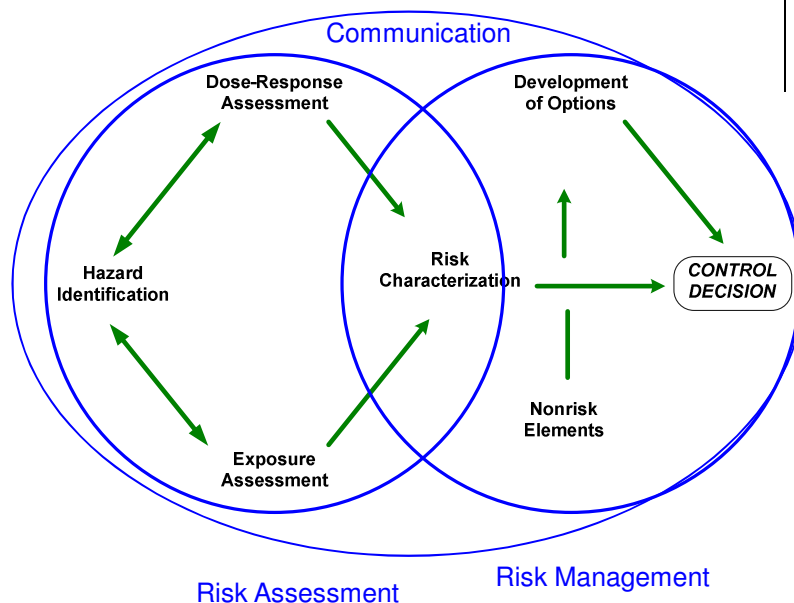


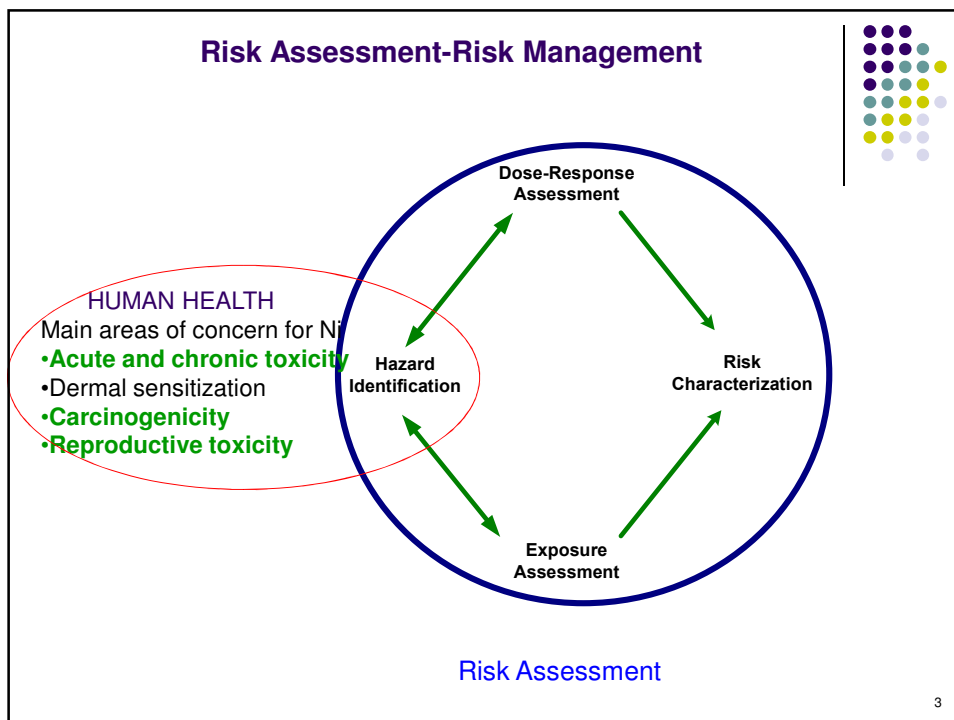
Nickel Scientific Research Programs Overview

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Rayetta G. Henderson, PhD
Hudson K. Bates PhD, DABT



Risk Assessment-Risk Management





Science Data Gaps: Nickel

- Many nickel compounds are currently considered as known human carcinogens by regulatory bodies. What about nickel metal?
 - Human studies have not shown an association between exposures to nickel metal and increased cancer risks. Could this be due to limited power of studies?
 - Animal studies by non relevant means of exposure show conflicting results
 - Proper animal inhalation study was lacking
- Some nickel compounds are associated with increased respiratory tumors. Can they also be systemic carcinogens?
 - Proper animal oral study was lacking

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Science Data Gaps: Nickel



- The respiratory carcinogenicity assessment of water soluble nickel compounds has been challenging.
 - Inconsistent animal and human results compared to the consistent results for some insoluble nickel compounds
 - Competing hypotheses: Ni ion theory versus bioavailability model
 - Understanding of the possible differences in mode of action for inhalation carcinogenicity of water soluble and insoluble nickel compounds was lacking
 - Uncertainties in methods used to assess exposure to specific chemical forms of nickel for attribution of cancer risks existed

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Science Data Gaps: Nickel



- Water soluble nickel compounds are considered as reproductive toxicants based on animal studies
 - Animal studies clearly indicate that exposures to nickel during pregnancy can cause adverse developmental effects in fetuses
 - Threshold for adverse effects observed in animals was not well defined
 - Some hints of possible reproductive effects in humans were reported but proper studies were lacking

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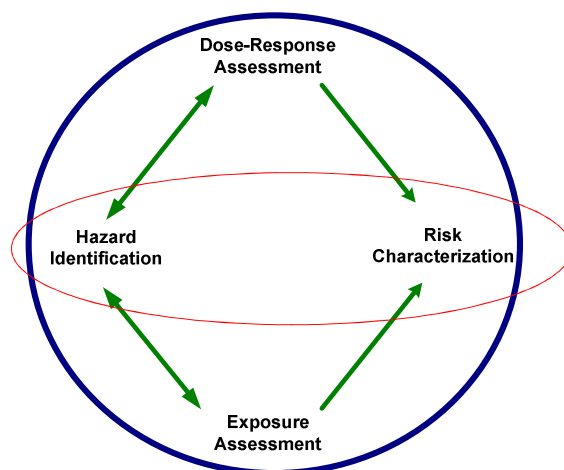
Overview of Human Health Scientific Research Program: Nickel



- Generate new data relevant to hazard, exposure assessment, & dose-response of source nickel substances
 - Carcinogenicity: nickel metal powder (inhalation)
 - Carcinogenicity: water soluble nickel compounds (oral) and mode of action (inhalation)
 - Exposure Assessment: improved discrimination between different chemical forms of nickel (i.e., speciation)
 - Particle size extrapolation from animal studies to workplace
 - Reproductive & Developmental Toxicity: animals & humans

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Risk Assessment-Risk Management



Risk Assessment

How to make predictions for data poor substances?

