ≥1.25 mg Co/m^3 as cobalt metal for 105 weeks (Behl et al. 2015; NTP 2014). Additional neoplastic changes observed following chronic-duration exposure to cobalt metal in rats included increased incidence of mononuclear cell leukemia in females at ≥1.25 mg Co/m^3 and bilateral benign pheochromocytoma in males at ≥1.25 mg Co/m^3 and in females at ≥2.5 mg Co/m^3 (Behl et al. 2015; NTP 2014).

No studies were identified regarding cancer effects in humans or animals after oral or dermal exposure to cobalt.

2.20 GENOTOXICITY

Available data indicate that cobalt has a low potential as a direct mutagenic agent; however, there is potential for indirect clastogenic and deoxyribonucleic acid (DNA) damaging effects based on findings from both *in vitro* and *in vivo* data. The overall weight of evidence indicates that cobalt and cobalt compounds are nonmutagenic in bacteria, yeast, and mammalian cells. The overall *in vitro* and *in vivo* evidence for the clastogenic potential of cobalt is mixed, with more recent *in vivo* studies (following guidelines with more rigorous criteria) demonstrating the absence of chromosomal damage following exposure to cobalt compounds. While *in vitro* results are mixed for DNA damage following exposure to various cobalt compounds, positive findings *in vivo* from human studies suggest that cobalt compounds have the potential to cause DNA damage, likely via reactive oxygen species. *In vitro* and *in vivo* studies of the genotoxic effects of cobalt are summarized in Tables 2-10 and 2-11, respectively.

Table 2-10. Genotoxicity of Cobalt *In Vitro*

Species (test system)	Endpoint	Results		Reference	Form
		With activation	Without activation		
Prokaryotic organisms					
Salmonella typhimurium (TA98)	Gene mutation	–	–	Kirkland et al. 2015	Cobalt metal powder
S. typhimurium (TA98, TA100)	Gene mutation	–	+ (TA98) +/– (TA100)	NTP 2014	Cobalt metal

Table 2-10. Genotoxicity of Cobalt *In Vitro*

Species (test system)	Endpoint	Results		Reference	Form
		With activation	Without activation		
Escherichia coli (WP2 *uvrA/*pKM101)	Gene mutation	–	–	NTP 2014	Cobalt metal
S. typhimurium (TA98, TA100, TA1535, TA1537, TA1538)	Gene mutation	Not tested	–	Arlauskas et al. 1985	Cobalt chloride
S. typhimurium (TA98, TA100, TA1535, TA1537, TA1538)	Gene mutation	Not tested	–	Kanematsu et al. 1980	Cobalt chloride
S. typhimurium (TA97a)	Gene mutation	–	–	Kirkland et al. 2015	Cobalt chloride
S. typhimurium (TA98, TA1538)	Gene mutation	–	Not tested	Mochizuki and Kada 1982	Cobalt chloride
S. typhimurium (TA1537, TA2367)	Gene mutation	Not tested	–	Ogawa et al. 1986	Cobalt chloride
S. typhimurium (TA97)	Gene mutation	Not tested	+	Pagano and Zeiger 1992	Cobalt chloride
S. typhimurium (TA100)	Gene mutation	Not tested	–	Tso and Fung 1981	Cobalt chloride
S. typhimurium (TA98, TA102, TA1535, TA1537)	Gene mutation	–	+ (TA98, TA1537) – (TA102, TA1535)	Wong 1988	Cobalt chloride
E. coli (WP2 uvrA p*Km* 101)	Gene mutation	Not tested	–	Arlauskas et al. 1985	Cobalt chloride
E. coli (B/r WP2 Try^-)	Gene mutation	Not tested	–	Kada and Kanematsu 1978	Cobalt chloride
E. coli (B/r WP2 *try*, WP2 *hcr try*)	Gene mutation (reversion)	Not tested	–	Kanematsu et al. 1980	Cobalt chloride
S. typhimurium (TA98, TA100, TA1535, TA1537, TA1538)	Gene mutation	Not tested	–	Kanematsu et al. 1980	Cobalt sulfate
S. typhimurium (TA100)	Gene mutation	–	–	Kirkland et al. 2015	Cobalt sulfate
S. typhimurium (TA100, TA1535, TA98)	Gene mutation	+ (TA100) – (TA1535, TA98)	+ (TA100) – (TA1535, TA98)	NTP 1991	Cobalt sulfate heptahydrate

