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.

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. LOAELs have been classified into “less serious” or “serious” effects. “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. “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). 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.

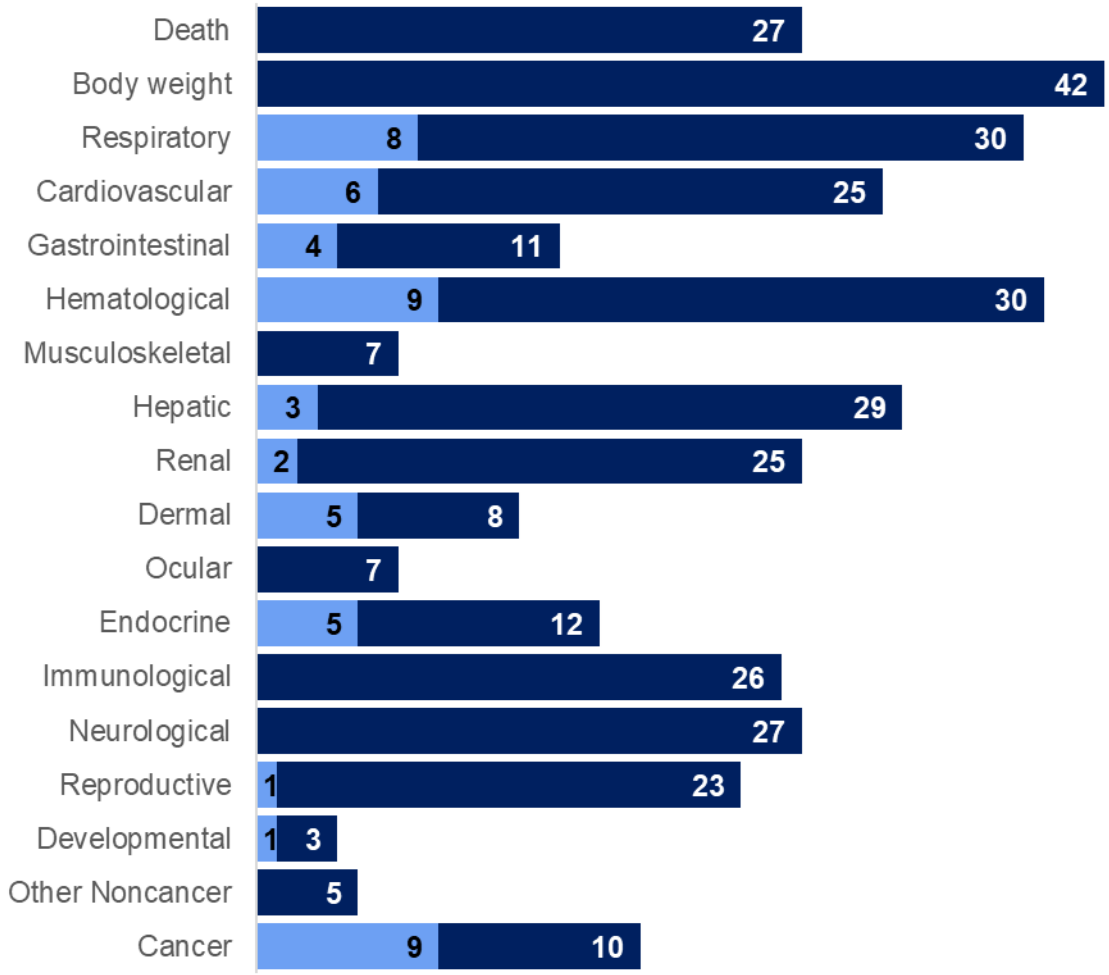
A User's Guide has been provided at the end of this profile (see Appendix D). This guide should aid in the interpretation of the tables and figures for LSEs and MRLs.

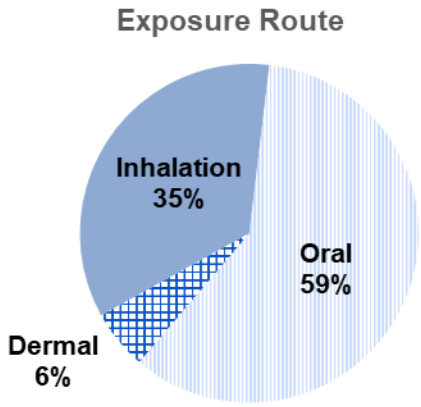
The health effects of cobalt were evaluated in laboratory animals and a few human occupational and controlled-exposure studies. As illustrated in Figure 2-1, most of the health effects data come from inhalation and oral studies in animals. Animal data are available for each exposure route and exposure duration category except for chronic-duration oral and chronic-duration dermal exposure. The effects on body weight are those that were examined most frequently in the literature followed by respiratory and hematological effects. There are few human studies that include control groups and occupational studies that examine health effects of exposure to cobalt. In those studies, both respiratory and hematological endpoints were identified as health effects following cobalt exposure. The cobalt database includes studies of its genotoxicity.

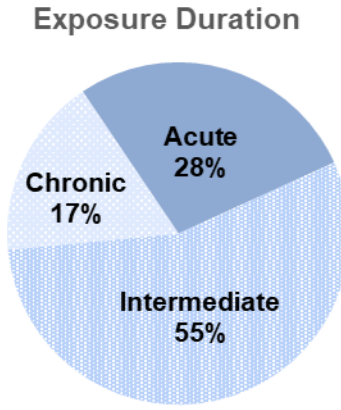
Figure 2-1. Overview of the Number of Studies Examining Cobalt Health Effects

***Most studies examined the potential body weight, hematological, and respiratory 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 100 studies (including those finding no effect) have examined toxicity. Studies may have examined more than one endpoint for health effects.

As outlined in Chapter 1, the most sensitive effects from inhaled cobalt exposure appear to be respiratory, hematological, renal, and cancer. Meanwhile those from ingested cobalt exposure appear to be gastrointestinal, endocrine, and hematological. Overall, respiratory and hematological effects are considered the most significant from a health perspective and are detailed below. The available human toxicity studies were primarily evaluated for the respiratory and hematological endpoints including in controlled-exposure studies. Observational and controlled-exposure cohort studies and population level studies have primarily examined respiratory, cardiovascular, gastrointestinal, and hematological endpoints. Animal studies have examined all endpoints following oral and inhalation exposure to cobalt. The respiratory and hematological endpoints were the most examined in animal studies. Animal studies have also examined body weight, renal, hepatic, and reproductive effects in oral animal studies. A very limited number of animal studies examined toxicity following dermal exposure. Respiratory and hematological effects were considered the most sensitive outcomes of cobalt exposure due to the frequency at which they were reported and the relatively low doses at which these health effects were observed (0.0151 and 0.8 mg Co/m^3, respectively), as health effects were observed even at low doses, and are commonly reported in case studies. Therefore, a systematic review (Appendix C) was conducted on these endpoints. The information in those human and animal studies indicate the following regarding potential targets of cobalt toxicity:

- ***Respiratory Effects***. Human and laboratory animal studies support respiratory toxicity as a sensitive endpoint following inhalation exposure to cobalt. Inhaled cobalt in humans was absorbed in the lungs and was associated with increases in chronic phlegm and decreases in spirometric parameters (Hamzah et al. 2014; Linna et al. 2003; Sauni et al. 2010; Walters et al. 2012). Chronic inhaled cobalt exposure is associated with decreased lung function in exposed workers, as well as increased cough sputum and dyspnea (Gennart and Lauwerys 1990; Kusaka et al. 1986a). Evidence from animal studies indicates that acute-duration cobalt inhalation particulate exposure causes pulmonary irritation, dose-dependent edema, and damage in the lungs (Palmes et al. 1959). Intermediate-duration inhalation of cobalt resulted in lesions and degeneration in respiratory tract tissues (Bucher et al. 1990; NTP 1991), including a 25% increase in lung weights, tissue inflammation with infiltrates of mainly neutrophils, lymphocytes, and eosinophils (Johansson et al. 1987; Johansson et al. 1992). Chronic-duration animal exposures caused inflammation in the nose, larynx, and lung combined with emphysema and lesions in the

respiratory tract (NTP 1998, 2014; Wehner et al. 1977). These findings in animal and human studies indicate inhalation exposure to cobalt can cause respiratory toxicity.

- ***Hematological Effects***. Several studies in animals and a few human studies lend support to hematological effects being a sensitive endpoint following both inhalation and oral exposures to cobalt. Some studies have called the effects polycythemia. When addressed in this profile, polycythemia refers to absolute polycythemia, which is an increase in red cell mass from exposure to a substance, such as cobalt. This profile does not address other forms or causes of polycythemia. Absolute polycythemia, when mentioned in this profile refers to polycythemia caused by cobalt toxicity. Other causes of erythrocytosis and polycythemia that will not directly be discussed in this profile include primary polycythemia (such as polycythemia vera or familial polycythemia), secondary polycythemia (elevated serum erythropoietin [EPO] as might be seen from a deficient oxygen supply), or relative polycythemia (plasma volume contraction as associated with dehydration).
  In the study by Lantin et al. (2011), the integrated exposure index (IEI) was significantly ($P< 0.001$) correlated with mean corpuscular hemoglobin concentration (MCHC) in both univariate and multivariate regression analyses, but there was no significant relationship between the IEI and the red cell count (red cell count was not affected), even after occupational exposure to inhaled cobalt (Lantin et al. 2011). Studies in laboratory animals examined higher concentrations of inhaled cobalt and identified changes in the levels of hemoglobin, basophils, and monocytes (NTP 1991, 2014; Palmes et al. 1959). Oral exposure to cobalt and cobalt compounds in humans and animals caused an increase in levels of erythrocytes (Davis and Fields 1958; Awoyemi et al. 2017). Davis and Fields (1958) reported an increase in erythrocyte levels that returned to normal upon cessation of cobalt exposure. The animal studies corroborated the effects seen in human studies. Acute-duration oral exposure to cobalt increased red blood cells, hematocrit, and hemoglobin (Awoyemi et al. 2017; Domingo and Llobet 1984; Shrivastava et al. 2010). Intermediate-duration oral exposure to cobalt also had effects in animals (Brewer 1940; Bryan and Bright 1973; Chetty et al. 1979; Corrier et al. 1985; Krasovskii and Fridlyand 1971) that were similar to those seen in the acute-duration studies. Pregnant dams exposed to cobalt orally showed unspecified changes in levels of hemoglobin and hematopoiesis (Gluhcheva et al. 2014). These findings in animal and human studies indicate oral exposure to cobalt can cause hematological toxicity.

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/m^3)	Parameters monitored	Endpoint	NOAEL (mg/m^3)	Less serious LOAEL (mg/m^3)	Serious LOAEL (mg/m^3)	Effects
ACUTE EXPOSURE									
Kusaka et al. 1986									**Cobalt Metal**
1	HUMAN 15M	6 hours	0, 0.038	CS OF	Resp		0.038		Non-dose related decrease in FVC
Palmes et al. 1959									**Cobalt Hydrocarbonyl**
2	RAT (Albino) 5M	30 minutes/day, Once	0, 7, 26, 47, 68, 78, 118, 191, 215, 222, 408	CS LE	Death			78	3/5 dead
					Resp	7	26	90	LOAEL: Edema (not otherwise described) observed at 26 mg/m^3 SLOAEL: Severe Edema (not otherwise described) observed at 90 mg/m^3
INTERMEDIATE EXPOSURE									
NTP 1991									**Cobalt Sulfate Heptahydrate**
3	RAT (F344/N) 10M, 10F	13 weeks, 5 days/week, 7 hours/day	0, 0.06, 0.21, 0.61, 2.09, 6.29	BC BI BW CS GN HE HP IX LE NX OF OW	Bd wt	6.29 F			
						2.09 M	6.29 M		17% decrease in body weight
					Resp	0.06 F	0.21 F		14% increase in lung weight in females
							0.06 M		7% increase in relative lung weight in males
					Cardio	6.29			

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Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/m^3)	Parameters monitored	Endpoint	NOAEL (mg/m^3)	Less serious LOAEL (mg/m^3)	Serious LOAEL (mg/m^3)	Effects
					Hemato	0.61 F	2.09 F		Polycythemia seen in female rats; the platelet count decreased in females (not otherwise described)
						0.21 M	0.61 M		Increases in erythrocytes, mean hemoglobin concentration, and hematocrit in male rats
					Renal	6.29 F			
							0.06 M		6% increase in kidney weight; increase in epithelial cells in urine (not otherwise described)
					Ocular	6.29			
					Endocr	0.61 F	2.09 F		Low T3 (not otherwise described)
						2.09 M	6.29 M		Low TSH (not otherwise described)
					Immuno	6.29			
					Neuro	6.29			
					Repro	6.29			
					Cancer			0.06 F	7/10 rats showed chronic inflammation of larynx and squamous metaplasia in the larynx (respiratory system)
								0.06 M	9/10 rats showed chronic inflammation of larynx and squamous metaplasia in the larynx (respiratory system)

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NTP 1991									**Cobalt Sulfate Heptahydrate**
4	RAT (F344/N) 5M,5F	16 days, 5 days/week, 6 hour/day	0, 0.02, 0.1, 0.99, 10.5, 41.72	BC BI BW CS GN HE HP IX LE NX OF OW	Death			41.72 F	5/5 died
								10.5 M	2/5 died
					Bd wt	0.99 F		10.5 F	22% decrease in bodyweight
						0.99 M		10.5 M	47% decrease in bodyweight
					Resp	0.99	10.5		Degeneration of olfactory epithelium, hyperplasia and squamous metaplasia in the epithelium of respiratory turbinates; inflammation in the nose and lungs (not otherwise described)
					Cardio	10.5			
					Hepatic		41.72		Congestion and necrosis of liver (not otherwise described)
					Ocular	0.99	10.5		Chromodacryorrhea (not otherwise described)
					Immuno	0.99	10.5		Necrosis of thymus and decrease in weights for both males and females (not otherwise described)
					Neuro	0.99	10.5		Congestion of vessels in brain and hypoactivity (not otherwise described)
					Repro	10.5 F			

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation

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						0.99 M	10.5 M		Atrophy in testes- decrease in number of cells in the seminiferous tubules and atypical germinal epithelial cells in the epididymal ducts (not otherwise described)
NTP 2014									**Cobalt Metal**
5	RAT (F344/N) 5M, 5F	16 days, 5 days/week, 6 hours + T90 (12 minutes)/day	0, 2.5, 5, 10, 20, 40	BC 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% less body weight than controls SLOAEL: 45% less body weight than controls
						5 M		20 M	20% less body weight than controls
					Resp		2.5 F	20 F	LOAEL: Significantly increased incidence of cytoplasmic vacuolization of bronchiole epithelium, and atrophy and necrosis of olfactory epithelium SLOAEL: respiratory epithelium necrosis

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/m^3)	Parameters monitored	Endpoint	NOAEL (mg/m^3)	Less serious LOAEL (mg/m^3)	Serious LOAEL (mg/m^3)	Effects
							2.5 M	20 M	LOAEL: Significantly increased incidence of cytoplasmic vacuolization of bronchiole epithelium, and atrophy of olfactory epithelium SLOAEL: abnormal breathing; increased incidence of lung hemorrhage and acute inflammation
					Cardio	20 F			
						10 M			
					Hepatic	10 F	20 F		Significant 15.9% increase in relative liver weight (in 2/5 rats) compared to controls
							2.5 M		Significant 12.8% decrease in relative liver weight compared to controls
					Renal	10 F	20 F		Significant 291% increase in urinary creatinine levels and 23.5% increase in relative kidney weight, compared to controls
						5 M	10 M		Significant 7.5% decrease of relative left kidney weight and 80% increase in urinary creatinine, compared to controls
					Endocr	20 F			
						10 M			
					Immuno	10 F	20 F		64% decrease in relative thymus weight
						10 M			

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					Neuro	20 F	40 F		Lethargy
						10 M	20 M		Lethargy
					Repro	10 M			
NTP 2014									**Cobalt Metal**
6	RAT (F344/N) 10M, 10F	14 weeks, 5 days/week, 6 hours + T90 (12 minutes)/day	0, 0.625, 1.25, 2.5, 5	BW CS GN HP OW RX	Bd wt	5			
					Resp			0.63	Increased incidence of chronic active inflammation in lung, pulmonary alveolar proteinosis, and increased relative lung weight (16-22%), all compared to controls
					Cardio	5			
					Gastro	5			
					Hemato	0.63 F	1.25 F		9% increase in hematocrit, hemoglobin, and erythrocyte levels, compared to controls
							0.63 M		4.5% increase in hemoglobin levels and 5.2% increase in erythrocytes, compared to controls
					Musc/skel	5			
					Hepatic	5			
					Renal	2.5 F	5 F		Significant 12.7% increase in relative right kidney weight, compared to controls

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/m^3)	Parameters monitored	Endpoint	NOAEL (mg/m^3)	Less serious LOAEL (mg/m^3)	Serious LOAEL (mg/m^3)	Effects
						2.5 M	5 M		23.5% increase in urinary creatinine by week 14 compared to controls
					Dermal	5			
					Ocular	5			
					Endocr	5			
					Immuno	5			
					Neuro	5			
					Repro	5 F			
						1.25 M		2.5 M	Significant decrease in % sperm motility by 5.6% and 10.1% increase in relative weight of right testis, all compared to controls
Palmes et al. 1959									**Cobalt Hydrocarbonyl**
7	RAT (Albino) 34-57 M	3 months, 5 days/week, 6 hours/day	0, 9	BW CS GN HE HP UR	Bd wt	9			
					Resp		9		Lung inflammation (not otherwise described)
					Hemato		9		10% increase in hemoglobin
NTP 1991									**Cobalt Sulfate Heptahydrate**
8	MOUSE (B6C3F1) 5M, 5F	16 days, 5 days/week, 6 hour/day	0, 0.02, 0.1, 0.99, 10.5, 41.72	BC BI BW CS GN HE HP IX LE NX OF OW	Death			10.5	4/5 male and 1/5 female mice died
					Bd wt	0.99		10.5	33% and 20% decrease in body weight in males and females, respectively

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Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/m^3)	Parameters monitored	Endpoint	NOAEL (mg/m^3)	Less serious LOAEL (mg/m^3)	Serious LOAEL (mg/m^3)	Effects
					Resp	0.1			
								0.99 F	Gray discoloration of lungs and fluid in larynx and trachea; 25% increase in relative lung weight; degeneration of olfactory epithelium
								0.99 M	Gray discoloration of lungs and fluid in larynx and trachea; 22% increase in relative lung weight; degeneration of olfactory epithelium
					Cardio	41.72			
					Gastro	41.72			
					Musc/skel	41.72			
					Hepatic	10.5 F			
						0.99 M	10.5 M		Necrosis of hepatocytes (not otherwise described)
					Renal	10.5			
					Dermal	10.5			
					Ocular	10.5	10.5		Chromodacryorrhea was observed (not otherwise described)
					Endocr	10.5			
					Immuno		10.5 M		Decrease in relative thymus weight (not otherwise described)
					Neuro	0.99	10.5		Hypoactivity and congestion of vessels in brain (not otherwise described)
					Repro	10.5			

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/m^3)	Parameters monitored	Endpoint	NOAEL (mg/m^3)	Less serious LOAEL (mg/m^3)	Serious LOAEL (mg/m^3)	Effects
					Cancer			10.5	Hyperplasia of the squamous epithelium in the larynx (not otherwise described)
NTP 1991									**Cobalt Sulfate Heptahydrate**
9	MOUSE (B6C3F1) 10M, 10F	13 weeks, 5 days/week, 7 hours/day	0, 0.06, 0.21, 0.61, 2.09, 6.29	BC BI BW CS GN HE HP IX LE NX OF OW	Death			6.29 M	2/10 died
					Bd wt	2.09			
								6.29 F	22% decrease in body weight
							6.29 M		14% decrease in body weight
					Resp	0.06 F		0.21 F	9/10 showed histiocytic infiltrates
								0.06 M	10/10 showed histiocytic infiltrates
					Gastro	6.29			
					Hemato		0.06 F		5% decrease in hemoglobin and 3% decrease in hematocrit
						0.06 M	0.21 M		4% decrease in platelet count
					Musc/skel	6.29			
					Hepatic	6.29			
					Renal	6.29			
					Dermal	6.29			
					Immuno	2.09	6.29		Lymph node hyperplasia (not otherwise described)
					Repro	2.09 F		6.29 F	Increased length estrous cycle in females by 19%
								0.61 M	Decreased sperm motility by 79%

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					Cancer			0.06	Larynx metaplasia in 7/10 male and 8/10 female mice
NTP 2014									**Cobalt Metal**
10	MOUSE (B6C3F1) 5M, 5F	17 days, 5 days/week, 6 hours + T90 (12 minutes)/day	0, 2.5, 5, 10, 20, 40	BW GN HP LE OW	Death			40	4/10 died
					Bd wt	10 F	20 F	40 F	LOAEL: 16.3% less body weight compared to controls SLOAEL: 37.5% less body weight than controls
						20 M		40 M	26.5% less body weight than controls
					Resp			2.5	Increased incidence of cytoplasmic vacuolization of bronchiole and respiratory epithelium, and atrophy of olfactory epithelium
					Cardio	20 F	40 F		Significant 39% increase of relative heart weight compared to controls
						40 M			
					Hepatic		2.5		Significant 10%-11% decrease in relative liver weight
					Renal	20			
					Endocr	40			

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					Immuno	2.5	5		Significantly increased incidence of minimal to moderate alveolar histiocytic cellular infiltration (accumulation of macrophages within the alveolar spaces and septa)
					Neuro	5 F	10 F		Lethargy
						10 M	20 M		Lethargy
					Repro	40 M			
NTP 2014									**Cobalt Metal**
11	MOUSE (B6C3F1) 10M, 9-10F	14 weeks, 5 days/week, 6 hours + T90 (12 minutes)/day	0, 0.625, 1.25, 2.5, 5, 10	BW CS GN HP OW RX	Bd wt	5	10		13%-14% decrease in body weight compared to controls
					Resp			0.63	Cytoplasmic vacuolization of bronchiole epithelium and squamous metaplasia of larynx in all mice
					Cardio	10			
					Gastro	10			
					Hemato	5 F	10 F		Significant 4.7% increase in erythrocytes compared to controls
						5 M	10 M		Significant 3% increase in hemoglobin and erythrocyte levels compared to controls
					Musc/skel	10			

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Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/m^3)	Parameters monitored	Endpoint	NOAEL (mg/m^3)	Less serious LOAEL (mg/m^3)	Serious LOAEL (mg/m^3)	Effects
					Hepatic	1.25 F	2.5 F		Significant 8.4% decrease of relative liver weight
						5 M	10 M		Significant 14.2% decrease of relative liver weight
					Renal	2.5	5		Significant 7.7%-12.4% decrease in relative kidney weight compared to controls
					Dermal	10			
					Ocular	10			
					Endocr	10			
					Immuno		0.63		Alveolar histiocytic cellular infiltration characterized by the presence of low to moderate numbers of histiocytes (macrophages)
					Neuro	10			
					Repro	5 F	10 F		Significantly longer estrous cycle
						1.25 M	2.5 M		Significant 4.7% decrease in % sperm motility compared to controls
Camner et al. 1993									**Cobalt Chloride**
12	GN PIG (Hartley) 6 F	6 hours/day, 7 days/week, 2 weeks	0, 2.4	BI CS OF	Resp			2.4	20% increase in lung weight, 53% increased retention of lavage fluid
Palmes et al. 1959									**Cobalt Hydrocarbonyl**
13	GN PIG 6-32M	3 months, 5 days/week, 6 hours/day	0, 9	CS GN HE HP LE UR	Hemato		9		5% increase in hemoglobin

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Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/m^3)	Parameters monitored	Endpoint	NOAEL (mg/m^3)	Less serious LOAEL (mg/m^3)	Serious LOAEL (mg/m^3)	Effects
Johansson et al. 1987									**Cobalt Metal**
14	RABBIT (NS) 8M	17 weeks, 5 days/week, 6 hours/day	0, 0.4, 2.0	CS HP OF OW	Resp		0.4	2	LOAEL: Moderate inflammation observed in lungs; accumulation of macrophages in lungs (not otherwise described) SLOAEL: Severe lung inflammation and accumulation of macrophages (not otherwise described); increase in weight of lower lung lobe by 25%
Johansson et al. 1991									**Cobalt Chloride**
15	RABBIT (NS) 8 M	4 months, 5 days/week, 6 hours/day	0, 0.5	BI CS GN HP	Resp	0.5			
Johansson et al. 1992									**Cobalt Chloride**
16	RABBIT (NS) 8 M	4 months, 5 days/week, 6 hours/day	0, 0.6	BI CS GN HP	Resp		0.6		Histologic alterations in pulmonary tissue; altered BAL parameters; 22% decrease in macrophages
Kerfoot 1975									**Cobalt Metal**
17	PIG 5NS	3 months, 5 days/week, 6 hour/day	0, 0.115, 0.991	GN HP CS UR	Bd wt		0.12		16% decrease in body weight
					Resp		0.12		29% decrease in specific compliance (a metric of mechanical ventilation)
					Cardio		0.12		14% increase in heartrate, 38% decrease in QRS amplitude

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Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/m^3)	Parameters monitored	Endpoint	NOAEL (mg/m^3)	Less serious LOAEL (mg/m^3)	Serious LOAEL (mg/m^3)	Effects
					Hemato	0.99			
					Hepatic	0.99			
					Renal	0.99			
					Immuno	0.99			
CHRONIC EXPOSURE									
Deng et al. 1991									**Cobalt Metal**
18	HUMAN 362	Occupational (occupational)	0, 0.0175	CS	Resp	0.02			
Kusaka et al. 1986									**Cobalt Metal**
19	HUMAN	3 years (occupational)	0, 0.85, 0.126	CS OF	Resp	0.09	0.13		2.7% decrease in FEV1% in exposed workers, suggestive of bronchial obstruction
Nemery et al. 1992									**Cobalt Metal**
20	HUMAN 212 M 41 F	Occupational (occupational)	0, 0.0053, 0.0151	CS OF UR	Resp	0.0053[b]	0.0151		Decreased FEV1(5%) and FVC (5%); increased cough (11/91), wheezing (4/91), and upper airway irritation (40/91) observed in the subjects
NTP 1998									**Cobalt Sulfate Heptahydrate**
21	RAT (Fischer-344) 50M, 50F	105 weeks, 5 days/week, 6 hours/day	0, 0.06, 0.21, 0.63	BW CS GN HP LE OW	Bd wt	0.63			
					Resp	0.06	0.21		Alveolar inflammation and lung lesions; metaplasia in the nose and epiglottis observed in 50/50 female and 49/50 male rats (not otherwise described)

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/m^3)	Parameters monitored	Endpoint	NOAEL (mg/m^3)	Less serious LOAEL (mg/m^3)	Serious LOAEL (mg/m^3)	Effects
					Cancer			0.06 F	Alveolar/bronchiolar neoplasms; hyperplasia in the adrenal medulla in 23/50 rats
								0.06 M	Alveolar/bronchiolar neoplasms
NTP 2014									**Cobalt Metal**
22	RAT (F344/N) 50M, 50F	105 weeks, 5 days/week, 6 hours + T90 (12 minutes)/day	0, 1.25, 2.5, 5	BW GN HP	Death			2.5 F	Decreased survivability compared to controls
					Bd wt	1.25 F	2.5 F	5 F	LOAEL: 11.6% less mean body weight than controls by exposure weeks 53-103 SLOAEL: 21.5% less mean body weight compared to controls by exposure weeks 53-103
						2.5 M		5 M	22.7% less mean body weight compared to controls by exposure weeks 53-103
					Resp			1.25	Significantly increased incidence of lung neoplasms and nonneoplastic lesions of lungs and nose; including hyperplasia of alveolar and bronchiole epithelium, chronic active inflammation (lung and nose), metaplasia and atrophy of olfactory epithelium
					Cardio	5			

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/m^3)	Parameters monitored	Endpoint	NOAEL (mg/m^3)	Less serious LOAEL (mg/m^3)	Serious LOAEL (mg/m^3)	Effects
					Gastro	5			
					Musc/skel	5			
					Hepatic	2.5 F	5 F		Increased incidence of basophilic focus (33/50 rats) compared to controls (16/50)
							1.25 M		Increased incidence of basophilic focus (17/50 rats) compared to controls (5/50 rats)
					Renal	5			
					Dermal	5			
					Ocular	5			
					Endocr		1.25 F	2.5 F	LOAEL: Increased incidence (27/50) of medullary hyperplasia in the adrenal gland compared to controls (12/50) SLOAEL: Increased incidence (8/50 rats) of bilateral benign pheochromocytoma compared to controls (2/50 rats)
								1.25 M	Significantly increased incidence (13/50) of bilateral benign pheochromocytoma, compared to controls (4/50)
					Immuno	2.5 F	5 F		Accumulation of macrophages around the alveolar/bronchiolar neoplasms
						5 M			
					Neuro	5			

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/m^3)	Parameters monitored	Endpoint	NOAEL (mg/m^3)	Less serious LOAEL (mg/m^3)	Serious LOAEL (mg/m^3)	Effects
					Repro	5 F			
						1.25 M		2.5 M	Increased incidence of testicular infarction (12/50 rats) compared to controls (1/50); unilateral with complete effacement of the parenchyma due to necrosis
					Cancer			1.25 F	Significantly increased incidence of mononuclear cell leukemia compared to controls (adjusted incidence rate: 62.4% in exposed, 35.7% in controls)
NTP 1998									**Cobalt Sulfate Heptahydrate**
23	MOUSE (B6C3F1) 50 M 50 F	105 weeks, 5 days/week, 6 hours/day	0, 0.06, 0.21, 0.63	BW CS GN HP LE OW	Bd wt	0.63			
					Resp		0.06		Non-neoplastic lesions on nose and larynx in 37 males and 45 females
					Hepatic	0.63			
					Endocr	0.63			
					Cancer			0.06 F	Combined alveolar/bronchiolar adenoma/carcinoma in 14/50 mice
								0.06 M	Combined alveolar/bronchiolar adenoma/carcinoma in 7/50

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/m^3)	Parameters monitored	Endpoint	NOAEL (mg/m^3)	Less serious LOAEL (mg/m^3)	Serious LOAEL (mg/m^3)	Effects
NTP 2014									**Cobalt Metal**
24	MOUSE (B6C3F1) 50M, 50F	105 weeks, 5 days/week, 6 hours + T90 (12 minutes)/day	0, 1.25, 2.5, 5		Death			2.5 M	Significantly less survival probability compared to controls
					Bd wt	2.5 F		5 F	25% less mean bodyweight than controls by weeks 53-103
							5 M		11.3% less mean body weight than controls by weeks 53-103
					Resp			1.25	Increased incidence of lung neoplasms and neoplastic lesions in the lung, nose, larynx and trachea, compared to controls. This includes hyperplasia and cytoplasmic vacuolization of alveolar/bronchiolar epithelium, alveolar/bronchiolar carcinoma, atrophy and hyperplasia of olfactory epithelium, and turbinate atrophy
					Cardio	5			
					Gastro	5			
					Musc/skel	5			
					Hepatic	5			
					Renal	5			
					Dermal	5			
					Ocular	5			

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/m^3)	Parameters monitored	Endpoint	NOAEL (mg/m^3)	Less serious LOAEL (mg/m^3)	Serious LOAEL (mg/m^3)	Effects
					Endocr	5			
					Immuno	2.5	5		Accumulations of small histiocyte/macrophage infiltrates aggregated adjacent to alveolar/bronchiolar neoplasms
					Neuro	5			
					Repro	5 F			
						2.5 M		5 M	Increased incidence (21/50 mice) of minimal to mild germinal epithelium degradation compared to controls (9/50 mice)
					Cancer			1.25	Increased rate of alveolar/bronchiolar carcinoma, multiple in exposed mice compared to controls (Adjusted rated in exposed: 79.4% in males, 53.8% in females; adjusted rates in controls: 22.8% in males, 11.3% in females)
Wehner et al. 1977									**Cobalt Oxide**
25	HAMSTER (ENG:ELA) 51M	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 observed (not otherwise described)
					Other noncancer	7.9			

Table 2-1. Levels of Significant Exposure to Cobalt – Inhalation

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/m^3)	Parameters monitored	Endpoint	NOAEL (mg/m^3)	Less serious LOAEL (mg/m^3)	Serious LOAEL (mg/m^3)	Effects

[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 a chronic inhalation minimal risk level (MRL) of 0.0001 mg/m^3; concentration adjusted for intermittent exposure and divided by an uncertainty factor of 10 (for human variability).

Studies listed in the table could potentially have examined more than one endpoint.

