3. Studies of Cancer in Experimental Animals

3.1 Inhalation exposure

3.1.1 *Mouse*

In a study undertaken by the National Toxicology Program (2002), groups of 50 male and 50 female B6C3F$_1$ mice, 6–7 weeks of age, were exposed to vanadium pentoxide particulate (light orange, crystalline solid; purity, ≈ 99%; MMAD, 1.2–1.3 μm; GSD, 1.9 μm) at concentrations of 0, 1, 2 or 4 mg/m^3 by inhalation for 6 h per day on 5 days per week for 104 weeks. Survival was significantly decreased in males exposed to 4 mg/m^3 compared with chamber controls (survival rates: 39/50 (control), 33/50 (low concentration), 36/50 (mid concentration) or 27/50 (high concentration) in males and 38/50, 32/50 30/50 or 32/50 in females, respectively; mean survival times, 710, 692, 704 or 668 days in males and 692, 655, 653 or 688 days in females, respectively). Mean body weights were decreased in females exposed to ≥ 1 mg/m^3 and in males exposed to ≥ 2 mg/m^3. Exposure to vanadium pentoxide caused an increase in the incidence of alveolar/bronchiolar neoplasms, but did not cause an increased incidence of neoplasms in other tissues. The incidence of neoplasms and non-neoplastic lesions of the respiratory system

Table 3. Occupational exposure limits and guidelines for vanadium (as V_2O_5 unless otherwise specified)

Country or region	Concentration (mg/m^3)	Classification[a]	Interpretation[b]
Australia	0.05 (respirable dust and fume)		TWA
Belgium	0.5		TWA
Canada			
Alberta	0.05 (respirable dust and fume)		TWA
	0.15 (respirable dust and fume)		STEL
Quebec	0.05 (respirable dust and fume)		TWA
China	0.05 (dust and fume, as V)		TWA
	0.15 (dust and fume, as V)		STEL
Finland	0.05 (dust, as V)		TWA
	0.5 (fume, as V)		TWA
France	0.05 (respirable dust and fume)		TWA
Germany	0.05 (respirable fraction)		TWA (MAC)
	0.05 (respirable fraction)		STEL
Hong Kong SAR	0.05 (respirable dust and fume)	A4	TWA
Ireland	0.04 (respirable dust, as V)		TWA
	0.05 (fume, as V)		TWA
	0.5 (total inhalable dust, as V)		TWA
Japan	0.1 (fume)		TWA
	0.5 (dust)		TWA (JSOH)
Malaysia	0.05		TWA
Mexico	0.5 (dust and fume)	A4	TWA
Netherlands	0.01		TWA
	0.03		STEL
New Zealand	0.05 (respirable dust and fume)		TWA
Poland	0.05 (dust and fume)		TWA
	0.1 (fume); 0.5 (dust)		STEL
Russian Federation	0.1 (fume)		MAC
	0.5 (dust)		NG
South Africa	0.05 (respirable dust and fume)		TWA (DOL-RL)
	0.5 (total inhalable dust)		TWA
Spain	0.05 (respirable dust and fume)		TWA
Sweden	0.2 (total dust, as V)		TWA
	0.05 (respirable dust, as V)		Ceiling
Switzerland	0.05		TWA
	0.05		STEL
United Kingdom	0.05		TWA (MEL)

Table 3 (contd)

Country or region	Concentration (mg/m^3)	Classification[a]	Interpretation[b]
USA[c]			
ACGIH	0.05 (respirable dust and fume)	A4	TWA (TLV)
NIOSH	0.05 (total dust and fume, as V)		Ceiling (REL)
OSHA	0.1 (fume); 0.5 (respirable dust)		Ceiling (PEL)

From Sokolov (1981); INRS (1999); Työsuojelusäädöksiä (2002); ACGIH Worldwide® (2003); Suva (2003)

[a] A4, not classifiable as a human carcinogen; the absence of any classification does not necessarily mean that vanadium pentoxide has been evaluated by individual organizations as non-carcinogenic to humans.

[b] TWA, time-weighted average; STEL, short-term exposure limit; MAC, maximum allowed concentration; JSOH, Japanese Society for Occupational Health; NG, not given; DOL-RL, Department of Labour-Recommended Limit; MEL, maximum exposure limit; TLV, threshold limit value; REL, recommended exposure limit; PEL, permissible exposure limit.

