

**ADDENDUM TO “UPDATE TO PERCHLORATE
TOXICOLOGICAL PROFILE AND HEALTH
ASSESSMENT”**

**REVIEW OF NEW STUDIES ON
PERCHLORATE**

In Support of:

**Perchlorate Maximum Contaminant Level (310 CMR 22.06)
Perchlorate Cleanup Standards (310 CMR 40.0000)**

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1.0 INTRODUCTION

During the internal review period of the Office of Research and Standards' (ORS) "Update to Perchlorate Toxicological Profile and Health Assessment" document (MassDEP, 2006), additional information related to perchlorate toxicity became available. This information includes results from human epidemiological studies and one animal study. In addition, the US EPA (2005) adopted an RfD for perchlorate; the Agency for Toxic Substances and Disease Registry (ATSDR, 2005) released a draft perchlorate toxicity assessment for public comment; and the New Jersey Department of Environmental Protection (NJ DEP, 2005) completed a toxicity assessment.

We have reviewed these documents to determine if the data and interpretations in these reports would alter our assessment of perchlorate's toxicity and the resulting reference dose recommended by ORS in its update document released concurrently with this document. The new reports and studies are discussed in the assessment below, which also reflects additional input and review from the MassDEP-DPH Advisory Committee on Health Effects.

2.0 EPIDEMIOLOGICAL STUDIES

New epidemiological studies include:

- (1) A study of pregnant women and their offspring chronically exposed to perchlorate in Chile (Tellez *et al.*, 2005);
- (2) An ecological study conducted of neonates born to perchlorate-exposed mothers in California (Buffler *et al.*, 2005);
- (3) An unpublished ecological study conducted of neonates born to perchlorate-exposed mothers in Israel (Amitai *et al.*); and,
- (4) An occupational study of workers chronically exposed to perchlorate (Braverman *et al.*, 2005).

2.1 TELLEZ *et al.*, 2005 (CHILEAN STUDY)

2.1.1 Study Overview

- Tellez *et al.* performed a longitudinal epidemiological study among pregnant women in three cities in Chile: Antofagasta, Chanaral, and Taltal with 0.5 µg/L, 6 µg/L and 114 µg/L perchlorate in drinking water, respectively.
- They investigated maternal and neonatal physical parameters and measures of thyroid function as well as urine and breast milk perchlorate and iodine levels.
- The authors concluded that perchlorate levels as high as 114 µg/L did not cause changes in maternal and neonatal thyroid function. However, ORS' evaluation of the study, as summarized below, indicates that the reported results should be interpreted cautiously due to a number of factors, which we believe preclude a firm conclusion of no perchlorate effect, and raise questions as to the generalizability of the results to the US population.

2.1.2 Summary Of ORS Critique Of Tellez *et al.*

ORS has reviewed the Chilean study and discussed it with the MassDEP/DPH Advisory Committee on Health Effects. ORS and the Committee found the overall study design to be of good quality and the statistical analyses appropriate. However, as discussed in more detail below, several issues raise questions about the study's applicability to US populations. In particular, alterations of some parameters associated with disrupted thyroid function were reported in the Chilean cohorts when compared to observations in other populations. Differences in iodide intake compared to the situation in the US and variable exposures to other thyroid active agents, such as lithium, between the study cohorts also complicate interpretation of the studies. At a minimum these observations raise questions about the comparability of the Chilean cohorts to those in the US and any conclusions regarding possible perchlorate effects. Some of the study results may also be suggestive of a perchlorate effect.

The data reported in the study indicate that the cohorts included are not comparable to US women with respect to their iodine intake, which was higher than in the US (i.e., NHANES III). In addition, a recent study in Boston highlights that 50% of the pregnant women studied had iodine intake below the U.S recommended daily allowance and 9% had levels consistent with iodine deficiency (Pearce *et al.*, 2004). Therefore, those in the US may be more susceptible to perchlorate effects.

The following sections summarize comments on specific components of the study by ORS and the Joint Committee on Health Effects.