BC = serum (blood) chemistry; BI = biochemical changes; BW or bd wt = body weight; Cardio = cardiovascular; CS = clinical signs; Endocr = endocrine; F = female(s); FVC = Forced vital capacity; Gastro = gastrointestinal; GN = gross necropsy; HE = hematological; Hemato = hematological; HP = histopathology; Immuno = immunological; IX = immune function; LE = lethality; LOAEL = lowest-observed-adverse-effect level; M = male(s); Musc/skel = Musculo/skeletal; Neuro = neurological; NOAEL = no-observed-effect-level; NS = not specified; NX = neurological function; OF = organ function; OW = organ weight; Resp = respiratory; T3 = Triiodothyronine; TSH = Thyroid-stimulating hormone; UR = urinalysis; polycythemia = author reported term associated with increased hemoglobin or erythrocyte count; RX = reproductive function; UR = urinalysis; (W) = water; WI = water intake

Figure 2-2. Levels of Significant Exposure to Cobalt–Inhalation
Acute (≤14 days)

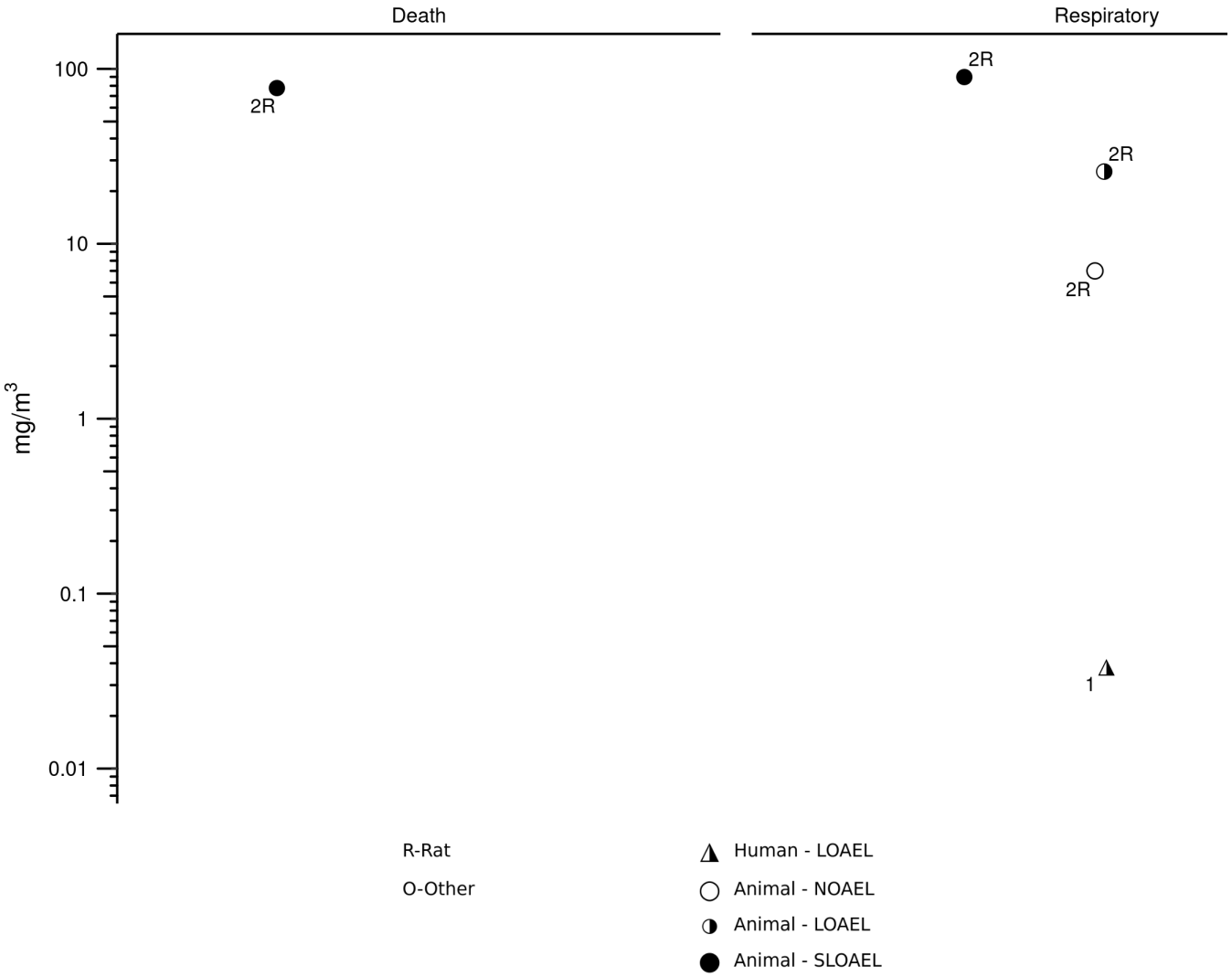


Figure 2-2. Levels of Significant Exposure to Cobalt–Inhalation
Intermediate (15-364 days)

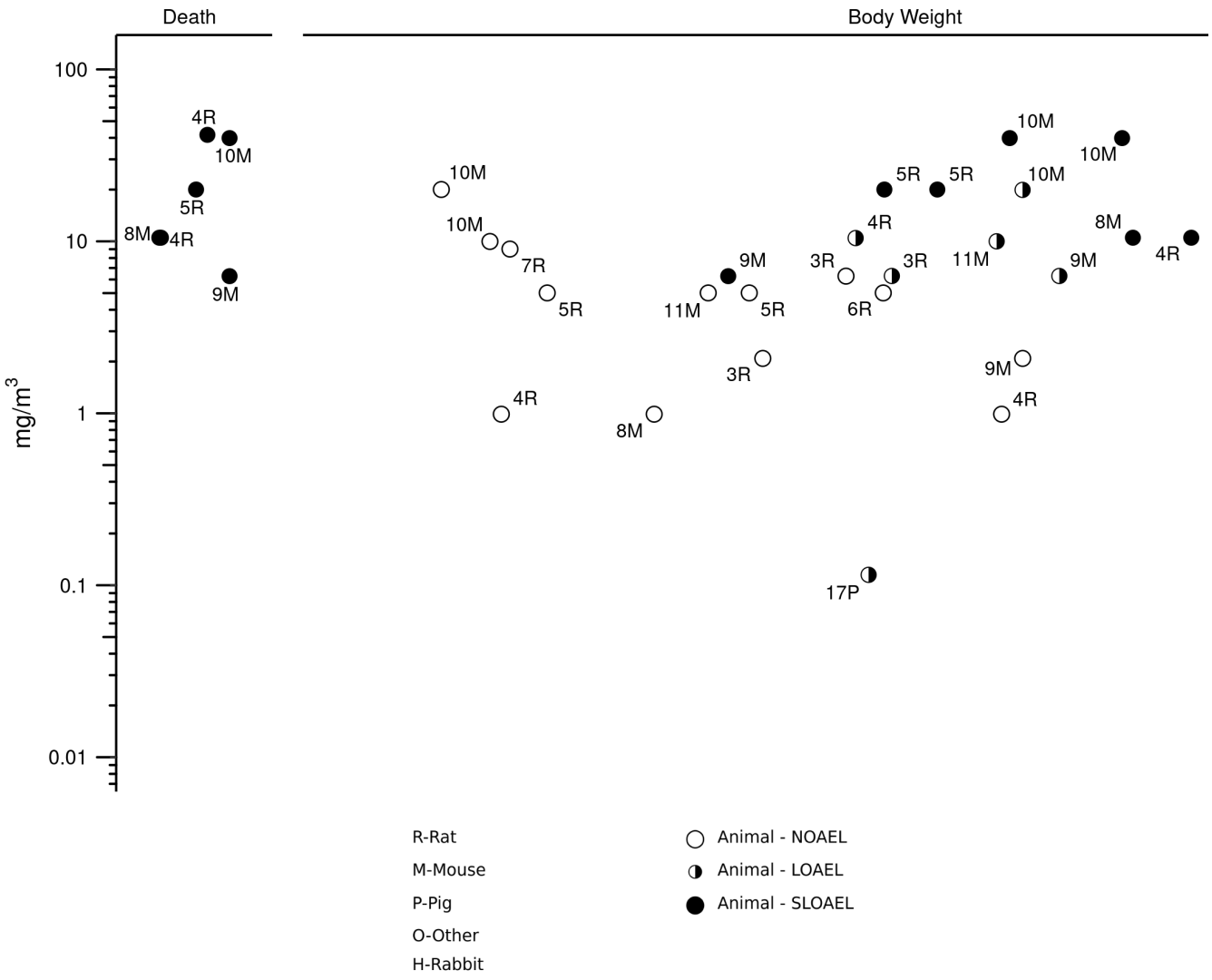


Figure 2-2. Levels of Significant Exposure to Cobalt–Inhalation
Intermediate (15-364 days)

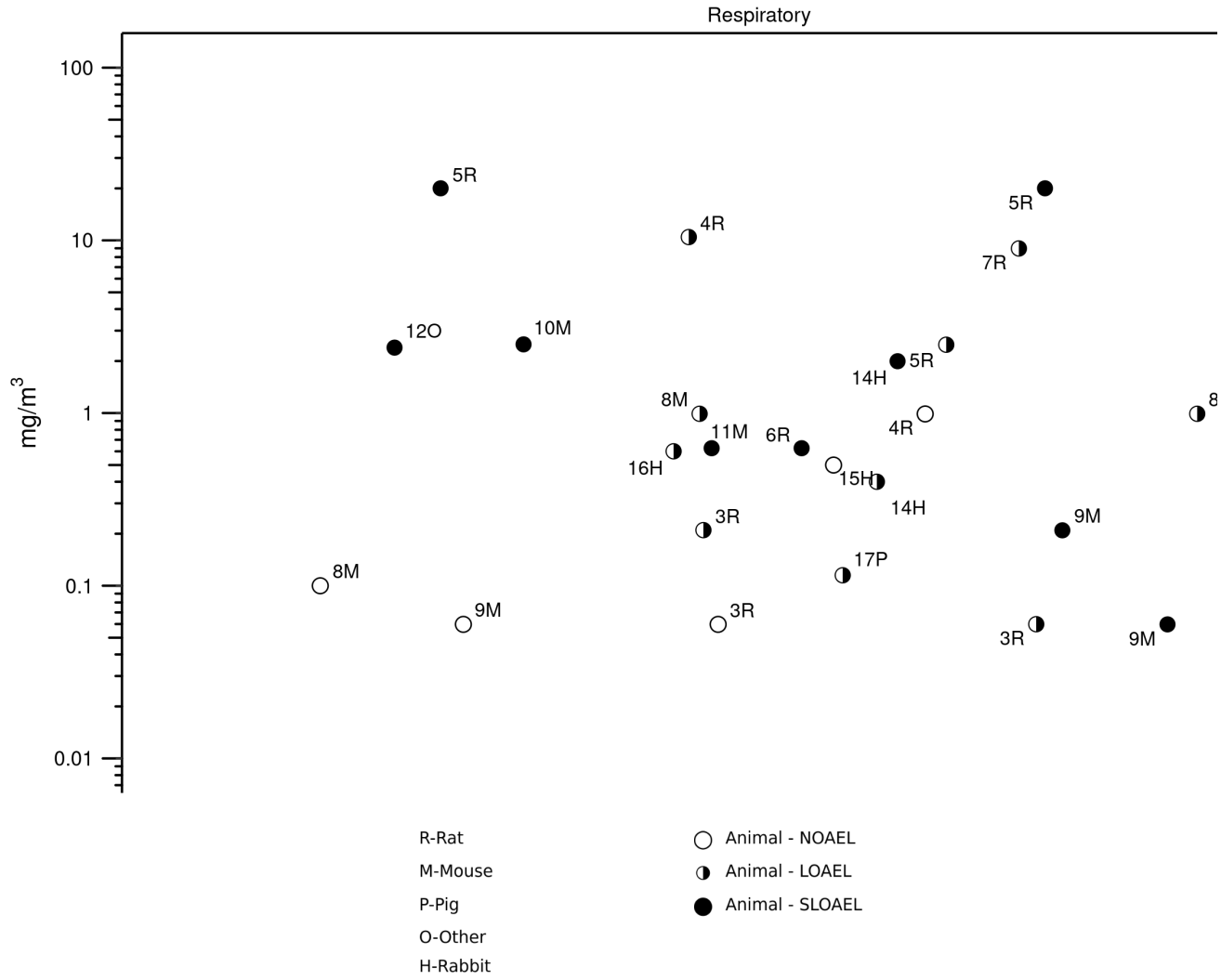


Figure 2-2. Levels of Significant Exposure to Cobalt–Inhalation
Intermediate (15-364 days)

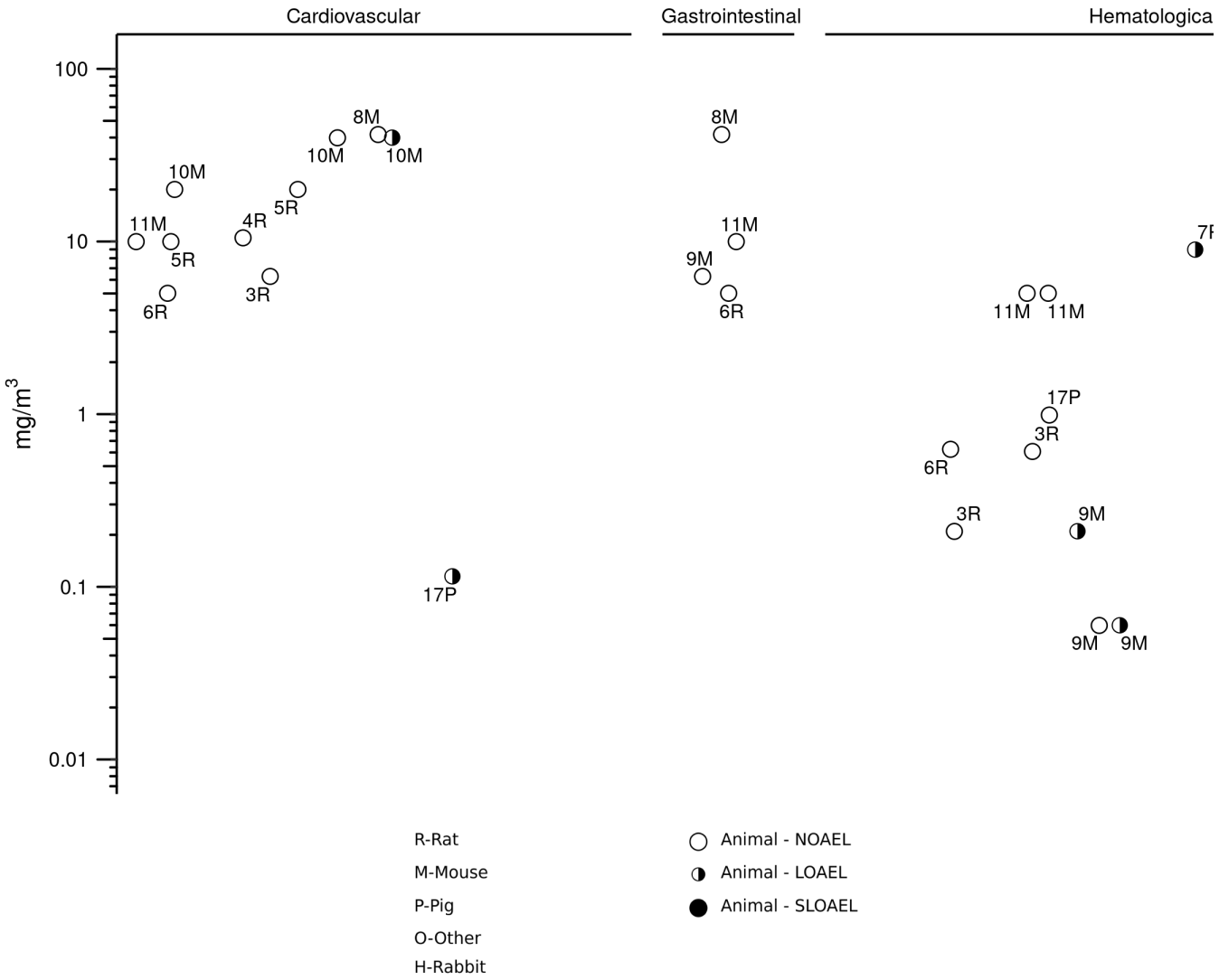


Figure 2-2. Levels of Significant Exposure to Cobalt–Inhalation
Intermediate (15-364 days)

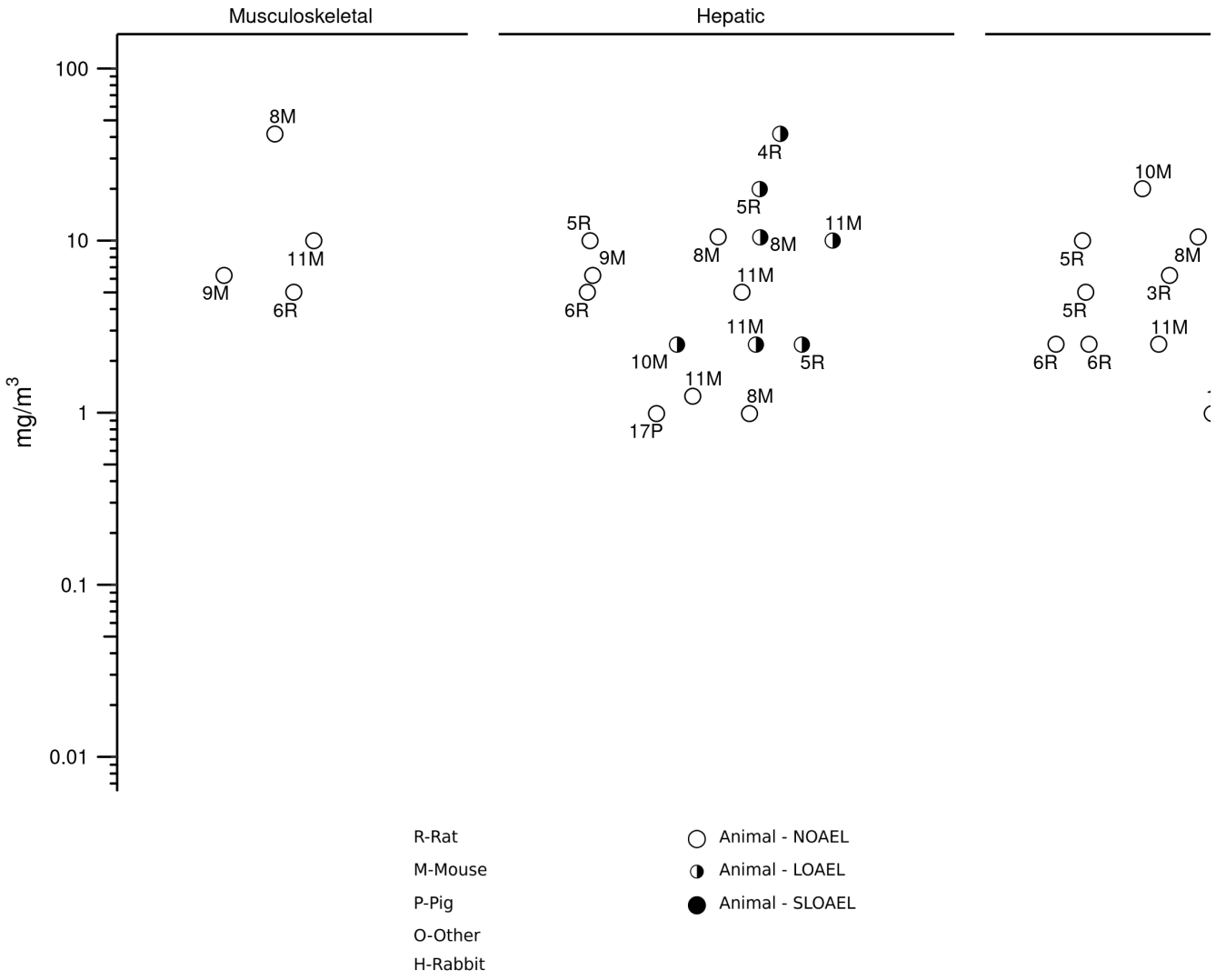


Figure 2-2. Levels of Significant Exposure to Cobalt–Inhalation
Intermediate (15-364 days)

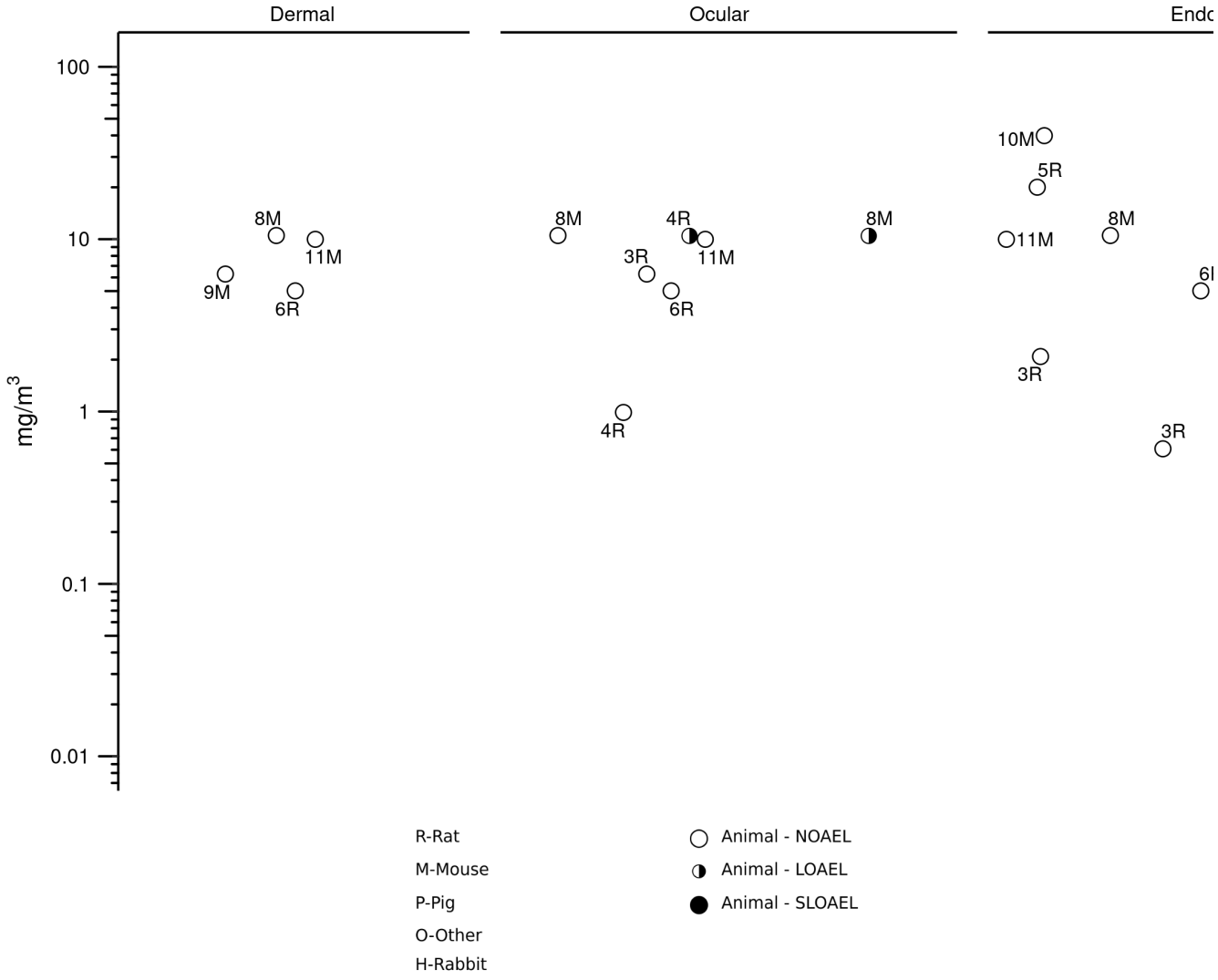


Figure 2-2. Levels of Significant Exposure to Cobalt–Inhalation
Intermediate (15-364 days)

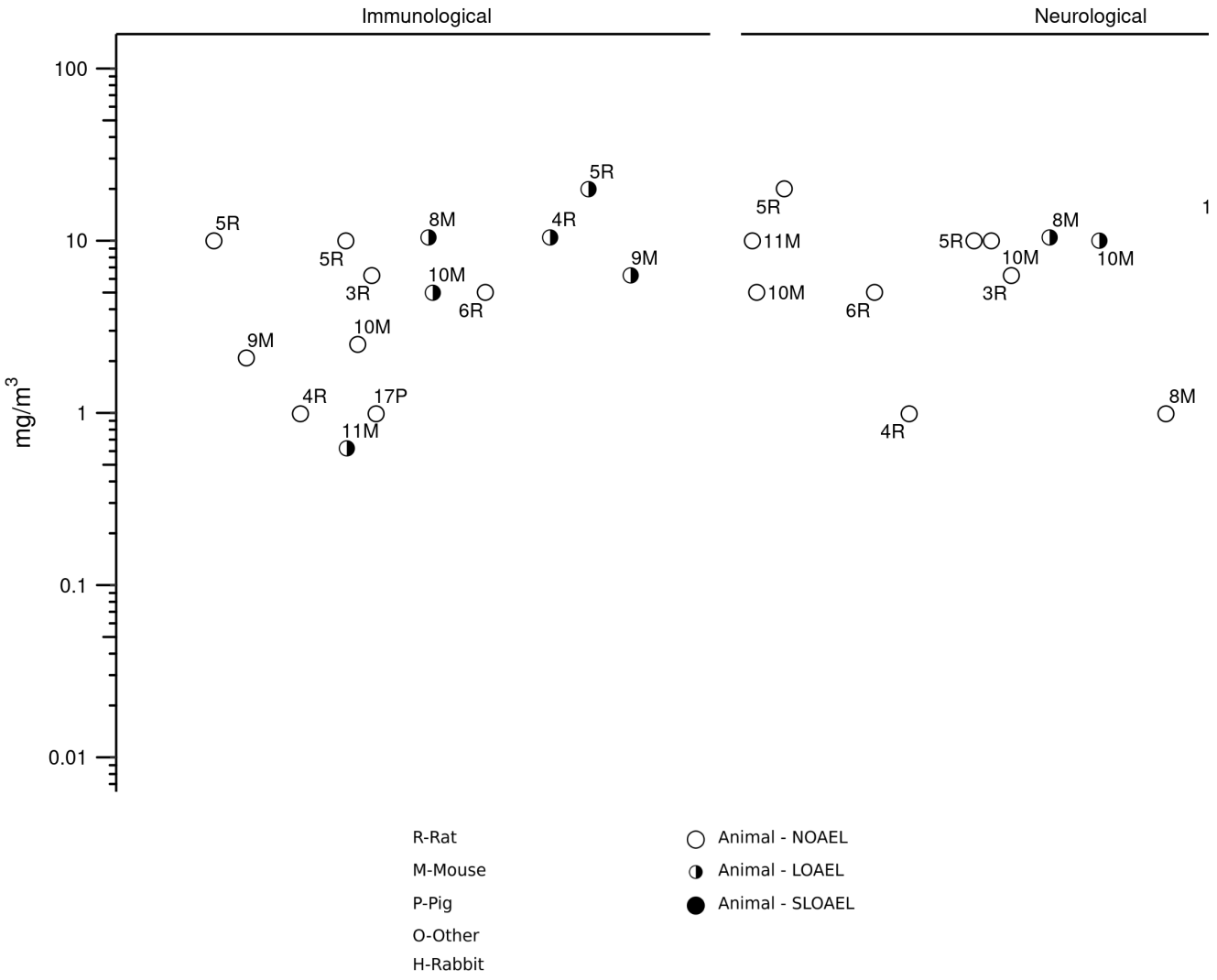


Figure 2-2. Levels of Significant Exposure to Cobalt–Inhalation
Intermediate (15-364 days)

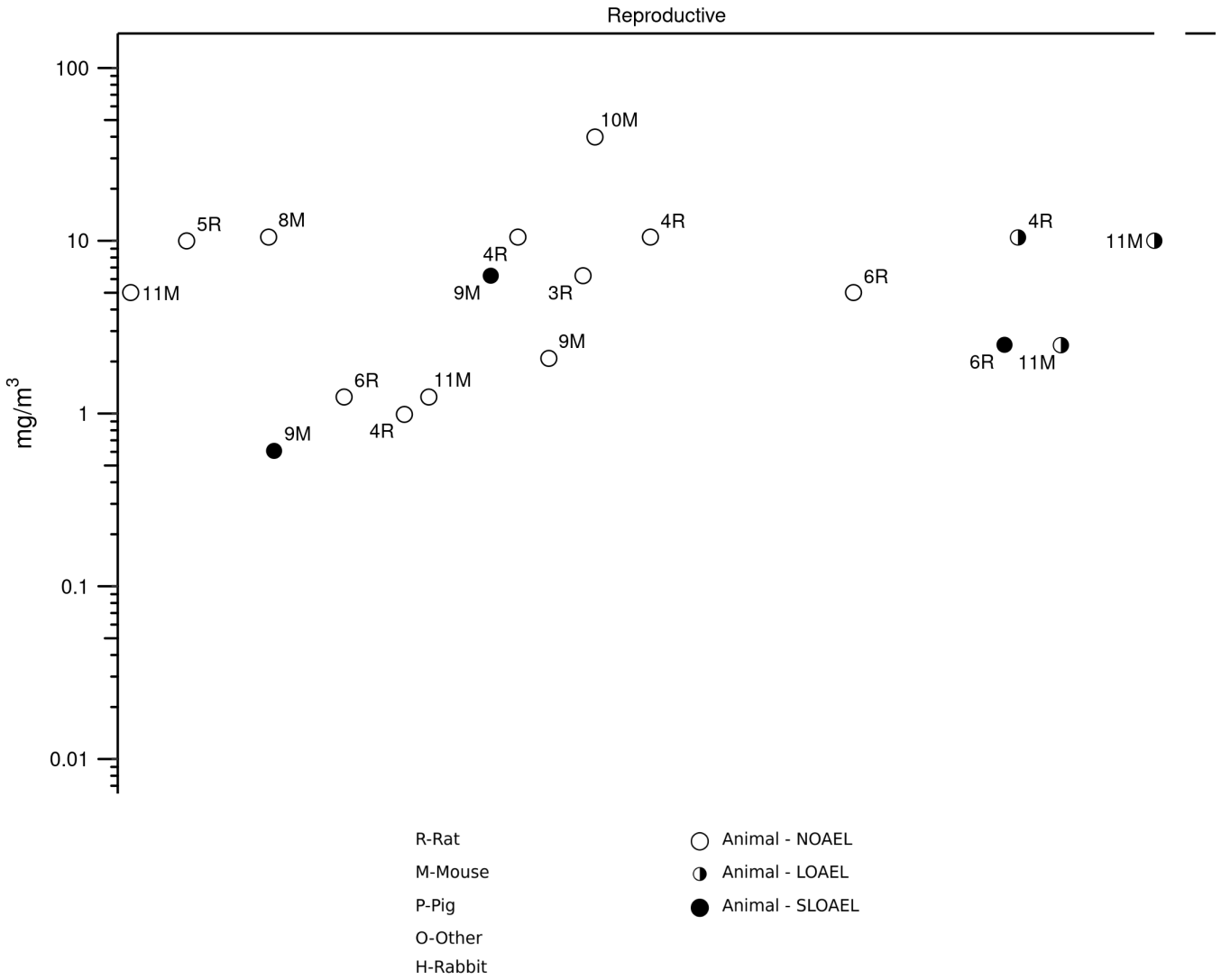


Figure 2-2. Levels of Significant Exposure to Cobalt–Inhalation
Chronic (≥365 days)