Table 2-10. Genotoxicity of Cobalt *In Vitro*

Species (test system)	Endpoint	Results		Reference	Form
		With activation	Without activation		
S. typhimurium (TA100, TA1535, TA98)	Gene mutation	+ (TA100) – (TA1535, TA98)	+ (TA100) – (TA1535, TA98)	NTP 1998	Cobalt sulfate heptahydrate
S. typhimurium (TA100, TA1535, TA98)	Gene mutation	(+) (TA100) – (TA1535, TA98)	–	Zeiger et al. 1992	Cobalt sulfate heptahydrate
E. coli (B/r WP2 *try*, WP2 *hcr try*)	Gene mutation (reversion)	Not tested	–	Kanematsu et al. 1980	Cobalt sulfate
S. typhimurium (TA100, TA1535, TA97, TA98,	Gene mutation	–	–	NTP 2018	Cobalt naphthenate
S. typhimurium (TA98, TA100, TA1535, TA1537, TA1538)	Gene mutation	Not tested	–	Kanematsu et al. 1980	Cobalt hydroxide
E. coli (B/r WP2 *try*, WP2 *hcr try*)	Gene mutation (reversion)	Not tested	–	Kanematsu et al. 1980	Cobalt hydroxide
S. typhimurium (TA98, TA100, TA1535, TA1537, TA1538)	Gene mutation	Not tested	–	Kanematsu et al. 1980	Cobalt carbonate
E. coli (B/r WP2 *try*, WP2 *hcr try*)	Gene mutation (reversion)	Not tested	–	Kanematsu et al. 1980	Cobalt carbonate
E. coli ($WP2_s$ λ)	DNA damage	Not tested	–	Rossman et al. 1984	Cobalt chloride
Bacillus subtilis (H17, M45)	DNA damage	Not tested	(+)	Kanematsu et al. 1980	Cobalt chloride
B. subtilis (H17, M45)	DNA damage	Not tested	–	Nishioka 1975	Cobalt chloride
B. subtilis (H17, M45)	DNA damage	Not tested	+	Kanematsu et al. 1980	Cobalt sulfate
B. subtilis (H17, M45)	DNA damage	Not tested	+	Kanematsu et al. 1980	Cobalt hydroxide
B. subtilis (H17, M45)	DNA damage	Not tested	+	Kanematsu et al. 1980	Cobalt carbonate
Eukaryotic organisms					
Saccharomyces cerevisiae (D7)	Gene mutation (reversion)	Not tested	–	Fukunaga et al. 1982	Cobalt chloride

Table 2-10. Genotoxicity of Cobalt *In Vitro*

Species (test system)	Endpoint	Results		Reference	Form
		With activation	Without activation		
S. cerevisiae (D7)	Gene mutation (cross-over)	Not tested	+	Fukunaga et al. 1982	Cobalt chloride
S. cerevisiae (D7)	Gene mutation (reversion)	Not tested	–	Kharab and Singh 1985	Cobalt chloride
S. cerevisiae (IL126-1C, IL8-8D, SBTD-2B, DP1-1B/514)	Gene mutation (induction of rho minus mutation)	Not tested	+	Prazmo et al. 1975	Cobalt chloride
S. cerevisiae (D7)	Gene mutation (reversion)	Not tested	–	Singh 1983	Cobalt chloride
S. cerevisiae (D7)	DNA repair (gene conversion)	Not tested	+	Fukunaga et al. 1982	Cobalt chloride
S. cerevisiae (D7)	DNA repair (gene conversion)	Not tested	+	Kharab and Singh 1985	Cobalt chloride
S. cerevisiae (D7)	DNA repair (gene conversion)	Not tested	(+)	Singh 1983	Cobalt chloride
Mammalian cells					
SHE cells	Cell transformation	Not tested	+	Procter and Gamble 1995	Cobalt sulfate hydrate
SHE cells	Cell transformation	Not tested	+	Costa et al. 1982	Cobalt sulfide (crystalline)[a]
SHE cells	Cell transformation	Not tested	–	Costa et al. 1982	Cobalt sulfide (amorphous)[b]
Mouse lymphoma L5178Y cells	Mutation at the HPRT locus	(+)	–	Kirkland et al. 2015	Cobalt metal powder
Mouse lymphoma L5178Y cells	Mutation at the HPRT locus	–	–	Kirkland et al. 2015	Cobalt metal powder (extract)[c]
Mouse lymphoma cells (L5178Y/TK$^{+/-}$)	Mutagenic activity	Not tested	–	Amacher and Paillet 1980	Cobalt chloride
Chinese hamster V79 cells	Mutation at the HPRT locus	Not tested	+	Hartwig et al. 1990	Cobalt chloride
Chinese hamster V79 cells	Mutation at the HPRT locus	Not tested	(+)	Hartwig et al. 1991	Cobalt chloride
Chinese hamster V79 cells	Mutation at the HPRT locus	Not tested	(+)	Miyaki et al. 1979	Cobalt chloride
Mouse lymphoma L5178Y cells	Mutation at the HPRT locus	–	–	Kirkland et al. 2015	Cobalt sulfate

Table 2-10. Genotoxicity of Cobalt *In Vitro*

Species (test system)	Endpoint	Results		Reference	Form
		With activation	Without activation		
Mouse lymphoma L5178Y cells	Mutation at the HPRT locus	–	–	Kirkland et al. 2015	Cobalt sulfide
Mouse lymphoma L5178Y cells	Mutation at the HPRT locus	–	–	Kirkland et al. 2015	Cobalt dihydroxide
Mouse lymphoma L5178Y cells	Mutation at the HPRT locus	–	–	Kirkland et al. 2015	Cobalt tetraoxide
Mouse lymphoma L5178Y cells	Mutation at the HPRT locus	–	–	Kirkland et al. 2015	Cobalt oxide
Human lymphocytes	Sister chromatid exchange	Not tested	+	Andersen 1983	Cobalt chloride
Mouse macrophage p388d1 cells	Sister chromatid exchange	Not tested	+	Andersen 1983	Cobalt chloride
Chinese hamster V79 cells	Sister chromatid exchange	Not tested	+	Hartwig et al. 1991	Cobalt chloride
Human lymphocytes	Chromosomal aberrations	Not tested	–	Olivero et al. 1995	Cobalt chloride
Human lung fibroblast cells	Chromosomal aberrations	Not tested	+	Smith et al. 2014	Cobalt chloride
Human bronchial epithelial cells	Chromosomal aberrations	Not tested	+	Xie et al. 2016	Cobalt chloride
Human lung fibroblast cells	Chromosomal aberrations	Not tested	+	Smith et al. 2014	Cobalt oxide
Human lymphocytes	Chromosomal aberrations	Not tested	–	Capomazza and Botta 1991	Cobalt chloride
Human lymphocytes	Chromosomal aberrations	Not tested	–	Olivero et al. 1995	Cobalt sulfate
Human lymphocytes	Chromosomal aberrations	Not tested	–	Olivero et al. 1995	Cobalt nitrate
Human lymphocytes	Chromosomal aberrations	Not tested	–	Paton and Allison 1972	Cobalt nitrate
Human diploid cells (WI38, MRC5)	Chromosomal aberrations	Not tested	–	Paton and Allison 1972	Cobalt nitrate
Human bronchial epithelial cells	Chromosomal aberrations	Not tested	+	Xie et al. 2016	Cobalt oxide
Human peripheral blood mononucleated cells	Micronuclei	Not tested	+	De Boeck et al. 2003	Cobalt metal

