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TO: Steve Roberts, Ph.D., Chair, SAB Perchlorate Advisory Board

FROM: Joanne Rovet, Ph.D., Grant Anderson, Ph.D., Nancy Carrasco, M.D., Paul Lipkin, M.D.

Subject: Preliminary response to Charge (Issue I)

Rationale for Issue 1: Sensitive Life Issues

Thyroid hormones (TH) are essential for the developing brain and are involved in a number of fundamental neurobiological processes such as neurogenesis, neuronal migration, process growth, synaptogenesis and myelination. TH's specific mode of action is to up- or down-regulate major brain genes that underlie these processes. Timing of need for TH within the brain varies among the different processes, with some (e.g., neurogenesis, neuronal migration) needing TH earlier than others (e.g., myelination). Additionally, different brain regions also vary in when they need TH both *in utero* and after birth. Since most aspects of brain development are complete by the second year of life, normal TH levels must be maintained from conception through to at least the age of two years. Also, as TH plays a role in later brain functioning by regulating key neurotransmitters, normal levels of TH are also required throughout life. Furthermore, because iodine is a key component of TH and it cannot be produced within the body, adequate dietary iodine is essential, particularly during pregnancy, when the need for iodine and TH increases by as much as 40-50%.

Iodide is actively concentrated in the thyroid by the sodium/iodide symporter (NIS), an intrinsic plasma membrane protein of 643 amino acids present on the basolateral surface of the thyroid follicular cells.. NIS-mediated iodide transport in the thyroid is the first and the key rate-limiting step in the biosynthesis of TH. NIS is also expressed in a few other tissues, such as lactating breast, salivary glands, placenta, and stomach, in all of which it also mediates active iodide transport.

Human autopsy evidence, including some from aborted fetuses, shows that TH receptors are present as early as the first trimester in human brain and that TH of maternal in origin are measurable in brain tissue. Because the fetal thyroid system has a protracted development, the fetus early in gestation has to rely totally on the maternal supply of TH transferred in a regulated fashion via the placenta. Furthermore, since the fetal thyroid does not secrete its own centrally regulated TH until the third trimester, an adequate maternal TH supply is therefore essential in the first half of pregnancy and also serves a supplementary role later until the fetal thyroid matures fully by term. In fact, by term, as much as 40% of fetal TH may still be from the mother. After birth, small amounts of TH continue to be transferred to the infant in breast milk. Conditions resulting in TH insufficiencies during critical stages of early brain development include clinical or subclinical hypothyroidism in the mother, whereby effects are presumably worse in early gestation; iodine deficiency, which affects both maternal and fetal/infant supplies of TH; congenital hypothyroidism, which typically begins at the end of gestation and continues postnatally for one to two months; and exposure to natural goitrogens or other chemicals that have thyroid disrupting effects on both the

mother's and the fetal/child thyroid gland. Conditions that disturb TH production later in childhood, when most TH-dependent brain development has occurred, include juvenile-acquired hypothyroidism as well as exposure to TH-disrupting environmental toxicants.

One such toxicant of current interest, and the topic for this review panel, is perchlorate, an anion present in drinking water and food. Perchlorate inhibits the NIS-mediated transport of iodide into the thyroid, as well as into the breast. Whereas perchlorate has long been known to be a competitive NIS inhibitor, and thought to be only a nontransported blocker of NIS, it has recently been demonstrated that perchlorate is actually a NIS substrate that is transported against its concentration gradient. Perchlorate readily crosses the placenta via NIS, and so can disrupt fetal thyroid production. Thus, perchlorate exposure at sensitive life stages will reduce availability of TH for both the developing and more mature brain. Exposure to perchlorate leads to reduced maternal TH availability in early pregnancy, less fetal and maternal TH in later pregnancy, and less TH from the child's own thyroid postnatally. The perchlorate-exposed breast-fed infant is doubly affected because less maternal iodide is received concurrent with its own thyroid production being limited by perchlorate in maternal milk. Thyroid inhibition can also occur in formula-fed infants, if their formula or the water added to powdered mixtures contains perchlorate; likewise, older infants and young children will be affected by the presence of perchlorate in dairy milk and certain foods.