2.1.2.1 Maternal Parameters Tested

Goiter Prevalence

- The authors stated that the percentage of subjects with goiter in the study populations was high in comparison to the US, especially in Chanaral and, post partum, in Taltal. The study authors mentioned that the goiter prevalence may be due to the high iodine levels in the recent past and did not have an explanation of the higher frequency of goiter in Chanaral (24 – 36%). The MassDEP/DPH Advisory Committee also noted that these rates were very high, raising concern about the comparability of the study populations to those in the US. Even though the study populations were iodide sufficient, as indicated by their urinary iodine levels (~20-40 µg/dL) and classification criteria for sufficiency, the results observed in Chanaral are similar to increases in goiter prevalence during pregnancy that have been observed in iodine deficient areas (Crooks *et al.*, 1967; Glinoe, 1990). The women in Taltal and Chanaral had increased goiter prevalence at the second prenatal visit relative to the first: Taltal's continued to increase at the postpartum visit relative to the second prenatal visit, while Chanaral's returned to the first prenatal value at the postpartum visit (Table 1). The observed pattern in goiter prevalence in Taltal is different than the patterns in

the other two cities (Table 1). Urinary iodine excretion (iodine/g creatinine) was also markedly decreased after delivery in the pregnant women in Taltal compared to the groups in Antofagasta and Chanaral. The decreased iodine excretion is consistent with a possible overcompensation phenomenon that could result from induction of the symporter protein by goitrogens, which ultimately increases iodide uptake into the thyroid. Excess iodide intake, like low iodide intake, is known to increase goiter prevalence (Pearce et al., 2002).

Table 1. Goiter Prevalence in Perchlorate Exposed Pregnant Women in Antofagasta, Chanaral or Taltal*

Measurement Period**	Antofagasta (CLO ₄ ⁻ < 4 µg/L)		Chanaral (CLO ₄ ⁻ 5.8 µg/L)		Taltal (CLO ₄ ⁻ 114 µg/L)		P-value	
	N	% with Goiter	N	% with Goiter	N	% with Goiter	Kruskal-Wallis	Regression
FP	64	8.7	52	24.4	65	9.4		0.04
SP	48	7.7	40	36.4	38	15.2		0.01
PP	42	11.1	26	24.4 ± 1.86	46	22.5		0.21

*Goiter prevalence was not found to be statistically significantly different between Antofagasta and Taltal at the first or second prenatal visit.

** FP = first prenatal visit; SP = second prenatal visit; PP = postpartum visit

Serum TSH Levels

- Tellez *et al.* measured TSH levels in their study populations. Elevated TSH values are indicative of iodide insufficiency or other factors that disrupt thyroid function. The mean TSH levels measured during pregnancy (excluding outliers in Antofagasta) in the Chilean pregnant women exposed to perchlorate (Table 2) ranged from 2.46 to 2.95 µUI/ml. Tellez *et al.* state that these TSH values were not statistically significantly different between cities nor gestational stage and that they fall within the range of 0.4 – 4.5 µUI/ml that has been cited as a “normal” range. However, current data (Hollowell *et al.*, 2002; Andersen *et al.*, 2002; Lee, 2003; Wartosky and Dickey, 2005) indicate that the optimal mean TSH should typically be less than 2 µUI/ml. Others argue the current reference range cited above continues to be appropriate (Surks *et al.*, 2005). Various studies (especially studies conducted recently using updated analytical techniques) in iodine sufficient pregnant women (Table 3) consistently support this view, with mean or median TSH values that do not typically exceed 1.4 µUI/ml in iodine sufficient populations with normal thyroid function. Values higher than 2 µUI/ml are observed in iodine deficient pregnant women.
- Thus, the TSH levels in the Chilean cohort are consistent with those observed in iodine deficient pregnant women even though the pregnant women in the three Chilean cities have sufficient iodine in their diets.
- In addition, about 6% of the pregnant women in all three cities were diagnosed with hypothyroidism during pregnancy and placed on thyroxin

treatment. This finding is in contrast to the reported 0.8 % hypothyroidism value for normal pregnant women tested during pregnancy in the United States (Walker et al., 2005).

- Taken together, these results (elevated TSH levels and increased rates of hypothyroidism despite sufficient dietary iodide) suggest that the pregnant women in the three Chilean cities may be experiencing factor(s) (perhaps including perchlorate) from water and food that perturb thyroid homeostasis.