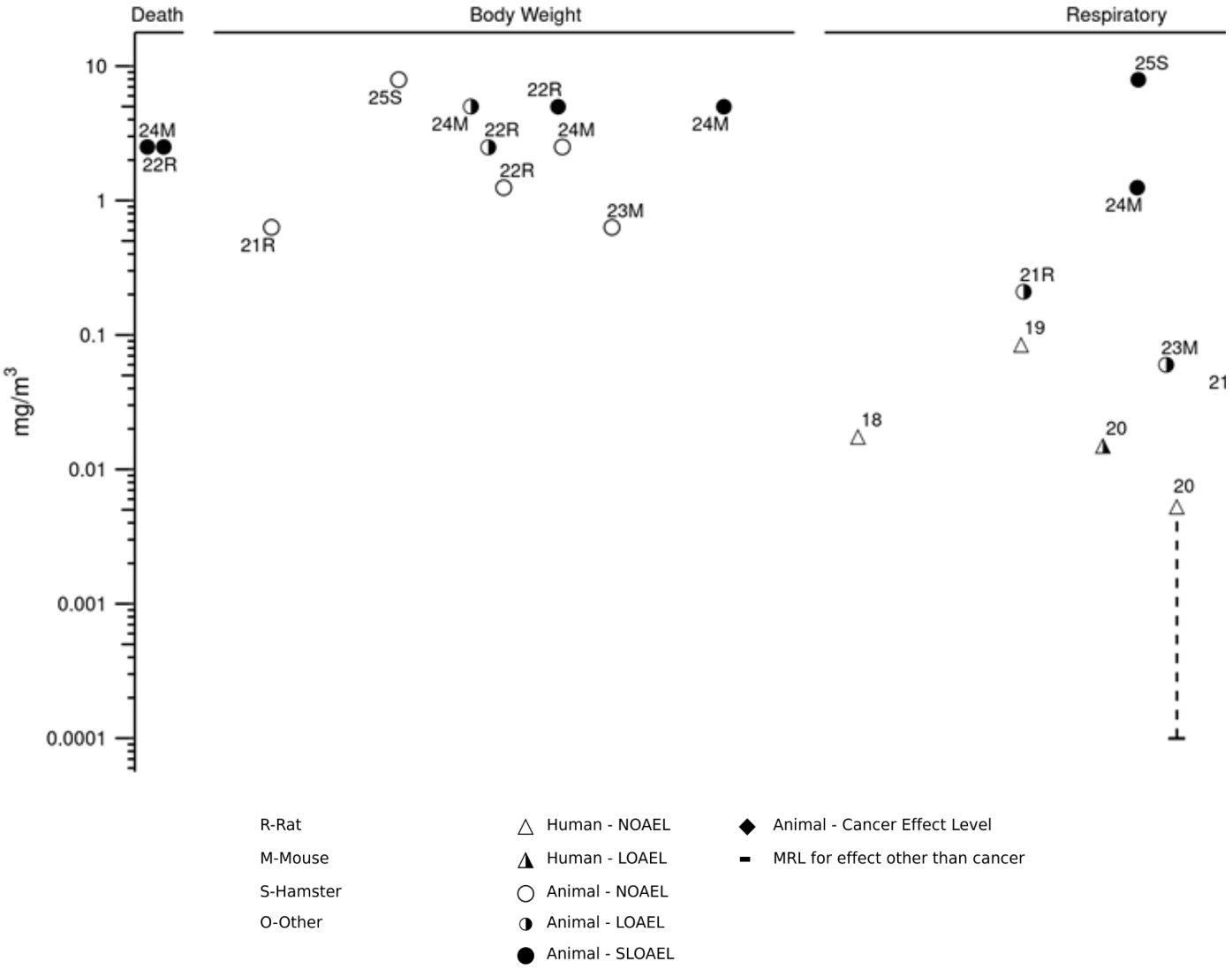


Figure 2-2. Levels of Significant Exposure to Cobalt–Inhalation
Chronic (≥365 days)

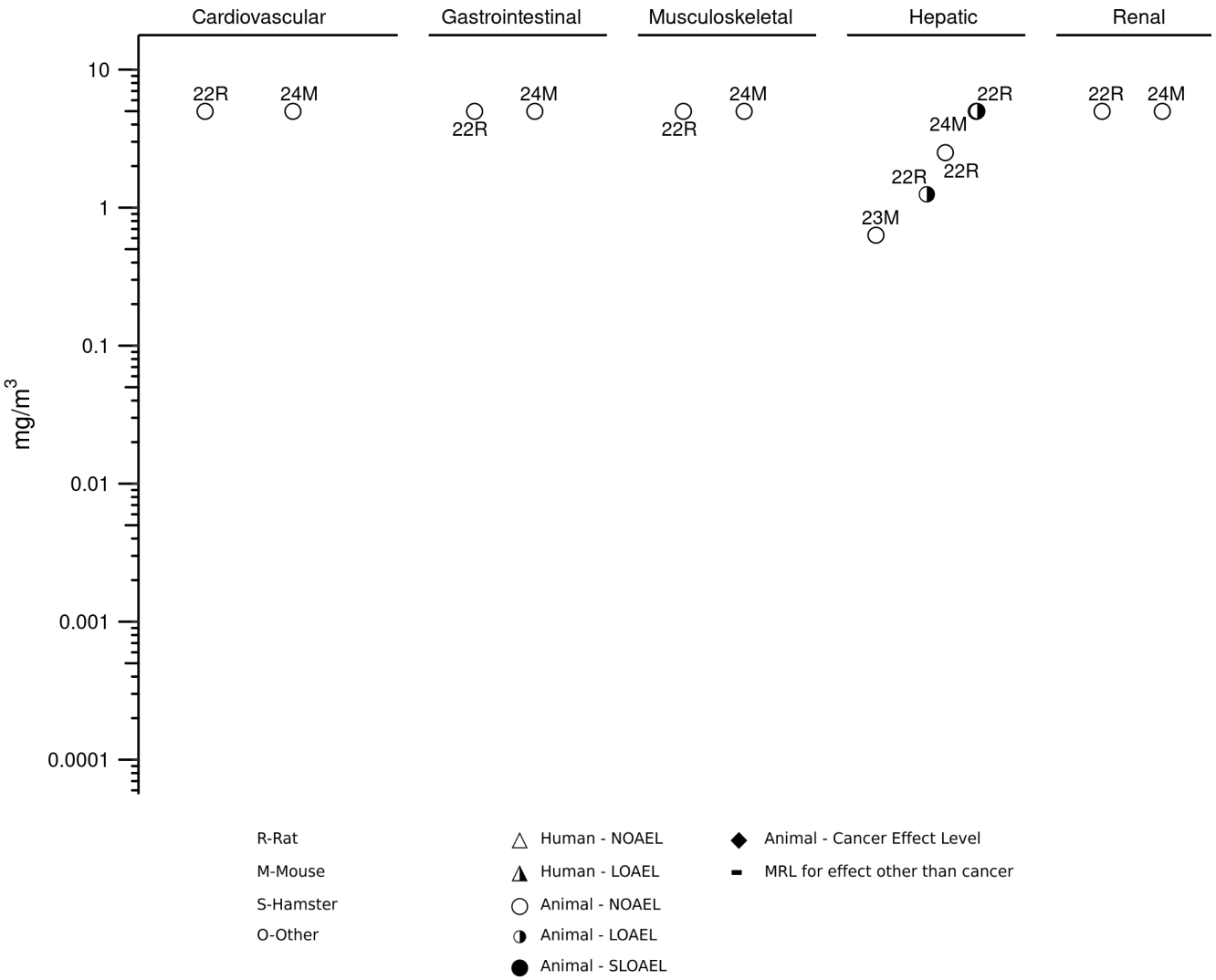


Figure 2-2. Levels of Significant Exposure to Cobalt–Inhalation
Chronic (≥365 days)

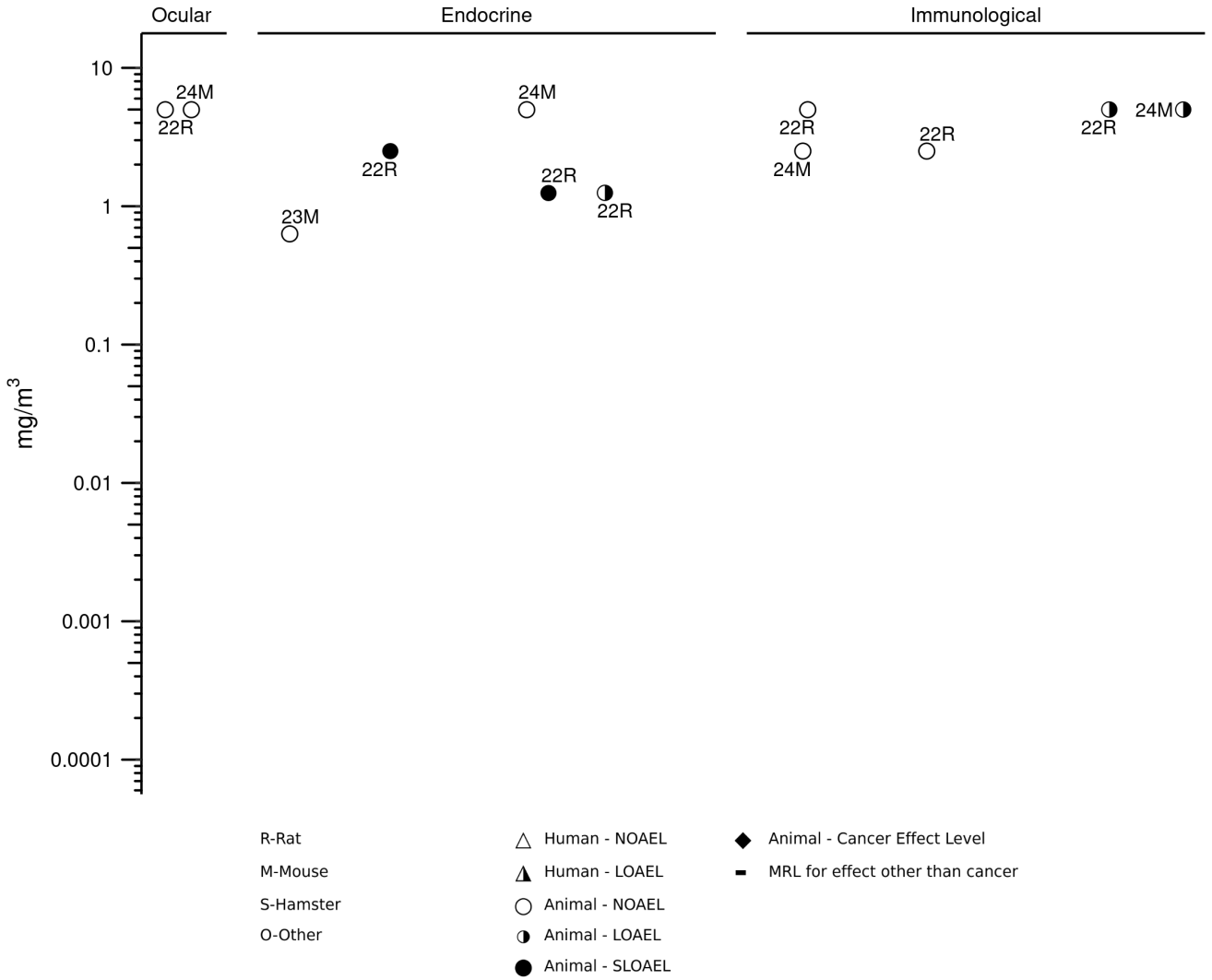


Figure 2-2. Levels of Significant Exposure to Cobalt–Inhalation
Chronic (≥365 days)

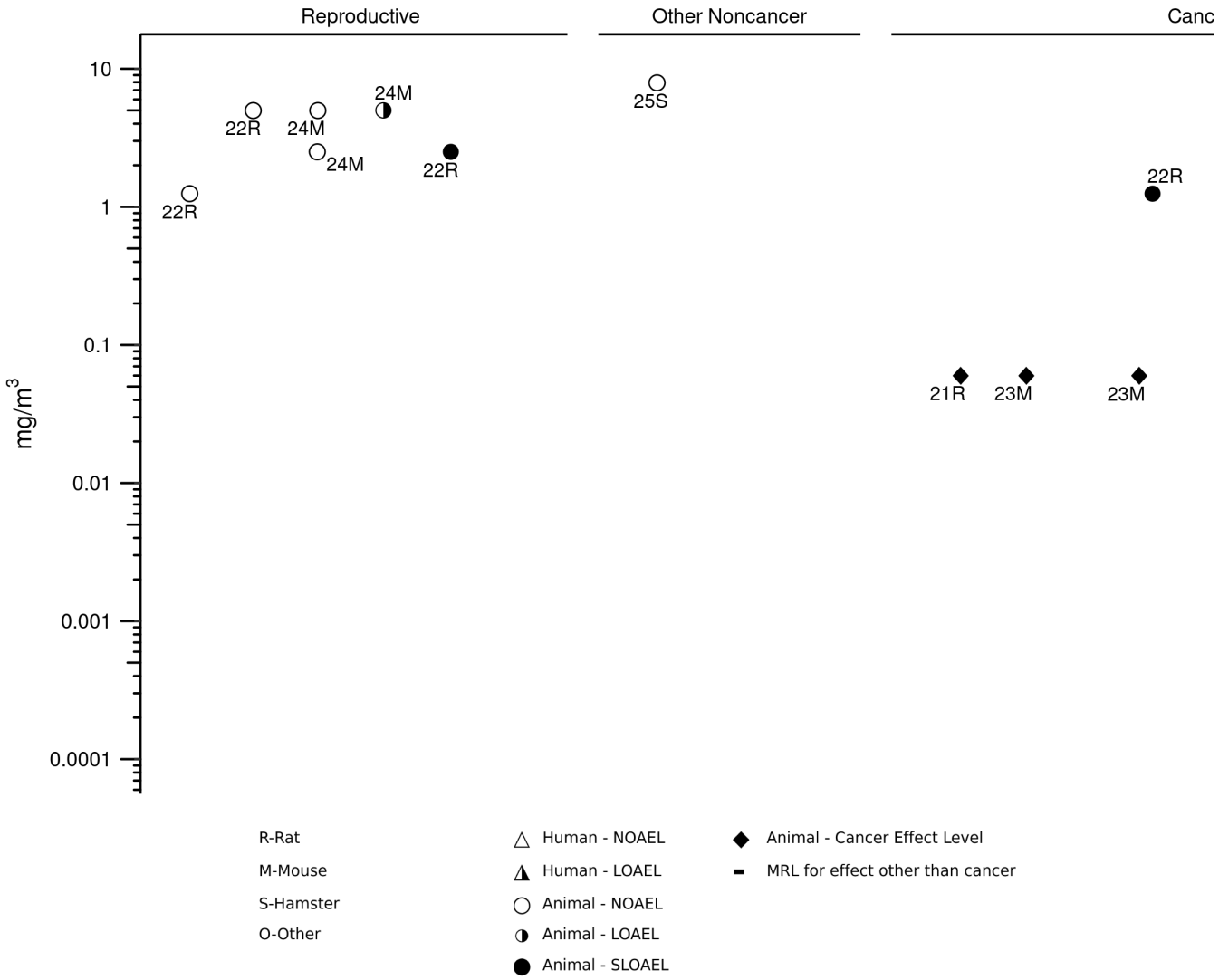


Table 2-2. Levels of Significant Exposure to Cobalt – Oral

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/kg/day)	Parameters monitored	Endpoint	NOAEL (mg/kg/day)	Less serious LOAEL (mg/kg/day)	Serious LOAEL (mg/kg/day)	Effects
ACUTE EXPOSURE									
Davis and Fields 1958									**Cobalt Chloride**
1	HUMAN 3M	6- 14 days, Daily (C)	0, 1	BC CS HE	Hemato		1[b]		Polycythemia, 8.7% increase in erythrocyte numbers
Paley et al. 1958									**Cobalt Chloride**
2	HUMAN 3M	10-14 days, Daily (C)	0, 0.54	BC CS OF	Gastro		0.54		Mild gastric distress
					Endocr		0.54		Decreased iodine uptake by thyroid in euthyroid subjects; ranged from 3.1/5.2=40% to 2.2/5.2=58% at 15 min in 3/3 subjects, and by 35/57=39% at 24 hr in 1/3 subjects
Ajibade et al. 2017									**Cobalt Chloride**
3	RAT (Wistar) 6M	2 weeks, Daily, 7days/week (GW)	0, 33.7	BI OF	Cardio		33.7		Cellular infiltration and cardiac cell swelling observed in the heart along with 67% increase in the NF-kB
					Renal		33.7		Histopathological study showed increased inflammation; 300% increase in NF-kB in the kidneys

Table 2-2. Levels of Significant Exposure to Cobalt – Oral

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/kg/day)	Parameters monitored	Endpoint	NOAEL (mg/kg/day)	Less serious LOAEL (mg/kg/day)	Serious LOAEL (mg/kg/day)	Effects
Akinrinde et al 2016b									**Cobalt(II) Chloride Hexahydrate**
4	RAT (Wistar) 7M	1 week, Daily, 7 days/week (W)	0, 19	BI HP OF	Cardio		19		Inflammation of the myocardium and areas of myocardial infarction; decreases of systolic blood pressure by 17%, diastolic blood pressure by 24%, and mean arterial pressure by 21%
					Renal		19		Inflammation in the peri- tubular and peri-vascular areas of kidney along with focal tubular necrosis
Akinrinde et al. 2016a									**Cobalt(II) Chloride Hexahydrate**
5	RAT (Wistar) 8M	2 weeks, Daily, 7days/week (W)	0, 18.4	BI GN HP OF	Cardio		18.38		Hemorrhagic lesions with congestion in the blood vessels along with inflammation in the myocardial cells; 12% decrease in systolic blood pressure and 150% increase in LDH compared to controls
					Renal		18.38		Loss of normal morphology, increased inflammation and vascular congestion in kidneys; increase in urea and creatinine by 33% and 19%
Akinrinde et al. 2016c									**Cobalt Chloride**
6	RAT (Wistar) 7M	7 days, Daily (W)	0, 19	BI HP IX OF	Gastro		19		Histopathology showed significant intestinal injury with depletion of absorptive epithelial cells; decrease in relative small intestine weight by 16%; decrease in GPx by 17%

Table 2-2. Levels of Significant Exposure to Cobalt – Oral

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/kg/day)	Parameters monitored	Endpoint	NOAEL (mg/kg/day)	Less serious LOAEL (mg/kg/day)	Serious LOAEL (mg/kg/day)	Effects
					Hepatic		19		Histology showed necrosis in the liver along with cytotoxicity in hepatocytes and other abnormal morphology; decrease in relative liver weight by 14%
					Immuno		19		Increase in TNF α by 60% and decrease in IL1β by 25%
Akinrinde et al. 2019									**Cobalt(II) Chloride Hexahydrate**
7	RAT (Wistar) 12M	1 week, Daily, 7days/week (GW)	0, 67.5	BC BI CS NX	Immuno			67.5	300% increase in IL-1β and 100% increase in TNF α
					Neuro			67.5	Battery of neurobehavioral tests showed poor performance in exposed rats and a 60% increase in AChE activity compared to controls
Awoyemi et al. 2017									**Cobalt(II) Chloride Hexahydrate**
8	RAT (Albino) 10M	1 week, Daily, 7 days/week, (W)	0, 6, 11, 22	BI BW CS HE OF	Hemato	6	11		~400% increase in the frequency of micronucleated polychromatic erythrocytes
					Hepatic		6	11	LOAEL: Alteration in liver enzyme levels (16% increase of ALT) SLOAEL: Hepatocytes with focal areas of moderate congestion of vessels, mild infiltration by inflammatory cells, focal area of necrosis and congestion of vessels

Table 2-2. Levels of Significant Exposure to Cobalt – Oral

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/kg/day)	Parameters monitored	Endpoint	NOAEL (mg/kg/day)	Less serious LOAEL (mg/kg/day)	Serious LOAEL (mg/kg/day)	Effects
Domingo and Llobet 1984									**Cobalt Chloride**
9	RAT (Sprague-Dawley) 20M	Once (GW)	0, 161	BC CS HE LE OF	Death			161	5/20 died
					Hemato		161		8% increase in hematocrit levels
					Hepatic	161			
					Renal			161	68% increase in urea and 57% decrease in uric acid
Domingo et al. 1985									**Cobalt(II) Chloride Hexahydrate**
10	RAT (Sprague-Dawley) 20M	Once (G)	0, 31, 67	LE	Death			37	10/20 died
Richardson et al. 2018									**Cobalt Chloride**
11	RAT (Sprague-Dawley) 5NS	5 days, Daily (W)	0, 12, 21, 37	OF	Gastro	21	37		Changes in gut microbiota composition (not otherwise described)
Shrivastava et al. 2009									**Cobalt(II) Chloride Hexahydrate**
12	RAT (Sprague-Dawley) 8M	1 week, Daily (G)	0, 12.5	BC BI BW HE HP OP OW	Bd wt	12.5			
					Resp	12.5			
					Cardio		12.5		

Table 2-2. Levels of Significant Exposure to Cobalt – Oral

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/kg/day)	Parameters monitored	Endpoint	NOAEL (mg/kg/day)	Less serious LOAEL (mg/kg/day)	Serious LOAEL (mg/kg/day)	Effects
					Hemato		12.5		60%, 10%, and 8% increase in RBC, hematocrit, and hemoglobin, respectively
					Hepatic	12.5			
					Renal	12.5			
					Immuno	12.5			
					Neuro	12.5			
Singh and Junnarkar 1991									**Cobalt Chloride**
13	RAT (Wistar) 5M, 5F	Once (GW)	0, 4.24	CS NX	Neuro		4.24		CNS depressant indicated by mild hypothermic effect and increased sleeping time by 31%
Singh and Junnarkar 1991									**Cobalt Sulfate**
14	RAT (Wistar) 5M, 5F	Once (GW)	0, 19.4	CS NX	Neuro		19.4		CNS depressant indicated by mild hypothermic effect and increased sleeping time by 19%
Wellman et al. 1984									**Cobalt Chloride**
15	RAT (Long-Evans) 7M	3 days, Daily (F)	0, 9, 45, 90	BW CS FI NX WI	Bd wt	90			
					Neuro	9	45		Taste aversion demonstrated by reduced saccharin consumption
					Other noncancer	9	45		~20% decrease in food consumption

Table 2-2. Levels of Significant Exposure to Cobalt – Oral

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/kg/day)	Parameters monitored	Endpoint	NOAEL (mg/kg/day)	Less serious LOAEL (mg/kg/day)	Serious LOAEL (mg/kg/day)	Effects
Bryan and Bright, 1973									**Cobalt(II) Chloride Hexahydrate**
16	MOUSE (Swiss-Webster) 3M	2 days, Once/day (W)	0, 763	BC BW HE	Hemato		763		Unspecified alterations in electrophoretic profile of serum proteins
Elbetieha et al. 2008									**Cobalt(II) Chloride Hexahydrate**
17	MOUSE (Swiss) 10M	12 days, Daily (W)	0, 6.4, 11.6, 23	BW CS HP OW RX WI	Death			11.6	1/10 mice died during the 10th week of exposure
					Bd wt		23		Significant 7.1% decrease in body weight gain compared to controls
					Repro			6.4	Significant 16.8% increase of relative preputial gland weight, 13.3% decrease in sperm count, and decreased male fertility compared to controls
Hassan et al. 2006									**Cobalt Chloride**
18	MOUSE (NS) 3M	5 days, Once/day (W)	0, 7, 14, 28	RX	Repro			7	126% increase in abnormal sperm
Seidenberg 1986									**Cobalt Chloride**
19	MOUSE 28F	5 days, Daily (Gd 8-12) (GW)	0, 81	CS FX MX DX	Bd wt			81	32% decrease in maternal weight gain
					Develop	81			

Table 2-2. Levels of Significant Exposure to Cobalt – Oral

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/kg/day)	Parameters monitored	Endpoint	NOAEL (mg/kg/day)	Less serious LOAEL (mg/kg/day)	Serious LOAEL (mg/kg/day)	Effects
Singh and Junnarkar 1991									**Cobalt Sulfate**
20	MOUSE (Swiss-Webster) 5M	Once (GW)	0, 12.3	CS NX	Neuro		12.3		CNS depression observed in mice (not otherwise described)
Singh and Junnarkar 1991									**Cobalt Chloride**
21	MOUSE (Swiss-Webster) 5M	Once (GW)	0, 8.9	CS NX	Neuro		8.9		CNS depression observed in mice (not otherwise described)
INTERMEDIATE EXPOSURE									
Davis and Fields 1958									**Cobalt Chloride**
22	HUMAN 2M	15-22 days, Daily (C)	0, 0.8, 1	BC CS HE	Hemato	0.8	1[c]		Polycythemia, 9.7% increase in erythrocyte numbers
Duckham and Lee 1976									**Cobalt Chloride**
23	HUMAN 6M 6F	12 weeks, 7days/week Twice/day (C)	0, 0.18	BC CS OF	Gastro		0.18 F		Nausea and constipation
					Hemato		0.18 F		Increased hemoglobin in anephric, hemoglobin deficient patients by 26-70%

Table 2-2. Levels of Significant Exposure to Cobalt – Oral

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/kg/day)	Parameters monitored	Endpoint	NOAEL (mg/kg/day)	Less serious LOAEL (mg/kg/day)	Serious LOAEL (mg/kg/day)	Effects
Holly 1955									**Cobalt Chloride**
24	HUMAN 20F	13 weeks, 7 days/week, Once/day (C)	0, 0.5-0.6	BC BI DX HE OF UR	Gastro		0.5		Gastric intolerance
					Hemato	0.6			
					Hepatic	0.6			
					Dermal		0.5		Skin rash in 1/20 disappeared when Co exposure was discontinued
					Endocr	0.5			
					Develop	0.5			
Paley et al. 1958									**Cobalt Chloride**
25	HUMAN 2F	21-25 days, Daily (C)	0, 0.54	CS OF	Gastro		0.54		Gastric distress
					Endocr		0.54		Decreases in Iodine uptake in hyperthyroid subjects of 24/28=14% and 2.3/12.5=92% at 15 min in 2/2 subjects
Taylor et al. 1977									**Cobalt Chloride**
26	HUMAN 8 NS	12-32 weeks, 7days/week (C)	0, 0.16, 0.32	BI CS HE	Hemato		0.16		Unspecified increase in hemoglobin

Table 2-2. Levels of Significant Exposure to Cobalt – Oral

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/kg/day)	Parameters monitored	Endpoint	NOAEL (mg/kg/day)	Less serious LOAEL (mg/kg/day)	Serious LOAEL (mg/kg/day)	Effects
Abdel-Rehman et al. 2019									**Cobalt Chloride**
27	RAT (Wistar) 10M	60 days, Daily (W)	0, 27		Death			27	4/10 rats died
					Neuro			27	Decrease in neurotransmitters levels- 23% decrease in serotonin, 26% decrease in norepinephrine, 48% decrease in dopamine, and 39% decrease in GABA; increases in encephalopathy were observed in cerebral cortex (not otherwise described); upregulation of microglial CD68 and neural caspase-3 in the brain (not otherwise described)
Bourg et al. 1985									**Cobalt Chloride**
28	RAT (Sprague-Dawley) 8M	57 days, Daily (W)	0, 20		Neuro		20		Increased latency during passive-avoidance retention testing by 342%; 972% increase in accumulation in the brain
Chetty et al. 1979									**Cobalt Chloride**
29	RAT (Sprague-Dawley) 8-12M	4 weeks, 7days/week, Daily (F)	0, 0.379, 1.9, 3.79, 7.59, 11.4	BC BW HE OF OW	Bd wt			0.38	45% reduction in body weight gain
					Cardio		11.4		
					Hemato	7.59	11.4		20% decrease in hemoglobin
					Hepatic	11.4			

Table 2-2. Levels of Significant Exposure to Cobalt – Oral

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/kg/day)	Parameters monitored	Endpoint	NOAEL (mg/kg/day)	Less serious LOAEL (mg/kg/day)	Serious LOAEL (mg/kg/day)	Effects
					Neuro		3.79		13% decrease in Na+-K+ ATPase activity
Clyne et al. 1988									**Cobalt Sulfate**
30	RAT (Sprague-Dawley) 5M	8 weeks, 7 days/week, Daily (F)	0, 4.2	BC BW GN HE OF WI	Bd wt			4.2	33% decrease in body weight gain
Corrier et al. 1985									**Cobalt Chloride**
31	RAT (Sprague-Dawley) 3M	14 weeks, 7 days/week, Once/day (F)	0, 20	BC HP OF RX	Hemato			20	41% increase in RBCs and 28% increase in hemoglobin
					Repro			20	Pronounced histologic alteration of seminiferous tubules (27%- 90%); sperm reserve dropped by 57%
Danzeisen et al. 2020									**Cobalt Tetraoxide**
32	Rat (Sprague-Dawley) 40M, 40F	90 days, once daily (G)	0, 0.74, 2.48, 7.44	BW FI HE, HP NX OW RX UR WI	Bd wt	220	744		Slight reduction in body weight gain (not otherwise reported)
					Hemato	73.4 F	220 F		Females showed a 5.9% increase in hemoglobin
						73.4 M	220 M		Males showed a 9.5% increase in hemoglobin, a 9.6% increase in red blood cells, and a 9.2% increase in hematocrit

Table 2-2. Levels of Significant Exposure to Cobalt – Oral

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/kg/day)	Parameters monitored	Endpoint	NOAEL (mg/kg/day)	Less serious LOAEL (mg/kg/day)	Serious LOAEL (mg/kg/day)	Effects
Danzeisen et al. 2020									**Cobalt(II) Chloride Hexahydrate**
33	Rat (Sprague-Dawley) 40M, 40F	90 days, once daily (G)	0, 0.74, 2.48, 7.44	BW FI HE HP NX, OW RX UR WI	Bd wt	2.48	7.44		The body weight at the end of the study at 90 days was reduced by 11% (males) and 9% (females), respectively
					Hemato	0.74			
							7.44 F		Females showed a 13.4% increase in hemoglobin, a 9.8% increase in red blood cells, and a 12% increase in hematocrit
							2.48 M		Males showed a 10.7% increase in hemoglobin, a 9.2% increase in red blood cells, and a 10.3% increase in hematocrit
Domingo et al. 1984									**Cobalt(II) Chloride Hexahydrate**
34	RAT (Sprague-Dawley) 20M	13 weeks, 7 days/week, Daily (W)	0, 30.2	BC BI CS FI GN HE OF OW UR WI	Bd wt	30.2			
					Resp		30.2		33% increase in relative lung weight
					Cardio		30.2		9.4% increase in relative heart weight
					Gastro	30.2			
					Hemato		30.2		29% increase in hematocrit, 31% increase in hemoglobin
					Musc/skel	30.2			
					Hepatic		30.2		30% decrease in liver enzyme (GPT)

Table 2-2. Levels of Significant Exposure to Cobalt – Oral

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/kg/day)	Parameters monitored	Endpoint	NOAEL (mg/kg/day)	Less serious LOAEL (mg/kg/day)	Serious LOAEL (mg/kg/day)	Effects
					Renal		30.2		35% decrease in urine volume
					Immuno			30.2	43% increase in relative spleen weight
					Repro		30.2		26% decrease in relative testicular weight
					Other noncancer		30.2		Significant 13% decrease in water consumption
Garoui et al. 2011									**Cobalt Chloride**
35	RAT (Wistar) 6F	4 weeks; Daily from day 14 of pregnancy to day 14 post-delivery (W)	0, 21	BI BW DX FI LE OF OW RX UR WI	Bd wt	21			
					Hepatic			21	Liver weight decreased in pups by 10%; increase in hepatic enzymes ALT and AST by 44% and 27%, respectively; oxidative damage observed in the liver: 31% increase in MDA; decrease in SOD, CAT, GPx, and GSH by 30%, 23%, 31%, and 20%, respectively
					Other noncancer			21	Significant reduction in water (32%) and food intake (29%) were observed

Table 2-2. Levels of Significant Exposure to Cobalt – Oral

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/kg/day)	Parameters monitored	Endpoint	NOAEL (mg/kg/day)	Less serious LOAEL (mg/kg/day)	Serious LOAEL (mg/kg/day)	Effects
Garoui et al. 2011									**Cobalt Chloride**
36	RAT (Wistar) 4M, 4F	4 weeks; Exposed through maternal dosing- in utero and lactation daily from day 14 of pregnancy to day 14 post-delivery (W)	0, 21	BC BI BW DX FI GN OF OW UR WI	Bd wt			21	40% decrease in body weight
					Hepatic			21	Increase in hepatic enzymes ALT and AST by 133% and 75%, respectively; hepatic injury was observed with the presence of vascular congestion and infiltration of mononuclear cells by histopathology; decrease in GPx and GSH by 39% and 35%, respectively
Garoui et al. 2012									**Cobalt Chloride**
37	RAT (Wistar) 5F	28 days, Daily from day 14 of pregnancy to day 14 post-delivery (W)	0, 20.3	BI BW CS FI GN HP OW UR WI	Bd wt	20.3			
					Renal		20.3		Vascular congestion, reduction of glomerular space, and infiltration of leukocyte cells between tubules; 15% increase in plasma creatinine, 34% decrease in urine creatinine, and slight reduction in relative kidney weight (4%), compared to controls
					Other noncancer		20.3		32% lower water intake and 29% lower food intake compared to controls

Table 2-2. Levels of Significant Exposure to Cobalt – Oral

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/kg/day)	Parameters monitored	Endpoint	NOAEL (mg/kg/day)	Less serious LOAEL (mg/kg/day)	Serious LOAEL (mg/kg/day)	Effects
Garoui et al. 2013									**Cobalt Chloride**
38	RAT (Wistar) 4M, 4F	4 weeks; Exposed through maternal dosing- in utero and lactation daily from day 14 of pregnancy to day 14 post-delivery; (W)	0, 20.3	BI BW CS DX NX OF OW	Neuro			20.3	AChE and BuChE levels decreased in cerebrum by 33% and 36%, respectively; AChE and BuChE levels decreased in cerebellum by 33% and 47%, respectively. Decreases in antioxidant enzymes in the brain were observed- in GSH and NPSH by 23% and 50% in the cerebrum, and by 16% and 25% in the cerebellum, respectively; co-treated rats exhibited poorly differentiated purkinje cells with frequent pyknotic cells, and their number of pyknotic cells was reduced (not otherwise described)
Grice et al. 1969									**Cobalt Sulfate**
39	RAT (Wistar) 30M	8 weeks, 7 days/week, Daily (F)	0, 26	CS OF	Cardio			26	Degeneration and swelling in myocardial cells accompanied by decrease in number of myofibrils in the cells based on histopathology; damaged mitochondria were identified with electron microscopy
Haga et al. 1996									**Cobalt Sulfate**
40	RAT (Sprague-Dawley) 10M	24 weeks, 7days/week, Daily (F)	0, 8.4	BW CS FI	Bd wt			8.4	31% decrease in body weight gain