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Science Data Gaps: Nickel



- Read-across for hazard assessment and risk characterization of nickel substances
 - Water solubility by itself is not robust
 - A more reliable paradigm, taking various routes of exposure and health endpoints into account, was lacking
 - A bioavailability model for cancer that could be used for read across was needed
- Read across from animals to humans for possible reproductive effects requires knowledge of toxicokinetics
 - Toxicokinetic data on blood and urine levels in humans that would be protective of possible reproductive effects observed in animals were not available

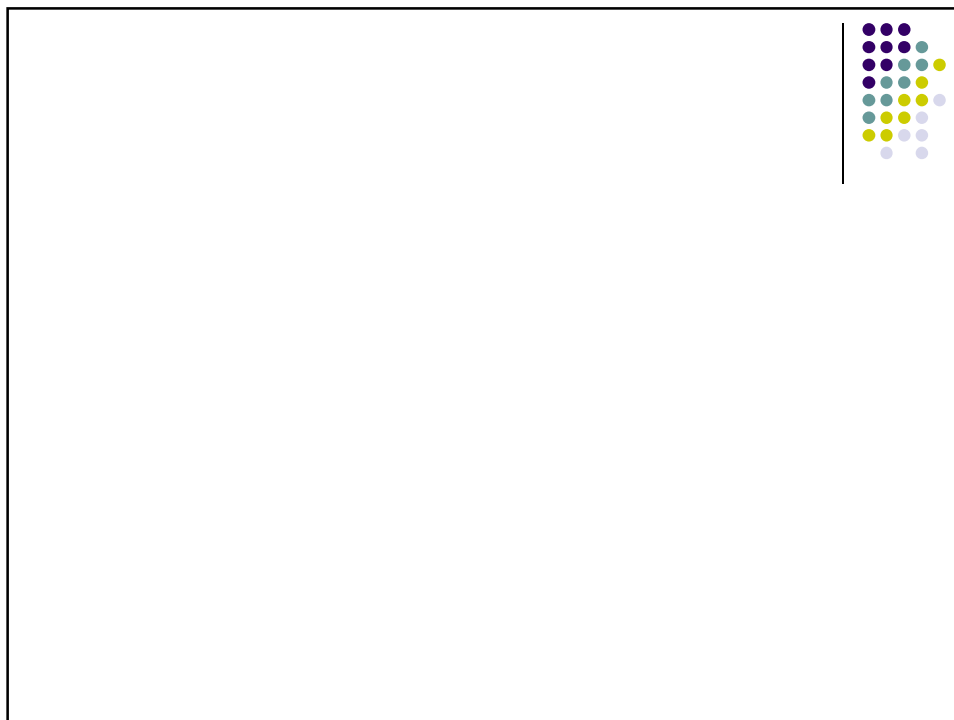
9

Overview of Human Health Scientific Research Program: Nickel



- Develop read-across for hazard assessment and risk characterization of nickel substances
 - Bioaccessibility studies in synthetic fluids, validation with in vivo studies, route of exposure specificity
 - Bioavailability model for respiratory cancer: expert workshop and planned research
 - Toxicokinetic human studies to define blood and urine levels corresponding to oral intake levels protective of reproductive effects observed in animals

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Science Update: Nickel



- Hazard assessment of source nickel substances
 - Carcinogenicity: nickel metal powder (inhalation)
 - Carcinogenicity: water soluble nickel compounds (oral) and mode of action (inhalation)
 - Exposure Assessment: discriminating between different chemical forms of nickel (i.e., speciation)
 - Particle size extrapolation from animal studies to workplace
 - Reproductive & Developmental Toxicity: animals & humans

Hazard assessment of source nickel substances: Carcinogenicity



Background:

- A clear association between exposure to nickel substances present during the processing and refining of sulfidic nickel ores was noted as early as 1958
- Both lung and nasal tumors have been linked to high exposures to certain nickel compounds
- Significant effort has been devoted over the years to understand the potential and/or potency of various forms of nickel to cause these cancers through:
 - epidemiological studies of different types of cohorts
 - speciation of aerosol exposures
 - creation of exposure matrices
 - assessment of cancer dose-response
 - animal inhalation studies with specific nickel substances



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Hazard assessment of Source Nickel Substances: Carcinogenicity



	Animal Data	Human Data	Weight Of Evidence as a Human Carcinogen
Inhalation			
Ni Subsulfide	++	+	+ (↑)
Ni Oxides	+	+/-	+ (↓)
Ni Metal	?	-	?
Water Soluble Ni compounds	-	+/-	+?
Oral			
Water Soluble Ni compounds	?	-	?

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Carcinogenicity: Nickel Metal powder (inhalation)



- Without a proper animal inhalation study, even though epidemiology has been negative, nickel metal remained classified as a suspect carcinogen
- A guideline compliant inhalation study in Wistar rats was negative for respiratory tumor induction (Oller et al., 2008)
 - Study was overseen by a group of external Experts and also included a Pathology Working Group
 - Study was accepted as negative by Denmark and Germany

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Carcinogenicity: Nickel Metal powder (inhalation)



- Epidemiological studies

1990 - ICNCM reviewed 10 cohorts (20,000 workers) and concluded: "There was no evidence that metallic nickel was associated with increased lung and nasopharyngeal cancer risks"

2005 - Sivulka reviewed an additional 10 cohorts and indicated "A review of numerous human studies published since 1990 leads to the conclusion that occupational exposure to metallic nickel does not appear to increase the risk of respiratory cancer at exposure levels that are as high or higher than those found in most current workplace environments."