ISSUE 1: SENSITIVE LIFE STAGES

1. Is there a need to have a sensitive life stage analysis?

This group unanimously concurs that a sensitive life stage analysis is critical to derive an MCLG for perchlorate based on the following evidence:

- The specific adverse effects on low thyroid hormone levels and inadequate iodide uptake on brain development vary at different life stages
- The infant is more susceptible to perchlorate exposure effects than adult, especially if exposure is acute. Although no data exist on the long-term adverse neurodevelopmental effects of perchlorate *per se*, the data on the adverse effects of thyroid hormone perturbations (a down stream target) on the developing brain justify the need for a life stage approach
- Other thyroidal insults (e.g., thiocyanate, iodine defciency) that may exacerbate the effects of perchlorate should be measured
- The evidence suggests that the most sensitive life stages for the permanent adverse effects of perchlorate for permanent effects on brain development are (i) the pregnant woman and her fetus, (ii) infancy and (iii) the lactating woman.
- At different life stages, there are multiple targets and depending on the life stage, those numbers may change. For example, in the third trimester, maternal thyroid, placenta, and fetal thyroid are all affected.

<u>Recommendation</u>: a sensitive life stage analysis needs to be conducted to determine MCLGs

2. How should the EPA consider differences in severity and permanence of potential adverse effects in neonates, infants, and young children compared with adults?

- Pregnancy is a sensitive life stage distinct from the non-pregnant woman and men in the handling of perchlorate.
- The pregnant woman is the sole source of fetal iodide and a primary source of fetal thyroxine throughout gestation. Because NIS is expressed in the placenta, exposure to perchlorate can block the direct transfer of iodide to the fetus as well as to the maternal thyroid leading to a state of mild to severe hypothyroxinemia and less available free T4 for the fetus. In addition, the mother's own thyroid status can affect the fetal brain beyond direct thyroid hormone effects.
- In the neonate and infant exposed to perchlorate via the water used in formula preparation, iodide transferred to the infant thyroid will be compromised leading to lower levels of thyroid hormone, which is critical for early brain development
- Breast milk additionally offers a distinct mode of delivery of perchlorate since the breast concentrates perchlorate to compete with the iodine contained in milk; this perchlorate will also be transferred through the milk to directly compete with iodide uptake into the infant thyroid
- Modest reductions in thyroid hormone, even as early as the first trimester, can have deleterious and potentially permanent effects on the developing brain. Therefore, each of these stages are critically important for considering the public health implications of exposure to this toxicant in the human population. Additionally as the woman in pregnancy is the source of both iodide and thyroid hormone to the developing fetus and, postnatally, the lactating mother provides iodide to the nursing infant, the pregnant and lactating mother should be considered a sensitive life stage

Recommendation: There should be distinct MCLGs for different life stages. It is also important to consider iodine status in mother and infant. Future studies need to measure perchlorate in water, milk, serum, fetal and maternal urine and take measurements throughout pregnancy; record newborn TSH screening levels; measure FT4 & TSH in infant serum and urine; Iodine measured in serum, urine, and milk

3. How should the EPA consider the effect of shorter half-life and lower reserves for thyroid hormone in infants compared to adults?

- The neonate and young infant have lower thyroid reserves. They also have a more rapid turn-over of iodide and thyroid hormone suggesting greater need for iodide than in the older person. Differences in half-life in thyroid hormones and lower reserves suggest greater vulnerability to acute and chronic exposures to perchlorate.

4. How should the EPA consider the impact of intrauterine exposure to perchlorate on thyroid status in fetuses?

- Because thyroid status in the fetus cannot be readily or safely determined, one needs to monitor maternal thyroid status as a biological indicator of perchlorate exposures. If the mother's thyroidal status is impacted then one can assume the fetal status is definitely affected. However, if the mother's thyroid status is not affected, one cannot assume that the fetal thyroid has not been affected. Therefore, it is critical to find other markers of fetal effects of perchlorate exposure.
- The neonatal screening value, albeit at parturition, is an indicator of thyroidal impact at the end of pregnancy.