Table 2-2. Levels of Significant Exposure to Cobalt – Oral

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/kg/day)	Parameters monitored	Endpoint	NOAEL (mg/kg/day)	Less serious LOAEL (mg/kg/day)	Serious LOAEL (mg/kg/day)	Effects
					Cardio			8.4	30% increase in the ratio of left ventricular weight to body weight; impaired ventricular function
					Hemato	8.4			
Haga et al. 1996									**Cobalt Sulfate**
41	RAT (Sprague-Dawley) 8M	16 weeks, 7days/week, Daily (F)	0, 8.4	BW CS FI	Bd wt			8.4	26% decrease in body weight
					Cardio	8.4			
					Hemato	8.4			
Holly 1955									**Cobalt Chloride**
42	RAT (Wistar) 8M	4 months, 7 days/week, 4 months, Daily (G)	0, 18	CS HE OF	Resp	18			
					Cardio	18			
					Gastro	18			
					Hemato		18		21% and 37% increases in red blood cell count and hemoglobin above controls; controls had an 18% increase in red blood cell count
					Hepatic	18			
					Renal	18			Tubular necrosis (not otherwise described)
					Endocr	18			
					Immuno	18			

Table 2-2. Levels of Significant Exposure to Cobalt – Oral

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/kg/day)	Parameters monitored	Endpoint	NOAEL (mg/kg/day)	Less serious LOAEL (mg/kg/day)	Serious LOAEL (mg/kg/day)	Effects
Khalil et al. 2020									**Cobalt(II) Chloride Hexahydrate**
43	RAT (Sprague-Dawley) 8M	4 weeks, Daily (W)	0, 68	BC CS LE NX OF	Hepatic		68		3.6-fold increase in LDH; 1.7, 4.5, and 1.7-fold increase in hepatic enzymes ALP, AST, and ALT, respectively; 1.9-fold increase in total bilirubin levels (these factors increase DNA damage in liver cells)
					Immuno		68		2.2-fold increase in immunoreactivity
					Neuro		68		Fatigue, lethargy, and dullness (not otherwise described)
Krasovskii and Fridlyand 1971									**Cobalt Chloride**
44	RAT (NS) 1-3NS	7 months, 6 days/week (GW)	0, 0.05, 0.5, 2.5	CS OF IX NX	Hemato	0.05	0.5		Unspecified increases in RBC, RBC diameter, and hemoglobin; mild transient polycythemia
					Hepatic	2.5			
					Renal	2.5			
					Endocr	2.5			
					Immuno	0.05	0.5		Unspecified decrease in phagocytic ability at 6-7 months
					Neuro	0.05	0.5	2.5	LOAEL: 0.5 mg/kg/day caused non-significant p<0.05 36% increase at 6-7 months SLOAEL: 2.5 mg/kg/day caused 47% increase in latent reflex at 4 months

Table 2-2. Levels of Significant Exposure to Cobalt – Oral

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/kg/day)	Parameters monitored	Endpoint	NOAEL (mg/kg/day)	Less serious LOAEL (mg/kg/day)	Serious LOAEL (mg/kg/day)	Effects
Mathur et al. 2011									**Cobalt(II) Chloride Hexahydrate**
45	RAT (Wistar) 8M	60 days, Daily (W)	0, 45	BC BI BW HP OW	Bd wt		45		Significant 8% decrease in body weight
					Hepatic		45		13% increase in relative liver weight; unspecified degradation, alteration in the morphology, and atrophy of liver cells; changes in liver biochemistry; 126% increase in SGOT and 122% increase in bilirubin
Mollenhauer et al. 1985									**Cobalt Metal**
46	RAT (Sprague-Dawley) 3M	14 weeks, Daily, 7 days/week (F)	0, 20	CS RX	Repro			20	Testicular degeneration and thickening of basal lamina and seminiferous tubules
Morvai et al. 1993									**Cobalt Chloride**
47	RAT (CFY) 8M	3 weeks, Daily, 7 days/week (G)	0, 12.4	BC HE HP NX OF	Bd wt	12.4			
					Cardio			12.4	~33% decrease in cardiac output; incipient, multifocal myocytolysis with degeneration of myofibrils (not otherwise described)
					Renal		12.4		8% decrease in relative kidney weight
					Neuro		12.4		10% decrease in relative brain weight

Table 2-2. Levels of Significant Exposure to Cobalt – Oral

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/kg/day)	Parameters monitored	Endpoint	NOAEL (mg/kg/day)	Less serious LOAEL (mg/kg/day)	Serious LOAEL (mg/kg/day)	Effects
Mutafova-Yambolieva et al. 1994									**Cobalt Nitrate**
48	RAT (Wistar) 6M	30 days, Daily (W)	0, 6.4	CS OF RX	Repro			6.4	275% increase in sympathetically-induced contractility of vas deferens (not otherwise described)
Nation et al. 1983									**Cobalt Chloride**
49	RAT (Sprague-Dawley) 6M	69 days, 7 days/week, Daily (F)	0, 5, 20	CS OW NX	Neuro	5	20		Lowered operant lever press rates (not otherwise described)
					Repro	5		20	Testicular atrophy seen in 42%
Pehrsson et al. 1991									**Cobalt Sulfate**
50	RAT (Sprague-Dawley) 12M	8 weeks, 7days/week, Daily	0, 8.4	BW CS HE OF	Bd wt			8.4	30% decrease in body weight
					Cardio	8.4			
					Hemato	8.4			
Saker et al. 1998									**Cobalt Chloride**
51	RAT (Sprague-Dawley) 6M	2 weeks, Daily (W)	0, 9.6	BC BI BW CS	Bd wt	9.6			
					Hepatic	9.6			
					Other noncancer	9.6			

Table 2-2. Levels of Significant Exposure to Cobalt – Oral

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/kg/day)	Parameters monitored	Endpoint	NOAEL (mg/kg/day)	Less serious LOAEL (mg/kg/day)	Serious LOAEL (mg/kg/day)	Effects
Umar et al. 2016									**Cobalt Chloride**
52	RAT (Wistar) 5B	28 days, Daily (NS)	0, 22.7	CS NX	Neuro	22.7			
Vassilev et al. 1993									**Cobalt Nitrate**
53	RAT (Wistar) 5M	30 days, Daily (W)	0, 6.44	CS NX OF	Bd wt	6.44			
					Neuro		6.44		Unspecified alterations in cholinergic sensitivity
Anderson et al. 1992									**Cobalt Chloride**
54	MOUSE (CD-1) 10M	13 weeks, 7 days/week, Daily (W)	0, 24.6	BW HP OF OW RX	Bd wt	24.6			
					Hepatic	24.6			
					Renal	24.6			
					Repro			24.6	Unspecified increase in the number of Leydig cells and changes in the peritubular area; increased folding in the germinal epithelium accompanied with changes in cell morphology
Anderson et al. 1993									**Cobalt Chloride**
55	MOUSE (CD-1) 10 M	13 weeks, 7days/week, Daily (W)	0, 43.4	BW HP OF OW RX	Bd wt		43.4		Significant 7% decrease in body weight

Table 2-2. Levels of Significant Exposure to Cobalt – Oral

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/kg/day)	Parameters monitored	Endpoint	NOAEL (mg/kg/day)	Less serious LOAEL (mg/kg/day)	Serious LOAEL (mg/kg/day)	Effects
					Repro			43.4	Irreversible testicular degeneration demonstrated by damage to seminiferous tubules and hypercellularity of the interstitial areas
Bryan and Bright, 1973									**Cobalt Chloride**
56	MOUSE (Swiss-Webster) 3 M	Once/day, 7 days/week, 13 weeks (W)	0, 763	BC BW HE	Hemato	763			
Bryan and Bright, 1973									**Cobalt Chloride**
57	MOUSE (Swiss-Webster) 3M	Once/day, 7 days/week, 3 weeks (W)	0, 763	BC BW HE	Hemato	763			
Gluhcheva et al. 2014									**Cobalt(II) Chloride Hexahydrate**
58	MOUSE (ICR) 7B	In utero for 2-3 days + 25 days via breastmilk + 65 days orally (W)	0, 18.6, 31	BW HE OW	Hemato		18.58		Unspecified hemoglobin changes and hematopoiesis
					Hepatic			18.58	Significant decrease (21.5%) of liver weight index in mice sacrificed on day 90, compared to controls

Table 2-2. Levels of Significant Exposure to Cobalt – Oral

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/kg/day)	Parameters monitored	Endpoint	NOAEL (mg/kg/day)	Less serious LOAEL (mg/kg/day)	Serious LOAEL (mg/kg/day)	Effects
					Renal		18.58	30.96	LOAEL: Increase (14.3%) of liver weight index in mice sacrificed on day 30, compared to controls at 18.58 SLOAEL: Significant increase (28.6%) of kidney weight index in mice sacrificed on day 30, compared to controls at dose 30.96
					Immuno			18.58	Significant decrease (43-53%) of spleen weight index (measure of relative weight) in mice sacrificed on day 60-90, compared to controls
Gluhcheva et al. 2020									**Cobalt(II) Chloride Hexahydrate**
59	MOUSE (ICR) 7-8B (W)	20-21 days, In utero and breastmilk; mothers exposed daily 2-3 days before birth and to post-natal day 18	0, 18.6	BI HE HP OW	Bd wt		18.57 F		17% decrease in body weight compared to controls
					Hemato		18.57 F		Statistically significant 17% increase in erythrocyte count; 19% decrease in mean corpuscular hemoglobin; and 10% decrease mean corpuscular volume, compared to controls
					Hepatic		18.57 F		Leukocyte infiltration; increased number of binucleated hepatocytes; abundant Kupffer cells; and apoptotic bodies in liver

Table 2-2. Levels of Significant Exposure to Cobalt – Oral

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/kg/day)	Parameters monitored	Endpoint	NOAEL (mg/kg/day)	Less serious LOAEL (mg/kg/day)	Serious LOAEL (mg/kg/day)	Effects
					Immuno		18.57 F		Reduced red pulp in spleen; 55% decrease in spleen weight index (measure of relative weight), compared to controls
Legostaeva et al. 2012									**Cobalt Chloride**
60	MOUSE (BALB/c) 8M, 8F	5 weeks, 7days/week, Daily (W)	0, 56	BI HE OF	Immuno		56		2-fold decrease in the concentration of the total blood protein and 1.5-fold decrease of total immunoglobulin G
Madzharova et al. 2010									**Cobalt Chloride**
61	MOUSE (BALB/c) 6-13 M	18 days, Daily	0, 34, 56	RX	Repro	56			
Pedigo and Vernon et al. 1993									**Cobalt Chloride**
62	MOUSE (B6C3F1) 10M	10 weeks, Daily (W)	0, 15	BW CS LE RX	Repro			15	Reduction in pregnancy in females by 57% when mated with males treated with Co; 28% decrease in implantation of embryos when mated with Co exposed males; 458% increase in preimplantation losses in pregnant females mated with Co-exposed males; sperm concentration decreased to 15.3% and sperm motility to 18.3%
Pedigo et al. 1988									**Cobalt Chloride**
63	MOUSE (B6C3F1) 20M	13 weeks, Daily (W)	0, 15	BW HE RX	Bd wt	15			

Table 2-2. Levels of Significant Exposure to Cobalt – Oral

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/kg/day)	Parameters monitored	Endpoint	NOAEL (mg/kg/day)	Less serious LOAEL (mg/kg/day)	Serious LOAEL (mg/kg/day)	Effects
					Hemato	15			
					Repro			15	Testicular weights as a % of body weight decreased to 14%, 21%, 57%, and 71% at 7, 9, 11, and 13 weeks, respectively; sperm concentration decreased to 81.3% after 9 weeks of treatment; 82% decrease in sperm motility after 11 weeks of exposure
Pedigo et al. 1988									**Cobalt(II) Chloride Hexahydrate**
64	MOUSE (B6C3F1) 5M	12 weeks, Daily (GW)	0, 23, 42, 72	BW CS HE OW RX	Bd wt	11	18		Significant decrease in body weight by 13%
					Hemato	6			
					Repro		6		Decrease in testicular weight (expressed as % of body weight) by 14%; 11% decrease in sperm concentration; 80% increase in serum testosterone
Shrivastava et al. 1996									**Cobalt Chloride**
65	MOUSE (Parkes) 6F	45 days, Daily (W)	0, 26	HP	Endocr			26	Time dependent effects of exposure; treated mice showed low epithelial lining with degenerated nuclei; degeneration in thyroid was observed 30 and 45 days after the exposure ceased; not otherwise described

Table 2-2. Levels of Significant Exposure to Cobalt – Oral

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/kg/day)	Parameters monitored	Endpoint	NOAEL (mg/kg/day)	Less serious LOAEL (mg/kg/day)	Serious LOAEL (mg/kg/day)	Effects
Zaksas et al. 2013									**Cobalt(II) Chloride Hexahydrate**
66	MOUSE (BALB/c) 19B	2-3 day in-utero + 25 days via breastmilk + 35 days orally (W)	0, 56.7	BC BW CS LE	Bd wt			56.7	33% decrease in average body weight by day 60, compared to controls
					Neuro	56.7			
Mohiuddin et al. 1970									**Cobalt Sulfate**
67	GN PIG 20M	5 weeks, Daily, 7 days/week (F)	0, 20	BW OB GN HP CS	Death			20	4/20 died
					Cardio		20		32% increase in relative heart weight; lesions observed in 75% of the samples examined

[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 0.03 mg Co/kg/day; dose was divided by an uncertainty factor of 10 for human variability and a modifying factor of 3 for limited database.
Studies listed in the table could potentially have examined more than one endpoint.

Anephric = without functioning kidneys; BC = serum (blood) chemistry; BI = biochemical changes; BW or bd wt = body weight; (C) = capsule; Cardio = cardiovascular; CS = clinical signs; DX = developmental toxicity; Endocr = endocrine; F = female(s); (F) = feed; FI = food intake; (G) = gavage – not specified; Gastro = gastrointestinal; GN = gross necropsy; (GW) = gavage – water intraperitoneal; HE = hematological; Hemato = hematological; HP = histopathology; Immuno = immunological; IX = immune function; LE = lethality; LOAEL = lowest-observed-adverse-effect level; M = male(s); Musc/skel = Musculo/skeletal; Neuro = neurological; NOAEL = no-observed-adverse- effect-level; NS = not specified; NX = neurological function; OF = organ function; OP = ophthalmology; OW = organ weight; Resp = respiratory; polycythemia = author reported term associated with increased hemoglobin or erythrocyte count; RX = reproductive function; UR = urinalysis; (W) = water; WI = water intake

Figure 2-3. Levels of Significant Exposure to Cobalt–Oral

Acute (≤14 days)

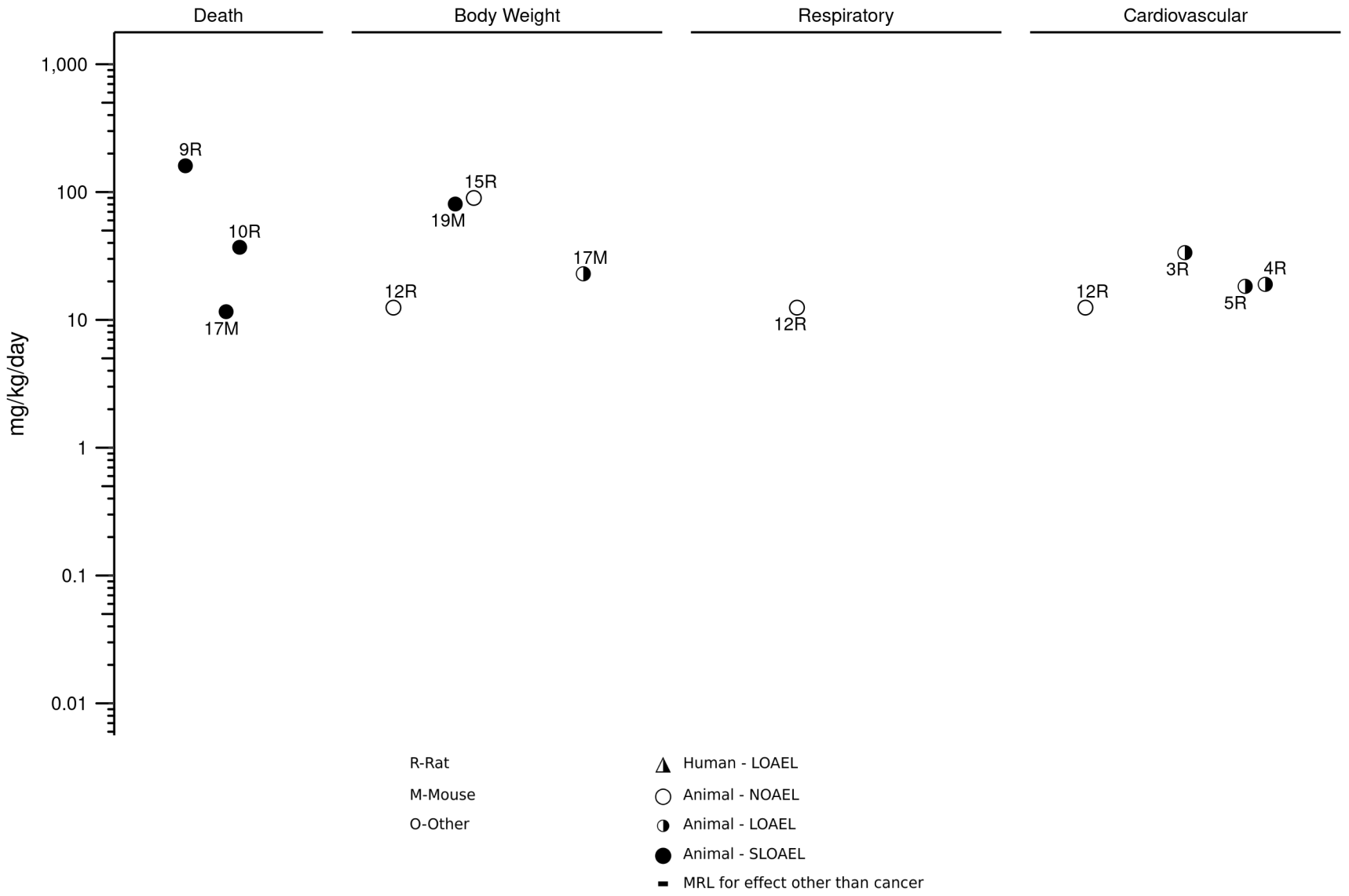


Figure 2-3. Levels of Significant Exposure to Cobalt–Oral

Acute (≤14 days)

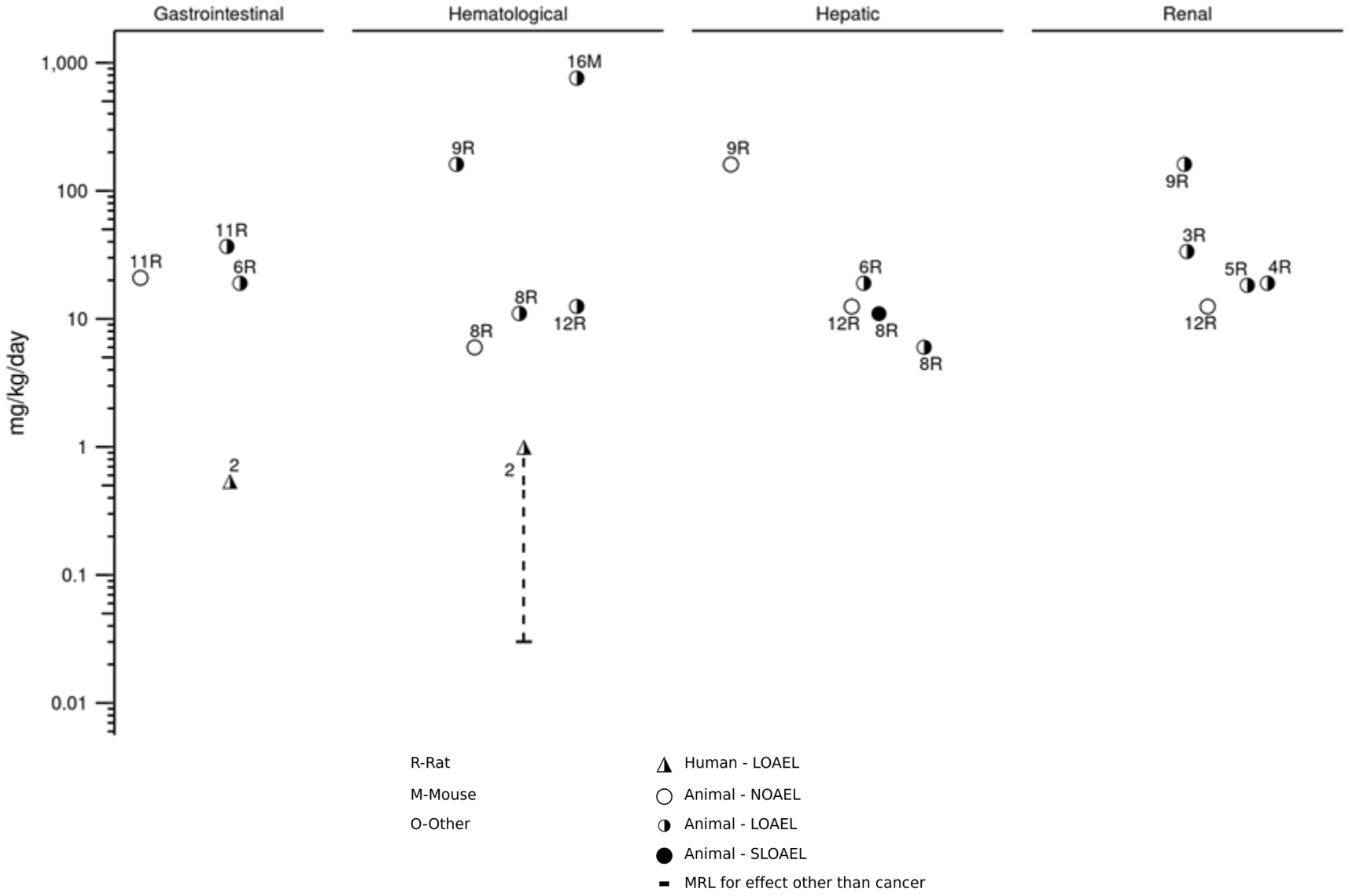


Figure 2-3. Levels of Significant Exposure to Cobalt–Oral

Acute (≤14 days)

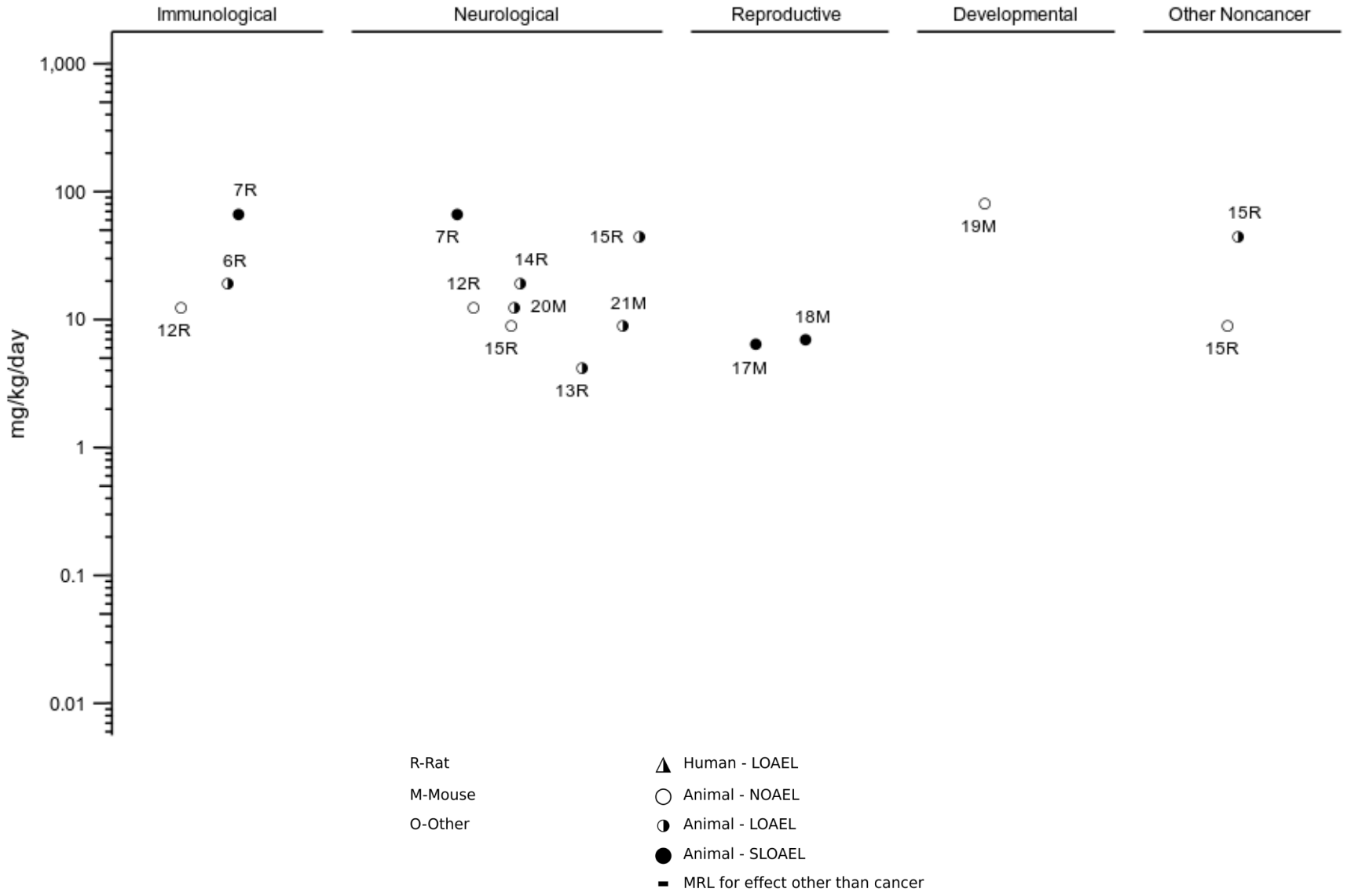


Figure 2-3. Levels of Significant Exposure to Cobalt–Oral
Intermediate (15-364 days)

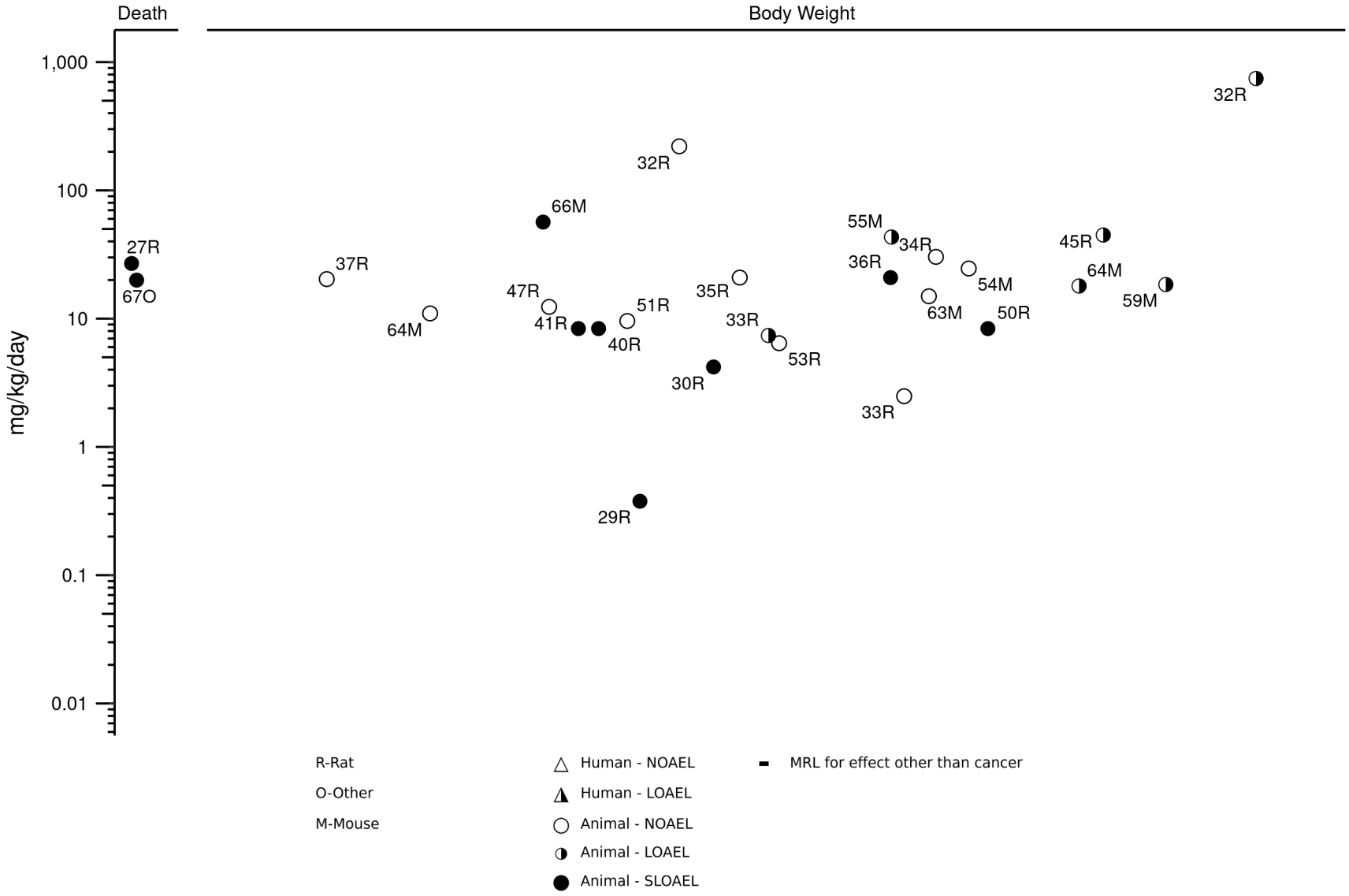


Figure 2-3. Levels of Significant Exposure to Cobalt–Oral

Intermediate (15-364 days)

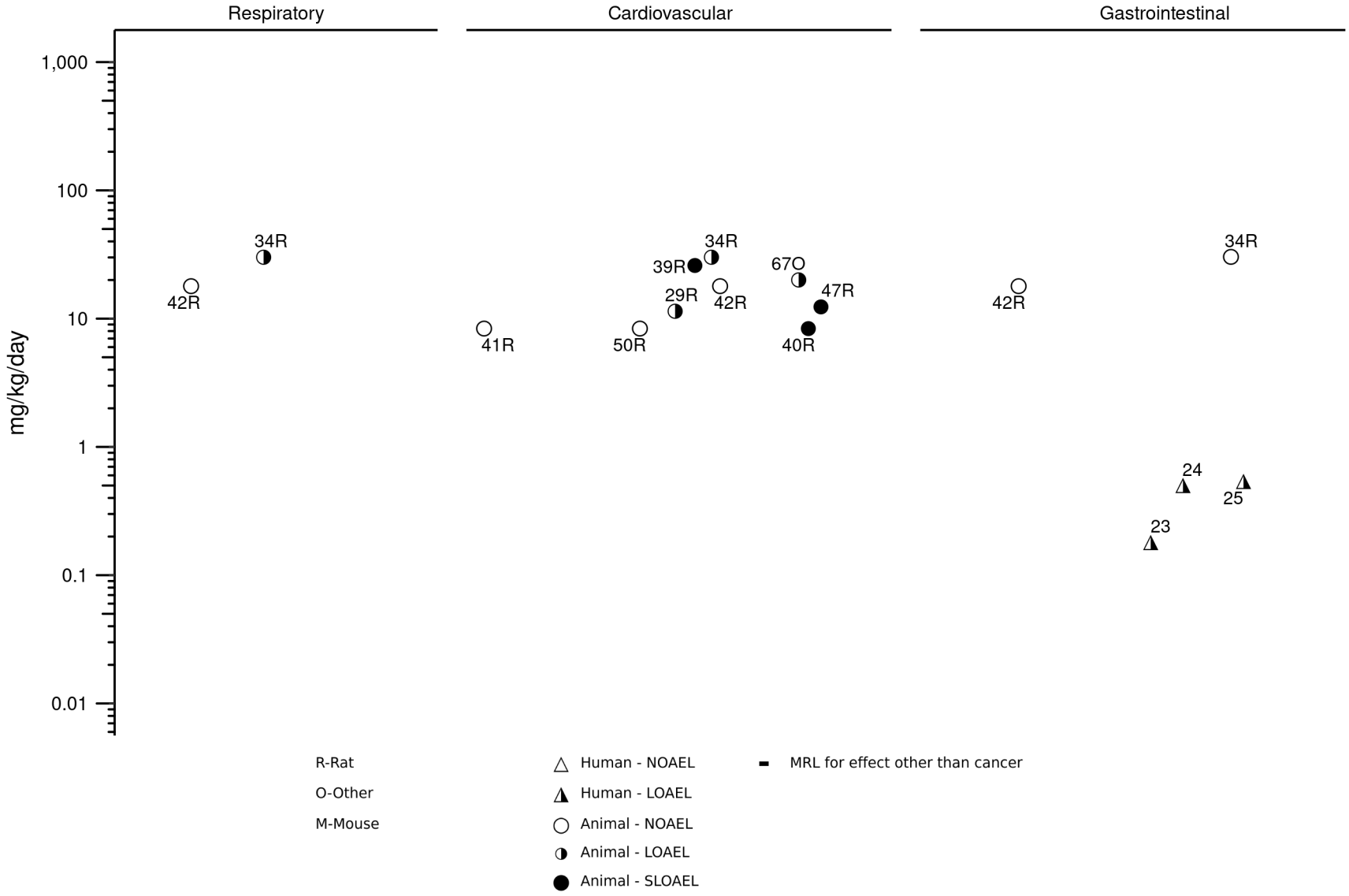


Figure 2-3. Levels of Significant Exposure to Cobalt–Oral
Intermediate (15-364 days)