are reported in Table 4. Alveolar/bronchiolar adenomas were typical of those that occur spontaneously in mice. Carcinomas had one or more of the following histological features; heterogeneous growth pattern, cellular pleomorphism and/or atypia, and local invasion or metastasis. A number of exposed males and females had multiple alveolar/bronchiolar neoplasms. This last finding is an uncommon response in mice and, in some cases, it was difficult to distinguish between multiplicity and metastases from other lung neoplasms. Mice are generally not considered to respond to particulate exposure by the development of lung neoplasms, even at high concentrations. There was a significantly-increased incidence of alveolar epithelial hyperplasia and bronchiolar epithelial hyperplasia in the lungs of exposed male and female mice. The hyperplasia was essentially a diffuse change with proliferation of epithelium in the distal terminal bronchioles and the immediately associated alveolar ducts and alveoli. The hyperplasia of the alveolar epithelium was pronounced and increased in severity with increasing exposure concentration, while the hyperplasia of the distal bronchioles was minimal to mild. Histiocytic infiltration occurred primarily within alveoli in close proximity to alveolar/bronchiolar neoplasms, particularly carcinomas (National Toxicology Program, 2002; Ress *et al*., 2003).

3.1.2 *Rat*

In a study undertaken by the National Toxicology Program (2002), groups of 50 male and 50 female Fischer 344/N rats, 6–7 weeks of age, were exposed to vanadium pentoxide particulate (light orange, crystalline solid; purity, ≈ 99%; MMAD, 1.2–1.3 μm; GSD, 1.9 μm) at concentrations of 0, 0.5, 1 or 2 mg/m^3 by inhalation for 6 h per day on 5 days per week for 104 weeks. No adverse effects on survival were observed in treated males or females compared with chamber controls (survival rates: 20/50 (control), 29/50 (low

Table 4. Incidence of neoplasms and non-neoplastic lesions of the respiratory system and bronchial lymph nodes in mice in a 2-year inhalation study of vanadium pentoxide

	No. of mice exposed to vanadium pentoxide at concentrations (mg/m^3) of			
	0 (chamber control)	1	2	4
Males				
Lung				
Total no. examined	50	50	50	50
No. with:				
Alveolar epithelium, hyperplasia	3 (3.0)[a]	41[b] (2.2)	49[b] (3.3)	50[b] (3.9)
Bronchiole epithelium, hyperplasia	0	15[b] (1.0)	37[b] (1.1)	46[b] (1.7)
Inflammation, chronic	6 (1.5)	42[b] (1.5)	45[b] (1.6)	47[b] (2.0)
Alveolus, infiltration cellular, histiocyte	10 (2.4)	36[b] (2.4)	45[b] (2.6)	49[b] (3.0)
Interstitial fibrosis	1 (1.0)	6 (1.7)	9[b] (1.2)	12[b] (1.7)
Alveolar/bronchiolar adenoma, multiple	1	1	11[b]	5
Alveolar/bronchiolar adenoma (includes multiple)	13	16	26[b]	15
Alveolar/bronchiolar carcinoma, multiple	1	10[b]	16[b]	13[b]
Alveolar/bronchiolar carcinoma (includes multiple)	12	29[b]	30[b]	35[b]
Alveolar/bronchiolar adenoma or carcinoma	22	42[b]	43[b]	43[b]
Larynx				
Total no. examined	49	50	48	50
No. with:				
Respiratory epithelium, epiglottis, metaplasia, squamous	2 (1.0)	45[b] (1.0)	41[b] (1.0)	41[b] (1.0)
Nose				
Total no. examined	50	50	50	50
No. witth:				
Inflammation, suppurative	16 (1.3)	11 (1.4)	32[b] (1.2)	23[c] (1.3)
Olfactory epithelium, atrophy	6 (1.0)	7 (1.6)	9 (1.3)	12 (1.2)
Olfactory epithelium, degeneration, hyaline	1 (1.0)	7[c] (1.0)	23[b] (1.1)	30[b] (1.2)
Respiratory epithelium, degeneration, hyaline	8 (1.1)	22[b] (1.0)	38[b] (1.2)	41[b] (1.4)
Respiratory epithelium, metaplasia, squamous	0	6[c] (1.2)	6[c] (1.3)	2 (1.5)
Lymph node, bronchial				
Total no. examined	40	38	36	40
No. with:				
Hyperplasia	7 (2.1)	7 (2.4)	12 (2.1)	13 (2.2)

Table 4 (contd)