Table 2. Mean Serum TSH (μ UI/ml)* Levels in Pregnant Women in Antofagasta, Chanaral, or Taltal

Measurement Period [†]	Antofagasta (CLO ₄ ⁻ < 4 μ g/L)		Chanaral (CLO ₄ ⁻ 5.8 μ g/L)		Taltal (CLO ₄ ⁻ 114 μ g/L)		P-value	
	N	Mean \pm SD	N	Mean \pm SD	N	Mean \pm SD	Kruskal-Wallis	Regression
FP	64	2.63 \pm 1.54	52	2.81 \pm 1.78	65	2.61 \pm 1.45	0.91	0.75
SP	48	2.46 \pm 1.44**	40	2.55 \pm 2.12	38	2.08 \pm 0.86	0.63	0.68
PP	42	2.39 \pm 2.8**	26	2.34 \pm 1.86	46	1.95 \pm 1.38	0.79	0.13

* Micro international unit

** These values calculated excluding outliers that developed hypothyroidism

[†] FP = first prenatal visit; SP = second prenatal visit; PP = postpartum visit

Table 3. Serum TSH Levels During Pregnancy in Iodine Sufficient and Deficient Populations in Different Parts of the World

TSH levels (μ UI/ml)	Iodine Status*	Country	References
1.37	Sufficient	Iran,	Soviad and Omrani (2000)
1.35	Sufficient	Japan	cited in Soviad and Omrane (2000)
1.37	Sufficient	Denmark	Weeke <i>et al.</i> , 1982
1.4	Sufficient	USA	Mitchell <i>et al.</i> , 2003
1.1	Sufficient	Holland	Berghout <i>et al.</i> , 1994
1.17 \pm 0.08 (during pregnancy)	Sufficient	Sweden	Soldin <i>et al.</i> , 2004
1.06 \pm 0.07 (1-year postpartum)			
2.9	Deficient	Chile (before iodine supplementation)	Silva and Silva, 1981
2.1	Deficient	Belgium	Glinoyer <i>et al.</i> , 1990

* Iodine status designations were made by the respective authors and in almost all cases was not accompanied by notation of the criteria for making such assignments.

Free T4 levels

- The authors found significant differences in Free T4 levels among the three cities at the second prenatal visit. However with regression analysis that controlled for differences in maternal age (mean maternal ages in Antofagasta, Chanaral, and Taltal respectively were 23.1 \pm 6.2, 28.2 \pm 6.3 and 25 \pm 6) among the three cities, the differences among cities were no longer significant. However, Sovaid and Omrani

(2000) found Free T4 to have no correlation with age, gestational age, or number of previous pregnancies in the population they studied. The age in the studied pregnant women in the Sovaid and Omrani study ranged from 15 to 45 years. Studies in various age groups also found no correlation of Free T4 with age (Penny *et al.*, 1983).

- The mean T4 level at the second prenatal visit was below the non-pregnant range (see Figure 3 in the Tellez paper) and was consistent with findings in iodine deficient pregnant women (Kung *et al.*, 2002 and Eltom *et al.*, 1999).

Serum Thyroglobulin (Tg) Levels

- Tg is the most abundant protein of the thyroid, providing the matrix for thyroid hormone synthesis, and is an indicator of the iodine status of the thyroid. Normally, small amounts are secreted or leak from the thyroid into the circulation. In comparative studies of areas that differed in terms of iodine status, lower serum thyroglobulin concentrations were reported in iodine sufficient areas than in iodine deficient areas (Gutekunst *et al.*, 1986; Szabolcs *et al.*, 1997). The mean Tg levels in perchlorate-exposed pregnant women in the three Chilean cities are presented in Table 4. These values range from 2.97 – 4.32 ng/ml (this range does not include postpartum values). ORS notes that these Tg levels are much lower than those reported in other studies. The mean Tg levels in iodine sufficient pregnant women in the first, second, and third trimesters, and 1 year after postpartum were reported to be 15.48 ± 1.96 , 14.92 ± 2.05 , 18.55 ± 2.8 , 13.95 ± 1.6 ng/ml, respectively, by Soldin *et al.*, (2004). In a second study, Tg levels in iodine deficient pregnant women in the first, second and third trimesters were 27.5, 25, and 30 ng/ml, respectively (Eltom *et al.*, 1999). The low Tg levels observed in the pregnant women in the three Chilean cities may be related to high iodine intake, which may inhibit thyroglobulin biosynthesis (Radvila *et al.*, 1976). Radvila *et al.* also showed that iodine has a very weak inhibitory action on thyroglobulin breakdown but strongly potentiates the inhibitory effect of lithium on thyroglobulin degradation.