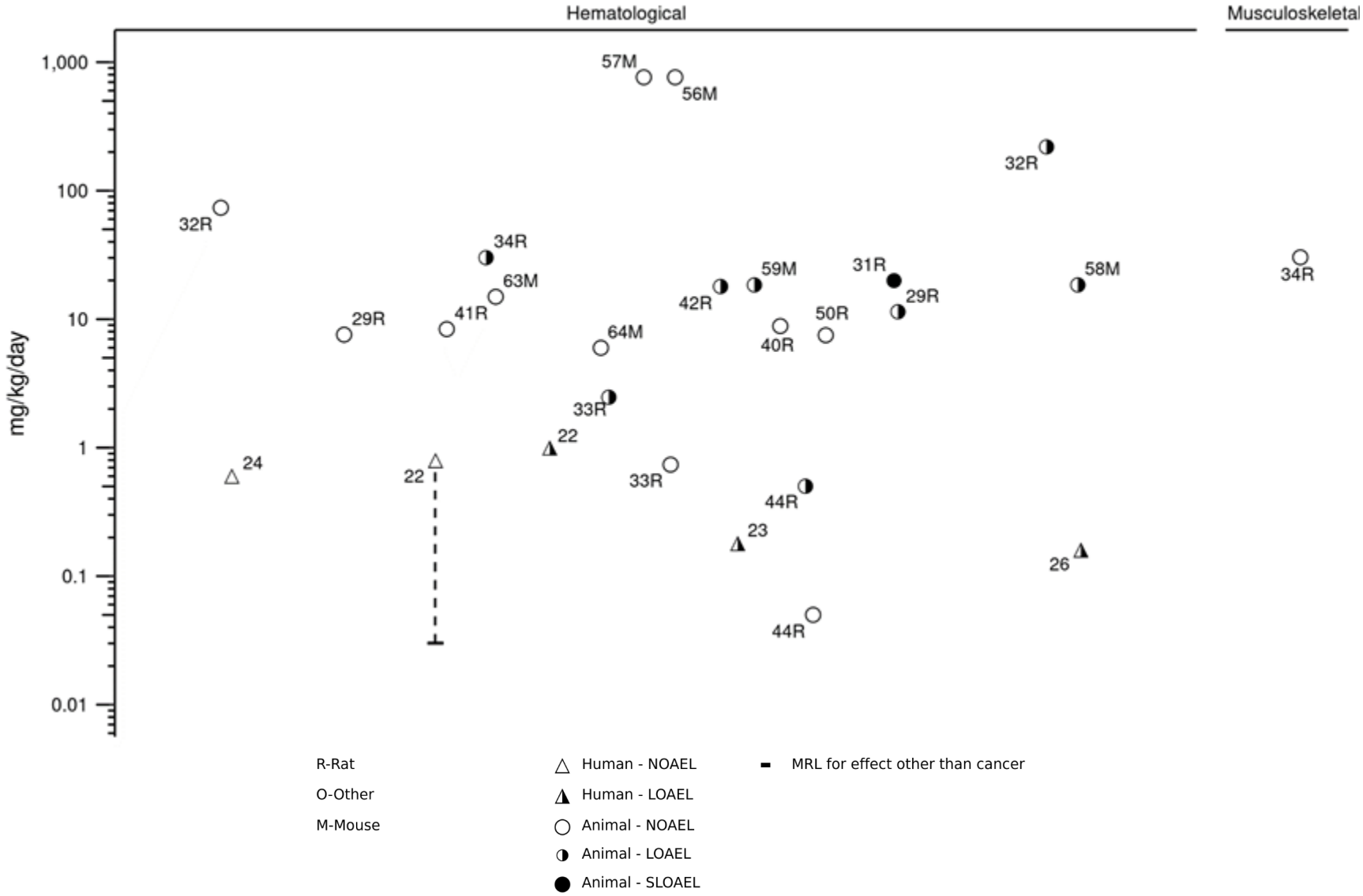


Figure 2-3. Levels of Significant Exposure to Cobalt–Oral

Intermediate (15-364 days)

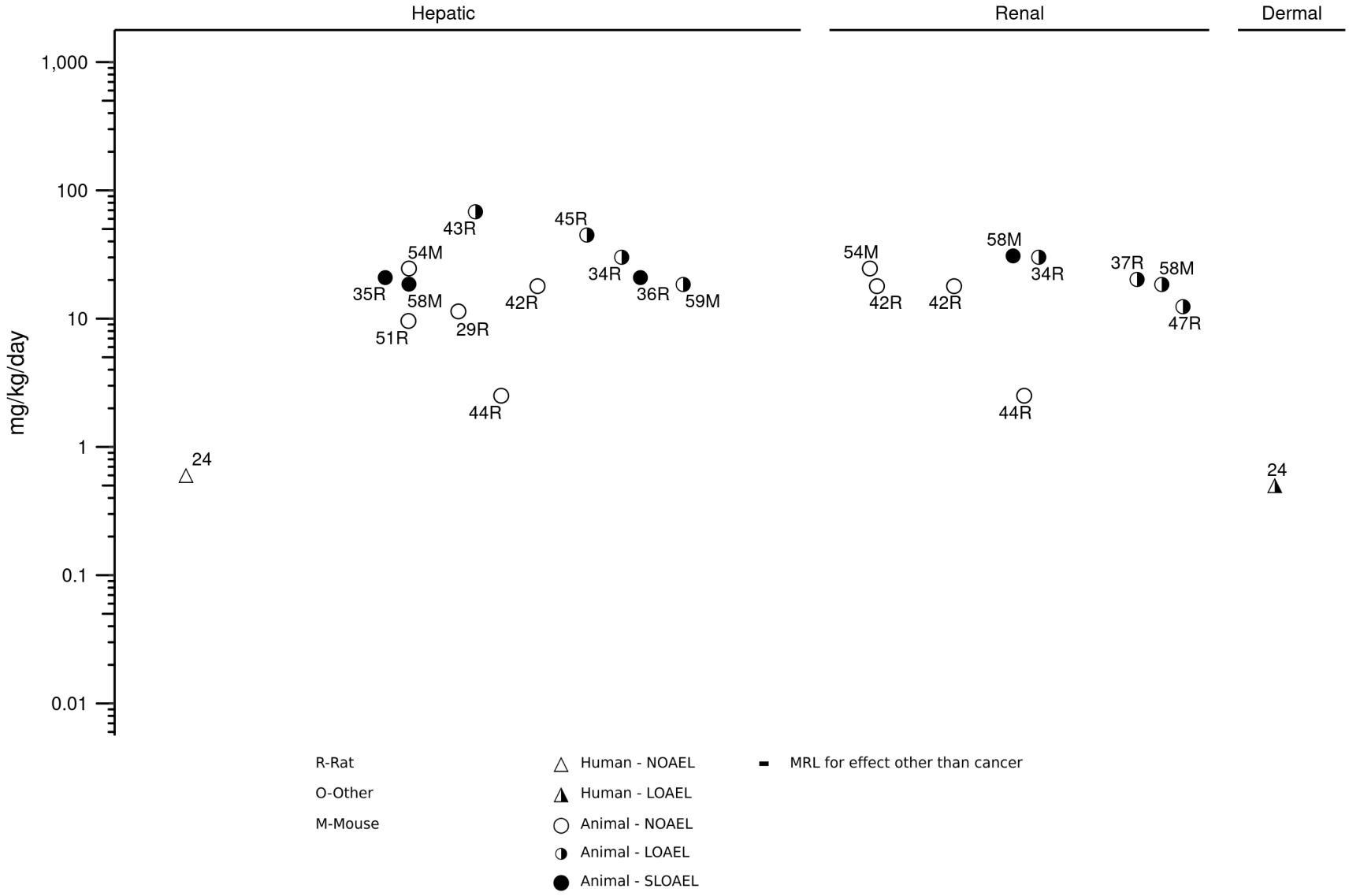


Figure 2-3. Levels of Significant Exposure to Cobalt–Oral

Intermediate (15-364 days)

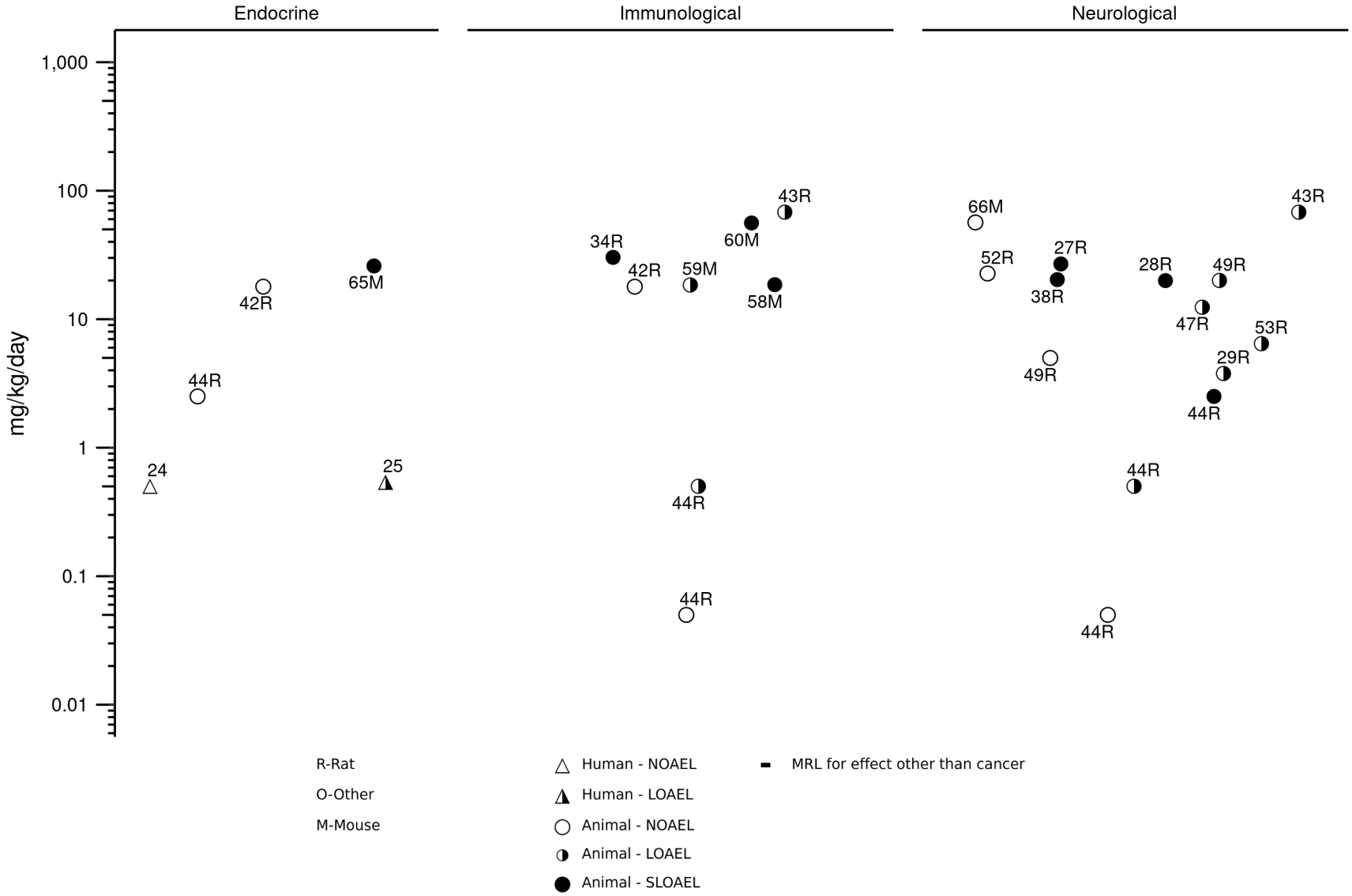


Figure 2-3. Levels of Significant Exposure to Cobalt–Oral

Intermediate (15-364 days)

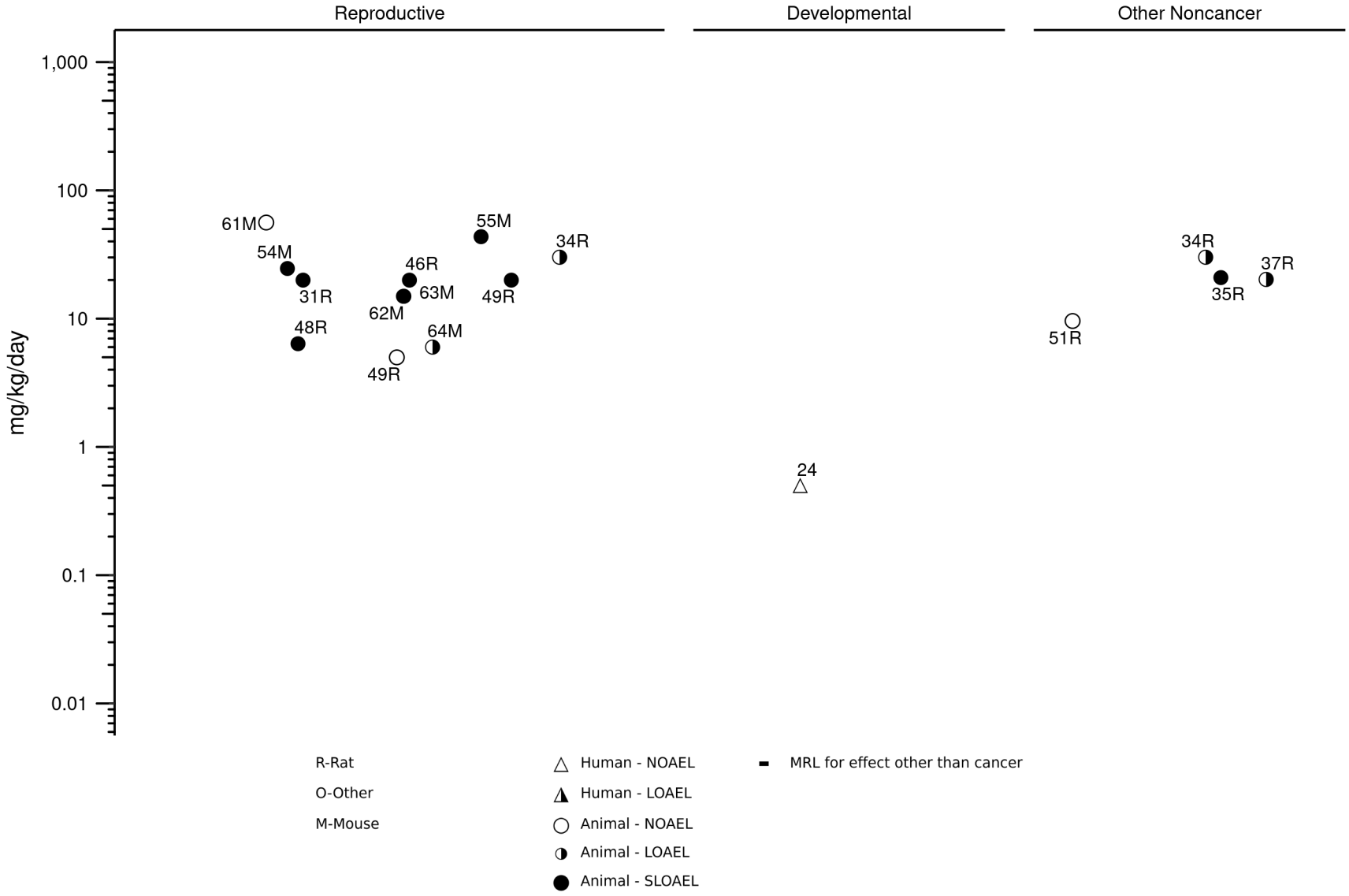


Table 2-3. Levels of Significant Exposure to Cobalt – Dermal

Figure key[a]	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**
1	RAT (Fischer-344) 3F	Once/day, 3 days w/v%	0, 0.5, 1, 2.5, 5%	CS IX OF	Immuno	1	2.5		Increased proliferation of lymphatic cells at dose of 2.5% w/v% solution of DMSO by a factor of 3.8
Ikarashi et al. 1992b									**Cobalt Chloride**
2	RAT (Fischer-344) 3F	Once/day, 3 days	0, 1, 5, 10%	CS IX OF	Immuno		1		Increased proliferation of lymphatic cells by factors of 1.5, 2.5, and 4.1 for 1%, 5%, and 10% doses, respectively
Ikarashi et al. 1992b									**Cobalt Chloride**
3	RAT (Fischer-344) 3F	Once/day 1 or 3 days in DMSO without or with abrasion	0, 5%	CS IX OF	Immuno		5		Increased proliferation of lymphatic cells by factor of 1.9 (after 1 dose) or 4.3 (after 3 doses without abrasion); pre-abrasion enhanced the 3-dose response another factor of 2.1
Bonefeld et al. 2015									**Cobalt Chloride**
4	MOUSE (NS) NS	Daily, 2 days	0, 10%	CS OF	Dermal		10		Swelling at the application site on the ears (increased ear thickness) by 1% for initial dose, 5% after challenge dose
					Immuno		10		Proliferation of B, CD4, and CD8 cells was approximately 40% for each cell type by initial plus challenge doses

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

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (%)	Parameters monitored	Endpoint	NOAEL (%)	Less serious LOAEL (%)	Serious LOAEL (%)	Effects
Ikarashi et al. 1992a									**Cobalt Chloride**
5	MOUSE (CBA/N) 3F	Once/day, 3 days	0, 0.5, 1, 2.5, 5 w/v%	CS IX OF	Immuno		0.5		Increased proliferation of lymphatic cells at a dose of 0.5 w/v% solution of DMSO by a factor of 2.1
Ikarashi et al. 1992a									**Cobalt Chloride**
6	GN PIG (Hartley) 3F	Once/day, 3 days	0, 0.5, 1, 2.5, 5% w/v%	CS IX OF	Immuno	2.5	5		Increased proliferation of lymphatic cells at dose of 5% w/v% solution of DMSO by a factor of 3.3
INTERMEDIATE EXPOSURE									
Kincaid et al. 1954									**Cobalt Dicobalt octacarbonyl**
7	GN PIG (NS) 3NS	Once/day, 5 days/week, 18 days	0, 2.4 %	CS	Dermal		2.4		Skin lesions, scabs, and denuded areas after dermal application at application site

CS = clinical signs; F = female(s); Immuno = immunological; IX = immune function; LOAEL = lowest-observed-adverse-effect level; NOAEL = no-observed-adverse-effect level; NS = not specified; OF = organ function

Table 2-4. Occupational Exposures to Cobalt and Health Outcome Associations

Reference, Study Type, and Study Population	Exposure, Inclusion/Exclusion Criteria, Covariates Considered and Adjustments	Outcomes and Limitations
Hematological		
Lantin et al. 2011 **Study Type:** Cross-sectional study of RBC and thyroid function in 249 workers in a cobalt refinery in Belgium, February 2008-August 2009.	**Exposure:** Blood and urine cobalt were assessed. Median blood and urine cobalt were 0.1 µg/100 mL and 3.9 µg/g creatinine, respectively. An integrated exposure index (IEI) based on historical biomonitoring records was used to assess long-term exposure, with a median of 106 µg/g creatinine × years. **Inclusion/Exclusion Criteria:** Retired workers were included only if they experienced substantial cobalt exposures in the past (minimum 5 to 8 years). Workers who were on chemotherapy or had hemochromatosis or hyperthyroidism were excluded. **Covariates Considered/Other Regression Adjustments:** Age, exercise, alcohol intake, cigarette smoking, and ethnicity.	**Outcomes:** No significant effects of cobalt exposure were observed on RBC measures or thyroid function. **Limitations:** Potential bias from residual confounding and healthy worker effect. Cross-sectional design precludes inferences of temporality.
Cardiovascular		
Lantin et al. 2013 **Study Type:** Cross-sectional study of cardiomyopathy in 237 workers in a cobalt refinery in Belgium, February 2008-August 2009.	**Exposure:** Recent exposures were assessed via urinary cobalt. Chronic exposure was assessed using an integrated exposure index (IEI) based on historical biomonitoring records. Median was approx. 4 µg/g creatinine for urinary cobalt and 100 µg/g creatinine × years for IEI. **Inclusion/Exclusion Criteria:** Participants with valvular heart disease, history of myocardial infarction, haemochromatosis or chemotherapy were excluded from the analysis. **Covariates Considered/Other Regression Adjustments:** Body mass index, height, age, heart rate, exercise, thyroid function, arterial hypertension,	**Outcomes:** Urinary cobalt was associated with decreased left ventricle volume, but not with any signs of dilated cardiomyopathy. No associations were found between IEI and echocardiographic or electrocardiographic parameters. **Limitations:** Potential bias from residual confounding and healthy worker effect. Cross-sectional design precludes inferences of temporality.

Table 2-4. Occupational Exposures to Cobalt and Health Outcome Associations

Reference, Study Type, and Study Population	Exposure, Inclusion/Exclusion Criteria, Covariates Considered and Adjustments	Outcomes and Limitations
	smoking, alcohol intake, diabetes, ferritin, abnormal ECG, emphysema, ethnicity, retirement status, and several types of heart disease.	
Linna et al. 2003a **Study Type:** Cross-sectional study of cardiotoxicity in 297 factory workers (203 exposed and 94 controls) in Finland, 1999	**Exposure:** Cumulative exposure was assessed using job-exposure matrices and ambient air measurements. Exposed workers had mean cobalt exposure of 0.82 mg-years. **Inclusion/Exclusion Criteria:** Employees with at least one year of exposure were included in the study. Workers with co-exposure to potential confounding chemicals (e.g., arsenic), history of myocardial infarction, or cardiac valvular disease were excluded. **Covariates Considered/Other Regression Adjustments:** Age, smoking status, blood pressure, alcohol overuse, physical activity, BMI, and heart rate.	**Outcomes:** Significant increases in left ventricular isovolumetric relaxation time and in deceleration time of the velocity of the early rapid filling wave were associated with cobalt exposure, indicating altered diastole. No differences in ECG findings, conduction parameters, or blood pressure were observed. **Limitations:** Relatively small number of participants in the control group. Control group was comprised of factory workers exposed to zinc.
Linna et al. 2020 **Study Type:** Cross-sectional study of cardiotoxicity in 142 factory workers (93 exposed and 49 controls) in Finland, 2006	**Exposure:** Cumulative exposure was assessed using job-exposure matrices and ambient air measurements. Exposed workers had mean cobalt exposure of 0.82 mg-years. **Inclusion/Exclusion Criteria:** Workers with co-exposure to potential confounding chemicals (e.g., arsenic), history of myocardial infarction, or cardiac valvular disease were excluded. **Covariates Considered/Other Regression Adjustments:** Age, smoking, BMI, hypertension, alcohol use, athleticism, and heart rate.	**Outcomes:** Prevalence of heart diseases, hypertension, and stroke were similar in exposed and unexposed workers. Exposed workers were more likely to report asthma and pulmonary diseases. No significant differences in blood pressure, heart rate, or ECG findings were observed by cobalt exposure. **Limitations:** Relatively small number of participants in the control group. Control group was comprised of zinc factory workers.

Table 2-4. Occupational Exposures to Cobalt and Health Outcome Associations

Reference, Study Type, and Study Population	Exposure, Inclusion/Exclusion Criteria, Covariates Considered and Adjustments	Outcomes and Limitations
Respiratory		
Hamzah et al. 2014 **Study Type:** Cross-sectional study of respiratory health in 436 factory workers in Malaysia, 2013	**Exposure:** Personal air sampling was conducted. TWA 8-hour cobalt concentrations in different job categories ranged from 0.01 mg/m^3 to 0.19 mg/m^3. **Inclusion/Exclusion Criteria:** Male factory workers aged 18 to 56 years old and over 1 year of employment were included. **Covariates Considered/Other Regression Adjustments:** Smoking status.	**Outcomes:** Exposure to cobalt was associated with significant increases in chronic phlegm and decreases in FVC and FEV1 ($p<0.05$). **Limitations:** Workers were co-exposed to high levels of chromium. Lung function tests may not have been able to identify all potential lung abnormalities, particularly in the small airways. Potential for bias from healthy worker effect.
Linna et al. 2003b **Study Type:** Cross-sectional study of cardiotoxicity in 110 factory workers (85 exposed and 25 controls) in Finland, 2006	**Exposure:** Cumulative exposure was assessed based on historical monitoring. Mean exposure to cobalt was 1.0 mg-years (range 0.1-4.6). **Inclusion/Exclusion Criteria:** Workers with a history of welding or work in other metallurgic plants were excluded. **Covariates Considered/Other Regression Adjustments:** Age and smoking status.	**Outcomes:** Symptoms of asthma such as phlegm, cough, and shortness of breath were more common in exposed participants. Compared to controls, the exposed group had significantly higher prevalence of suspected asthma (17.3% vs. 5.8%, $p<0.01$) and work-related asthma (14% vs. 3%, $p<0.008$). Respiratory flow rates at 50% and 25% of vital capacity were significantly lower in exposed smokers than in unexposed smokers. **Limitations:** Workers were co-exposed to irritants such as SO_2. Relatively small number of participants in the control group.
Sauni et al. 2010 **Study Type:** Case study of occupational asthma in cobalt plant workers in Finland, 1967-2003	**Exposure:** Mean air concentrations of cobalt in different departments ranged from 0.03 to 0.15 mg/m^3. **Inclusion/Exclusion Criteria:** None. **Covariates Considered/Other Regression Adjustments:** None.	**Outcomes:** In departments with higher air concentrations of cobalt, incidence of asthma was higher and latency period before symptoms occurred was shorter. Upon bronchial challenge tests for cobalt, workers displayed immediate, delayed, and dual asthmatic reactions. **Limitations:** Cases were diagnosed over a long-time span; challenge tests were not standard over

Table 2-4. Occupational Exposures to Cobalt and Health Outcome Associations

Reference, Study Type, and Study Population	Exposure, Inclusion/Exclusion Criteria, Covariates Considered and Adjustments	Outcomes and Limitations
		the study period. Co-exposures to irritant gases such as SO_2 occurred.
Walters et al. 2013 **Study type:** Case report of occupational asthma (n=4) and cross-sectional study (n=62) of factory workers in Alabama, 2010	**Exposure:** Urinary cobalt concentrations and excretion were assessed. Mean values were 0.6 µg/L and 0.6 µg/g creatinine, respectively. **Inclusion/Exclusion Criteria:** None. **Covariates Considered/Other Regression Adjustments:** None.	**Outcomes:** One case was attributed to cobalt exposure. Urinary cobalt concentrations were significantly higher in workers with probable/definite occupational asthma than in asymptomatic workers ($t<0.001$). No associations between cobalt exposure and occupational rhinitis were observed. **Limitations:** Workers were co-exposed to metals including chromium. Potential bias from confounding and healthy worker effect. Cross-sectional design precludes inferences of temporality.
Dermal		
Wahlqvist et al. 2020 **Study Type:** Cross-sectional study of 71 metal factory workers in Sweden, March 2017 to October 2018	**Exposure:** Air sampling of inhalable dust and biological sampling of blood, urine, and skin. Geometric mean breathing air cobalt concentrations ranged from 0.0001 to 0.019 mg/m^3. Mean blood cobalt concentrations were 6.2 nmol/L pre-shift, 6.9 nmol/L post-shift, and 6.6 nmol/L after 2 days. **Inclusion/Exclusion Criteria:** None. **Covariates Considered/Other Regression Adjustments:** None.	**Outcomes:** Many workers reported dry skin (42%). Prevalence of eczema on hands, face, and arms was 6-7%. **Limitations:** Dermal effects assessed via self-report. High percentage of exposed workers (14%) had eczema as a child.

BMI = body mass index, ECG = electrocardiogram, FEV1 = forced expiratory volume in one second, FVC = forced vital capacity, HDL = high-density lipoprotein, LDL = low-density lipoprotein, RBC = red blood cell, TWA = time-weighted average

Table 2-5. Environmental Exposure to Cobalt and Health Outcome Associations in Human Studies

Reference, Study Type, and Study Population	Exposure, Inclusion/Exclusion Criteria, Covariates Considered and Adjustments	Outcomes and Limitations
Hematological		
Jefferson et al. 2002 **Study Type:** Case-control study of cobalt levels and excessive erythrocytosis in 80 Peruvian men (21 high-altitude cases, 25 high-altitude controls, and 28 sea-level controls).	**Exposure:** Measured serum cobalt. Normal cobalt concentrations were considered to be 1.7 to 5.1 nmol/L. In the subset of cases with packed-cell volume >75%, concentrations ranged from 22 to 71 nmol/L. **Inclusion/Exclusion Criteria:** Men who smoked more than five cigarettes per day or had phlebotomy conducted within the past year were excluded from the study. **Covariates Considered/Other Regression Adjustments:** Altitude.	**Outcomes:** Serum cobalt was significantly elevated in cases (defined as packed-cell volume >65%) as compared to sea-level controls ($p = 0.002$) and high-altitude controls ($p = 0.002$). In cases, serum cobalt was correlated with packed-cell volume (erythrocytosis) ($r = 0.4$, $p = 0.01$). **Limitations:** Small sample size. Serum cobalt is only indicative of recent exposure; controls may have had past cobalt exposure.

Table 2-6. Oral Exposure to Cobalt and Health Outcome Associations in Human Studies

Reference, Study Type, and Study Population	Exposure, Inclusion/Exclusion Criteria, Covariates Considered and Adjustments	Outcomes and Limitations
Hematological		
Finley et al. 2013 **Study Type:** Human controlled exposure study of cobalt ingestion in 10 adults in the United States	**Exposure:** Volunteers ingested 1.0 mg/day of cobalt (0.08-0.19 mg/kg/day) for 31 days. Male and female participants had mean serum cobalt concentrations of 16 µg/L and 33 µg/L, respectively. **Inclusion/Exclusion Criteria:** Exclusion criteria were use of vitamins or dietary supplements; history of cobalt allergy; prior total joint replacement; history of cardiovascular, thyroid, kidney, or liver disease; insulin-dependent diabetes; weight <45 kg; and pregnancy or lactation. **Covariates Considered/Other Regression Adjustments:** None.	**Outcomes:** Cobalt ingestion was not associated with overt adverse health effects or biochemical indicators of thyroid, cardiac, liver, or kidney functions. In males only, there was a non-clinically significant (<5%) increase in hemoglobin, hematocrit, and RBC counts at 7 days after dose termination. **Limitations:** Small sample size and homogenous sample of healthy adults.
Tvermoes et al. 2014 **Study Type:** Human controlled exposure study of cobalt ingestion in 10 adults in the United States	**Exposure:** Volunteers ingested 1.0 mg/day of cobalt (0.08-0.19 mg/kg/day) for up to 90 days. Mean serum cobalt concentrations in men and women were 25 µg/L and 71 µg/L, respectively. **Inclusion/Exclusion Criteria:** Exclusion criteria were use of vitamins or dietary supplements; history of cobalt allergy; prior total joint replacement; history of cardiovascular, thyroid, kidney, or liver disease; insulin-dependent diabetes; weight <45 kg; and pregnancy or lactation. **Covariates Considered/Other Regression Adjustments:** None.	**Outcomes:** No significant changes in hematological parameters or biomarkers of cardiac, liver, or kidney function were observed. One female participant had elevated TSH and decreased T4 levels. Cobalt was not associated with cardiac, auditory, or visual changes. Some participants showed non-significant decreases in sensory nerve conduction velocity and amplitude. **Limitations:** Small sample size and homogenous sample of healthy adults.

RBC = red blood cell, TSH = thyroid-stimulating hormone, T4 = free thyroxine

2.2. DEATH

No studies were identified regarding death in humans after inhalation, oral, or dermal exposure to cobalt at any duration.