Table 2-10. Genotoxicity of Cobalt *In Vitro*

Species (test system)	Endpoint	Results		Reference	Form
		With activation	Without activation		
Human osteosarcoma cells	Micronuclei	Not tested	+	Miller et al. 2001	Cobalt metal powder
Human lymphocytes	Micronuclei	Not tested	+	Van Goethem et al. 1997	Cobalt metal powder
Human lymphocytes	Micronuclei	Not tested	+	Capomazza and Botta 1991	Cobalt chloride
Human lymphocytes	Micronuclei	Not tested	+	Olivero et al. 1995	Cobalt chloride
Human bronchial epithelial cells	Micronuclei	Not tested	+	Uboldi et al. 2016	Cobalt chloride
Mouse fibroblasts (Balb/3T3)	Micronuclei	Not tested	–	Ponti et al. 2009	Cobalt chloride
Human lymphocytes	Micronuclei	Not tested	–	Olivero et al. 1995	Cobalt sulfate
SHE cells	Micronuclei	Not tested	+	Gibson et al. 1997	Cobalt sulfate hydrate
Human lymphocytes	Micronuclei	Not tested	–	Olivero et al. 1995	Cobalt nitrate
Human bronchial epithelial cells	Micronuclei	Not tested	+	Uboldi et al. 2016	Cobalt oxide
Mouse fibroblasts (Balb/3T3)	Micronuclei	Not tested	+	Ponti et al. 2009	Cobalt nanoparticles
Human lymphocytes	DNA damage	Not tested	+	De Boeck et al. 1998	Cobalt metal
Human peripheral blood mononucleated cells	DNA damage	Not tested	–	De Boeck et al. 2003	Cobalt metal
Human lymphocytes	DNA damage	Not tested	+	Van Goethem et al. 1997	Cobalt metal powder
Embryonic mouse stem cells	DNA damage	–	–	Derr et al. 2022	Cobalt metal
Human lymphocytes	DNA damage	Not tested	+	De Boeck et al. 1998	Cobalt-tungsten carbide alloy
Human lymphocytes	DNA damage	Not tested	+	Van Goethem et al. 1997	Cobalt-tungsten carbide alloy
Human sub mandibular gland ductal cells	DNA damage	Not tested	+	Akita et al. 2007	Cobalt chloride
Human hepatocarcinoma (HepG2) cells	DNA damage	Not tested	+	Alarifi et al. 2013	Cobalt chloride

Table 2-10. Genotoxicity of Cobalt *In Vitro*

Species (test system)	Endpoint	Results		Reference	Form
		With activation	Without activation		
Human T lymphocyte (Jurkat) cells	DNA damage	Not tested	+	Caicedo et al. 2008	Cobalt chloride
Human lymphocytes	DNA damage	Not tested	+	De Boeck et al. 1998	Cobalt chloride
Human diploid fibroblasts	DNA damage	Not tested	+	Hamilton-Koch et al. 1986	Cobalt chloride
Human HeLa cells	DNA damage	Not tested	+	Hartwig et al. 1990	Cobalt chloride
Human hepatoblastoma cells	DNA damage	Not tested	–	Kopp et al. 2018	Cobalt chloride
Human epithelial colorectal adenocarcinoma cells	DNA damage	Not tested	–	Kopp et al. 2018	Cobalt chloride
Human HeLa cells	Inhibition of DNA synthesis	Not tested	+	Painter and Howard 1982	Cobalt chloride
Human bronchial epithelial cells	DNA damage	Not tested	+	Uboldi et al. 2016	Cobalt chloride
Embryonic mouse stem cells	DNA damage	–	–	Derr et al. 2022	Cobalt chloride
Mouse fibroblasts (Balb/3T3)	DNA damage	Not tested	+	Ponti et al. 2009	Cobalt chloride
CHO cells	DNA damage	Not tested	+	Hamilton-Koch et al. 1986	Cobalt chloride
Embryonic mouse stem cells	DNA damage	–	–	Derr et al. 2022	Cobalt sulfide
Embryonic mouse stem cells	DNA damage	–	–	Derr et al. 2022	Cobalt carbonate
Embryonic mouse stem cells	DNA damage	–	–	Derr et al. 2022	Cobalt dihydroxide
Embryonic mouse stem cells	DNA damage	–	–	Derr et al. 2022	Cobalt tetraoxide
Human epithelial colorectal adenocarcinoma cells	DNA damage	Not tested	–	Kopp et al. 2018	Cobalt oxide
Human hepatoblastoma cells	DNA damage	Not tested	–	Kopp et al. 2018	Cobalt oxide
Human bronchial epithelial cells	DNA damage	Not tested	+	Uboldi et al. 2016	Cobalt oxide
Embryonic mouse stem cells	DNA damage	–	–	Derr et al. 2022	Cobalt oxide
Human hepatocarcinoma (HepG2) cells	DNA damage	Not tested	+	Alarifi et al. 2013	Cobalt oxide nanoparticles