Table 4. Serum Tg Levels (ng/mL) Measured in Perchlorate Exposed Pregnant Women in Three Chilean Cities

Measurement Period*	Antofagasta (CLO ₄ ⁻ < 4 µg/L)		Chanaral (CLO ₄ ⁻ 5.8 µg/L)		Taltal (CLO ₄ ⁻ 114 µg/L)		P-value	
	N	Mean ± SD	N	Mean ± SD	N	Mean ± SD	Kruskal-Wallis	Reference
FP	58	4.32 ± 0.3.63	45	3.67 ± 3.49	58	3.64 ± 3.31.13	0.30	Tellez, <i>et al.</i> , 2005
SP	47	2.97 ± 2.05	38	2.99 ± 2.39	35	3.70 ± 2.78	0.23	
PP	41	3.80 ± 5.31	20	6.50 ± 8.05	44	5.78 ± 10.03	0.16	

* FP = first prenatal visit; SP = second prenatal visit; PP = postpartum visit

- The reason for this dramatic difference between Tg levels in all three cities and those reported in other populations is unknown but may be indicative of complex

interactions between various factors impacting thyroid function (e.g. high iodide intake; perchlorate; lithium etc). In any case, this observation raises concerns about the representativeness of the study populations for those in the U.S.

Serum T3 levels

- The women in Chanaral had statistically higher T3 levels (Table 5) than women from the other two cities at both prenatal visits, however no differences were observed at the post partum visit, which could possibly be explained by the increased secretion of perchlorate into the breast milk, as evidenced by decreased urinary excretion of perchlorate after delivery (Table 8).
- This increase in T3 during pregnancy in Chanaral women is consistent with the results observed in studies of populations in iodine deficient areas, where T3 is preferentially synthesized and released by the thyroid to preserve the diminished iodide supply (T3) (Glinoe *et al.*, 1990, 1995). The same response was not observed in Taltal. Although this lack of a perchlorate-related dose response may indicate of a lack of perchlorate effect, it is important to note that nonlinear dose response relationships, which can vary depending on the endpoint under consideration, are observed, especially in hormone mediated systems with complex feedback regulatory systems. In this case it is possible that at the higher dose situation in Taltal, compensatory mechanisms may have been effectively activated. Thus, we do not think it is appropriate to discount the significant effect on this parameter observed in Chanaral. Also, the coexistence of perchlorate with levels of other goitrogens like lithium, nitrate, arsenic (Table 6) in the drinking water in the three Chilean cities could complicate interpretation of the dose-response data and the observed effects. Although the authors have indicated that the levels of the various goitrogens are within acceptable ranges, the lithium value differs considerably between the cities.

Table 5. Serum T3 Levels (ng/dL) Measured in Perchlorate Exposed Pregnant Women in Three Chilean Cities

Measurement Period*	Antofagasta (CLO ₄ ⁻ < 4 µg/L)		Chanaral (CLO ₄ ⁻ 5.8 µg/L)		Taltal (CLO ₄ ⁻ 114 µg/L)		P-value	
	N	Mean ± SD	N	Mean ± SD	N	Mean ± SD	Kruskal -Wallis	Regression
FP	64	183 ± 35.7	52	207 ± 38.5	65	187 ± 36.1	0.001	0.0004
SP	48	196 ± 46.3	40	206 ± 40.7	38	173 ± 39.9	0.003	0.006
PP	42	115 ± 40.3	26	114 ± 50.2	46	107 ± 28.1	0.74	0.48

* FP = first prenatal visit; SP = second prenatal visit; PP = postpartum visit

Table 6. Water Analysis for Perchlorate and Other Potential Goitrogens

Parameter	Antofagasta		Chanaral		Taltal	
	N	Mean ± SD	N	Mean ± SD	N	Mean ± SD
Perchlorate, µg/L	66	ND	53	5.82 ± 0.63	62	113.9 ± 13.3
Iodide, µg/L ^a	5	156 ± 35	5	87 ± 18	5	114 ± 14
Arsenic, µg/L ^b	5	11 ± 6	5	ND	5	15 ± 6
Lithium, µg/L ^c	5	848 ± 205	5	370 ± 72	5	75 ± 10
Nitrate nitrogen mg/L ^d or	6	0.61 ± 0.47	6	0.64 ± 0.4	6	2.06 ± 0.84
Nitrate, µg/L		2700		2800		9123

^a iodide: urinary iodide excretion was not different among the cities

^b arsenic current standard is 10 µg/L

^c lithium: average diet contains 2 mg/day. Therapeutic dose is 100 mg/day

^d nitrate nitrogen: US standard is 10 mg/L

Body weight

- Body weights of the women studied in Chanaral and Taltal were higher than those observed in Antofagasta (Table 7). The authors mentioned these variable body weights in the three cities but did not assess whether age or other factors could account for the observed results. Of interest is the fact that increased body weight is one of the symptoms of an under active thyroid gland.