Inhalation

In laboratory animal studies, high dose acute- and intermediate-duration exposure to cobalt appeared to cause mortality but chronic-duration exposure to lower concentrations had no effect on survival. Acute inhalation of cobalt hydrocarbonyl was examined in albino rats by Palmes et al (1959) and it established that the LC_{50} for a two-week exposure was 165 mg Co/m^3. A 30-minute exposure to 78 mg Co/m^3 resulted in death of 3/5 albino rats (Palmes et al. 1959). In NTP (2014), lethal effects were seen in F344/N rats and B6C3F_1 mice exposed to 20 and 40 mg Co/m^3, respectively, after a 60-hour exposure. At 20 mg Co/m^3 5/5 males and 3/5 female rats died and at 40 mg Co/m^3, 4/10 mice died (NTP 2014). Intermediate-duration exposure to 6.29 mg Co/m^3 for 13 weeks was lethal only to B6C3F_1 mice and not to F344/N rats (Bucher et al. 1990; NTP 1991). A 3-month exposure to 9 mg Co/m^3 did not cause death in albino rats or guinea pigs (species not specified) (Palmes et al. 1959), and exposure to cobalt sulfate heptahydrate for 13 weeks did not affect the survival of either male and female Fisher-344 rats (Bucher et al. 1990). After a 16-day exposure to 19 mg Co/m^3 of cobalt sulfate, 2/5 male rats and 0/5 female rats died, while 5/5 male and 5/5 female mice died (NTP 1991). Chronic exposure for 105 weeks to cobalt sulfate heptahydrate did not have a significant effect on death in F344/N rats at 0.63 mg Co/m^3 (Bucher et al. 1999; NTP 1998). No increase in mortality in F344/N rats or B6C3F_1 mice of either sex was seen following 104-weeks of exposure to 1.14 mg Co/m^3 as cobalt sulfate (Bucher et al. 1999; NTP 1998). Chronic-duration exposure to cobalt metal for 105 weeks showed a reduced survival probability in male mice and female (F344/N) rats exposed to 2.5 mg Co/m^3, compared to controls (NTP 2014). Lethal levels for each species and duration category are presented in Table 2-1 and plotted in Figure 2-2.

Oral

In animals, acute-duration oral administration of cobalt at high doses caused death. Doming and Llobet (1984) showed that a single oral exposure to 161 mg Co/kg caused death in 5/20 Sprague-Dawley rats. Acute oral exposure to cobalt chloride tetrahydrate at a dose of 149 mg Co/kg/day killed 10 out of 20 Sprague-Dawley rats in the treatment group (Domingo et al. 1985a).

Oral intermediate-duration exposure to cobalt compounds in animals resulted in death. In an intermediate-duration exposure study, 4/10 Wistar rats died following exposure to 27 mg Co/kg/day for 60 days (Abdel-Rahman Mohamed et al. 2019). Elbetieha et al. (2008) demonstrated that a 12-week exposure to

11.61 mg Co/kg/day as cobalt chloride hexahydrate caused the death of 1 Swiss mouse during the 10th week (Elbetieha et al. 2008). The authors in Elbetieha et al. (2008) did not specify whether this death was treatment related. No death was observed at 20.3 mg Co/kg/day as cobalt chloride in Wistar rat dams and pups exposed for 2 weeks during gestation and then additionally for 2 weeks of lactation (Garoui et al. 2013). Death of 1 mouse was observed during the 10th week in a study where ICR mice were orally exposed to 6.4 mg Co/kg/day in water for 12 weeks (Gluhcheva et al. 2020). The authors of Gluhcheva et al. (2020) did not specify whether this effect was treatment related. 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 death was reported among BALB/c mice exposed to cobalt in utero, via breastmilk, and then orally (Zaksas et al. 2013). In this study, dams were exposed to 56.7 mg Co/kg/day as cobalt chloride hexahydrate for 2-3 days during gestation, followed by 25 days during breastfeeding, and then offspring were orally exposed for 35 days through drinking water (Zaksas et al. 2013). A 4-week exposure to 68 mg Co/kg/day as cobalt chloride hexahydrate did not cause any death in Sprague-Dawley rats (Khalil et al. 2020).

Dermal

Two studies in animals reported no death following dermal exposures to cobalt compounds. Acute 3-day dermal exposure to 0.5-10% cobalt chloride did not cause death in mice (Bonefeld et al. 2015; Ikarashi et al. 1992b; Ikarashi et al. 1992a). Intermediate 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 (species not specified) (Kincaid et al. 1954).

Other

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 tetrahydrate at a dose of 12 mg Co/kg/day in the form of 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). Doming 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.

Inhalation

Several studies in animals indicate that inhalation exposure to cobalt and cobalt compounds results in decreased body weight. Intermediate exposure to 20 mg Co/m^3 for 16 days resulted in 45% and 20% decreases in body weight in female and male F344/N rats, respectively, compared to control rats (NTP 2014). Similar effects were seen in B6C3F_1 mice, where females showed a 37% decrease, and males a 26% decrease in body weight after exposure to 40 mg Co/m^3 (NTP 2014). Inhalation exposure to 30 mg Co/m^3 as cobalt sulfate heptahydrate for 13 weeks, 5 days a week, 6 hours/day reduced the mean body weights in male F344/N rats; however, no differences were seen in female rats when compared to control animals (Bucher et al. 1990). A 3-month exposure to cobalt metal for 5 days a week, 6 hours/day at 0.1 mg Co/m^3 resulted in a 16% decrease in body weight in pigs (strain not specified) (Kerfoot 1974). B6C3F_1 mice exposed to 19 mg Co/m^3 as cobalt sulfate heptahydrate for 16 days, 5 days a week, 6 hours/day showed a 33% and 20% decrease in body weight in males and females, respectively (NTP 1991). A 13-week 5 days a week, 6 hours/day exposure to 6.29 mg Co/m^3 as cobalt sulfate heptahydrate caused a 22% decrease in female B6C3F_1 mice and a 14% decrease in male mice (NTP 1991). A 14-week, 5 days a week, 6 hours/day exposure to cobalt metal at 10 mg Co/m^3 decreased body weight by 13-14% in B6C3F_1 mice (NTP 2014). No weight loss was seen in albino rats or guinea pigs (strain not specified) exposed for 3 months to cobalt at a level of 9 mg Co/m^3 as cobalt hydrocarbonyl (Palmes et al. 1959).

Continuous chronic-duration exposure to 5 mg Co/m^3 for 105 weeks (5 days a week, 6 hours/day) caused a decrease in body weight in both male (~23%) and female (21%) F344/N rats and in male (11%) and female (25%) B6C3F_1 mice (NTP 2014). Lifetime continuous exposure, (5 days a week, 7 hours/day) to 7.9 mg Co/m^3 as cobalt oxide did not result in decreased body weight gain in hamsters (ENG:ELA) (Wehner et al. 1977).

Oral

Decreased body weight was commonly observed in animals orally exposed to cobalt and its compounds. Intermediate-duration exposure of 30 days to 0.379 mg Co/kg/day as cobalt chloride caused a 45% decrease in body weight gain in male Sprague-Dawley rats (Chetty et al. 1979). Rats showed a 33% decrease in body weight gain after exposure to 4.2 mg Co/kg/day as cobalt sulfate for 4 weeks (Clyne et al. 1988). Exposure of Sprague-Dawley rats to 8.4 mg Co/kg/day as cobalt sulfate for 8-24 weeks resulted in a 30% to 31% decrease in body weight (Haga et al. 1996; Pehrsson et al. 1991). At 9.6 mg Co/kg/day as cobalt chloride for 2 weeks there were no effects on body weight in Sprague-Dawley rats (Saker et al. 1998). Elbetieha et al. (2008) demonstrated that a 12 week exposure to 23 mg Co/kg/day as cobalt chloride hexahydrate induced a significant 7% increase of body weight in Swiss mice, which is not

considered biologically significant based on ATSDR guidelines (ATSDR 2018). A 7.1% decrease in body weight gain compared to controls was observed in a study where ICR mice were orally exposed to 6.36 mg Co/kg/day in water for 12 weeks (Gluhcheva et al. 2020). An in utero dose of 56.73 mg Co/kg/day as cobalt chloride hexahydrate for 2-3 days, followed by an equivalent dose for 25 days via breastmilk, followed by the same dose for 35 days orally via drinking water significantly decreased the average body weight of offspring BALB/c mice by 33% by day 60 of exposure when compared to control animals (Zaksas et al. 2013). The body weight at autopsy was reduced by 11% (males) and 9% (females), respectively, at 7.44 mg Co/kg bw/day as $CoCl_2$. At the end of the 4-week recovery period (test day 118), the body weight of the male and female animals exposed to the highest dose was still reduced by 17% or by 13%, respectively, compared with the control group (Danzeisen et al. 2020). Danzeisen et al. (2020) also examined the effects of oral exposure to Co_3O_4 at the dose of 734 mg Co/kg/day for 90 days and observed there were marginal effects on body weight in male and female rats.

Dermal

Dermal exposure to cobalt did not induce body weight changes in laboratory animal studies. Acute-duration dermal exposure to 0.5-10% cobalt chloride (dissolved in DMSO) did not induce any changes in body weight in mice (Bonefeld et al. 2015; Ikarashi et al. 1992b; Ikarashi et al. 1992a). No body weight changes were observed after intermediate-duration dermal exposure to 51.70 mg Co/kg/day as dicobalt octacarbonyl in methyl ether ketone in guinea pigs (Kincaid et al. 1954). Bonefeld et al. (2015) and Ikarashi et al. (1992a, 1992b) tested an unspecified strain of mouse and ICR mice, respectively, and did not observe any differences.

Other

Acute-duration exposure by subcutaneous injection to 45 mg Co/kg/day as dicobalt octacarbonyl did not affect body weight in guinea pigs (strain not specified) (Kincaid et al. 1954). A daily 6-week subcutaneous injection study in albino rats caused a 24% weight loss following administration of cobalt chloride at a dose of 2.5 mg Co/kg/day (Stanley et al. 1947).

2.4. RESPIRATORY

Inhalation

Inhalation exposure to cobalt and its compounds resulted in altered spirometry and increased evidence of pulmonary irritation and dyspnea in human occupational exposure studies. In laboratory animal studies, acute-duration inhalation exposure to cobalt increased inflammation, edema, and necrosis in the lungs.

Intermediate-duration exposure caused an increase in lung weight and inflammation and chronic-duration exposure increased neoplasms and hyperplasia along with lung inflammation.

Several occupational studies have found respiratory effects associated with cobalt exposures. In Malaysian factory workers exposed to 8-hour cobalt concentrations ranging from 0.01 to 0.19 mg/m^3, exposure to cobalt was associated with significant increases in chronic phlegm and decreases in forced vital capacity and forced expiratory volume ($p<0.05$) (Hamzah et al. 2014). Cobalt exposure has also been linked to increased risk of occupational asthma (Linna et al. 2003a; Sauni et al. 2010; Walters et al. 2013). Compared to controls, exposed factory workers in Finland had significantly higher prevalence of suspected asthma (17.3% vs. 5.8%, $p<0.01$) and work-related asthma (14% vs. 3%, $p<0.008$) (Linna et al. 2003a). Sauni et al. (2010) conducted a case study of occupational asthma in cobalt plant workers in Finland from 1967-2003 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 (SO_2) and ammonia (NH_3) that are known respiratory irritants (Andersson et al. 2006; ATSDR 1998; Huber and Loving 1991). After 1987, cobalt was produced using by-products of the metallurgic industry as raw material which eliminated the co-exposure to the irritant gases and the incidence of asthma decreased to only 1 case between 1987-2003 compared to 21 cases between 1967-1987 (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. Walters et al. (2013) found that urinary cobalt concentrations were significantly higher in workers with probable or definite occupational asthma than in asymptomatic workers ($t<0.001$). Morfeld et al. (2017) reported an increase in non-malignant respiratory diseases in a cohort working in a German hard metal industry after occupational exposure. In the studies detailed above, all the factory workers were subjected to co-exposures with other metals like nickel and chromium and irritant gases, therefore the health effects observed might not be caused by cobalt alone (Hamzah et al. 2014; Linna et al. 2003a; Sauni et al. 2010; Walters et al. 2013). Respiratory effects of exposure to cobalt, tungsten, and nickel were evaluated in an international cohort of hard metal production workers (Marsh et al. 2017a; Marsh et al. 2017b).Workers from 3 companies, 17 sites among 5 countries, including the United States, Austria, Germany, Sweden, and the United Kingdom were evaluated. Information on respiratory parameters were obtained from various national datasets, and phone interviews were completed for participants when possible. These interviews provided information on demographics and lifestyle factors. The exposed workers showed chronic obstructive pulmonary disease, bronchitis, emphysema, and asthma (Marsh et al. 2017a; Marsh et al. 2017b).

An acute 6-hour exposure to cobalt dust at 0.038 mg Co/m^3 decreased lung function in exposed workers compared to non-exposed workers (Kusaka et al. 1986a). Chronic 3-year exposure to 0.126 mg Co/m^3 caused a 2.7% decrease in lung function, specifically FEV1% in exposed workers, while 0.085 mg Co/m^3 did not affect the pulmonary function (Kusaka et al. 1986a). Occupational exposure to cobalt metal for unspecified periods at 0.0152 and 0.1355 mg Co/m^3 decreased lung parameters FEV1 and FVC by ~10% and increased cough, sputum, and dyspnea in exposed workers, and these parameters correlated with their urinary cobalt levels (Gennart and Lauwerys 1990). In this study by Gennart and Lauwerys (1990), cobalt air concentrations were measured from 2 rooms that workers moved freely between during the work shift and no individual worker stay times or exposures were provided. The absence of this information did not allow accurate estimation of the average exposure per worker. However, at a similar concentration of 0.0175 mg Co/m^3, there were no effects observed in workers after a chronic occupational exposure for 3 years (Deng et al. 1991). Occupational exposure to cobalt at the concentration of 0.0151 mg Co/m^3 decreased FEV1 (5%) and FVC (5%) in exposed workers. The exposed workers also exhibited increased incidence of cough (11/91), wheezing (4/91), and upper airway irritation (40/91) (Nemery et al. 1992). Among the workers subjected to work-related exposure, upper airway effects were seen in 30% of controls, 26% of low dose individuals, and 43% of high dose individuals. Work-related cough was not observed in the control subjects, but was observed in 4% of low exposure individuals and in 12% of high exposure individuals. While the respiratory effects appeared at a greater rate in individuals who were exposed to higher concentrations of Co, 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, 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, control. The lower exposure concentration of 0.0053 mg Co/m^3 did not alter pulmonary function in the exposed workers.

Animal studies have also observed respiratory effects consistent with the effects seen in human studies. Following a 30-minute exposure to 90 mg Co/m^3 all albino rats showed pulmonary irritation with dose-dependent edema and damage in the lungs (Palmes et al. 1959). The albino rats also exhibited labored breathing and disturbed respiration (Palmes et al. 1959). In this study, all animals (albino rats and guinea pigs) exposed to concentrations ≥106 mg Co/m^3 showed lung inflammation. A 2-week exposure to cobalt chloride at 2.4 mg Co/m^3 increased lung weight by 20% and retention of lavage fluid by 53% in female

Hartley guinea pigs (Camner et al. 1993). As per the authors, the significance of retained lavage fluid is unclear.

Male albino rats exposed to 9 mg Co/m^3 intermittently for 3 months showed lung inflammation, edema, congestion, and bronchitis (Palmes et al. 1959). In F344/N rats exposed to cobalt concentrations as low as 0.21 mg Co/m^3 for 13 weeks, relative lung weights increased by 14% compared to controls (Bucher et al. 1990; NTP 1991). B6C3F1 mice exposed to cobalt at 19 mg/m^3 for 16 days showed lesions and degeneration of the olfactory epithelium, squamous metaplasia in the respiratory epithelium, inflammation in the nose, and metaplasia of the trachea (Bucher et al. 1990; NTP 1991). Necrosis and inflammation of the respiratory tract epithelium (nasal turbinates, larynx, trachea, and bronchioles) were reported in F344/N rats exposed to 19 mg Co/m^3 and in mice exposed to concentrations ≥1.9 mg Co/m^3 for 16 days (Bucher et al. 1990; NTP 1991).

Intermittent exposure of F344/N rats and $B6C3F_1$ mice to cobalt as cobalt sulfate for 13 weeks, resulted in adverse effects on all parts of the respiratory tract, with the larynx being the most sensitive part (Bucher et al. 1990; NTP 1991). NTP (1991) observed an increase in lung weights in both male (7%) and female (14%) rats along with histiocytic infiltrates in the lung reported at similar levels in both the rats and mice. Severe edema and lung inflammation were observed in albino rats following exposure to a concentration of 90 mg Co/m^3 for 3-months (Palmes et al. 1959). A continuous intermediate 3-month exposure to 0.1 mg Co/m^3 as cobalt metal in pigs decreased specific respiratory compliance by 29% (Kerfoot 1974). At a concentration of 20 mg Co/m^3, F344/N rats of both sexes showed abnormal breathing, increased incidence of lung hemorrhage, and acute inflammation (Behl et al. 2015; NTP 2014). An intermediate 16-day intermittent inhalation exposure to cobalt metal at 2.5 mg Co/m^3 in F344/N rats and B6C3F1 mice of both sexes induced necrosis in the respiratory epithelium and atrophy of olfactory epithelium (Behl et al. 2015; NTP 2014). A 14-week intermittent intermediate exposure to 0.625 mg Co/m^3 caused an increase in the incidence of chronic active inflammation in lungs and an increase in relative lung weight in both male and female F344/N rats (Behl et al. 2015; NTP 2014). Both male and female B6C3F1 mice exposed to 0.625 mg Co/m^3 for 14 weeks showed cytoplasmic vacuolization of the bronchiole epithelium (Behl et al. 2015; NTP 2014). A 17-week intermittent exposure in male rabbits to 0.4 mg Co/m^3 caused inflammation in lungs, accumulation of macrophages, and at 2 mg Co/m^3, caused severe inflammation and an increase in lung lobe weight by 25% (Johansson et al. 1987). Johansson et al. (1992) also observed alterations in pulmonary tissues, alterations in BAL parameters, and a 22% decrease in macrophages in male rabbits after a 4-month intermittent exposure to cobalt metal.

In F344/N rats, chronic exposure to cobalt sulfate for 105 weeks caused inflammation of the larynx at ≥0.21 mg Co/m^3, and more severe effects on the nose, larynx, and lung were reported at concentrations

≥0.21 mg Co/m^3 (NTP 1998). In B6C3F1 mice, acute inflammation of the nose was observed at ≥0.63 mg Co/m^3 along with atrophy, metaplasia, or hyperplasia in the larynx and olfactory epithelium (NTP 1998). Exposure of F344/N rats and mice to aerosols of cobalt (as cobalt sulfate) at concentrations from 0.06 to 0.63 mg Co/m^3 for 2 years resulted in a spectrum of inflammatory, fibrotic, and proliferative lesions in the respiratory tract of males and females (Bucher et al. 1999; NTP 1998). A 2-year intermittent chronic exposure to cobalt metal at 1.25 mg Co/m^3 increased the incidence of lung neoplasm and non-neoplastic lesions in the lungs and nose in rats and mice of both sexes (Behl et al. 2015; NTP 2014). 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).

Oral

No studies examined respiratory toxicity in humans following oral exposure to cobalt.

A significant 33% increase in the weight of the lungs, without morphological or histological changes, was observed in Sprague-Dawley rats that received 30.2 mg Co/kg/day as cobalt chloride in drinking water for 3 months, as compared with controls (Domingo et al. 1984). No morphological changes were seen in the lungs of Wistar rats treated with 18 mg Co/kg/day for 4 months (Holly 1955).

Dermal

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

2.5. CARDIOVASCULAR

Inhalation

Several studies were identified that examined cardiovascular effects in humans after occupational inhalation exposure to cobalt which provided contradictory evidence of cardiovascular toxicity. A study of cobalt refinery workers in Belgium, Lantin et al. (2013) found no association between cumulative cobalt exposures and any changes in echocardiographic or electrocardiographic parameters. Increased urinary cobalt was associated with decreased left ventricle volume, but not with any signs of dilated cardiomyopathy (Lantin et al. 2013). In a study of Finnish factory workers and a 6-year follow-up, no differences in electrocardiogram findings or blood pressure were observed from cobalt exposure (Linna et al. 2003b, 2020). Exposed workers did show significant changes in left ventricular relaxation and filling, indicating altered diastole (Linna et al. 2003b). However, at follow-up, prevalence of heart disease, hypertension, and stroke were similar in exposed and unexposed workers (Linna et al. 2020). Morfeld et

al. (2017) reported an increase in heart disease in a cohort working in a German hard metal industry after occupational exposure. Cardiovascular effects of exposure to cobalt, tungsten, and nickel were evaluated in an international cohort of hard metal production workers (Marsh et al. 2017a; Marsh et al. 2017b). Workers from 3 companies, 17 sites among 5 countries, including the United States, Austria, Germany, Sweden, and the United Kingdom were evaluated. Information on cardiovascular parameters were obtained from various national datasets, and phone interviews were completed for participants when possible. These interviews provided information on demographic and lifestyle factors. The exposed workers showed increased incidences of cardiovascular diseases as a result of occupational exposure (Marsh et al. 2017a; Marsh et al. 2017b). Exposure to 20 mg Co/m^3 did not cause cardiovascular effects in F344/N rats after a 16-day intermittent exposure (NTP 2014). In contrast, female B6C3F1 mice similarly exposed showed a 39% increase in heart weight at 40 mg Co/m^3 (NTP 2014). Intermittent exposure to cobalt metal at 0.1 mg Co/m^3 for 3 months caused a 14% increase in heart rate, a 38% decrease in QRS amplitude, and electrocardiogram abnormalities that may reflect ventricular impairment in pigs (strain not specified) (Kerfoot 1974). No signs of cardiovascular toxicity were observed in experimental studies where animals were exposed to concentrations ranging from 0.625 to 19 mg Co/m^3 for intermediate and chronic-durations in F344/N rats and 0.625 to 41.72 mg Co/m^3 for intermediate and chronic-duration exposures in B63F1 mice (NTP 1991, 1998, 2014).

Oral

No studies were identified that examined cardiovascular effects in humans after oral exposure to cobalt.

Animal studies indicate that oral exposure to cobalt induces cardiovascular effects for multiple animal species exposed to cobalt for acute and intermediate durations. In Wistar rats, oral exposure to 650 mg Co/L of cobalt chloride in drinking water (34 mg Co/kg/day) induced effects on the cardiovascular system (Ajibade et al. 2017). In this study, exposure to 33.7 mg Co/kg/day decreased glutathione (GSH) and glutathione peroxidase (GPx) expression in the heart by 2.2% and 11%, respectively (Ajibade et al. 2017). Cobalt exposure also increased mean blood pressure by 50% and Nf-kB expression by 67% (Ajibade et al. 2017). Acute-duration oral exposure to 18.38 mg Co/kg/day in Wistar rats caused cardiac damage in a study by Akinrinde et al. (2016). This study examined histopathology of the heart and observed hemorrhagic lesions with congestion of coronary blood vessels and mild infiltration of the myocardium and atrium by inflammatory cells. Additionally, a 12% decrease in systolic blood pressure, a 150% increase in lactate dehydrogenase, and a 67% increase in AST were also observed (Akinrinde et al. 2016b). A second acute-duration exposure study by Akinrinde et al. (2016) also corroborated their previous study as exposure to 19 mg Co/kg/day caused inflammation of the myocardium and areas of

myocardial infarction along with a 17, 24, and 21% decrease in systolic blood pressure, diastolic blood pressure, and mean arterial pressure, respectively, in Wistar rats (Akinrinde et al. 2016c).

Clyne et al. (2001) reported that exposure of Sprague-Dawley rats to 8.4 mg Co/kg/day, as cobalt sulfate, in the diet for 24 weeks resulted in significant reductions in a number of enzymes in cardiac tissues, including manganese-superoxide dismutase, succinate-cytochrome c oxidase, NADH-cytochrome c reductase, and cytochrome c oxidase, as well as reducing the mitochondrial ATP production rate (Clyne et al. 2001). Exposure of Sprague-Dawley rats to 8.4 mg Co/kg/day as cobalt sulfate resulted in left ventricular hypertrophy and impaired left ventricular systolic and diastolic functions (Haga et al. 1996). Conversely, in Sprague-Dawley rats exposed to 8.4 mg Co/kg/day as cobalt sulfate for 8 weeks, there were no effects on cardiac function (Pehrsson et al. 1991). A 3-week exposure to 12.4 mg Co/kg/day as cobalt chloride in male CFY rats resulted in cardiac damage, presenting as multifocal myocytolysis with myofibril degeneration (Morvai et al. 1993). Two to three months of daily exposure to 26 to 30.2 mg Co/kg/day in drinking water resulted in degenerative heart lesions (Grice et al. 1969) and a 9.4% increase in relative heart weight (Domingo et al. 1984) in Wistar and Sprague-Dawley rats, respectively.

An oral exposure to 20 mg Co/kg/day for 5 weeks as cobalt sulfate in guinea pigs (strain not specified) 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 histopathologically. Exposure also caused an increase in relative bradycardia, decrease in QRS voltage, and a significant increase in abnormal ECG findings (Mohiuddin et al. 1970). While there were no control groups included, Wistar rats exposed to a single dose of 176.6 mg Co/kg administered by gavage as cobalt fluoride or a single dose of 795 mg Co/kg administered as cobalt oxide showed a proliferation of interstitial tissue, swollen muscle fibers, and focal degeneration in the cardiac tissues in males and females (Speijers et al. 1982).

Dermal

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

No cardiovascular effects have been observed after dermal exposure to cobalt in animals. Acute-duration dermal exposure to 0.5-10% cobalt chloride did not induce any cardiovascular effects in mice (Bonefeld et al. 2015; Ikarashi et al. 1992b; Ikarashi et al. 1992a). No cardiovascular effects were observed after intermediate-duration dermal exposure to 51.70 mg Co/kg/day as dicobalt octacarbonyl in guinea pigs (Kincaid et al. 1954).

Other

Acute-duration exposure by subcutaneous injection of 45 mg Co/kg/day as dicobalt octacarbonyl did not cause cardiovascular toxicity in guinea pigs (Kincaid et al. 1954).

2.6. GASTROINTESTINAL

No studies were identified that examined gastrointestinal effects in humans after inhalation, oral, or dermal exposure to cobalt.

Inhalation

No histological lesions were reported in the esophagus, stomach, duodenum, ileum, jejunum, cecum, colon, or rectum of rats or mice of both sexes exposed to 0.4 mg Co/m^3 to 41.72 mg Co/m^3 for 16 days, 0.06 to 6.29 mg Co/m^3 for 13-14 weeks, and 5 mg Co/m^3 and 0.63 mg Co/m^3 for 105 weeks (Behl et al. 2015; NTP 1991, 1998, 2014).

Oral

Studies showed that oral exposure to cobalt resulted in gastric distress in humans and changes in gut microbiota along with signs of intestinal injury in animals. Twice a day, daily oral exposure to 0.18 mg Co/kg/day for 12 weeks in humans caused gastric distress manifested as nausea and constipation in anephric patients (with non-functioning kidneys) (Duckham and Lee 1976). Intestinal injury in the form of loss of epithelial cells in the intestines were observed in Wistar rats exposed to 29 mg Co/kg/day as cobalt chloride (Akinrinde et al 2016). Acute-duration oral exposure to cobalt chloride at 37 mg Co/kg/day altered the gut microbiota composition in treated Sprague-Dawley rats which were quantified using 16s rRNA amplicon sequences (Richardson et al. 2018). No morphological changes in the gastrointestinal system were observed following exposure of 20 Sprague-Dawley male rats exposed to 30.2 mg Co/kg/day in drinking water for 3 months (Domingo et al. 1984) or in Wistar rats exposed to 18 mg Co/kg/day for 4 months (Holly 1955).

Dermal

No gastrointestinal effects have been observed after dermal exposure to cobalt in animals. Acute-duration dermal exposure to 0.5-10% cobalt chloride (in DMSO) did not induce gastrointestinal effects in mice (Bonefeld et al. 2015; Ikarashi et al. 1992b; Ikarashi et al. 1992a). No gastrointestinal effects were observed after intermediate dermal exposure for 18 days, 5 days/week, once/day to 2.4% cobalt as dicobalt octacarbonyl (in methyl ethyl ketone) in guinea pigs (Kincaid et al. 1954).

Other

Acute exposure by subcutaneous injection of 45 mg Co/kg/day as dicobalt octacarbonyl did not cause any gastrointestinal illness or alter physiology in the gastrointestinal tract of guinea pigs (Kincaid et al. 1954).

2.7. HEMATOLOGICAL

Inhalation

One occupational study examined hematological effects in humans from inhalation exposure to cobalt. Chronic-duration exposure to cobalt in refinery workers was not associated with changes in red blood cell parameters such as hemoglobin and hematocrit even though exposure resulted in increased urinary cobalt levels. The workers used protective masks since 2002 which lowered the urinary cobalt levels compared to workers without protective gear (Lantin et al. 2011).

Inhalation exposure of cobalt induced changes in hematological parameters mainly in erythrocytes, hematocrit, and hemoglobin. An intermittent 3-month intermediate-duration exposure to 9 mg Co/m^3 as cobalt hydrocarbonyl increased levels of hemoglobin, numbers of basophils, and monocytes in albino rats and guinea pigs (strain not specified), but not in dogs (beagles) (Palmes et al. 1959). Bucher et al. (1990) showed that male and female F344/N rats developed polycythemia (as reported in the study) following exposure to 10 and 3 mg/m^3, respectively, after 13 weeks of intermittent exposure. Both sexes also had increased hemoglobin and hematocrit levels at 10 mg/m^3. The reticulocyte count increased only in female rats exposed to 30 mg/m^3 (Bucher et al. 1990). No hematological effects were seen in pigs (strain not specified) after a 3-month exposure to cobalt metal (Kerfoot 1974). Polycythemia (as reported in the study) was reported in F344/N rats, but not B6C3F1 mice, exposed to 1.14 mg Co/m^3 as cobalt sulfate for 13 weeks (NTP 1991). A 5% decrease in hemoglobin, a 3% decrease in hematocrit, and a 4% decrease in platelet count were seen in female and male B6C3F1 mice after a 13-week exposure (NTP 1991). After a 14-week intermittent exposure, female F344/N rats showed a 9% increase in hematocrit, hemoglobin, and erythrocyte levels, compared to controls at 1.25 mg Co/m^3 and male F344/N rats showed a 4.5% increase in hemoglobin levels and 5.2% increase in erythrocytes, compared to controls at 0.625 mg Co/m^3. Female and male B6C3F1 mice showed a significant 4.7% and 3% increase in erythrocytes, respectively (Hong et al. 2015).

Oral

Oral exposure to cobalt altered hematological parameters including hemoglobin, hematocrit, and erythrocytes. Finley et al. (2013) administered 1.0 mg/day of cobalt (0.08-0.19 mg/kg/day) to volunteers (n=10) for 31 days. At 7 days post-exposure, a non-clinically significant (<5%) increase in hemoglobin,

hematocrit, and red blood cell counts was observed in males only (Finley et al. 2013). Tvermoes et al. (2014) found no significant changes in hematological parameters following 90-day exposure to 1.0 mg/day of cobalt (0.08-0.19 mg/kg/day) in 10 volunteers. Acute oral exposure to 1 mg Co/kg/day for 6-14 days increased red blood cells by 8.7% in humans and intermediate exposure to 0.8 mg Co/kg/day for 15 days did not influence the levels of red blood cells but 1 mg Co/kg/day for 22-23 days increased the red blood cells by 9.7% (Davis and Fields 1958). Acute oral exposure to 1 mg Co/kg/day for 6-14 days increased red blood cells by 8.7% in humans and intermediate exposure to 0.8 mg Co/kg/day for 15 days did not have an effect on the red blood cells, but 1 mg Co/kg/day for 22-23 days increased the red blood cell count by 9.7% (Davis and Fields 1958). Twice a day, daily oral exposure to 0.18 mg Co/kg/day for 12 weeks in anephric (with non-functioning kidneys), hemoglobin deficient patients increased hemoglobin in male and female human subjects by 26-70% and eliminated the need for transfusions in 4/12 individuals (Duckham and Lee 1976). Daily oral exposure to 0.16 mg Co/kg/day in humans for 12-32 weeks caused an increase in hemoglobin which was not quantified in the study (Taylor et al. 1977). A 13 week daily oral exposure to 0.6 mg Co/kg/day in humans did not cause any hematological effects (Holly 1955). A study by Roche and Layrisse (1956) examined iodine uptake in 12 euthyroid (normal thyroid) patients who were orally exposed to 1mg Co/kg-day (assuming a body weight of 70 kg) for 2 weeks which resulted in a greatly reduced uptake of 48-hour radioactive iodine by the thyroid when measured after 1 week of exposure to cobalt. The decreased uptake is likely resulting from cobalt blocking the organic binding of iodine (Paley et al. 1958). This effect was reversed by the second week of exposure nearly completely (Roche and Layrisse, 1956). No other clinical details (e.g., including effects on thyroid stimulating hormone [TSH]) were provided for the human subjects in this study, therefore, the mechanism for the effect of cobalt on thyroidal iodine uptake cannot be ascertained.