Table 2-10. Genotoxicity of Cobalt *In Vitro*

Species (test system)	Endpoint	Results		Reference	Form
		With activation	Without activation		
Mouse fibroblasts (Balb/3T3)	DNA damage	Not tested	+	Ponti et al. 2009	Cobalt nanoparticles

[a]Cobalt sulfide typically has a crystalline structure.
[b]An amorphous form of cobalt sulfide was produced by precipitation of the sulfide salt from an ultrapure solution of cobalt chloride in water in the presence of an excess of ammonium sulfide.
[c]Cobalt metal powder was extracted in RPMI 1640 medium containing 5% heat-inactivated horse serum, plus 100 units/mL penicillin, 100 mg/mL streptomycin, 2.5 mg/mL amphotericin B, and 0.5 mg/mL pluronic for 72 hours and then undissolved solid was removed via centrifugation prior to testing.

– = negative result; + = positive result; (+) = weakly positive result; +/– = equivocal (an increase in revertants that is not dose related, not reproducible, or not of sufficient magnitude to support a determination of mutagenicity); CHO = Chinese hamster ovary; DNA = deoxyribonucleic acid; HPRT = hypoxanthine-guanine-phosphoribosyltransferase; SHE = Syrian hamster embryo

Table 2-11. Genotoxicity of Cobalt *In Vivo*

Species (exposure route)	Endpoint	Results	Reference	Form
Mammals				
Human (occupational)	Micronuclei (lymphocytes)	–	De Boeck et al. 2000	Cobalt dust
Human (occupational)	DNA damage (lymphocytes)	–	De Boeck et al. 2000	Cobalt dust
Human (environmental)	Oxidative DNA damage (urinary 8-OhdG)	+ (children) – (adults)	Banza Lubaba Nkulu et al. 2018	Cobalt dust
Human (occupational)	Oxidative DNA damage (urinary 8-OhdG)	–	De Boeck et al. 2000	Cobalt dust
Human (occupational)	Sister chromatid exchanges (lymphocytes)	+	Gennart et al. 1993	Cobalt metal
Human (occupational)	DNA damage (mononuclear blood cells)	+	Hengstler et al. 2003	Cobalt metal
Human (occupational)	Micronuclei (lymphocytes)	+	Iarmarcovai et al. 2005	Cobalt metal
Mice (inhalation)	Micronuclei (peripheral blood erythrocytes)	–	NTP 2014	Cobalt metal
Human (occupational)	DNA damage (lymphocytes)	+	Iarmarcovai et al. 2005	Cobalt metal
Human (occupational)	Oxidative DNA damage (blood 8-OHdG)	+	Berniyanti et al. 2020	cobalt metal
Rat (oral)	Chromosomal aberrations (spermatogonial cells)	–	Kirkland et al. 2015	cobalt chloride

Table 2-11. Genotoxicity of Cobalt *In Vivo*

Species (exposure route)	Endpoint	Results	Reference	Form
Mice (oral)	Chromosomal aberrations (bone marrow, spermatocytes)	+	Hassan et al. 2006	cobalt chloride
Mice (oral	Chromosomal breaks and aberrations (bone marrow)	+	Palit et al. 1991a, 1991b, 1991c, 1991d	cobalt chloride
Hamsters (i.p.)	Chromosomal aberrations (bone marrow)	+	Farah 1983	cobalt chloride
Hamsters (i.p.)	Chromosomal aberrations (testicular cells)	+	Farah 1983	cobalt chloride
Rat (oral)	Micronuclei (polychromatic erythrocytes)	+	Awoyemi et al. 2017	cobalt chloride hexahydrate
Mice (i.p.)	Micronuclei (bone marrow)	+	Rasgele et al. 2013	cobalt chloride
Mice (i.p.)	Micronuclei (bone marrow)	+	Suzuki et al. 1993	cobalt chloride
Rat (oral)	Chromosomal aberrations (bone marrow)	–	Kirkland et al. 2015	cobalt sulfate
Rat (oral)	Chromosomal aberrations (bone marrow)	–	Kirkland et al. 2015	cobalt oxide
Rat (oral)	Chromosomal aberrations (bone marrow)	–	Kirkland et al. 2015	cobalt tetraoxide
Rat (inhalation)	Oxidative DNA damage (lung tissue)	–	Burzlaff et al. 2022a	cobalt tetraoxide
Rat (i.p.)	Oxidative DNA damage (liver, kidney, lung)	+	Kasprzak et al. 1994	cobalt acetate
Nonmammalian eukaryotic organisms				
Drosophila melanogaster	Somatic mutation and recombination	+	Ertuğrul et al. 2020	Cobalt chloride
D. melanogaster	Somatic mutation and recombination	+	Kaya et al. 2002	Cobalt chloride
D. melanogaster	DNA damage (single-strand breaks)	+	Ertuğrul et al. 2020	Cobalt chloride
D. melanogaster	Somatic mutation and recombination	+	Ertuğrul et al. 2020	Cobalt nanoparticles
D. melanogaster	DNA damage (single-strand breaks)	+	Ertuğrul et al. 2020	Cobalt nanoparticles