2008 - Oller et al. latest update confirms lack of increased cancer risks associated with metallic nickel exposures at a Canadian nickel refinery

Consistent negative animal and human results!

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Carcinogenicity: Nickel Metal powder (inhalation)



- Why does nickel metal behave differently than insoluble nickel compounds?
 - Negative or very weak genotoxicity in vivo (same for Mo^0 , Cr^0)
 - Surface properties of Ni metal lead to low cellular uptake and low bioavailability of Ni ion at critical cellular respiratory sites
 - **surface passivation layer is different from NiO!**

- Negative animal study together with negative epidemiological studies confirm the lack of carcinogenic potential of nickel metal

- Planned work on bioavailability model testing and manuscript on differences in physicochemical properties between Ni metal and NiO will add to weight of evidence assessment for nickel metal

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Carcinogenicity: Water Soluble Nickel compounds



- **Oral:** A guideline compliant oral study with Ni sulfate hexahydrate in Fischer rats was negative for tumor induction (Heim et al., 2007)
 - Accepted as negative by U.S. EPA and EU Commission

- Negative animal study together with negative epidemiological studies (ICNCM, 1990) confirm the lack of oral carcinogenic potential of water soluble nickel compounds

[water soluble Ni = worst case for all nickel-containing substances]

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Consistency of Carcinogenicity Evidence



	Animal Data	Human Data	Weight Of Evidence as a human carcinogen
Inhalation			
Ni Sub sulfide	++	+	+
Ni Oxides	+	+/-	+
Ni Metal	-	-	-
Water Soluble Ni compounds			
Oral			
Water Soluble Ni compounds	-	-	-

Inconsistent one

Different MOA?

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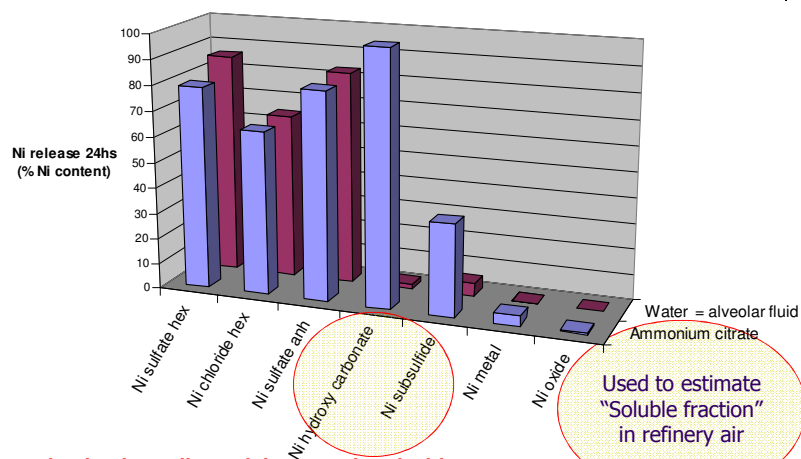
Carcinogenicity: Soluble Nickel compounds



- Inhalation: Inconsistencies in data. Negative animal data; mostly positive epidemiological studies
 - In 2004 a group of EU Experts looked just at epi data and considered it sufficient to indicate a known human carcinogen
 - However, inconsistent results among cohorts and uncertainties in speciation of exposures and attribution of risks have been worrisome
 - Speciation methods may have assigned sulfidic and some oxidic Ni exposures to the soluble aerosol fraction
- Weight of evidence evaluation by Goodman et al. (Crit Rev Tox, 2009) concluded that Soluble Ni can be considered as a suspect human respiratory carcinogen, with a different mode of action than that of insoluble Ni compounds
- Planned work on bioavailability model testing should help clarify the existence of a different mode of action for soluble nickel compounds

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Exposure Assessment: Speciation



Can mistakenly attribute risks associated with these compounds to water soluble compounds