Table 7. Body Weight (kg) of Perchlorate Exposed Pregnant Women in Three Chilean Cities

Measurement Period*	Antofagasta (CLO ₄ ⁻ < 4 µg/L)		Chanaral (CLO ₄ ⁻ 5.8 µg/L)		Taltal (CLO ₄ ⁻ 114 µg/L L)		P-value Kruskal -Wallis
	N	Mean ± SD	N	Mean ± SD	N	Mean ± SD	
FP	46	62.1 ± 13.5	45	67.6 ± 13.1	60	66.8 ± 14.6	0.04
SP	38	69.2 ± 14.4	31	74.3 ± 13.0	46	75.3 ± 12.2	0.02
PP	38	60.3 ± 10.2	43	68 ± 12.3	48	68.1 ± 13.2	0.01

* FP = first prenatal visit; SP = second prenatal visit; PP = postpartum visit

Urine Iodine Levels

- Urine iodine levels between visits were not statistically discussed by the paper's authors. Means appeared to have increased from the first to the second prenatal visit in Chanaral and Taltal, respectively (Table 8). The urine iodine levels in Antofagasta may have decreased from the first to second prenatal visits. It is interesting to note that urine iodine levels are lower in Taltal than Antofagasta and Chanaral. Taltal, however, has higher water concentration (114 µg/L) of iodine than Chanaral (87 µg/L). Urine levels of iodine also remained depressed in Taltal after delivery and were 75% of the first prenatal visit value while the levels in Chanaral and Antofagasta bounced back close to the first prenatal visit values. It is possible that iodine in the diet could be lower in Taltal, or the thyroid glands of the pregnant women in Taltal may be trapping iodine efficiently to compensate

for iodine loss due to higher chronic perchlorate exposure compared to the other two cities, thus attenuating the effects of perchlorate on hormone levels.

Table 8. Urine Iodide Levels (Iodine/g creatinine) Measured in Perchlorate Exposed Pregnant Women in Three Chilean Cities

Measurement Period*	Antofagasta (ClO ₄ ⁻ < 4 µg/L)		Chanaral (ClO ₄ ⁻ 5.8 µg/L)		Taltal (ClO ₄ ⁻ 114 µg/L)		P-value	
	N	Mean ± SD	N	Mean ± SD	N	Mean ± SD	Kruskal -Wallis	
FP	62	407 ± 191	39	363 ± 140	62	323 ± 157		0.02
SP	47	368 ± 192	27	422 ± 123	37	358 ± 120		0.08
PP	34	382 ± 488	25	334 ± 95	39	244 ± 114		0.003

* FP = first prenatal visit; SP = second prenatal visit; PP = postpartum visit

Urine perchlorate

- Urine perchlorate levels were measurable and significantly different between cities at the first and second prenatal visits (increasing with higher perchlorate in drinking water). Perchlorate in urine decreased substantially in both Chanaral and Taltal at the postpartum visit (Table 9). The decreased urine levels of perchlorate after delivery suggest another route of perchlorate loss, which may be secretion into breast milk, consequently lowering maternal perchlorate levels. This may be the reason why various altered thyroid parameters diminished after delivery.

Table 9. Urine Perchlorate Levels (µg ClO₄⁻¹/g creatinine) Measured in Perchlorate Exposed Pregnant Women in Three Chilean Cities

Measurement Period*	Antofagasta (ClO ₄ ⁻ < 4 µg/L)		Chanaral (ClO ₄ ⁻ 5.8 µg/L)		Taltal (ClO ₄ ⁻ 114 µg/L)		P-value	
	N	Mean ± SD	N	Mean ± SD	N	Mean ± SD	Kruskal -Wallis	Regression
FP	61	28.4 ± 22	53	80.2 ± 129.6	59	135.5 ± 95		<0.0001
SP	35	23.5 ± 18.2	36	71.9 ± 148.9	27	192.1 ± 138.6		<0.0001
PP	6	49.7 ± 88.8	4	23 ± 18	16	54 ± 36.9		0.19

* FP = first prenatal visit; SP = second prenatal visit; PP = postpartum visit

2.1.2.2 Neonatal Parameters

Cord Serum T3 and Tg

- In the neonatal cord serum, T3 and Tg were significantly less in Chanaral than Antofagasta and Taltal (Table 10). The mean cord serum Tg levels reported in iodine deficient and iodine sufficient areas are 25.8 ng/ml and 43.4 ng/ml, respectively (Sava *et al.*, 1986). These values are higher than the mean cord serum Tg levels (14.03 – 18.11ng/ml) measured in the three Chilean cities.