– = negative result; + = positive result; 8-OhdG = 8-hydroxydeoxyguanosine; DNA = deoxyribonucleic acid; i.p. = intraperitoneal

Mutagenicity. In general, most studies reported that both soluble and poorly soluble/insoluble forms of cobalt were nonmutagenic to *Salmonella typhimurium* with or without metabolic activation (see Table 2-10 for references). However, a few studies reported positive or equivocal results. For cobalt metal, NTP (2014) reported equivocal results without metabolic activation in *S. typhimurium* strain

TA100 and weak mutagenicity (lacking dose response) in strain TA98 without metabolic activation. Cobalt chloride was mutagenic in strains TA97, TA98, and TA1537 without metabolic activation (Pagano and Zeiger 1992; Wong 1988). Cobalt sulfate was mutagenic in strain TA100 with and without metabolic activation in two NTP studies (NTP 1991, 1998). Zeiger et al. (1992) also reported mutagenicity for cobalt sulfate in strain TA100 with metabolic activation only. Soluble forms of cobalt were nonmutagenic in *Escherichia coli* without metabolic activation (Kanematsu et al. 1980). Cobalt chloride was nonmutagenic in *Saccharomyces cerevisiae* (Fukunaga et al. 1982; Kharab and Singh 1985; Singh 1983), but mutagenic in assays that examined gene cross-over and rho minus mutations (Fukunaga et al. 1982; Prazmo et al. 1975).

Mutations at the hypoxanthine-guanine-phosphoribosyltransferase (HPRT) locus were primarily negative for soluble and poorly soluble/insoluble cobalt forms tested in mouse lymphoma cells with or without metabolic activation (Amacher and Paillet 1980; Kirkland et al. 2015). A weak mutagenic response was reported for cobalt metal powder with metabolic activation in mouse lymphoma cells; however, these results were not reproducible with extracts of the powder, and were therefore not considered to be biologically relevant (Kirkland et al. 2015). Cobalt chloride induced HPRT mutations in Chinese hamster V79 cells without metabolic activation, with weakly positive results reported in two studies (Hartwig et al. 1991; Miyaki et al. 1979).

No *in vivo* mutagenicity data were identified.

Clastogenicity. *In vitro* results are mixed for the induction of chromosomal aberrations in various cultured human cells with soluble and poorly soluble/insoluble cobalt forms. Several studies reported negative results for chromosomal aberrations in human lymphocytes and human diploid cells exposed to soluble and poorly soluble/insoluble cobalt forms (Capomazza and Botta 1991; Olivero et al. 1995; Paton and Allison 1972). Conversely, cobalt chloride and cobalt oxide induced dose-dependent chromosomal aberrations in human bronchial epithelial and lung fibroblast cells (Smith et al. 2014; Xie et al. 2016). Increased sister chromatid exchanges were reported for cobalt chloride in human lymphocytes, mouse macrophage cells, and Chinese hamster V79 cells (Andersen 1983; Hartwig et al. 1991). Results for micronuclei induction in mammalian cells are generally positive for cobalt chloride, cobalt metal, and other soluble/insoluble cobalt forms (Capomazza and Botta 1991; De Boeck et al. 2003; Gibson et al. 1997; Miller et al. 2001; Olivero et al. 1995; Ponti et al. 2009; Uboldi et al. 2016; Van Goethem et al. 1997). Three studies reported negative results for micronuclei induction in mouse fibroblasts and human

lymphocytes treated with cobalt chloride, cobalt sulfate, or cobalt nitrate (Olivero et al. 1995; Ponti et al. 2009).

In vivo findings for clastogenicity of cobalt and cobalt compounds are mixed from both occupational studies in humans and laboratory studies in animals. Sister chromatid exchanges and micronuclei were increased in the lymphocytes of workers occupationally exposed to cobalt metal in the metal powder production and welding industries, respectively (Gennart et al. 1993; Iarmarcovai et al. 2005). Micronuclei were not increased in the lymphocytes of nonsmoking workers occupationally exposed to cobalt dust from cobalt refineries and hard metal plants (De Boeck et al. 2000). Conversely, micronuclei were elevated in the lymphocytes of cobalt dust-exposed workers who smoked (De Boeck et al. 2000). In laboratory animals, cobalt chloride administered orally in mice and intraperitoneally in hamsters resulted in chromosomal aberrations in bone marrow and reproductive cells (Farah 1983; Hassan et al. 2006; Palit et al. 1991a, 1991b, 1991c, 1991d). However, more recent studies following updated (and more stringent) study guidelines reported that cobalt chloride and various poorly soluble/insoluble cobalt forms administered orally in rats did not induce chromosomal aberrations in bone marrow or spermatogonial cells (Kirkland et al. 2015). NTP (2014) reported negative findings for micronuclei in the peripheral blood erythrocytes of mice following inhalation exposure to cobalt metal. Cobalt chloride induced micronuclei in the polychromatic erythrocytes and bone marrow of orally and intraperitoneally exposed rats and mice, respectively (Awoyemi et al. 2017; Rasgele et al. 2013; Suzuki et al. 1993). However, the increased micronuclei findings in the bone marrow are potentially due to erythropoiesis stimulation, rather than direct genotoxic action of cobalt (Kirkland et al. 2015).