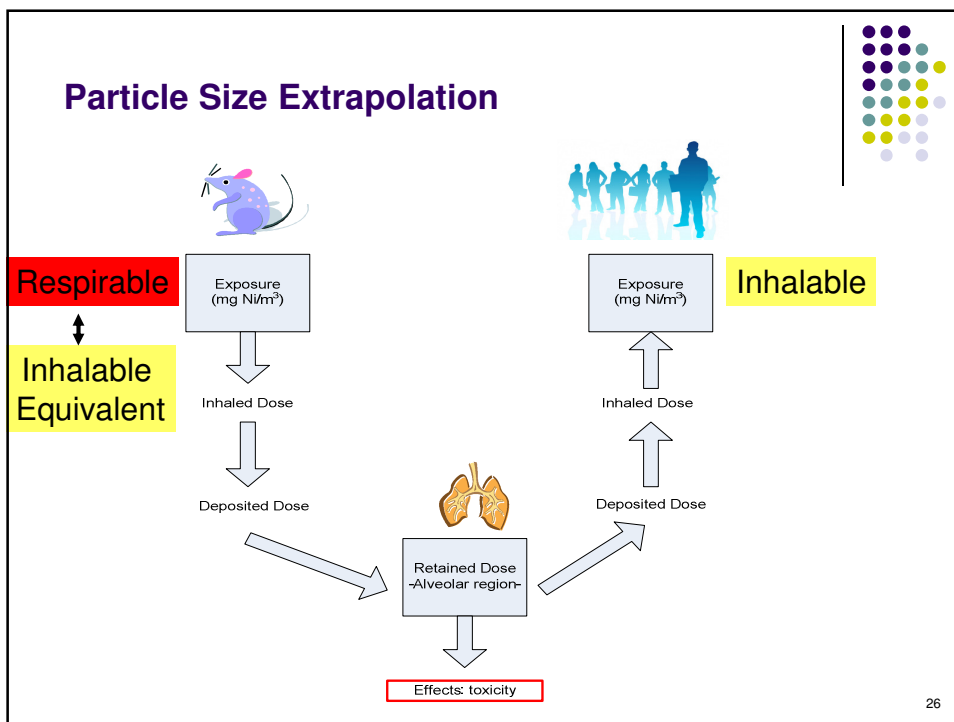
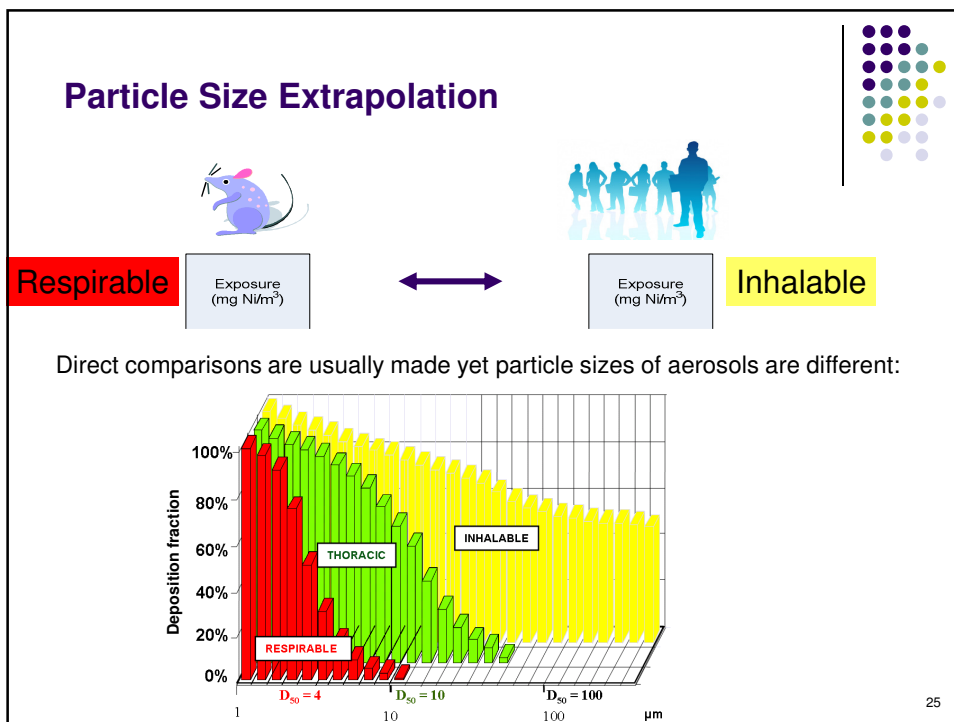
Oller et al. 2009

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Exposure Assessment: speciation

- Combined use of XANES and XRD will yield more reliable speciation results (Tirez et al., 2010; Vale ongoing work)
- Current results indicate that there are problems with the leaching method for assessing workplace exposure to various forms of nickel:
 - overestimation of some chemical forms (e.g., soluble nickel) and underestimation of other forms (e.g., oxidic nickel)
- Recently completed work aims at developing a combination of physical and chemical methods (e.g., using water to extract the soluble fraction) to assess workplace exposure to particular forms of nickel.
- This work will lead to a re-examination of the influence of speciation on attribution of cancer risks


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Particle Size Extrapolation

Without Particle size Adjustment...

No tumors




0.1 mg Ni/m³

Exposure (mg Ni/m³)

↔


Tumors



0.06-0.8 mg Ni/m³

Exposure (mg Ni/m³)

It may be concluded that rats exposed to nickel sulfate did not get tumors because their exposures were lower than those experienced by workers.




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Particle Size Extrapolation

With Particle size Adjustment...

No tumors



0.7 mg Ni/m³


Exposure (mg Ni/m³)

Inhaled Dose

↓

Deposited Dose

Tumors




0.06-0.8 mg Ni/m³

Exposure (mg Ni/m³)

Inhaled Dose

↑

Deposited Dose




Retained Dose -Alveolar region-

↓

Effects: toxicity

When equivalent exposures are compared, another explanation for the different results is needed (e.g., tumor promoter?)



Oller and Oberdörster, 2010

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Reproductive & Developmental Toxicity: animals & humans



- **Background:** Oral exposure to water soluble Ni can cause increased perinatal mortality in rats (developmental toxicity)
- The Siglin (2000) one- and two-generation studies identified a threshold of 2.2 mg Ni/kg bw/day for these effects
- Are the effects reported in rats also observed in humans? Is the window of susceptibility or mode of action relevant for humans?

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Reproductive & Developmental Toxicity: animals & humans



- A preliminary report from a Russian Ni refinery (Chaschin et al., 1994) indicated increased reproductive risks in female workers that the authors attributed to nickel exposure
 - A well-conducted epidemiological study (E Nieboer -McMaster Univ) did not show an association between inhalation exposure to Ni (systemic dose) and a variety of adverse reproductive and developmental outcomes (Vaktskjold et al., 2006, 2007, 2008a,b)
- Risks predicted based on animal data did not realize in heavily exposed nickel workers!
- Planned research in next 5 years looking at mode of action associated with adverse outcomes in rats (dose-response, timing, cell-specificity) will determine whether effects observed in rats are relevant to humans

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Science Update: Nickel

- Read-across for hazard assessment and risk characterization of nickel substances
 - Bioaccessibility studies in synthetic fluids, validation with in vivo studies, route of exposure specificity
 - Bioavailability model for respiratory cancer: expert workshop and planned research
 - Toxicokinetic human studies to define blood and urine levels corresponding to oral intake levels protective of reproductive effects observed in animals

Bioaccessibility studies in synthetic fluids, in vivo validation, route of exposure specificity