Acute-duration oral exposure to cobalt has also led to hematological effects in rats. A single oral exposure to cobalt chloride of 161 mg Co/kg caused an 8% increase in hematocrit levels in Sprague-Dawley rats (Domingo and Llobet 1984). Acute 5-day exposure to cobalt in albino rats caused a dose-dependent increase in the frequency of micronucleated polychromatic erythrocytes with a 400% increase at 11 mg Co/kg/day (Awoyemi et al. 2017). The NOAEL dose of 6 mg Co/kg/day used in Awoyemi et al. (2017) where no effects were observed is a dose that is rather high for humans to be exposed to cobalt via oral exposure. The average daily intakes are often in the microgram range. Oral acute-duration exposure to 12.5 mg Co/kg/day resulted in 60%, 10%, and 8% increased red blood cells, hematocrit, and hemoglobin, respectively, in Sprague-Dawley rats (Shrivastava et al. 2008; Shrivastava et al. 2010). A single oral dose of 161 mg Co/kg caused significant increases in erythrocyte count (polycythemia, as reported by the study authors), hematocrit, and hemoglobin in Sprague- Dawley rats (Domingo et al. 1984).

Intermediate-duration oral exposure to cobalt caused hematological effects in rats and mice. Rats were exposed to 0.74, 2.48, and 7.44 mg Co/kg/day as cobalt chloride hexahydrate orally daily for 90 days (Danzeisen et al. 2020). In this study, 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 was a 10.7%, 9.2%, and 10.2% increase in hemoglobin, erythrocytes, and hematocrit, respectively. While the male rats were more sensitive and showed changes in hematological parameters at lower doses, female rats showed an increase of 13.4% and 9.8% in hemoglobin and erythrocytes, respectively, only at a dose of 7.44 mg Co/kg/day (Danzeisen et al. 2020). Danzeisen et al. (2020) also examined effects of Co_3O_4 on hematological parameters. They observed that a daily oral dose of 220 mg Co/kg/day increased hemoglobin, erythrocytes, and hematocrit by 9.5%, 9.6%, and 9.2%, respectively, in male rats, and a 5.9% increase in hemoglobin level in female rats. At the highest dose of 734 mg Co/kg/day, males and female rats showed an increase in hemoglobin (25.4% males and 16.4% females), erythrocytes (22.7% males and 12.9% females), and hematocrit (24.2% males and 13.9% females) (Danzeisen et al. 2020). Krasovskii and Fridyland (1971) exposed groups of rats to 0, 0.05, 0.5 and 2.5 mg Co/kg/day as cobaltous chloride, daily for 7 months. The group treated with 2.5 mg Co/kg/day showed a persistent increase in erythrocytes, the 0.5 mg Co/kg/day group showed a transient increase, and the lowest exposure group showed no effect. This study provided qualitative findings but did not report numerical data and their statistical significance. An intermediate exposure of cobalt chloride in dogs for a 4-week period to daily doses of 5- 30 mg Co/kg/day and a dose ≤ 15 mg Co/kg/day caused a significant increase in erythrocyte number and hemoglobin level, when compared to preexposure levels (Brewer 1940). 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 76.4 mg Co/kg as cobalt chloride in the drinking water (Bryan and Bright 1973). Exposure for an intermediate-duration (3 weeks or 3 months) to 76.4 mg Co/kg as cobalt chloride in the drinking water resulted in no alterations in serum proteins examined in Swiss mice (Bryan and Bright 1973). Intermediate exposure to 18 and 0.5 mg Co/kg/day for 4 and 7 months, respectively, in water caused an increase in red blood cells and hemoglobin (Holly 1955; Krasovskii and Fridlyand 1971), and induced polycythemia in Wistar rats (reported as mild transient polycythemia by study authors) (Krasovskii and Fridlyand 1971). A 30-day exposure to 11.4 mg Co/kg/day as cobalt chloride caused a 20% decrease in hemoglobin in male Sprague-Dawley rats (Chetty et al. 1979). Hematological parameters in Sprague-Dawley rats exposed to 20 to 30 mg Co/kg/day as cobalt chloride for 13 to 14 weeks in food or drinking water resulted in an increase in red blood cells (41%), hemoglobin (28-31%) and hematocrit (29%) (Corrier et al. 1985; Domingo et al. 1984). Pregnant ICR mouse dams were orally treated with 18.6 mg Co/kg/day as cobalt chloride hexahydrate which resulted in the offspring being exposed in utero for 2-3 days followed by 25 days via breastmilk; after weaning they were exposed orally for 65 days which

altered levels of hemoglobin and hematopoiesis (Gluhcheva et al. 2014). Pedigo et al. (1988) observed that a 6 mg Co/kg/day exposure resulted in no hematological effects in male mice after a daily exposure for 13 weeks in B6C3F1 mice (Pedigo et al. 1988). A 6-week study in albino rats showed a dose- and time-related increase in erythrocyte number following oral administration of 2.5 mg Co/kg/day (Stanley et al. 1947).

Dermal

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

No hematological effects have been observed after dermal exposure to cobalt in animals. Acute dermal exposure to 0.5-10% cobalt chloride (in DMSO) did not induce hematological effects in mice (Bonefeld et al. 2015; Ikarashi et al. 1992b; Ikarashi et al. 1992a). No hematological effects were observed after intermediate dermal exposure to 51.70 mg Co/kg/day as dicobalt octacarbonyl (in methyl ethyl ketone) in guinea pigs (Kincaid et al. 1954). Bonefeld et al (2015) and Ikarashi et al. (1992a, 1992b) tested an unspecified strain of mouse and ICR mice, respectively, and did not observe any differences.

Other

Acute-duration exposure to cobalt chloride by 10 subcutaneous injections in a controlled exposure human study (9 days gap between 2 blocks of 5 consecutive injections) of 18 mg Co/kg/day increased triglycerides by 49%, caused lipemia, and 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 ($r = 0.4$, $p = 0.01$). Doming 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 6-week subcutaneous injection study in albino rats showed an increase in erythrocyte number following administration of cobalt chloride at a dose of 0.6 mg Co/kg/day (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.

Inhalation

No histological lesions were reported in the sternebrae (unpaired segments of the sternum), including the bone marrow, of rats or mice exposed to 0.06-41.72 mg Co/m^3 as cobalt sulfate for 16 days, 0.06 to 6.29 mg Co/m^3 for 13 weeks, and 0.06 to 0.63 mg Co/m^3 for 104 weeks (NTP 1991, 1998, 2014).

Oral

No morphological changes were found in the skeletal muscle of rats exposed to 30.2 mg Co/kg/day as cobalt chloride in the drinking water for 3 months (Domingo et al. 1984).

Dermal

No musculoskeletal effects have been observed after dermal exposure to cobalt in animals. Acute dermal exposure to 0.5-10% cobalt chloride (in DMSO) did not induce musculoskeletal effects in mice (Bonefeld et al. 2015; Ikarashi et al. 1992b; Ikarashi et al. 1992a). No musculoskeletal effects were observed after intermediate-duration dermal exposure to 51.70 mg Co/kg/day as dicobalt octacarbonyl (in methyl ethyl ketone) in guinea pigs (Kincaid et al. 1954). Bonefeld et al (2015) and Ikarashi et al. (1992a, 1992b) tested an unspecified strain of mouse and ICR mice, respectively, and did not observe any differences.

Other

Acute-duration exposure by a single subcutaneous injection of 45 mg Co/kg/day as dicobalt octacarbonyl did not cause musculoskeletal effects in guinea pigs (Kincaid et al. 1954).

2.9. HEPATIC

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

Inhalation

Intermediate-duration inhalation exposure to cobalt in rats and mice altered liver weight, caused necrosis and congestion. Sixteen days of intermittent exposure to 20 mg Co/m^3 increased relative liver weight by 13% and 16% in male and female F344/N rats, respectively, and in both male and female B6C3F1 mice by 10-11% (NTP 2014). Necrosis and congestion of the liver were observed in both F344/N rats and B6C3F1 mice that died following intermittent exposure to 19 mg Co/m^3 as cobalt sulfate over 16 days (NTP 1991). A significant decrease in relative liver weights were observed in female and male B6C3F1 mice at 2.5 and 10 mg Co/m3, respectively, after 14 weeks of intermittent inhalation exposure (NTP 2014). 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). No effects on hepatic function were seen in F344/N rats following intermittent exposure to 5 mg Co/m^3 for 14 weeks (NTP 2014). In NTP (1998), a chronic intermittent exposure of 105 weeks to cobalt sulfate heptahydrate did not alter hepatic function in both F344/N rats and B6C3F1 mice (NTP 1998). Chronic intermittent 105-week exposure to 1.25 and 5 mg Co/m^3/day increased the incidence of basophilic focus in male and female F344/N rats, respectively, but had no effect in B6C3F1 mice (NTP 2014).

Oral

Acute-duration oral exposure to cobalt caused necrosis, congestion, changes in liver weight, and inflammation of the liver in animal studies. Domingo and Llobet (1984) showed that a single oral exposure to 161 mg Co/kg did not alter hepatic function in Sprague-Dawley rats (Domingo and Llobet 1984). Akinrinde et al. (2016) observed necrosis, cytotoxicity, and abnormal morphology in Wistar rat hepatocytes following exposure to 29 mg Co/kg/day as cobalt chloride. This study also noted a 14% decrease in relative liver weight, and 50% and 40% decrease in GSH and GPx, respectively (Akinrinde et al. 2016a). An acute 1-week exposure to 6 mg Co/kg/day in albino rats caused moderate congestion in the hepatocytes as well as very mild infiltration by inflammatory cells, focal areas of necrosis, and congestion of vessels in rats (Awoyemi et al. 2017). Awoyemi et al. (2017) reported statistically significant changes in several hepatic parameters, e.g., GSH, SPx, and ALT, at doses ranging from 6 to 22 mg Co/kg/day. Additionally, immunohistochemistry of liver in albino rats showed a dose dependent increase in hepatic expressions of cyclooxygenase 2 (COX-2) and BCL-2- associated protein (BAX) relative to the control. Changes in oxidative stress markers along with altered liver enzymes were observed, namely a 19% increase in H_2O_2, 5% decrease in GSH, 37% decrease in GPx, and 16% increase in ALT in Wistar rats (Awoyemi et al. 2017).

Intermediate-duration oral exposure to cobalt altered liver enzymes and caused inflammation of the liver in animals. A 13-week exposure to 30.2 mg Co/kg/day as cobalt chloride in the drinking water caused a 30% decrease in liver enzymes in rats (Domingo et al. 1984). A dose of 9.6 mg Co/kg/day for 2 weeks produced no effects on hepatic function in Sprague-Dawley rats (Saker et al. 1998). Garoui et al (2011) demonstrated that daily exposure, 2 weeks during gestation and 2 weeks post-delivery, to 21 mg Co/kg/day as cobalt chloride decreased liver weight in Wistar rat pups by 10%, and the dams (rats) showed an increase in hepatic enzymes ALT and AST by 44% and 27%, respectively, and a decrease in SOD, CAT, GPx and GSH by 30%, 23%, 31%, and 20%, respectively (Garoui el et al. 2011). The same study showed that, in pups exposed to cobalt in utero and through lactation, there was an increase in hepatic enzymes ALT and AST by 133 and 75%, respectively, and hepatic injury was observed with the presence of vascular congestion and infiltration of mononuclear cells by histopathology, along with a

decrease in GPx and GSH by 39% and 35%, respectively (Garoui el et al. 2011). No morphological or enzymatic changes were found in the livers of rats (Sprague-Dawley, Wistar, and albino, respectively) exposed to doses of 2.5 to 30.2 mg Co/kg/day as cobalt chloride by gavage or as cobalt chloride in the drinking water for 3 to 7 months (Domingo et al. 1984; Holly 1955; Krasovskii and Fridlyand 1971). At a dose of 18.6 mg Co/kg/day as cobalt chloride hexahydrate in utero for 2-3 days, followed by 25 days via breastmilk, and lastly 65 days orally, there was a significant decrease (21.5%) of liver weight index in ICR mice sacrificed on day 90, compared to controls (Gluhcheva et al. 2014). Mathur et al. (2011) performed a 60-day exposure to 45 mg Co/kg/day as cobalt chloride hexahydrate in Wistar rats, which showed an increase in relative liver weight by 13% along with degradation and alteration in the morphology and atrophy of liver cells (Mathur et al. 2011). This study showed alterations in liver biochemistry which 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 water as cobalt chloride hexahydrate increased LDH by 3.6-fold; hepatic enzymes ALP, AST, and ALT by 1.7, 4.5, and 1.7-fold, respectively, and total bilirubin levels by 1.9-fold, all of which contributed to an increase in DNA damage in Sprague-Dawley hepatocytes (Khalil et al. 2020). 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 administered a single dose of 68.2 mg Co/kg as cobalt fluoride or a single dose of 157.3 mg Co/kg as cobalt oxide (Speijers et al. 1982).

Dermal

No hepatic effects have been observed after dermal exposure to cobalt in animals. Acute-duration dermal exposure to 0.5-10% cobalt chloride (in DMSO) did not induce hepatic effects in mice (Bonefeld et al. 2015; Ikarashi et al. 1992b; Ikarashi et al. 1992a). No hepatic effects were observed after intermediate-duration dermal exposure to 51.70 mg Co/kg/day as dicobalt octacarbonyl (in methyl ethyl ketone) in guinea pigs (Kincaid et al. 1954). Bonefeld et al (2015) and Ikarashi et al. (1992a, 1992b) tested an unspecified strain of mouse and ICR mice, respectively, and did not observe any differences.

Other

In guinea pigs and rats exposed to a single intraperitoneal injection of 27 and 36 mg Co/kg, respectively, altered antioxidant markers like malondialdehyde increased by approximately 160% and 36%, GSH levels decreased by 12% and 25%, and increased glutathione reductase levels by 36% and 27% were seen in guinea pigs and rats, respectively (Christova et al. 2002).

2.10. RENAL

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

Inhalation

Inhalation exposure to cobalt caused changes in kidney weight and creatinine levels in rats and mice. Sixteen-day intermittent exposure to 10 and 20 mg Co/m^3 as cobalt metal caused a significant 7.5% decrease of relative left kidney weight and an 80% increase in urinary creatinine in male F344/N rats, and a 291% increase in urinary creatinine levels and 23.5% increase in relative kidney weight in female rats, and had no effect in B6C3F1 mice (NTP 2014). 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). A significant increase in the relative weight of the kidneys was reported in male rats exposed to ≥0.06 mg Co/m^3 for 13 weeks (NTP 1991). An intermediate intermittent exposure for 14 weeks to cobalt metal at 5 mg Co/m^3 increased kidney weights in female F344/N rats and increased 24% urinary creatinine in males (NTP 2014). No effects were observed upon histological examination of the kidneys in F344/N rats or B6C3F1 mice following intermittent exposure to ≤41.72 mg Co/m^3 as cobalt sulfate for 16 days, up to 6.29 mg Co/m^3 for 13 weeks, or ≤5 mg Co/m^3 for 104 to 105 weeks (NTP 1991, 1998, 2014).

Oral

Acute oral exposure to cobalt caused changes in kidney weight, altered renal morphology and physiology in rats. Domingo and Llobet (1984) showed that a single oral exposure to 161 mg Co/kg caused a 68% increase in urea and a 57% decrease in uric acid, which indicates alterations in renal function in the Sprague-Dawley rats exposed to cobalt (Domingo and Llobet 1984). Oral exposure to 33.7 mg Co/kg/day of cobalt chloride in Wistar rats induced effects on the renal system (Ajibade et al. 2017). Oral exposure to 33.7 mg Co/kg/day decreased GPx expression in the kidneys by 15%. Co exposure also increased the Nf-κB expression by 300% (Ajibade et al. 2017). Acute-duration oral exposure to 18.4 mg Co/kg/day of cobalt chloride hexahydrate in Wistar rats caused renal damage (Akinrinde et al. 2016a). This study examined histopathology of the kidney and observed a severe loss of normal morphology, loss of tubular and glomerular outlines with marked peri-tubular inflammatory cell infiltration, and vascular congestion. The authors also noted an approximate increase in urea by 33% and creatinine by 19% (Akinrinde et al. 2016b). A second acute-duration exposure study by Akinrinde et al. (2016b) also corroborated their previous study where 19 mg Co/kg/day caused inflammation in the peri- tubular and peri-vascular areas along with focal tubular necrosis (Akinrinde et al. 2016c).

Intermediate-duration oral exposure to cobalt caused changes in kidney weight, altered renal morphology, function, and physiology in rats and mice. Abdel-Daim et al. (2020) showed altered renal function in Sprague-Dawley rats after a 4 week oral exposure to 16.2 mg Co/kg/day caused an increase in urea and creatinine by 105% and 137%, respectively, and a decrease in GSH by 63% (Abdel-Daim et al. 2020). A single oral dose of 161 mg Co/kg caused alterations in renal function by increasing urea production by 68% and decreasing uric acid by 57% in Sprague-Dawley rats (Domingo et al. 1984). Morvai et al. (1993) observed a 10% decrease in relative kidney weight change after exposure to 12.4 mg Co/kg/day as cobalt chloride for 3 weeks in CFY rats (Morvai et al. 1993). After a 13-week exposure to 30.2 mg Co/kg/day as cobalt chloride in the drinking water, a 35% decrease in urine volume in Sprague-Dawley rats was seen (Domingo et al. 1984). Garoui et al (2012) observed that daily exposure, 2 weeks during gestation and 2 weeks post-delivery, to 20.26 mg Co/kg/day in water as cobalt chloride caused vascular congestion, reduction of glomerular space, and infiltration of leukocyte cells between tubules based on histology. A 15% increase in plasma creatinine, 34% decrease in urine creatinine, and a slight reduction in relative kidney weight (4%), compared to controls were also reported in Wistar rats (Garoui el et al. 2012). ICR mice were exposed to doses of 18.6 and 31 mg Co/kg/day in utero for 2-3 days followed by 25 days via breastmilk, and lastly 65 days orally (Gluhcheva et al. 2014). The lower dose group of 18.6 mg Co/kg/day showed an increase (14.3%) in kidney weight index in mice sacrificed on day 30, compared to controls. There were serious effects observed at 31 mg Co/kg/day where there was a 28.6% of kidney weight index in mice sacrificed on day 30, compared to controls (Gluhcheva et al. 2014). Histopathology of the kidney revealed peritubular and periglomerular inflammation and focal glomerular necrosis following Co exposure at 18.6 mg Co/kg/day (Gluhcheva et al. 2014). While there are no control animals in this study, renal injury, evidenced by histologic alteration of the proximal tubules, was observed in Wistar rats after a single oral exposure to 42 mg Co/kg as cobalt fluoride (Speijers et al. 1982) and after exposure to 10 to 18 mg Co/kg/day as cobalt chloride for 4 to 5 months (Holly 1955; Murdock 1959). A slightly decreased urinary output was observed in Wistar rats exposed to 19.4 mg Co/kg as cobalt sulfate, but not in Wistar rats exposed to 4.24 mg Co/kg as cobalt chloride (Singh and Junnarkar 1991).

Dermal

No renal effects have been observed after dermal exposure to cobalt in animals. Acute-duration dermal exposure to 0.5-10% cobalt chloride (in DMSO) did not induce renal effects in mice (Bonefeld et al. 2015; Ikarashi et al. 1992b; Ikarashi et al. 1992a). No renal effects were observed after intermediate dermal exposure to 2.4% Co as dicobalt octacarbonyl (in methyl ethyl ketone) in guinea pigs (Kincaid et al. 1954). Bonefeld et al (2015) and Ikarashi et al. (1992a, 1992b) tested both an unspecified strain of mouse and ICR mice, respectively, and did not observe any differences.

Other

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 increase in excretion of methemoglobin within 3 hours from the renal tissues in Wistar rats (Horiguchi et al. 2004). An intermediate exposure to 1.6 mg Co/kg/day as subcutaneous injections of cobalt nitrate for 4 weeks caused glomerulo-tubular nephrosis with degenerative changes and was toxic to the renal tubule cells in albino rats (Hanafy and Soltan 2004).

2.11. DERMAL

Inhalation

One study examined dermal effects in humans after occupational inhalation exposure to cobalt. Metal factory workers (n=71) exposed to air cobalt concentrations ranging from 0.0001 to 0.019 mg/m^3 had high self-reported prevalence of dry skin (42%) and eczema (6-7%) (Wahlqvist et al. 2020).

Oral

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

Dermal

Dermal exposure to cobalt has been associated with eczema and contact dermatitis in several case reports (Alinaghi et al. 2019, 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 over 10% cobalt by weight (Alinaghi et al. 2019). 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).

In an intermediate 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). Bonefeld et al. (2015) showed acute exposure to 10% cobalt chloride (in petrolatum) in mice (strain not specified) caused no swelling on the ears where it was applied but the ears showed inflammation with the same dose after being sensitized with a dose of 10% $CoCl_2$.

Other

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. Additionally, no studies in animals examined ocular effects following oral exposure to cobalt.

Inhalation

Inhalation studies of cobalt inhalation exposure in animals showed mixed results for ocular effects. Intermediate-duration exposure to 19 mg Co/m^3/day as cobalt sulfate heptahydrate caused chromodacryorrhea in F344/N rats and B6C3F1 mice after 16 days of intermittent exposure (NTP 1991). No histological lesions were reported in the eyes of F344/N rats or B6C3F1 mice intermittently exposed to ≤41.72 mg Co/m^3 as cobalt sulfate for 16 days, up to 6.29 mg Co/m^3 for 13 weeks, up to 0.63 mg Co/m^3 for 104 weeks (5 days/week, 6 hours/day), or up to 5 mg Co/m^3 for 105 weeks (Behl et al. 2015; NTP 1991, 1998, 2014).

Dermal

No ocular effects have been observed after dermal exposure to cobalt in animals. Acute-duration dermal exposure to 0.5-10% cobalt chloride (in DMSO) did not induce ocular effects mice (Bonefeld et al. 2015; Ikarashi et al. 1992b; Ikarashi et al. 1992a). No ocular effects were observed after intermediate-duration dermal exposure to 2.4% as dicobalt octacarbonyl (in methyl ethyl ketone) in guinea pigs (Kincaid et al. 1954). Bonefeld et al (2015) and Ikarashi et al. (1992 & 1992b) tested an unspecified strain of mouse and ICR mice, respectively, and did not observe any differences.

Other

Acute-duration exposure by a single subcutaneous injection of 45 mg Co/kg as dicobalt octacarbonyl did not cause ocular effects in guinea pigs (species not specified) (Kincaid et al. 1954).

2.13. ENDOCRINE

Inhalation

Two studies examined endocrine effects in humans after inhalation exposure to cobalt and showed alterations in thyroid function. In a study of cobalt refinery workers in Belgium, Lantin et al. (2013) found

no association between cumulative cobalt exposures and thyroid function. One volunteer out of 10 who ingested 1.0 mg Co/day for up to 90 days showed elevated TSH and decreased T4 (Tvermoes et al. 2013).

In animal studies minimal effects of acute-duration cobalt exposure were seen on the endocrine system. No effects were observed on the endocrine system in an intermediate-duration exposure in F344/N rats for up to 20 mg Co/m^3 and in B6C3F1 mice for up to 40 mg Co/m^3, exposed as cobalt metal after a 16 day intermittent exposure (NTP 2014). Intermediate-duration exposure to 4 mg Co/m^3 lowered T3 in female F344/N rats and 11 mg Co/m^3 lowered TSH in male rats after a 13-week intermittent exposure (NTP 1991).

Intermediate-duration exposure to cobalt caused no effects on endocrine function while chronic-duration exposure to cobalt altered adrenal morphology. No effects were seen after a 14-week intermittent exposure to 5 and 10 mg Co/m^3 as cobalt metal in F344/N rats and B6C3F1 mice, respectively (NTP 2014). No effects were observed after 16 days of intermittent exposure to 19 mg Co/m^3 as cobalt sulfate heptahydrate in F344/N rats (NTP 1991). Chronic intermittent exposure for 105 weeks to 1.25 and 2.5 mg Co/m^3 increased the incidence (13/50 males and 8/50 females) of bilateral benign pheochromocytoma in F344/N rats (NTP 2014). An increased incidence of medullary hyperplasia in the adrenal gland was seen in 27 female F344/N rats after chronic exposure to 1.25 mg Co/m^3 (NTP 2014). Exposure to 0.63 mg Co/m^3 as cobalt sulfate heptahydrate intermittently for 105 weeks and 5 mg Co/m^3 as cobalt metal for 105 weeks did not alter endocrine function in B6C3F1 mice (NTP 1998, 2014).

Oral

No studies were identified regarding endocrine effects in humans after oral exposure to cobalt.

A single study was identified that examined endocrine effects in animals after oral exposure to cobalt in animals. Female Parkes mice exposed to 26 mg Co/kg-day as cobalt chloride in the drinking water for 45 days showed histopathological changes to the thyroid gland i.e. low epithelial lining with degenerated nuclei (Shrivastava et al. 1996). The study also observed a time-dependent effect on the degeneration seen within the thyroid and the degradation persisted 30 and 45 days after the exposure ceased (Shrivastava et al. 1996).

Dermal

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

No endocrine effects have been observed after dermal exposure to cobalt in animals. Acute-duration dermal exposure to 0.5-10% (in DMSO) cobalt chloride did not induce endocrine effects in mice

(Bonefeld et al. 2015; Ikarashi et al. 1992b; Ikarashi et al. 1992a). No endocrine effects were observed after intermediate-duration dermal exposure to 2.4% cobalt as dicobalt octacarbonyl (in methyl ethyl ketone) in guinea pigs (Kincaid et al. 1954). Bonefeld et al. (2015) and Ikarashi et al. (1992 & 1992b) tested an unspecified strain of mouse and ICR mice, respectively, and did not observe any differences.

Other

Fifteen day intermediate 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 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 exposure to cobalt nitrate salts subcutaneously was detrimental to the alpha cells in the pancreas in guinea pigs (strain not specified) (Van Campenhout 1955). An acute-duration 10-day subcutaneous exposure in ICR mice to 0.59 mg Co/kg/day as cobalt chloride resulted in increased adipocyte 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.

2.14. IMMUNOLOGICAL

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

Inhalation

Inhalation exposure to cobalt reduced thymus weight, caused accumulation of macrophages, and resulted in necrosis. Sixteen-day intermittent exposure to 20 mg Co/m^3 decreased thymus weight in female rats by 64%, but had no effect in male F344/N rats. In the lung, at the lower exposure of 5 mg Co/m^3, the incidence of minimal to moderate alveolar histiocytic cellular infiltration (accumulation of macrophages within the alveolar spaces and septa) was seen in 5/5 male and 5/5 female B6C3F1 mice (NTP 2014).

No effects were seen in pigs (strain not specified) after a 3 month intermittent exposure to 0.1 mg Co/m^3 (Kerfoot 1974). No effects were seen in F344/N rats, but B6C3F1 mice showed hyperplasia in lymph nodes after an intermittent exposure of 11 mg Co/m^3 for 13 weeks (NTP 1991). At a higher dose of 19 mg Co/m^3 as cobalt sulfate heptahydrate intermittently for 16 days, F344/N rats showed necrosis and decreased weight of the thymus in both males and females, whereas B6C3F1 mice only showed a decrease in thymus weight (NTP 1991).

Alveolar histiocytic cellular infiltration characterized by the presence of low to moderate numbers of histiocytes (macrophages) were observed after a chronic exposure to 0.625 mg Co/m^3 for 104 weeks (NTP 2014). Macrophages accumulated around alveolar/bronchial neoplasms after chronic exposure to 5 mg Co/m^3 as cobalt metal intermittently for 105 weeks in female F344/N rats and in B6C3F1 mice (NTP 2014). Tests of immunological function, however, were not performed on the rats or mice.

Oral

Acute-duration oral exposure to cobalt altered the immune response and thymus weight in rats. Akinrinde et al. (2019) showed that 1-week oral exposure to cobalt chloride hexahydrate caused a 300% increase in 1L-1β and a 100% increase in TNF- α at 67.5 mg Co/kg/day in Wistar rats (Akinrinde and Adebiyi 2019). In an acute 7-day oral exposure study by Akinrinde et al (2016), Wistar rats exposed to 19 mg Co/kg/day as cobalt chloride showed an increase in TNF- α by 60% and a decrease in IL1- β by 25% (Akinrinde et al. 2016a). Abdel-Daim et al. (2020) revealed alterations in immune function after a 4 week intermediate oral exposure to 16.24 mg Co/kg/day as cobalt chloride hexahydrate, this caused a 1400% increase in TNF-α in Sprague-Dawley rats (Abdel-Daim et al. 2020). Atrophy of the thymus was reported in male Sprague-Dawley rats exposed to 3.8 mg Co/kg/day as cobalt chloride in the feed for 4 weeks (Chetty et al. 1979).

Intermediate-duration oral exposure to cobalt altered the immune response and spleen weight in rats and mice. After a 13 week exposure to 30.2 mg Co/kg/day as cobalt chloride in the drinking water, a 43% decrease in spleen weight in Sprague-Dawley rats was seen (Domingo et al. 1984). At doses of 18.6 and 31 mg Co/kg/day as cobalt chloride hexahydrate in utero for 2-3 days, followed by 25 days via breastmilk, and then 65 days orally, ICR mice showed a significant decrease in the spleen weight index by 43-53% (measure of relative weight) on day 60-90, compared to controls (Gluhcheva et al. 2014). A 2-fold decrease in the concentration of the total blood protein and a 1.5-fold decrease of total immunoglobulin G was observed in both male and female BALB/c mice at 56 mg Co/kg/day after an oral exposure for 5 weeks (Legostaeva et al. 2013). A 4-week exposure to 68 mg Co/kg/day in water as cobalt

chloride hexahydrate caused a 2.2-fold increase in immunoreactivity in Sprague-Dawley rats (Khalil et al. 2020).

Dermal

Dermal exposure to cobalt altered the immune response in Guinea pigs and mice. Acute 3 day dermal exposure to 0.5-5 % cobalt chloride (in DMSO) in mice caused an increase in cellular proliferation in the local lymph node assay in a dose dependent manner (Ikarashi et al. 1992b; Ikarashi et al. 1992a). Three consecutive exposures to increasing doses of cobalt chloride in Balb/c mice and Hartley guinea pigs elicited lymph node cell proliferation (Ikarashi et al. 1992b). Bonefeld et al. (2015) showed that sensitizing the mice (strain not specified) with 10% cobalt chloride (in petrolatum) and exposing them to a dermal challenge with the same dose of cobalt chloride, elicited an immune response in the mice and caused an increase in proliferation of B cells and T cells (Bonefeld et al. 2015).

2.15. NEUROLOGICAL

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

Inhalation

Inhalation exposure to cobalt caused congestion in the cranial vasculature but did not alter neurological function in rats and mice. A 16 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). Congestion in the vessels of the brain in rats and mice were observed after a 16 day intermittent exposure to 19 mg Co/m^3 as cobalt sulfate heptahydrate (NTP 1991). No effects were seen in F344/N rats and B6C3F1 mice exposed to 11 mg Co/m^3 for 13 weeks as cobalt sulfate heptahydrate (NTP 1991). A 14 week intermittent exposure to 5 and 10 mg Co/m^3 as cobalt metal did not affect neurological function in F344/N rats and B6C3F1 mice, respectively (NTP 2014). A chronic 105 week intermittent exposure to 5 mg Co/m^3 as cobalt metal did not affect neurological function in either F344/N rats or B6C3F1 mice (NTP 2014).

Oral

Acute-duration oral exposure to cobalt produces neurological effects in animals. A study by Wellman et al. (1984) that examined acute-duration oral exposure to 45 mg Co/kg/day as cobalt chloride showed increased saccharin aversion in Long-Evans rats in an operant chamber task during the training phase. These effects in Wellman et al. (1984) were also accompanied with significant food aversion, which

resulted in a decrease in body weight in the rats treated with cobalt (Wellman et al. 1984). Acute-duration oral exposure to cobalt sulfate in both Wistar rats and Swiss-Webster mice at a dose of 19.4 mg Co and 12.3 mg Co per kg body weight, respectively, caused a decrease in motor activity and impairments in reflexes, ultimately resulting in anoxic convulsions (Singh and Junnarkar 1991). The dose also had CNS depressant effects which were indicated by mild hypothermic effects, moderate reduction in spontaneous activity, muscle tone, touch response, and respiration, and increased sleeping time by 19% in rats (Singh and Junnarkar 1991).

Similar effects were seen when both Wistar rats and Swiss-Webster mice were exposed to cobalt chloride at respective doses of 4.2 and 8.9 mg Co/kg body weight (Singh and Junnarkar 1991). The doses also had CNS depressant effects which were indicated by mild hypothermic effects and increased sleeping time by 31% in rats (Singh and Junnarkar 1991). Akinrinde et al. (2019) showed that exposure to cobalt chloride hexahydrate caused deficits in performance on a battery of neurobehavioral tests along with an increase in expression by 60% of AChE activity as compared to controls at 67.5 mg Co/kg/day in Wistar rats (Akinrinde and Adebiyi 2019). Abdel-Rehman et al. (2019) conducted an intermediate-duration exposure study at a dose of 27 mg Co/kg/day in Wistar rats where there was a 251% increase in Co accumulation in the brain accompanied by decreases in neurotransmitters levels- 23% in serotonin, 26% in norepinephrine, 48% in dopamine, and 39% in GABA. The authors observed an increase in encephalopathy of the cerebral cortex. This was also accompanied with an upregulation of microglial CD68 and neural caspase-3 in the brain which indicates that there was an upregulation of inflammatory response in the brain (Abdel-Rahman Mohamed et al. 2019).