DNA Interactions and Damage. *In vitro* findings in bacteria demonstrated increased DNA damage from exposure to cobalt chloride and poorly soluble/insoluble cobalt forms (Kanematsu et al. 1980). Two studies showed negative results for DNA damage in bacteria treated with cobalt chloride (Nishioka 1975; Rossman et al. 1984). Several studies reported positive findings for DNA damage in human lymphocytes treated with cobalt metal, cobalt metal powder, and cobalt-tungsten carbide alloy (De Boeck et al. 1998; Van Goethem et al. 1997). In contrast, De Boeck et al. (2003) and Derr et al. (2022) did not observe DNA damage in human peripheral blood mononucleated cells or in embryonic mouse stem cells following treatment with cobalt metal. Multiple studies of cobalt chloride revealed DNA damage in cultured human and rodent cells (see Table 2-10 for references). However, more recent studies conducted in cultured human cells and embryonic mouse stem cells treated with cobalt chloride (or poorly soluble/insoluble forms of cobalt) did not result in DNA damage (Derr et al. 2022; Kopp et al. 2018). Positive findings for DNA damage were reported for cobalt oxide, cobalt oxide nanoparticles, and cobalt

nanoparticles following treatment in cultured human cells and mouse fibroblasts (Alarifi et al. 2013; Ponti et al. 2009; Uboldi et al. 2016). Additional *in vitro* assessments of cobalt chloride were either positive or weakly positive for DNA repair via gene conversion in bacteria (Fukunaga et al. 1982; Kharab and Singh 1985; Singh 1983) and positive for inhibition of DNA synthesis in human HeLa cells (Painter and Howard 1982).

DNA damage and oxidative DNA damage *in vivo* have been demonstrated in mononuclear blood cells, serum, and lymphocytes obtained from workers occupationally exposed to cobalt metal (Berniyanti et al. 2020; Hengstler et al. 2003; Iarmarcovai et al. 2005). De Boeck et al. (2000) reported negative findings for DNA damage and oxidative DNA damage in the lymphocytes and urine, respectively, of nonsmoking workers occupationally exposed to cobalt dust from cobalt refineries and hard metal plants. Conversely, oxidative DNA damage was increased in the lymphocytes of cobalt dust-exposed workers who smoked (De Boeck et al. 2000). Increased levels of a urinary biomarker for oxidative damage (8-hydroxydeoxy-guanosine) was also associated with increased urinary cobalt levels in children residing in mining regions of the Democratic Republic of the Congo (D.R. Congo) (Banza Lubaba Nkulu 2018). Similar associations were not observed in adults; however, median urinary cobalt levels in children were nearly 3 times the levels observed in adults. Increased exposure in children may be due to hand-to-mouth activities recognized to occur with greater frequency in children compared to adults; Banza Lubaba Nkulu (2018) did not address this confounding factor.

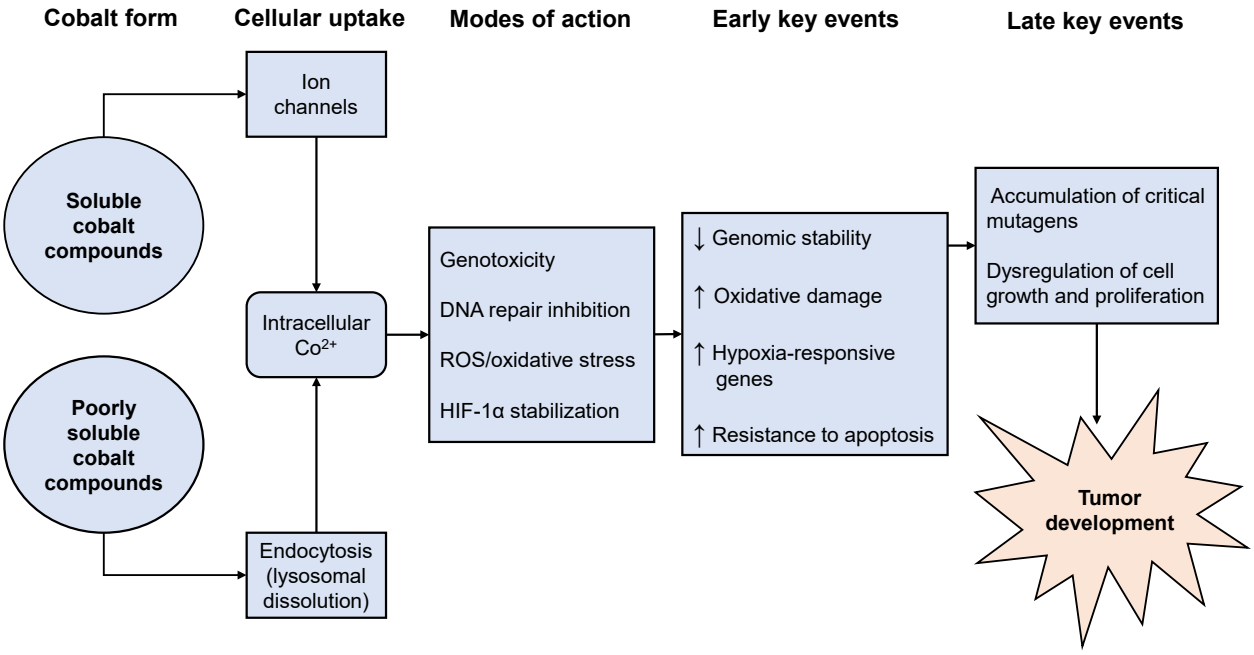
In *in vivo* studies in animals, cobalt tetraoxide administered to rats via inhalation did not result in oxidative damage in lung tissue (Burzlaff et al. 2022a). However, oxidative DNA damage increased in the liver, kidney, and lung of rats following intraperitoneal administration of cobalt acetate (Kasprzak et al. 1994). Positive findings were reported for DNA damage and somatic mutations/recombinations in *Drosophila melanogaster* following exposure to cobalt chloride and cobalt nanoparticles (Ertuğrul et al. 2020; Kaya et al. 2002).