Background

- What is read-across?
 - It is the process by which an assessment/prediction of hazard for data-poor substance is made. It uses existing hazard information from source substances and some measure of the likelihood that the data-poor substance will behave in the same manner as the source substance.
- What are water solubility and bioaccessibility studies?
 - Water solubility of nickel substances provides information on the relative release of Ni ion in pure distilled water. Bioaccessibility studies provide information on the relative release of Ni ion in synthetic biological fluids relevant to different routes of exposure. [Bioaccessibility provides an estimate of bioavailability](#)
 - Sweat = dermal
 - Stomach/ intestinal = oral
 - Lung fluid/ interstitial = inhalation

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Bioaccessibility studies in synthetic fluids, in vivo validation, route of exposure specificity



- In EU, the Danish EPA read-across for hazard identification/classification from source compounds to data-poor Ni substances based on water solubility alone [all endpoints]
- Under REACH, the nickel industry developed a more scientifically justified read-across method based on bioaccessibility in biological fluids, route of exposure specificity, and in vivo validation.
- Analyses of 4 source reference substances and ~10 data poor substances have been completed (SOT, 2011; manuscripts in preparation)

Data-rich substance = reference substance = source substance
 Data-poor substance = target substance = substance being studied

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Bioaccessibility studies in synthetic fluids, in vivo validation, route of exposure specificity



- In several instances this approach suggests that changes to hazard identifications (i.e., classifications) that were based on water solubility read-across are warranted:
 - More stringent classifications (e.g., Ni hydroxycarbonate, acute inhalation toxicity)
 - Less stringent classifications (e.g., Ni dihydroxide, acute oral toxicity)
- In some instances read-across for one route of exposure was different from read-across for another route
 - Nickel hydroxycarbonate read across from:
 - Ni sulfate for oral exposure,
 - Ni subsulfide for inhalation exposure
 - Ni oxide for dermal exposure

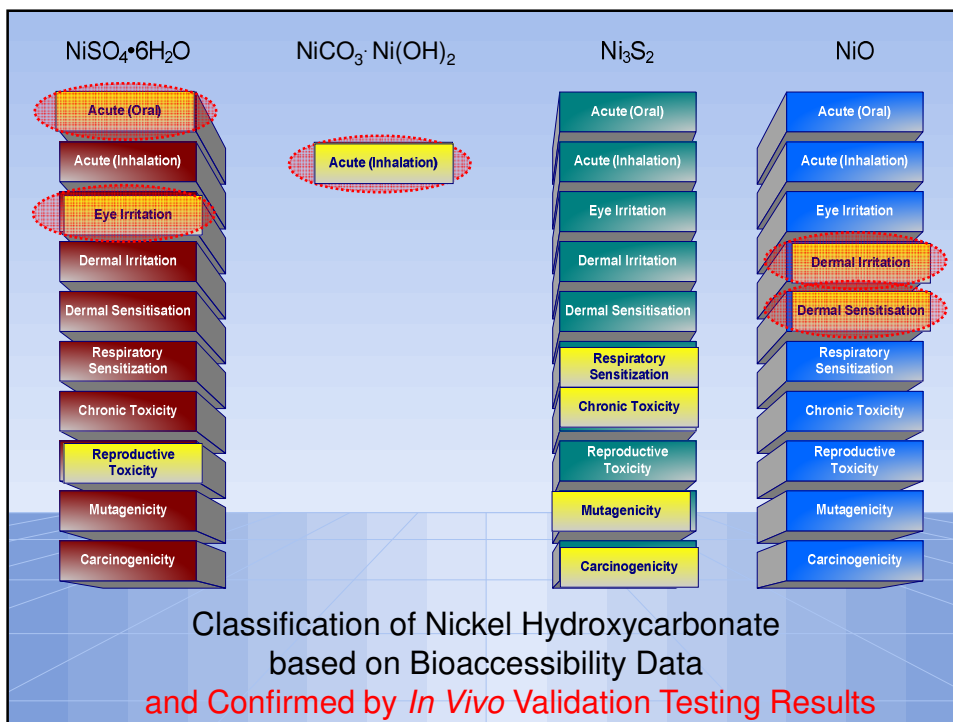
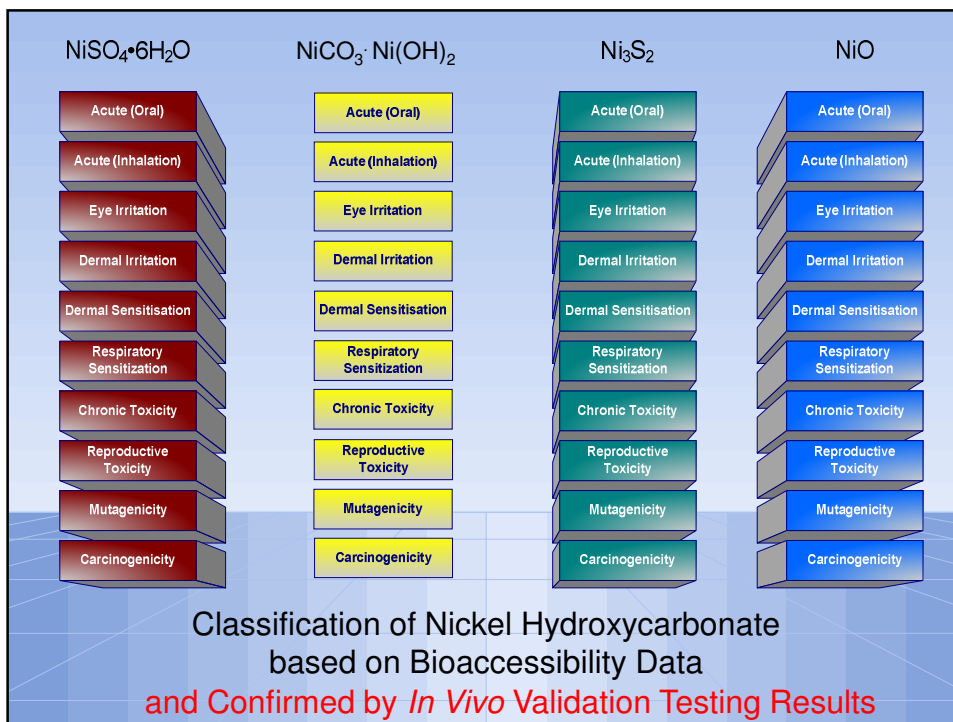
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Bioaccessibility studies in synthetic fluids, in vivo validation, route of exposure specificity