- Other altered neonatal parameters in the study include decreased median cord blood TSH to median maternal TSH ratio, and increased mean cord Tg to maternal serum Tg ratio compared to literature values. The percentage of males is also significantly higher in Taltal than in Chanaral and Antofagasta.
- The MassDEP/DPH Advisory Committee also noted that some of the neonatal parameters measured such as, length, weight, and head circumference are insensitive measures of developmental effects.

Table 10. Neonatal Parameters

Parameter	Antofagasta (CLO ₄ ⁻ < 4 µg/L)		Chanaral (CLO ₄ ⁻ 5.8 µg/L)		Taltal (CLO ₄ ⁻ 114 µg/L)		P-value	
	N	Mean ± SD	N	Mean ± SD	N	Mean ± SD	Kruskal -Wallis	Regression
T ₃ (ng/dL)	35	79 ± 13.4	42	73 ± 17.9	28	82 ± 20.6	0.03	0.003*
Tg ng/mL)	30	16.79 ± 9.93	36	14.03 ± 10.98	28	18.11 ± 12.82	0.29	0.01**
% male	55	49.1	48	54.2	55	72.7	0.03	
Cord serum perchlorate (µg/L) ^{***}	4	< 4	1	< 4	14	19.9 ± 5.0	0.005	
% vaginal birth	56	60.7	49	69.4	57	40.4	0.01	

*T₃ was not different between Antofagasta and Taltal (p = 0.76)

** Tg was not different between Antofagasta and Taltal (p = 0.95)

*** There was no attempt to measure serum perchlorate in all samples

2.1.2.3 Breast Milk

- Perchlorate was found in breast milk. Higher concentrations were associated with increasing perchlorate in drinking water and with increasing urinary levels during the first prenatal visit (Table 11). It is interesting to note that the postpartum urine perchlorate levels in Chanaral and Taltal are lower than the first prenatal visit values, consistent with perchlorate secretion into the breast milk. The urine levels of perchlorate in Antofagasta at postpartum visit are close to the levels measured in Taltal and highly variable. It is not clear if the woman with high levels of perchlorate in her urine (230 µg/g creatinine) was included in the analysis.
- Serum levels of perchlorate were reported only for postpartum women from Antofagasta, precluding any assessment of the relationship between serum and breast milk levels. As previously noted, rodent data and PBPK modeling suggest that perchlorate may be concentrated into breast milk.

Table 11. Perchlorate Concentration in Breast Milk, Drinking Water and Urine

Parameter	Antofagasta (CLO ₄ ⁻ < 4 µg/L)		Chanaral (CLO ₄ ⁻ 5.8 µg/L)		Taltal (CLO ₄ ⁻ 114 µg/L)		P-value Kruskal-Wallis
	N	Mean ± SD	N	Mean ± SD	N	Mean ± SD	
Median ^{**} breast milk CLO ₄ ⁻ (µg/L)	14	<4	16	19	25	114	<0.0001
Mean ^{**} breast milk CLO ₄ ⁻ (µg/L)	14	81.6 ± 277.1	16	18.3 ± 17.7	25	95.6 ± 54.6	<0.0001
Mean water CLO ₄ ⁻ (µg/L)	66	< 4	53	5.82 ± 0.63	62	113.9 ± 13.3	
Mean urine CLO ₄ ⁻ /g creatinine (FP)	61	28.4 ± 22	53	80.2 ± 129.6	59	135.5 ± 95	<0.0001
Mean urine CLO ₄ ⁻ /g creatinine (PP)	6	49.7 ± 88.8	4	23 ± 18	16	54 ± 36.9	0.19
Mean Serum CLO ₄ ⁻ (PP) (µg/L)	4	<4	0	Not reported	0	Not reported	

PP = postpartum visit; FP = first prenatal visit

^{**} There is an extreme outlier in the data set, which skews the mean and does not significantly affect the median.