	No. of mice exposed to vanadium pentoxide at concentrations (mg/m^3) of			
	0 (chamber control)	1	2	4
Females				
Lung				
Total no. examined	50	50	50	50
No. with:				
Alveolar epithelium, hyperplasia	0	31[b] (1.6)	38[b] (2.0)	50[b] (3.3)
Bronchiole epithelium, hyperplasia	0	12[b] (1.0)	34[b] (1.0)	48[b] (1.5)
Inflammation, chronic	4 (1.0)	37[b] (1.3)	39[b] (1.8)	49[b] (2.0)
Alveolus, infiltration cellular, histiocyte	0	34[b] (2.4)	35[b] (2.4)	45[b] (2.7)
Interstitial fibrosis	0	1 (2.0)	4[c] (2.5)	8[b] (1.5)
Alveolar/bronchiolar adenoma, multiple	0	3	5[c]	6[c]
Alveolar/bronchiolar adenoma (includes multiple)	1	17[b]	23[b]	19[b]
Alveolar/bronchiolar carcinoma, multiple	0	9[b]	5[c]	5[c]
Alveolar/bronchiolar carcinoma (includes multiple)	0	23[b]	18[b]	22[b]
Alveolar/bronchiolar adenoma or carcinoma	1	32[b]	35[b]	32[b]
Larynx				
Total no. examined	50	50	50	50
No. with:				
Respiratory epithelium, epiglottis, metaplasia, squamous	0	39[b] (1.0)	45[b] (1.0)	44[b] (1.1)
Nose				
Total no. examined	50	50	50	50
No. with:				
Inflammation, suppurative	19 (1.1)	14 (1.2)	32[b] (1.2)	30[b] (1.3)
Olfactory epithelium, atrophy	2 (1.5)	8[c] (1.3)	5 (1.0)	14[b] (1.3)
Olfactory epithelium, degeneration, hyaline	11 (1.2)	23[b] (1.0)	34[b] (1.2)	48[b] (1.3)
Respiratory epithelium, degeneration, hyaline	35 (1.3)	39 (1.5)	46[b] (1.7)	50[b] (1.8)
Respiratory epithelium, metaplasia, squamous	0	3 (1.3)	7[b] (1.1)	8[b] (1.1)
Respiratory epithelium, necrosis	0	0	1 (2.0)	7[b] (1.4)
Lymph node, bronchial				
Total no. examined	39	40	45	41
No. with:				
Hyperplasia	3 (2.0)	13[b] (1.8)	14[b] (2.3)	20[b] (2.3)

From National Toxicology Program (2002)

[a] Average severity grade of lesions in affected animals: 1, minimal; 2, mild; 3, moderate; 4, marked

[b] Significantly different ($p \leq 0.01$) from the chamber control group by the Poly-3 test

[c] Significantly different ($p \leq 0.05$) from the chamber control group by the Poly-3 test

concentration), 26/50 (mid concentration) or 27/50 (high concentration) in males and 33/50, 24/50, 29/50 or 30/50 in females, respectively; mean survival times: 668, 680, 692 or 671 days in males and 688, 678, 679 or 683 days in females, respectively). Mean body weights were slightly decreased in females exposed to 2.0 mg/m^3 throughout the study compared with chamber controls. Although there was a marginally increased incidence of alveolar/bronchiolar neoplasms in female rats, the increase was not statistically significant, did not occur in a concentration-related fashion and was in the historical control range. Thus, it was uncertain whether the increased incidence observed was exposure-related. Exposure to vanadium pentoxide caused an increase in the incidence of alveolar/bronchiolar neoplasms in male rats. Although not statistically significant, the incidence of alveolar/bronchiolar adenoma in males exposed to 0.5 mg/m^3 and of alveolar/bronchiolar carcinoma and alveolar/bronchiolar adenoma or carcinoma (combined) in males exposed to 0.5 and 2 mg/m^3 exceeded the historical ranges in controls (all routes) given NTP-2000 diet and inhalation controls given NIH-07 diet. This response was considered to be related to exposure to vanadium pentoxide. However, exposure to vanadium pentoxide did not cause increased incidence of neoplasms in other tissues. The incidence of neoplasms and non-neoplastic lesions of the respiratory system in male rats is reported in Table 5. Alveolar bronchiolar adenomas, typical of those occurring spontaneously, were generally distinct masses that compressed surrounding tissue. Component epithelial cells were generally uniform in appearance and were arranged in acinar and/or irregular papillary structures and occasionally in a solid cellular pattern. Alveolar/bronchiolar carcinomas had similar cellular patterns but were generally larger and had one or more of the following histological features; heterogeneous growth pattern, cellular pleomorphism and/or atypia, and local invasion or metastasis. Three male rats exposed to 0.5 mg/m^3, one male rat exposed to 1 mg/m^3 and three male rats exposed to 2 mg/m^3 developed alveolar/bronchiolar carcinomas, one of which metastasized. There were no primary lung carcinomas in the chamber control rats. Alveolar/bronchiolar adenomas and especially carcinomas with metastases from the site of origin are uncommon in rats (Hahn, 1993). Exposure to vanadium pentoxide caused a spectrum of inflammatory and proliferative lesions in the lungs that were similar in male and female rats. There was a significantly-increased incidence of alveolar epithelial hyperplasia in the lungs of males exposed to 0.5 mg/m^3 or greater and females exposed to 1 or 2 mg/m^3. Squamous metaplasia of the alveolar epithelium occurred in 21/50 male and 6/50 female rats exposed to 2.0 mg/m^3 vanadium pentoxide. Squamous epithelium is not a normal component of the lung parenchyma. It is a more resilient epithelium and its occurrence in the lung generally represents a response to injury (National Toxicology Program, 2002; Ress *et al*., 2003).