- These changes affected health endpoints like dermatitis, respiratory sensitization, acute toxicity, and reproductive toxicity
- These changes **did not affect carcinogenicity** hazard identification since at the moment all nickel compounds are considered as known human carcinogens. However, proper read-across based on bioaccessibility data and the bioavailability model can be used for more accurate risk characterization for cancer

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Changes in EU/CLP Classifications Based on New Data Generated Under REACH



- REACH work plan is in place to implement new self-classifications in 2012 (for endpoints/substances where there is no existing classification)
- Changes in existing classifications will require proposal by a Member State Competent Authority (MSCA). This activity falls under a separate NI initiative.
- Successful support for changing classifications **will require acknowledgement and acceptance of the bioaccessibility-based read-across methodology at the regulatory level**
- NI/NiPERA continues to work with other commodities to advance this initiative

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Examples of Current Activities Supporting the Bioaccessibility-Based Approach



- **Bio-elution roadmap**
 - Communication program to inform regulators about bioaccessibility work and its utility in assessments of various metals issues (substances, alloys, ores & concentrates, etc.)
 - Lead by Eurometaux
- **ECHA Training Workshops: “Metals Evaluation Under REACH”**
 - First workshop held in March 2011, organized by Eurometaux
 - Aim to familiarize ECHA and Member State evaluators with assessment needs, methodologies and approaches for metals and inorganic metal compounds based on proven cases and examples in order to optimize future evaluations

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Examples of Current Activities Supporting the Bioaccessibility-Based Approach



- **Inter-laboratory validation study**
 - Program to validate the bio-elution test methodology between laboratories
 - Includes metal, metal substance, alloy, and ore/concentrate samples
 - Managed by Eurometaux + 4 sponsoring associations, including NiPERA
- **HERAG Alloys Fact Sheet, Guidance on Ores & Concentrates, etc.**
 - Guidance documents seeking to establish a unified approach for specific types of materials
 - Lead by ICMM

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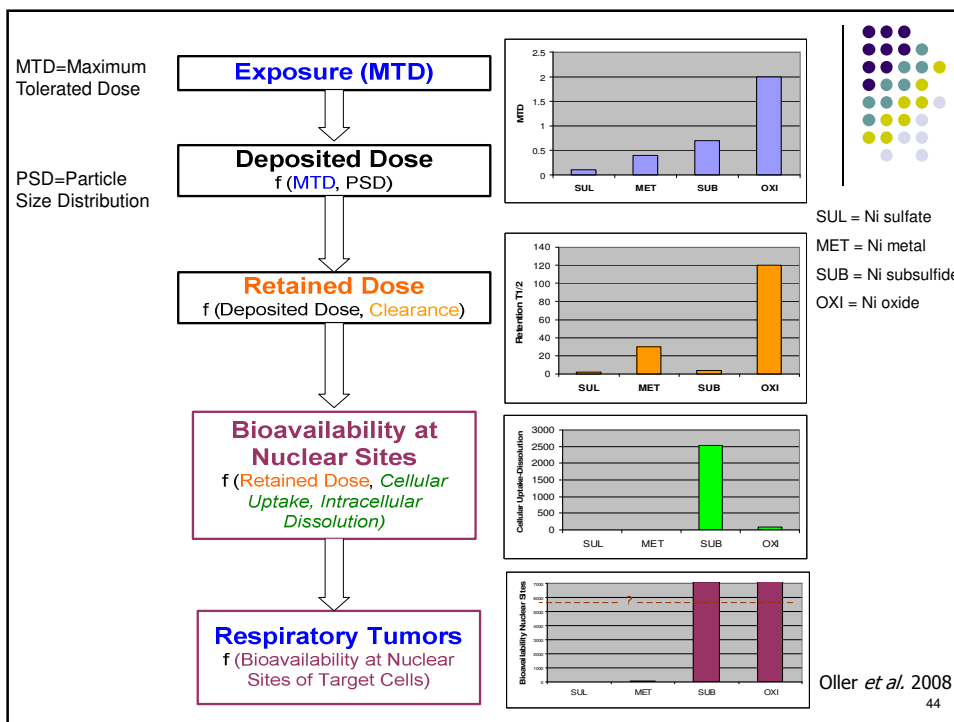
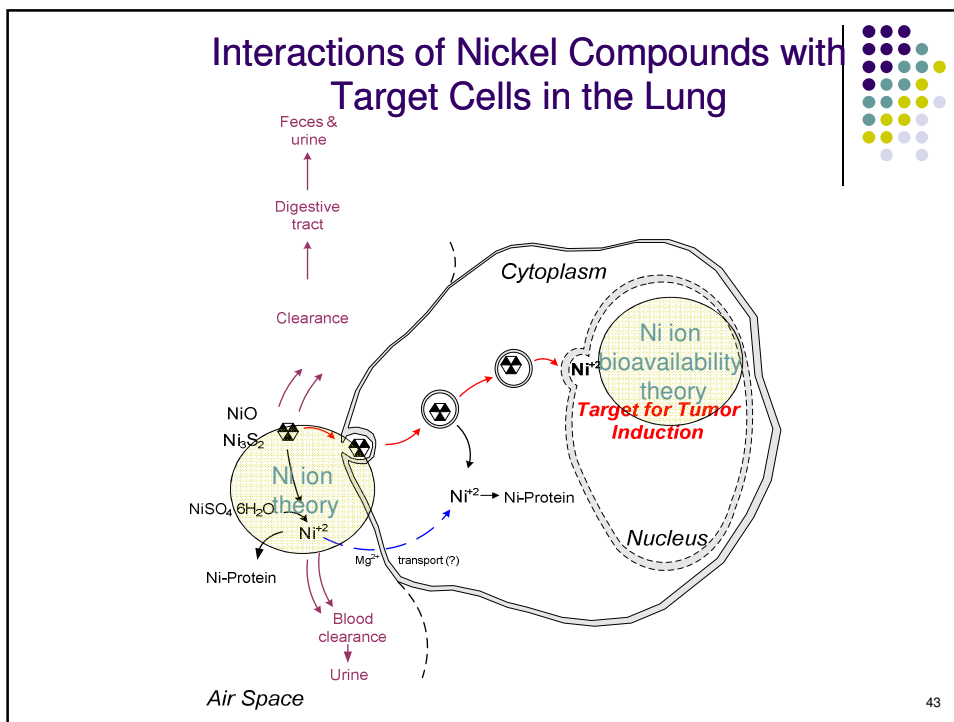
Bioavailability model for respiratory cancer: expert workshop, manuscript, and planned research