2.2 BUFFLER *et al.*, 2005 (NEONATAL STUDY IN CALIFORNIA)

This study investigates the prevalence of primary congenital hypothyroidism and high TSH levels among California newborns born to mothers putatively exposed to perchlorate in drinking water at concentrations ≤ 5 µg/L or > 5 µg/L. The main issues with this study include:

- High potential for exposure misclassification due to uncertain assessment of perchlorate levels in consumed water, which introduces a bias toward null hypothesis of no relationship;
- Confounding due to dietary perchlorate exposures, which also biases toward null;
- Use of insensitive measures of effect. The MassDEP/DPH Advisory Committee particularly emphasized this point, noting that there is no reason to expect any relationship between overt clinical hypothyroidism and perchlorate exposure from drinking water. Additionally, the cutoff for classifying infant TSH levels as being elevated was also established at the high end of the range of the data, reducing the ability of the study to detect any potential perchlorate relationship;
- Questionable censoring of data that might in fact suggest a relationship between perchlorate in drinking water and early TSH levels in newborns. Data presented in Table 1 of the published study indicate that elevated TSH levels were observed in the > 5 µg/l perchlorate group (0.29% with elevated TSH) vs. the < 5 µg/l perchlorate group (0.18% with elevated TSH). The authors dismiss this data and claim that the TSH data obtained from screenings performed < 24 hr after birth

should not be included in the analysis because of the normal surge in TSH levels that occurs post birth. When the analysis is restricted to screenings > 24 hrs post birth, the prevalence odds ratios (PORs) are less than 1 for both normal and low birth weight infants (adjusted for race, sex, and other factors). The information that would allow one to calculate PORs is not presented for the <24 hr screening data. Using all the data (>24 hr and < 24 hr samples) an *unadjusted* POR of 1.59 can be calculated from the information in Table 1 in Buffler *et al.* This suggests that TSH values in the < 24 hr samples in the > 5 µg/L perchlorate group were significantly higher than in the < 5 µg/L group. Whether this difference is attributable to differences in the relative sampling times between the groups cannot be determined based on the data presented.

- Additionally, altered thyroid and pituitary hormone levels at screening times < 24 hours post birth, although unstable, were also observed by Brechner *et al.* (2000) and Schwartz *et al.* (2001).

2.3 AMITAI *et al.* (NEONATAL STUDY IN ISRAEL)

- In an unpublished study presented as an abstract by Amitai *et al.* (year of publication unknown), T4 values obtained from a National Screening Program for Congenital Hypothyroidism were compared between newborns whose mothers resided in suburbs of Israel with different concentrations of perchlorate in drinking water at levels ranging from <14 – 1,100 ppb.
- The authors found no difference in T4 levels between the neonates that were exposed to different concentrations of perchlorate. They also compared T4 levels in neonates whose mothers were exposed to highest concentration of perchlorate with neonates whose mothers drank bottled water and found no difference in the T4 levels. However, the authors did not indicate if the bottled waters were analyzed for perchlorate.

It is hard to critique this paper, as it is available in an abstract form with no details on study protocols, exposure assessment, neonatal and maternal health conditions, etc. Moreover, T4 is not the most sensitive parameter to measure neonatal thyroid conditions (Buffler *et al.*, 2005)

2.4 BRAVERMAN *et al.*, 2005 (OCCUPATIONAL STUDY)

- This study was conducted in a limited number of workers that were exposed to perchlorate intermittently (3 days out of the week).
- Urinary excretion levels of iodide in these workers indicate that the study cohorts were iodine sufficient.
- Statistically significant decreased RAIU and increased thyroid hormone levels (T3, T4) and free T4 index were observed in the exposed workers. The authors did not consider these effects as adverse. However, some literature reports indicate that sustained elevation or fluctuating levels of thyroid

hormones may be associated with adverse outcomes (Osman *et al.*, 2002; Biondi *et al.*, 2002; Squizzato *et al.*, 2005; Lakatos, 2003).

- This study is viewed by ORS and the MassDEP/DPH Advisory Committee to be of limited value as a quantitative basis for the derivation of a reference dose for the general population and sensitive subgroups that are exposed to perchlorate on continuous basis.