In an intermediate-duration oral exposure study where Sprague-Dawley rats were exposed to 20 mg Co/kg/day as cobalt chloride for 80 days in water, there was increased latency during memory retention testing by 342% (Bourg and Nation 1985). Intermediate-duration exposure to cobalt chloride for 7 months in water caused a significant increase in the conditioned latent reflex at 2.5 mg Co/kg/day and a pronounced neurotropic effect (disturbed conditioned reflexes and loss of memory retention) in albino rats as investigated by the motor-alimentary method (Krasovskii and Fridlyand 1971). The decrease in memory retention observed in the rats was determined by the authors to be a function of dose and exposure duration. Morvai et al. (1993) observed a 10% decrease in relative brain weight after exposure to 12.4 mg Co/kg/day as cobalt chloride for 3 weeks in CFY rats (Morvai et al. 1993). Sprague-Dawley rats exposed to 20 mg Co/kg/day via food for 69 days showed varied deficits in neurobehavioral tasks. For example, there was a slower rate of lever pressing than controls, but no change in behavioral reactivity to stress (Nation et al. 1983). Taken together, the differences in the study are driven by variability in the control group. Wistar rats exposed to 6.44 mg Co/kg/day as cobalt nitrate in the drinking

water for 30 days showed an increased sensitivity and decreased maximal response to a cholinergic agonist (Vassilev Peter P. et al. 1993). Intermediate exposure of 30 days to 3.79 mg Co/kg/day as cobalt chloride caused a 13% decrease in Na+- K+ ATPase activity in male Sprague-Dawley rats (Chetty et al. 1979).

Wistar pups were exposed in-utero and during lactation for 2 weeks each to 20.3 mg Co/kg/day as cobalt chloride which caused a decrease in the levels of AChE and BuChE in the cerebrum by 33% and 36%, respectively, and the cerebellum by 33% and 47%, respectively (Garoui et al. 2013). The authors of this study also observed a decrease in antioxidant enzymes in the brain, namely GSH and NPSH by 23% and 50% in the cerebrum and by 16% and 25% in the cerebellum, respectively, in the Wistar rat pups (Garoui et al. 2013). Purkinje cells in the cerebellum of the Wistar rat pups exposed to Co in utero and via lactation were poorly differentiated with frequent pyknotic cells and had fewer cells (Garoui et al. 2013). At a dose of 56.73 mg Co/kg/day as cobalt chloride hexahydrate in utero for 2-3 days followed by 25 days via breastmilk and lastly 35 days orally did not alter neurological function in both male and female Balb/c mice (Zaksas et al. 2013). Khalil et al. (2020) showed that a 4-week exposure to 68 mg Co/kg/day in water as cobalt chloride hexahydrate caused fatigue, lethargy, and dullness in the treated Sprague-Dawley rats (Khalil et al. 2020). No neurological effects were seen at 22.7 mg Co/kg/day as cobalt chloride when dosed for 28 days in Wistar rats (Umar et al. 2016).

Dermal

Dermal exposure to cobalt did not produce neurological effects in animals. Acute-duration dermal exposure to 0.5-10% cobalt chloride did not induce neurological effects in mice (Bonefeld et al. 2015; Ikarashi et al. 1992b; Ikarashi et al. 1992a). No neurological effects were observed after intermediate-duration dermal exposure to 52 mg Co/kg/day as dicobalt octacarbonyl in guinea pigs (Kincaid et al. 1954). Bonefeld et al (2015) and Ikarashi et al. (1992 & 1992b) tested an unspecified strain of mouse and ICR mice, respectively, and did not observe any differences.

Other

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 Co is potentially ototoxic (Lee et al. 2016). 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). 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).

2.16. REPRODUCTIVE

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

Inhalation

Inhalation exposure to cobalt produces reproductive effects in some animals studied. Intermittent exposure to 10 and 40 mg Co/m^3 as cobalt metal in male and female rats and mice, respectively, for 5 hr/day, 6 days/week, for 16 days did not affect reproductive function (NTP 2014). Another 16 day intermittent exposure to 19 mg Co/m^3 as cobalt sulfate heptahydrate did not alter reproductive function in female F344/N rats and B6C3F1 mice, but in male rats it caused testicular atrophy along with a decrease in number of cells in the seminiferous tubules and atypical germinal epithelial cells in the epididymal ducts (NTP 1991). Intermediate-duration exposure to 11 mg Co/m^3 as cobalt sulfate heptahydrate intermittently for 13 weeks did not affect reproductive function in male and female F344/N rats, but in female B6C3F1 mice the same exposure increased the length of the estrous cycle by 19%; sperm motility decreased by 79% in male mice at 1.1 mg Co/m^3 (NTP 1991). Male F344/N rats and B6C3F1 mice showed decreased reproductive function after intermittent exposure to 2.5 mg Co/m^3 for 14 weeks; however, only female mice showed longer estrous cycles (NTP 2014). Bucher et al. (1990) demonstrated that an intermediate-duration inhalation exposure of cobalt sulfate heptahydrate caused reproductive deficits in male B6C3F1 mice. There was a marked decrease in sperm motility in male B6C3F1 mice at 3 mg Co/m^3 and increased numbers of abnormal sperm were observed at 3, 10, and 30 mg Co/m^3 after 13 weeks of intermittent exposure (Bucher et al. 1990; NTP 1991). Intermittent exposure at a higher dose of 30 mg Co/m^3 for 13 weeks was associated with decreased epididymal and testis weights (Bucher et al. 1990; NTP 1991). Estrous cycle length increased to 5 days in female B6C3F1 mice exposed to 30 mg Co/m^3 (Bucher et al. 1990). Chronic intermittent 105-week exposure to cobalt-containing aerosols caused deficits in reproductive functioning and resulted in effects on reproductive end points. A chronic intermittent 105 week exposure to 2.5 and 5 mg Co/m^3 was associated with decreased reproductive function in both male F344/N rats and B6C3F1 mice, respectively, but not in the female rats or mice (NTP 2014).

Oral

Oral exposure to cobalt produces reproductive effects in animals. An acute-duration 5 day exposure to 7 mg Co/kg/day as cobalt chloride increased abnormal sperm in male Swiss mice by 126% (Hassan et al. 2006). Intermediate-duration 13 week oral exposure to 24.6 mg Co/kg/day as cobalt chloride in water induced reproductive effects in male CD-1 mice which included an unspecified increase in the number of Leydig cells, degeneration in the peritubular area of the seminiferous tubules, and increased folding in the germinal epithelium of seminiferous tubules of the testicles were accompanied with changes in epithelial cell morphology (Anderson et al. 1992). In a follow up study by Anderson et al. (1993) with intermediate-duration exposure for 13 weeks to 43.4 mg Co/kg/day in water, irreversible testicular degeneration occurred demonstrated by damage to seminiferous tubules and hypercellularity of the interstitial areas in CD-1 mice (Anderson et al. 1993).

Mollenhauer et al. (1985) demonstrated that an exposure to cobalt metal through food at 20 mg Co/kg/day for 14 weeks 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 (Mollenhauer et al. 1985). Testicular atrophy was observed at a dose of 20 mg Co/kg/day in Sprague-Dawley rats after exposure for nearly 2 months via food (Nation et al. 1983). A 30 day exposure to 6.4 mg/Co/kg/day as cobalt nitrate in water caused a 275% increase in sympathetically-induced contractility of the vas deferens (Mutafova-Yambolieva et al. 1994). Testicular degeneration and atrophy which includes alteration in seminiferous tubules (27-90%), drop in sperm reserves (57%), and a marked decrease in testicular weight (26%) have been reported in Sprague-Dawley rats exposed to 20-30 mg Co/kg/day as cobalt chloride for 13-14 weeks in food or drinking water (Corrier et al. 1985; Domingo et al. 1984). Elbetieha et al. (2008) demonstrated that a 12 week exposure to 6.36 mg Co/kg/day as cobalt chloride hexahydrate in drinking water induced a significant 16.8% increase of relative preputial gland weight, a 13.3% decrease in sperm count, and decreased male fertility compared to controls in Swiss mice (Elbetieha et al. 2008). When males Swiss mice exposed to 6.3 mg Co/kg/day were mated with females, the number of implantations was reduced in those females and the number of viable fetuses also decreased significantly (Elbetieha et al. 2008). Additionally, the number of resorptions and the number of mice with resorptions were increased in females mated with the exposed males at all three concentrations of cobalt chloride (Elbetieha et al. 2008). A significant 16.8% increase of relative preputial gland weight, 13.3% decrease in sperm count, and decreased male fertility compared to controls was observed in a study where ICR mice were orally exposed to 6.36 mg Co/kg/day in water for 12 weeks (Gluhcheva et al. 2020). Pedigo et al. (1988) observed that a dose of 6 mg Co/kg/day as cobalt chloride hexahydrate in

drinking water decreased testicular weight (expressed as % of body weight) by 14% with an 11% decrease in sperm concentration, and an 80% increase in serum testosterone in B6C3F1 mice (Pedigo et al. 1988). In the same study, a dose of 15 mg Co/kg/day for 13 weeks caused a decrease in testicular weights as a % of body weight by 14%, 21%, 57%, and 71% at 7, 9, 11, and 13 weeks, respectively, a reduction in sperm concentration to 81.3% after 9 weeks of treatment, and an 82% decrease in sperm motility after 11 weeks of exposure (Pedigo et al. 1988). A subsequent 10 week study by Pedigo et al. (1993) where male B6C3F1 mice were exposed to 15 mg Co/kg/day showed a reduction in pregnancy in females by 57% when mated with males treated with Co, a 28% decrease in implantation of embryos when mated with Co exposed males, a 458% increase in preimplantation losses in pregnant females mated with Co- exposed males, a decrease in sperm concentration to 15.3%, and a decrease in sperm motility to 18.3% (Pedigo and Vernon 1993).

No reproductive function was altered in in male Balb/c mice after exposure to 56 mg Co/kg/day for 18 days (Madzharova et al. 2014).

Dermal

No studies were identified regarding reproductive effects in humans after dermal exposure to cobalt for any duration.

Acute-duration dermal exposure to 0.5-10% cobalt chloride did not induce reproductive effects in mice (Bonefeld et al. 2015; Ikarashi et al. 1992b; Ikarashi et al. 1992a). No reproductive effects were observed after intermediate-duration dermal exposure to 52 mg Co/kg/day as dicobalt octacarbonyl in guinea pigs (Kincaid et al. 1954). Bonefeld et al (2015) and Ikarashi et al. (1992 & 1992b) tested an unspecified strain of mouse and ICR mice, respectively, and did not observe any differences.

Other

No studies were identified regarding reproductive effects in animals after intermediate- and chronic-duration parenteral 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 following inhalation, oral, or dermal exposure to cobalt.

Inhalation

No studies were identified regarding developmental effects in animals after inhalation exposure to cobalt for any duration.

Oral

Oral exposure to cobalt produces developmental effects in animals. Acute-duration oral exposure to 81 mg Co/kg/day as cobalt chloride for 5 days during gestation days 8–12 was reported to have no effect on fetal growth or mortality in the pups, but did decrease maternal weight gain by 32% in ICR mice (Seidenberg et al. 1986). Oral exposure of female Sprague-Dawley rats to cobalt chloride at 5.4 or 21.8 mg Co/kg/day from gestation day 14 through lactation day 21 resulted in stunted growth and decreased survival, respectively, of newborn pups (Domingo et al. 1985a). Maternal toxicity was observed at these same doses making it unclear if the observed findings were due to a potential indirect effect of maternal toxicity or a direct effect of cobalt on the fetus (Domingo et al. 1985a).

In a study without a control group, no effects on fetal growth or survival were found following exposure to 24.8 mg Co/kg/day as cobalt chloride during gestation days 6–15 in Sprague-Dawley rats (Paternian and Domingo 1988).

Dermal

No developmental effects were observed after dermal exposure to cobalt. Acute-duration dermal exposure to 0.5-10% cobalt chloride did not induce developmental effects in mice (Bonefeld et al. 2015; Ikarashi et al. 1992b; Ikarashi et al. 1992a). No developmental effects were observed after intermediate-duration dermal exposure to 2.4% cobalt as dicobalt octacarbonyl in guinea pigs (Kincaid et al. 1954). Bonefeld et al (2015) and Ikarashi et al. (1992 & 1992b) tested an unspecified strain of mouse and ICR mice, respectively, and did not observe any differences.

Other

Acute-duration exposure by subcutaneous injection of 45 mg Co/kg/day as dicobalt octacarbonyl did not cause developmental effects in guinea pigs (Kincaid et al. 1954).

2.18. OTHER NONCANCER

No studies were identified that examined other noncancer effects in humans following inhalation, oral, or dermal exposure to cobalt.

Inhalation

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

Oral

Oral exposure to cobalt produced other noncancerous effects in animals. 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 30.2 mg Co/kg/day as cobalt chloride hexahydrate for 13 weeks decreased water intake in Sprague-Dawley rats (Domingo et al. 1984). Garoui et al (2012) observed decreased water and food intake in Wistar rats exposed to 20.26-21 mg Co/kg/day as cobalt chloride (Garoui el et al. 2011; Garoui el et al. 2012).

Dermal

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

2.19. CANCER

No studies were identified that reported significant cancerous effects in humans following inhalation, oral, or dermal exposure to cobalt.

Inhalation

EPA has not classified cobalt for carcinogenicity. IARC has classified cobalt as *Group 2B- possibly carcinogenic to humans.* 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 the US cohort which included eight sites, there was no increased lung cancer mortality risk or trends in SMRs from long term exposure to cobalt or from the other metals studied (Marsh et al. 2017a). Standardized mortality ratios were not statistically higher by sex and while two plants observed excess lung cancer mortality, this was not statistically significant (Marsh et al. 2017a). Study authors state that the lung cancer risks were higher in females than in males in

Germany, the US, 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 nor cumulative exposure to cobalt impacted lung cancer mortality risk. In other studies conducted at hard metal production factories in the United Kingdom and Europe, the study authors found no significant exposure-response relationship between cancer and inhalation exposure to cobalt (McElvenny et al. 2017; Morfeld et al. 2017; Sauni et al. 2017; Wallner et al. 2017; Westberg et al. 2017a; Westberg et al. 2017b).

Inhalation exposure to cobalt metal produces cancerous effects in animals. An intermediate-duration 13 week intermittent exposure to 0.06 mg Co/m^3 caused squamous metaplasia in the larynx in F344/N rats and B6C3F1 mice (Bucher et al. 1990; NTP 1991). At concentrations ≥0.06 mg Co/m^3 for 13 weeks, rats and mice also developed squamous metaplasia of the larynx (Bucher et al. 1990; NTP 1991). Chronic intermittent exposure to cobalt sulfate heptahydrate for 105 weeks at 0.06 mg Co/m^3 caused alveolar/bronchiolar neoplasms along with granulomatous inflammation and metaplasia in the nose and epiglottis in F344/N rats and B6C3F1 mice of both sexes (NTP 1998). Chronic intermittent exposure at 0.06 mg Co/m^3 for 105 weeks also caused hyperplasia in the adrenal medulla in F344/N rats of both sexes (NTP 1998). Increased incidence of alveolar/bronchiolar neoplasms was noted following lifetime exposure of male rats to 0.63 mg Co/m^3 and in female F344/N rats exposed to 0.21 mg Co/m^3 (Bucher et al. 1999; NTP 1998). Statistical analysis revealed that tumors occurred with significantly positive trends in both sexes of rats (NTP 1998). Similarly, B6C3F1 mice of both sexes exposed to 0.63 mg Co/m^3 showed an increase in alveolar/bronchiolar neoplasms, again with lung tumors occurring with significantly positive trends (NTP 1998). Another chronic study with intermittent exposure (105 weeks) to cobalt metal caused a significantly increased incidence of mononuclear cell leukemia in female rats at 1.25 mg Co/m^3 compared to controls (adjusted incidence rate: 62.4% in exposed, 35.7% in controls) (Behl et al. 2015; NTP 2014). In B6C3F1 mice of both sexes, exposure at 1.25 mg Co/m^3 for 105 weeks intermittently increased the rate of alveolar/bronchiolar carcinoma in exposed mice compared to controls (Adjusted rates in exposed: 79.4% in males, 53.8% in females; adjusted rates in controls: 22.8% in males, 11.3% in females) (Behl et al. 2015; Hong et al. 2015; NTP 2014).

Oral

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

Dermal

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

2.20. GENOTOXICITY

No studies were identified regarding genotoxic effects in humans following oral or dermal exposure to cobalt. No studies were identified regarding genotoxic effects in animals following inhalation and dermal exposure to cobalt.

Gennart et al. (1993) examined a cohort of 26 male workers who had been occupationally-exposed to cobalt and observed that analysis of variance on sister-chromatid exchange rank values revealed that exposure status (exposed vs. controls) had statistically significant effects (Gennart et al. 1993). De Boeck et al. (2000) reported no significant change in the comet assay on lymphocytes from nonsmoking workers who had been occupationally exposed to cobalt (De Boeck et al. 2000). The genotoxic effects of Co showed that metallic Co induced a statistically significant concentration dependent increase in micronucleated binucleates quantified by the comet assay in blood samples from two donors (De Boeck et al. 2003). In Hengsteler et al. (2003) blood samples from 78 subjects (62 men and 16 women) exposed to cobalt in occupational settings were collected. The concentration of cobalt in air was quantified 6 h immediately before blood samples were collected for DNA –SSB analysis and the air concentration of cobalt of the work areas of the 78 individuals examined in this study varied widely ranging from 0 to 10 µg /m3 for cobalt (Hengstler et al. 2003). Hengstler et al. (2003) showed a correlation between increased air concentration of cobalt and levels of single stranded DNA binding protein (DNA- SSB). Mateuca et al. (2005) collected blood from 21 cobalt exposed and 26 matched controls to examine the genotoxic effects of cobalt exposure on lymphocytes by using a Comet assay. The workers who were exposed to cobalt showed chromosomal rearrangements resulting from chromosome loss or acentric fragments assessed as micronucleated mononucleates and binucleates (Mateuca et al. 2005). Welders exposed to cobalt in occupational settings showed a significant increase of OTMχ^2 distribution along with a significant induction of DNA strand breaks (Iarmarcovai et al. 2005). The micronucleus assay showed that the exposed welders had higher frequency of chromosomal damage, in particular, the XRCC1 variant allele coding Gln amino acid at position 399 was found to be associated with a higher number of DNA breaks as revealed by the comet assay (Iarmarcovai et al. 2005). Uboldi et al (2016) showed a dose- dependent increase in genotoxic effects caused by $CoCl_2$ in human bronchial epithelial cells. The underlying cause for the observed genotoxic effects in the bronchial epithelial cells is oxidative DNA damage as evidenced by modification in FPG (DNA-formamidopyrimidine glycosylase) and hOGG1 enzymes (Uboldi et al. 2016). Xi et al (2016) exposed human bronchial epithelial cells to cobalt oxide and cobalt chloride and

observed a dose dependent increase in cytotoxicity and genotoxicity. Cobalt oxide and cobalt chloride induced chromosome damage in human bronchial epithelial cells where the greatest aberration observed for both were chromatid lesions (Xie et al. 2016). Exposing lung fibroblast cells to cobalt oxide and cobalt chloride hexahydrate resulted in increased cytotoxic effects by cobalt chloride hexahydrate, but they both had similar effects on genotoxicity in a study by Smith et al. (2014). Increased chromosome damage, i.e. increased percent of metaphases and total aberrations in 100 metaphases, was observed when the cells were exposed to cobalt chloride hexahydrate which is more soluble than cobalt oxide (Smith et al. 2014). Cobalt chloride induced DNA fragmentation in both a dose- and time- dependent manner in human submandibular gland cells (Akita et al. 2007). γH2AX is an early and sensitive marker of genotoxicity in Hep G2 (liver cells) and LS-174T (colon cells) cell lines that did not show any changes upon exposure to cobalt chloride and cobalt oxide (Kopp et al. 2018). An acute 1-week oral exposure to cobalt chloride in albino rats showed a dose- dependent increase in frequency of micronucleated polychromatic erythrocytes. This oral administration led to hepatic damage through induction of oxidative stress, inflammation, and apoptosis (Awoyemi et al. 2017). Genotoxic effects of *in vivo* exposure to cobalt are presented in Table 2-7.

Single oral exposures of male Swiss mice to 0, 4.96, 9.92, or 19.8 mg Co/kg as cobalt chloride resulted in significantly increased percentages of both chromosomal breaks and chromosomal aberrations in bone marrow cells with significant linear trends toward increased aberrations with increased exposure (Palit et al. 1991a, 1991b; Palit et al. 1991c; Palit et al. 1991d). Thirty hours following a single intraperitoneal injection of cobalt(II) chloride in BALB/c mice, an increase in micronucleus formation was seen at doses of 12.4 or 22.3 mg Co/kg (as cobalt chloride), but not at 6.19 mg/kg (Suzuki et al. 1993). Single intraperitoneal injection of 50 mg Co/kg (as cobalt chloride) resulted in significantly increased micronucleus formation at 24 hours post-injection, but not at 12, 48, 72, or 96 hours (Suzuki et al. 1993). Two or 10 days following intraperitoneal injection of male and female F344 rats with 3 or 6 mg Co/kg, increased levels of oxidatively-damaged DNA bases were noted in the liver, kidney, and to a lesser extent, the lung (Kasprzak et al. 1994). *Drosophila melanogaster* exposed to cobalt chloride showed mutagenic activity resulting in malformed wings (Kaya et al. 2002). Oral exposure to cobalt compounds studied by Kirkland et al. (2015) did not elicit any chromosomal aberrations in the bone marrow or sperm.

Cobalt was found to be non-mutagenic in bacteria (*Salmonella typhimurium*, *Escherichia coli*) and yeast (Arlauskas et al. 1985; Fukunaga et al. 1982; Kanematsu et al. 1980; Kharab and Singh 1985; Ogawa et al. 1986; Singh 1983; Tso and Fung 1981). A very weak mutagenic response was found with *Bacillus subtilis* (Kanematsu et al. 1980). A mutagenic response to cobalt was found when compounds with a valence state of III were tested in *S. typhimurium* and *E. coli* (Schultz et al. 1982). The authors suggested

that this may be due to the formation of cobalt(III) complexes that are inert to ligand substitution, allowing optimal interaction of cobalt with genetic material (Schultz et al. 1982). Other studies have shown cobalt to be a co-mutagen in combination with 4-substituted pyridines in *S. typhimurium* (Ogawa et al. 1988). It also has been reported that cobalt acts as an anti-mutagen in bacterial (*S. typhimurium*, *B. subtilis*, *E. coli*) and yeast test systems. (*Saccharomyces cerevisiae*) (Inoue et al. 1981; Kada et al. 1986; Kuroda and Inoue 1988). A possible explanation is that cobalt acts by correcting the error-proneness of deoxyribonucleic acid (DNA) replicating enzymes by improving their performance during DNA synthesis (Inoue et al. 1981; Kada et al. 1986; Kuroda and Inoue 1988). However, cobalt has also been shown to increase the frequency of genetic conversions in *S. cerevisiae* (Kharab and Singh 1985; Singh 1983). The reasons for this apparent dichotomy in yeast cells is not known.

In contrast to the results seen in bacteria, cobalt compounds were generally found to be genotoxic or mutagenic in mammalian assay systems. Exposure to cobalt compounds (metal, salts, or hard metal) can produce clastogenic effects in mammalian cells, including human lymphocytes (Anard et al. 1997; Hamilton-Koch et al. 1986; Painter and Howard 1982); transformation in hamster cells (Costa et al. 1982); sister chromatid exchanges in human lymphocytes (Andersen 1983); and micronucleus formation in rodent bone marrow cells (Suzuki et al. 1993) and human lymphocytes (Capomazza and Botta 1991; Olivero et al. 1995; Van Goethem et al. 1997). Hard metal is generally more genotoxic in *in vitro* tests than other cobalt compounds. Cobalt ions are thought to inhibit DNA repair in mammalian cells by interaction with zinc-finger proteins involved in DNA excision repair (De Boeck et al. 1998; Hartwig et al. 1991; Kasten et al. 1997; Sarkar 1995). Genotoxic effects of *in vitro* exposure to cobalt are represented in Table 2-8.

Table 2-7. Genotoxicity of Cobalt In Vivo

Species (test system)	End Point	Results	Reference
Drosophila Melanogaster (wing spot test)	Clastogenicity resulting in malformed wings	+	Kaya et al. 2002
Sprague-Dawley rats	Clastogenicity	+	Kirkland et al. 2015
Sprague-Dawley rats	Chromosomal aberration	+	Kirkland et al. 2015
Albino rats	DNA damage	+	Awoyemi et al. 2017
Human peripheral blood mononucleated cells	DNA damage	+	DeBoeck et al. 2003
Human mononuclear blood cells	DNA damage	+	Hengstler et al. 2003
Human lymphocytes	DNA damage	+	Mateuca et al. 2005
Human lymphocytes	Breakage of DNA strands	+	Iarmarcovai et al. 2005

– = negative result; + = positive result; (+) = weakly positive result; DNA = deoxyribonucleic acid;

Table 2-8. Genotoxicity of Cobalt In Vitro

Species (test system)	End Point	Results		Reference
		With Activation	Without Activation	
Prokaryotic organisms				
Salmonella typhimurium (plate incorporation)	Gene mutations	No data	-	Tso and Fung 1981
Salmonella typhimurium (plate incorporation)	Gene mutations	No data	-	Arlauskas et al. 1985
Salmonella typhimurium (plate incorporation)	Gene mutations	No data	-	Ogawa et al. 1986
Salmonella typhimurium (plate incorporation)	Gene mutations	No data	+	Schultz et al. 1982
Salmonella typhimurium (plate incorporation)	Gene mutations	-	-	Kirkland et al. 2015
Salmonella typhimurium (pre incubation)	Gene mutations	-	-	Kirkland et al. 2015
Bacillus subtilis (rec assay)	Gene mutations	No data	(+)	Kanematsu et al. 1980
Escherichia coli (reversion assay)	DNA damage	No data	-	Kanematsu et al. 1980
E. coli (repair assay)	Reversion	No data	+	Schultz et al. 1982
Eukaryotic organisms				
Saccharomyces cerevisiae (plate assay)	Reversion	No data	-	Kharab and Singh 1985
S. cerevisiae (plate assay)	Reversion	No data	-	Fukunaga et al. 1982
S. cerevisiae (plate assay)	Conversion	No data	-	Singh 1983
S. cerevisiae (plate assay)	Conversion	No data	+	Kharab and Singh 1985
S. cerevisiae (plate assay)	Conversion	No data	+	Fukunaga et al. 1982
S. cerevisiae (plate assay)	Conversion	No data	+	Singh 1983
Mammalian cells				
Hamster ovary cells	Clastogenic effects	No data	+	Hamilton-Koch et al. 1986
Hamster embryo cells	Transformation	No data	+	Costa et al. 1982
Mouse lymphoma cells	Clastogenic effects	+	+	Kirkland et al. 2015
Human lymphocytes	Sister chromatid exchange	No data	+	Andersen 1983
Human lymphocytes	Gene mutations	-	-	Kirkland et al. 2015

Table 2-8. Genotoxicity of Cobalt In Vitro

Species (test system)	End Point	Results		Reference
		With Activation	Without Activation	
Human lymphocytes	Chromosomal aberration	+	+	Kirkland et al. 2015
Human HeLa cells	Inhibition of DNA synthesis	No data	+	Painter and Howards 1982
Human diploid fibroblasts	DNA damage	No data	+	Hamilton-Koch et al. 1986
Human bronchial epithelial cells	DNA damage	No data	+	Uboldi et al 2016*
Human bronchial epithelial cells	Chromosome aberration and chromatid lesions	No data	+	Xie et al. 2016
Human lung fibroblast cells	Induces cell cycle arrests and absence of metaphase	No data	+	Smith et al 2014
Human sub mandibular gland ductal cell line	DNA fragmentation	No data	+	Akita et al. 2007
Human hepatoblastoma cells	DNA damage	No data	-	Kopp et al. 2018
Human epithelial colorectal adenocarcinoma cells	DNA damage	No data	-	Kopp et al. 2018

– = negative result; + = positive result; (+) = weakly positive result; DNA = deoxyribonucleic acid; *= hOGG1- 8-Oxoguanine glycosylase also known as OGG1 is a DNA glycosylase enzyme that, in humans, is encoded by the OGG1 gene. It is involved in base excision repair. It is found in bacterial, archaeal, and eukaryotic species; FPG-DNA-formamidopyrimidine glycosylase is a base excision repair enzyme which recognizes and removes a wide range of oxidized purines from correspondingly damaged DNA

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 thorough the ion channels or via endocytosis of poorly soluble cobalt compounds. The poorly soluble cobalt compounds are then solubilized in the acidic environment and then 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.* 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).

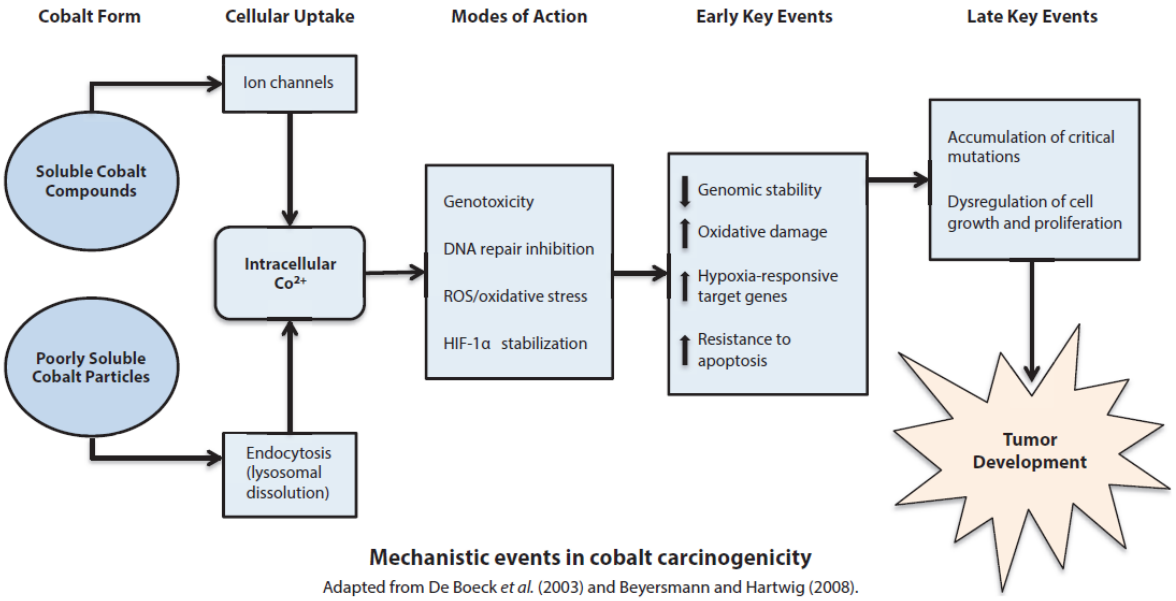
Calcium influx in cells is known to be altered by soluble cobalt when it blocks the inorganic calcium channels in cells harvested from rodent models (Henquin and Lambert 1975; Moger 1983; Yamatani et al.

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 $CoCl_2$ on postnatal day 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 the hypoxia inducible factor α (HIF- α) and degrading it, thus, exposure to cobalt can often mimic hypoxic conditions in *in vitro* models (Yuan et al. 2003). The testicular degeneration seen as a result of cobalt exposure is often 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). Cobalt ions are also responsible for stabilizing HIF-1α and HIF-2α and thus increasing the production of red blood cells, and increasing hemoglobin concentrations in human male participants (Hoffmeister et al. 2018).

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, glutathione peroxidase, 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).

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



Source: Beyersmann and Hartwig 2008; De Boeck et al. 2003; NIEHS 2016

2.22. COBALT NANOPARTICLES

The following section provides a brief overview of cobalt nanoparticle 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 cobalt nanoparticles (CoNPs) were identified. Increased levels of Co ions in serum and testis were observed in male rats after *in vivo* exposure of 500 µg/kg bodyweight 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; Alinovi et al. 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 reactive oxygen species 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 caused an increase in relative brain, kidney, and liver weights, along with increases in erythrocyte and hemoglobin counts (Ali 2019). 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 3 rats in the treatment group (Brown et al. 2018). Transgenic mice (gpt delta) were intratracheally instilled with 50 μg CoNPs per mouse and examined on day 1, 3, 7, and 28 after exposure in a study by Wan et al. (2017). This study identified toxic effects in the respiratory system that included lung inflammation, oxidative stress, injury, and cell proliferation, which further resulted in DNA damage and DNA mutation (Wan et al. 2017). In Hansen et al. (2006), Sprague-Dawley rats underwent subcutaneous implantation of CoNPs and 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. Exposure to CoNPs from inhalation, dermal, and oral routes, as well as via prosthetics and therapeutics needs 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.