Cell Transformation. Cell transformation assays performed in Syrian hamster embryo cells were positive for cobalt sulfate and crystalline cobalt sulfide, while negative results were reported for amorphous cobalt sulfide (Costa et al. 1982; Procter and Gamble 1995). Crystalline is the natural structure for cobalt sulfide; amorphous cobalt sulfide was generated by precipitation of the sulfide salt from an ultrapure solution of cobalt chloride in water in the presence of an excess of ammonium sulfide.

2.21 MECHANISM OF ACTION

Soluble and insoluble forms of cobalt give rise to toxicity and carcinogenicity in animal models following cellular uptake of the metal and subsequent release of cobalt ions from its salts. These ions elicit a cascade of downstream biological effects. The extracellular release of cobalt ions from water-soluble compounds is transported into the cells through the ion channels or via endocytosis of poorly soluble cobalt compounds. The poorly soluble cobalt compounds are then solubilized in the acidic environment and released as ionic cobalt in the intracellular space. While the exact mechanism(s) for the transport of cobalt cations through cellular membranes are unknown, the natural resistance-associated macrophage protein 2 (NRAMP 2)/divalent metal transporter 1 (DMT1) can play a role in this transport (Forbes and Gros 2003). There are several plausible ways through which these ions can cause toxicity *in vivo* (Figure 2-4). These include inhibition of DNA repair, genotoxicity, generation of reactive oxygen species (ROS) resulting in oxidative damage, and stabilization of hypoxia-inducible factor 1α (HIF-1α), a protein that increases the expression of genes that promote survival of cells when they receive less oxygen (NTP 2016).

Figure 2-4. Mechanistic Events Associated with Cobalt Toxicity and Carcinogenicity



Source: NTP (2016)

Calcium influx in cells is known to be altered by soluble cobalt when it blocks the inorganic calcium channels in cells harvested from rodent models (Henquin and Lambert 1975; Moger 1983; Yamatani et al. 1998). Blocking these channels is associated with a decrease in steroidogenesis in mouse Leydig cells (Moger 1983). The ubiquitous calcium channels in liver cells harvested from rats (Yamatani et al. 1998) and pancreatic cells harvested from mice (Henquin and Lambert 1975) also get blocked by cobalt. Cobalt also affects neuromuscular calcium transmission because muscle tissues have an abundance of calcium ion channels in an *in vitro* sartorius nerve muscle preparation (Weakly 1973). An *in vitro* study of cobalt chloride on PND 3 rat cochlear organotypic cultures reported damage to cochlear hair cells and peripheral auditory nerve fibers, along with loss of spiral ganglion neurons that were concentration and duration dependent; these occurred along with increased expression of superoxide radicals and increased expression of caspase-3 in hair cells indicative of apoptotic mediation (Li et al. 2015).

Cobalt is also known to interfere with normal homeostatic degradation of HIF-1α under normal cellular oxygen conditions; thus, exposure to cobalt can often mimic hypoxic conditions in *in vitro* models (van den Brule et al. 2022; Yuan et al. 2003). This stabilization of HIF-1α triggers a cascade of cellular responses to hypoxia (despite normoxic conditions). Interaction of cobalt compounds with HIF-1α is highly dependent upon the bioaccessibility of cobalt; compounds with high intracellular bioaccessibility have a greater potential to stabilize HIF-1α (cobalt chloride, cobalt sulfate, cobalt nitrate, cobalt oxide, cobalt hydroxide, cobalt acetate, cobalt metal, cobalt hydroxide) compared to compounds with low intracellular bioaccessibility (cobalt carbonate, cobalt sulfide, cobalt tetraoxide) (van den Brule et al. 2022; Verougstraete et al. 2022). Stabilization of HIF-1α and activation of downstream targets (e.g., erythropoietin) has been proposed as the underlying mechanisms for several cobalt-associated health effects, particularly polycythemia (Hoffmeister et al. 2018; NTP 2016). Hoffmeister et al. (2019) demonstrated single or repeated oral exposure to low doses of cobalt (<1 mg Co/kg/day) increased erythropoietin levels in humans. The testicular degeneration seen as a result of cobalt exposure can be a result of the testis itself becoming hypoxic due to blockage of veins and arteries by increases in the number of red blood cells, alterations in permeability due to thickening of basal lamina and basement membranes, and enlargement of interstitial Leydig cells in a rodent model (Elbetieha et al. 2008; Mollenhauer et al. 1985). Hypoxia can also be observed in other tissues such as cardiac, brain, liver, and renal from rats and mice (Mayfield et al. 1994; Morelli et al. 1994). However, repeated low-level exposure to cobalt may lead to increased tolerance of hypoxic conditions via activation of HIF-1α and its downstream gene targets, facilitating oxygen delivery via angiogenesis, vasodilation, glucose transport, and scavenging of oxygen radicals (Shrivastava et al. 2008).