Background:

- **Ni ion theory:** suggests that Ni (II) ion is responsible for respiratory tumor formation; all substances containing nickel should be considered as carcinogenic – *the more Ni a substance releases, the more carcinogenic it will be*
- **Ni ion bioavailability theory:** proposes that only those Ni substances that can result in levels of Ni (II) at target sites of respiratory cells that are sufficiently high to initiate tumors should be considered as carcinogenic (*complete carcinogens*)

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Bioavailability model for respiratory cancer: expert workshop, manuscript, and planned research



- A draft paper comparing and discussing these hypotheses provided the basis for discussions at an expert workshop organized by *TERA* in Feb 2010
- Experts represented academia, industry and regulatory bodies (MAK, SCOEL, EPA)
- Experts unanimously agreed that [the bioavailability model](#) is a refinement of the Ni ion theory that better represents our current knowledge about Ni and cancer
- The bioavailability model helps integrate negative and positive animal results for various nickel substances although both hypotheses are compatible with the epidemiological data

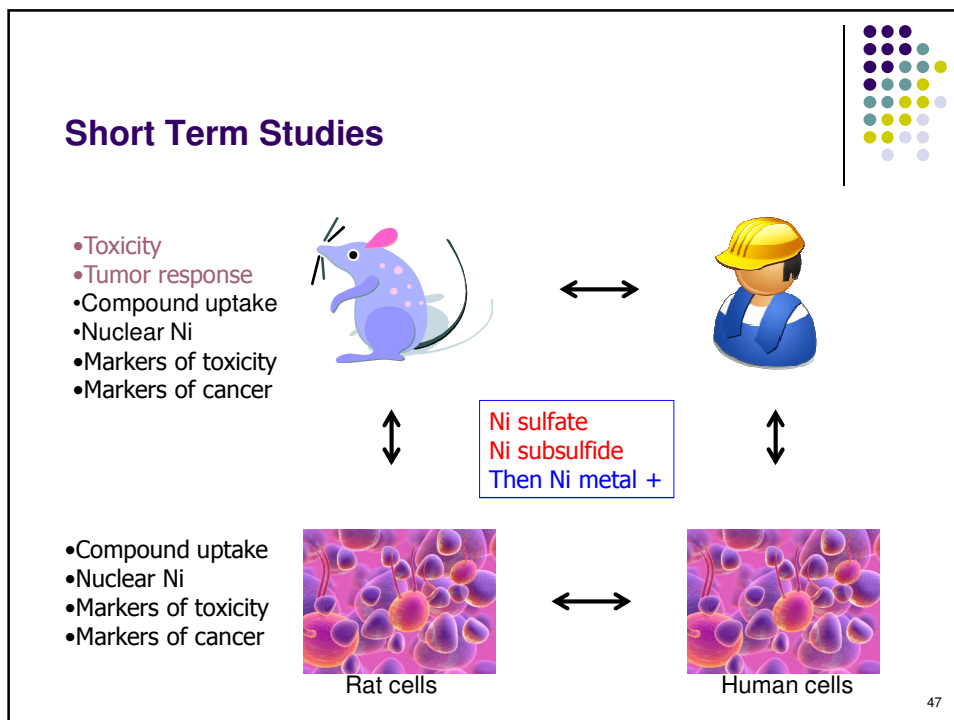
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Bioavailability model for respiratory cancer: expert workshop, manuscript, and planned research



- The TERA workshop helped identify data gaps in the model and made suggestions for future research
- This research will evaluate whether more than one mode of action for increased respiratory cancer risk by nickel compounds is possible
- The report from this workshop is available at TERA's website. Experts' comments were incorporated into Goodman et al. paper (Crit Rev Tox, 2011)

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Bioavailability model for respiratory cancer: expert workshop, manuscript, and planned research

- Contracts signed 1Q 2011. Work will include:
 - In vitro air/liquid interface studies of Ni uptake and Ni biomarkers
 - Comparisons among Ni substances, among animal species, between in vitro and in vivo conditions

• Results will allow the development of a kinetic model that will help assess cancer risks for data-poor Ni substances and mixtures

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Toxicokinetic human studies



- Reproductive effects are the only systemic effects of concern for workers
- An oral reference value of **20 µg Ni/kg body weight** for the daily amounts of nickel to which females can be exposed for a lifetime without ill effects can be calculated based on the reproductive effects observed in animals
- Some regulatory bodies calculate 10 µg Ni/kg body weight
- These values are relevant to workers and to the general public