3.1.3 *Comparison of findings from the rat and mouse inhalation studies*

A wide range of proliferative lesions in the lungs were observed in rats and mice exposed to vanadium pentoxide for 2 years. The incidence of hyperplasia of the alveolar and bronchiolar epithelium was increased in exposed rats and mice. Although given

Table 5. Incidence of neoplasms and non-neoplastic lesions of the respiratory system and bronchial lymph nodes in male rats in a 2-year inhalation study of vanadium pentoxide

	No. of rats exposed to vanadium pentoxide at concentrations (mg/m^3) of			
	0 (chamber control)	0.5	1	2
Lung				
Total no. examined	50	49	48	50
No. with:				
Alveolar epithelium, hyperplasia	7 (2.3)[a]	24[b] (2.0)	34[b] (2.0)	49[b] (3.3)
Bronchiole epithelium, hyperplasia	3 (2.3)	17[b] (2.2)	31[b] (1.8)	49[b] (3.3)
Alveolar epithelium, metaplasia, squamous	1 (1.0)	0	0	21[b] (3.6)
Bronchiole epithelium, metaplasia, squamous	0	0	0	7[b] (3.7)
Inflammation, chronic active	5 (1.6)	8 (1.8)	24[b] (1.3)	42[b] (2.4)
Interstitial fibrosis	7 (1.4)	7 (2.0)	16[c] (1.6)	38[b] (2.1)
Alveolus, infiltration cellular, histiocyte	22 (1.3)	40[b] (2.0)	45[b] (2.3)	50[b] (3.3)
Alveolus, pigmentation	1 (2.0)	0	2 (1.5)	28[b] (2.1)
Alveolar/bronchiolar adenoma, multiple	0	2	0	0
Alveolar/bronchiolar adenoma (includes multiple)	4	8	5	6
Alveolar/bronchiolar carcinoma, multiple	0	1	0	0
Alveolar/bronchiolar carcinoma (includes multiple)	0	3	1	3
Alveolar/bronchiolar adenoma or carcinoma	4	10	6	9
Larynx				
Total no. examined	49	50	50	49
No. with:				
Inflammation, chronic	3 (1.0)	20[b] (1.1)	17[b] (1.5)	28[b] (1.6)
Respiratory epithelium, epiglottis, degeneration	0	22[b] (1.1)	23[b] (1.1)	33[b] (1.5)
Respiratory epithelium, epiglottis, hyperplasia	0	18[b] (1.5)	34[b] (1.5)	32[b] (1.9)
Respiratory epithelium, epiglottis, metaplasia, squamous	0	9[b] (1.7)	16[b] (1.8)	19[b] (2.1)
Nose				
Total no. examined	49	50	49	48
No. with:				
Goblet cell, respiratory epithelium, hyperplasia	4 (1.8)	15[b] (1.8)	12[c] (2.0)	17[b] (2.1)

From National Toxicology Program (2002)

[a] Average severity grade of lesions in affected animals: 1, minimal; 2, mild; 3, moderate; 4, marked

[b] Significantly different ($p \leq 0.01$) from the chamber control group by the Poly-3 test

[c] Significantly different ($p \leq 0.05$) from the chamber control group by the Poly-3 test

distinct diagnoses, the lesions were considered to be one pathogenic process. The authors concluded that this hyperplastic change was striking and appeared more prominent than had been observed in other National Toxicology Program inhalation studies. Although the exact pathogenesis was not determined in this study, the hyperplasia of the alveolar and bronchiolar epithelium was consistent with bronchiolization, a process in which bronchiolar epithelium proliferates and migrates down into alveolar ducts and adjacent alveoli. Although there was clearly proliferation, it was thought primarily to represent a metaplastic change. Whether this represented a precursor lesion for development of pulmonary neoplasms is not known. The lung tumour response in rats and mice following exposure to vanadium pentoxide was not concentration-related; there was a flat dose response. Several dose metrics and lung-burden data were used to aid in interpretation of lung pathology in exposed rats and mice. In the case of all dose metrics, rats received more vanadium than mice. In mice, the total 'dose' was similar in the groups exposed to 1 mg/m^3 and 2 mg/m^3 and this may help explain the flat dose response in the lung neoplasms in male and female mice. The total dose does not explain the differences in neoplasms in rats compared with mice. However, when the total dose is corrected for body weight, mice received a three- to five-fold higher dose of vanadium than rats at comparable exposure concentrations of 1 and 2 mg/m^3. Therefore, on a body weight basis, mice received considerably more vanadium than rats, and this may help explain the differences in responses between the species (National Toxicology Program, 2002; Ress *et al*., 2003).