Cobalt ions can damage DNA by inhibiting DNA polymerization, thus affecting DNA repair in human fibroblasts (Kasten et al. 1997). It can also cause induction of oxidative damage in a mouse model and human lung fibroblast cells (Lison 2015; Smith et al. 2014). Changes in hepatic enzymes like superoxide dismutase, catalase, GPx, and heme oxygenase are associated with an increase in lipid peroxidation in the liver, which is a direct result of an increase in oxidative damage in *in vivo* animal models (Akinrinde et al. 2016a; Awoyemi et al. 2017; Christova et al. 2001, 2002). Several authors reported elevated markers of oxidative stress in rats following oral exposure to cobalt associated with toxic effects in various organs, including the heart, intestines, liver, kidney, and brain; some studies demonstrated that co-administration with an antioxidant attenuates toxic effects (Ajibade et al. 2017; Akinrinde et al. 2016a, 2016b, 2016c, 2019; Clyne et al. 2001; Garoui et al. 2011, 2012, 2013; Oria et al. 2022).

2.22 COBALT NANOPARTICLES

This section provides a brief overview of cobalt nanoparticle (CoNP) toxicity and focuses on highlighting key findings from experimental animal studies and *in vitro* studies using human and animal cell lines. No epidemiologic studies focusing on the health effects of exposure to CoNPs were identified. Increased levels of cobalt ions in serum and testes were observed in male rats after *in vivo* exposure of 500 μg/kg body weight via an intra-articular injection (Wang et al. 2013). *In vivo* exposure to CoNPs at a dose of 20 mg/kg body weight via intravenous exposure in New Zealand rabbits demonstrated accumulation of CoNPs in lung, liver, and kidney tissues after a histopathological examination (Hanini et al. 2016). No other toxicokinetic studies examining the absorption, metabolism, or excretion of CoNPs were identified. *In vitro* models using human cell lines have demonstrated that CoNPs induce metabolic impairment, oxidative stress, and cytotoxicity (Alinovi et al. 2015, 2017; Bastian et al. 2009). Research on the effects of CoNPs in animals is limited but generally suggests that CoNPs are toxic in laboratory animals. Several *in vivo* and *in vitro* studies have demonstrated that CoNPs increase the production of ROS and reactive nitrogen species, which have both been previously shown to be associated with inflammation, genotoxicity, cytotoxicity, and reproductive toxicity (Hussien and Mohamed 2018; Moche et al. 2015; Monteiller et al. 2007).

Primary target organs for CoNPs toxicity include the testicles, brain, and lungs. Male rats exposed to CoNPs at a dose of 500 μg/kg body weight via an intra-articular injection, once per week for 10 consecutive weeks, suffered from testicular damage; reduced epididymal sperm motility, viability, and concentration; and increased abnormal sperm rate (Wang et al. 2013). In male Wistar rats, significant neural damage was observed in both the hippocampus and the cortex of the temporal lobe at a dose of

2 mg/kg body weight administered intraperitoneally once per day for 20 days (Zheng et al. 2019). Zheng et al. (2019) also compared the neurotoxic potential of cobalt chloride and CoNPs and identified that the nanoparticles showed greater neurotoxic potency. Male albino rats exposed to a single oral dose of 1 g/kg body weight of CoNPs via food had an increase in relative brain, kidney, and liver weights, along with increases in erythrocyte and hemoglobin counts (Ali 2019). Acute-duration inhalation exposure to 2.12 mg/m^3 CoNPs for 4 days (5 hours/day) caused slight damage to respiratory tissues in rats when the lungs were assessed by electron microscopy; no histopathological changes were observed using standard light microscopy (Kyono et al. 1992). No damage was observed after a single 5-hour exposure to 2.72 mg/m^3 CoNPs (Kyono et al. 1992). No respiratory effects were observed 24 hours post treatment in male Sprague-Dawley rats exposed to a single dose of 62.5 µg CoNPs intratracheally; however, this study included only three rats in the treatment group (Brown et al. 2018). In transgenic mice (gpt delta) that were intratracheally instilled with 50 µg CoNPs per mouse and examined on days 1, 3, 7, and 28 after exposure, toxic effects identified in the respiratory system included lung inflammation, oxidative stress, injury, and cell proliferation, which further resulted in DNA damage and DNA mutation (Wan et al. 2017). Sprague-Dawley rats that underwent subcutaneous implantation of CoNPs developed subcutaneous and intramuscular nodules; toward the end of the study period (6 months), all treated animals developed handicapping tumors (Hansen et al. 2006).

The overall database for CoNPs in mammals is limited to a few studies in rats, mice, and rabbits. While CoNPs are becoming increasingly useful for various healthcare-related applications, the toxicity profile and toxicokinetics for these CoNPs need to be studied further. More studies need to be conducted to examine how CoNPs affect the physiology in each organ system. Exposures to CoNPs from inhalation, dermal, and oral routes, as well as via prosthetics and therapeutics, need to be studied. Since CoNPs have distinct physical and chemical properties that are different from other cobalt compounds, a focused effort should be made on developing a complete toxicological profile to better understand the health effects and toxicokinetics of these unique chemicals.