3.0 ANIMAL STUDY (DOD, 2005; PERSONAL COMMUNICATION)

- Female rats were made iodine deficient and then treated with perchlorate. The author reported that iodine sufficient rats were more sensitive to perchlorate-induced iodide uptake inhibition than were the iodine deficient rats due to the up-regulation of the iodide transport protein. No thyroid or pituitary hormone levels were measured in perchlorate treated iodide sufficient or deficient animals. Also, the study was not conducted in the most sensitive subgroup, the pregnant rat and its fetus.
- Two studies conducted by Versloot *et al.* (1997) and Schroder–van Der Elst *et al.* (2001) on pregnant rats showed that iodine deficiency affected the thyroid activity of the iodine deficient pregnant rat more than the iodine deficient non-pregnant rat, and perchlorate treatment aggravated the observed thyroid effects in these rats. Moreover, the fetus was more sensitive than the mother to iodine deficiency and to the combined effects of iodine deficiency and perchlorate treatment as measured by iodide uptake inhibition and thyroid and pituitary hormone measurements.

4.0 REGULATORY AGENCY ASSESSMENTS

4.1 US EPA (2005)

A few weeks after the NAS (NAS, 2005) recommended a reference dose for perchlorate, the US EPA adopted this value, a chronic RfD of 0.0007 mg/kg/day (USEPA, 2005). US EPA has been criticized (Brown, 2005) for finalizing this value and placing the NAS RfD in its IRIS database without the usual internal agency review, public notice, and comment period.

4.2 ATSDR (2005)

ATSDR released a draft toxicological profile for perchlorate in September 2005 for public review. ATSDR also proposes to adopt the NAS/EPA RfD as a chronic Minimal Risk Level (MRL). However, ATSDR is soliciting comment on how to derive acute and intermediate duration MRLs, since the NAS RfD is based on an acute exposure period (14-days) with no adjustment for chronic exposure duration.

4.3 New Jersey (2005)

New Jersey has chosen to use the NAS/EPA (2005) RfD. They have used this value as the basis for proposing an MCL of 5 µg/L for perchlorate, with a relative source contribution factor of 20%. In their assessment, NJ notes that one member of their advisory committee recommended use of the MassDEP approach. In their document they discuss uncertainties relating to breast milk perchlorate concentrations. ORS notes that if the current limited breast milk data is confirmed, a standard lower than that proposed by NJ could be supported.

5.0 CONCLUSIONS

The information in the new studies reviewed in this addendum is deemed insufficient to warrant any change to ORS's perchlorate assessment or the proposed RfD. The MassDEP/DPH Health Effects Advisory Committee unanimously concurred with this conclusion.

The dietary intake of iodine in the study populations in the Tellez study is higher than that of the US population. Thus, the cohorts would be expected to be less sensitive, overall, to perchlorate effects.

The cohorts in that study exhibited thyroid function parameters that differ from those reported in other studies. Notably,

- Although the study populations were iodine sufficient, mean TSH levels were none-the-less in the range reported for iodine deficient groups.
- The percent of subjects with hypothyroidism in the study populations during pregnancy was high (6%) in comparison the US pregnant population (0.8%).
- The goiter rate was also high in comparison to the US.
- Tg levels were much lower compared to literature values for pregnant women and for cord blood.

These observations, at a minimum, raise questions as to the comparability of the Chilean study populations to those in the US. Furthermore, the values for the first three thyroid parameters are consistent with those observed in iodine deficient areas. The pregnant women in the three Chilean cities, however, are iodine sufficient, suggesting that either perchlorate from food and/or water can alter thyroid function even in iodine sufficient populations or that the pregnant women in the three Chilean cities may be experiencing other factors, such as excess iodine intake, lithium intake or exposures to other agents in the diet or water that perturb normal thyroid function.

Although the observed thyroid-related parameters were not linearly related to perchlorate exposure in the study, this is not unexpected in complex and dynamic systems like the hypothalamus-pituitary-thyroid axis. The complexity of the hypothalamus-pituitary-thyroid axis function is illustrated in a well-designed study conducted by Elnagar *et al.*

(1998). Pregnant women who were mildly iodine deficient exhibited greater thyroid function alterations than pregnant women who were moderately iodine deficient. The authors proposed that compensatory mechanisms may be responsible for the attenuated thyroid effects in the moderately iodine deficient group.

The occupational and animal bioassay studies are also limited in their applicability for reference dose derivation.

In conclusion ORS does not believe that these studies warrant any change to its proposed RfD value. The MassDEP/DPH Advisory Committee of Health Effects was consulted and concurred with this determination.

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