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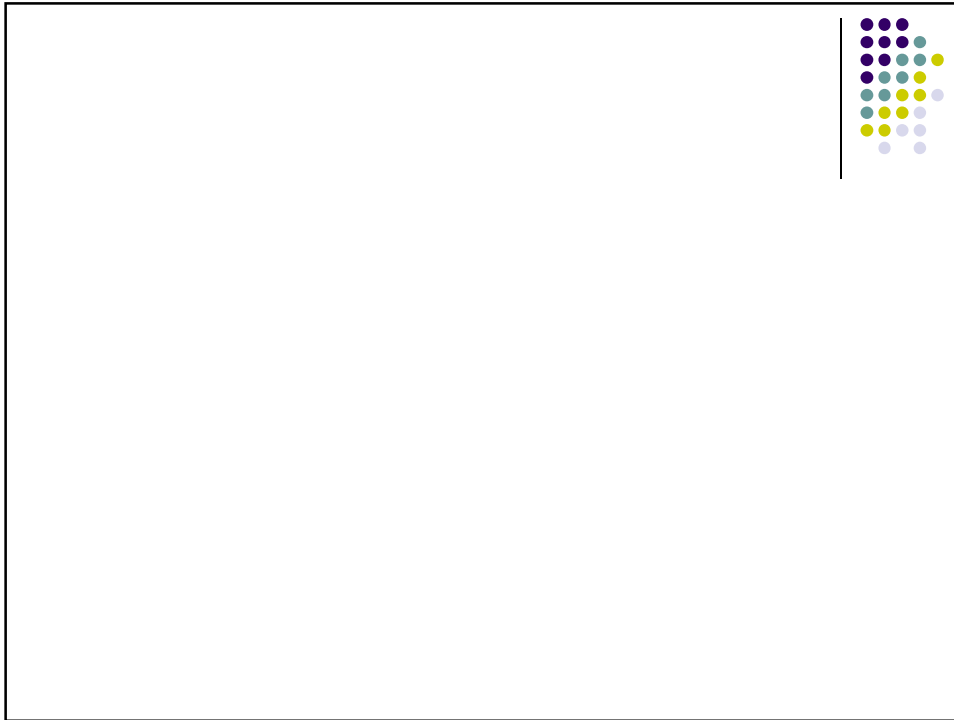
Toxicokinetic human studies



- Can a biological action or reference value be derived for the workplace based on the oral reference values of **10 or 20 µg Ni/kg body weight** and human toxicokinetics data?

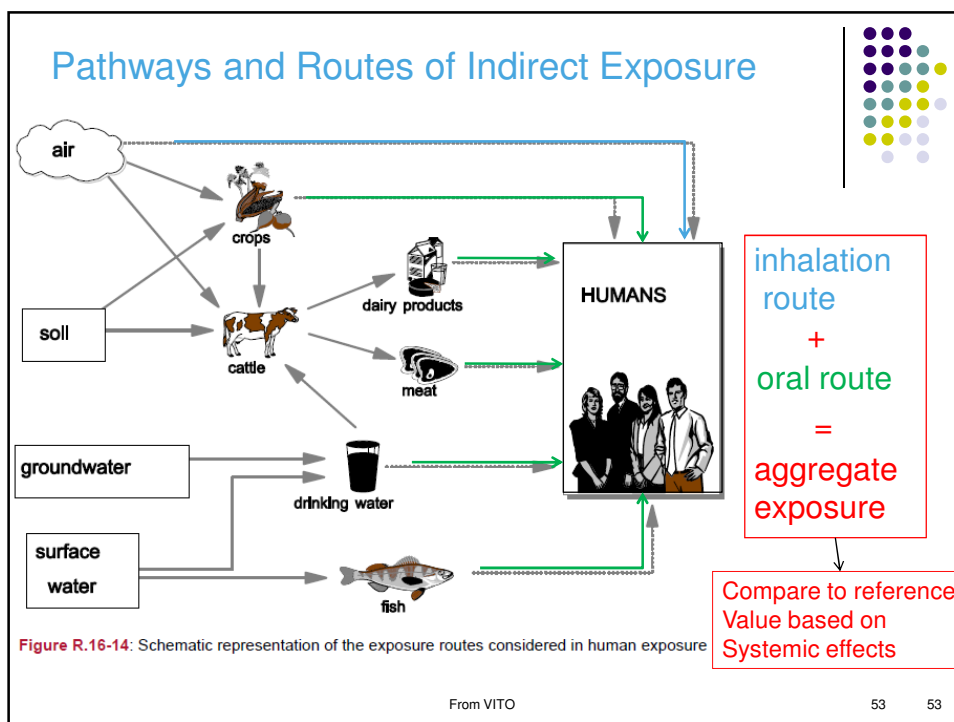
- An ongoing study is looking at the blood and urinary nickel levels of volunteers ingesting a single dose of 5, 10, and 20 µg Ni/kg. This will allow the derivation of a urinary nickel value that will be protective of workers regarding any possible reproductive effects and/or systemically-elicited dermal effects

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Agenda

- What recent research could impact the setting of reference values for the general public from a Human Health point of view?
 - Drinking water
 - Ambient air



Reference Value for Water

- WHO in 2005 set a drinking water standard of 70 $\mu\text{g Ni/liter}$ based on Tolerable Daily Intake (TDI) of 11-12 $\mu\text{g Ni/kg body weight/day}$, considering two different endpoints:
 - Oral elicitation of dermatitis in sensitized individuals
 - Human study
 - Sensitive subpopulation
 - LOAEL: 12 $\mu\text{g Ni/kg body weight/day}$
 - Adverse reproductive effects in rats after oral exposure
 - Animal study
 - Serious health endpoint
 - NOAEL: 1.1 $\text{mg Ni/kg body weight/day}$

Reference Value for Water



- California OEHHA in 2011 set a chronic oral Reference Exposure Level (REL) = 11 μg Ni/kg body weight/day
 - Adverse reproductive effects in rats after oral exposure
 - Animal study
 - Serious health endpoint
 - NOAEL: 1.1 mg Ni/kg/day

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Reference Value for Water



- Completed or Ongoing research of relevance: none
- Datagaps:
 - What is conclusive NOAEL for reproductive effects in rats?
 - Reproductive study to clarify if the conclusive NOAEL is 1.1 or 2.2 mg Ni/kg/day
 - Scheduled to start in 2012
 - Could affect some but not all drinking water values

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Natural Sources of Nickel in the Atmosphere



- Natural source contributions of nickel to the ambient atmosphere range from 15% to 35% of total global atmospheric emissions
- Nickel compounds of natural origin are likely to be complex silicate-oxides which are insoluble in water
- Natural sources of nickel in the atmosphere come from windblown dusts, volcanic fallout, and burning of vegetation (e.g., forest fires)



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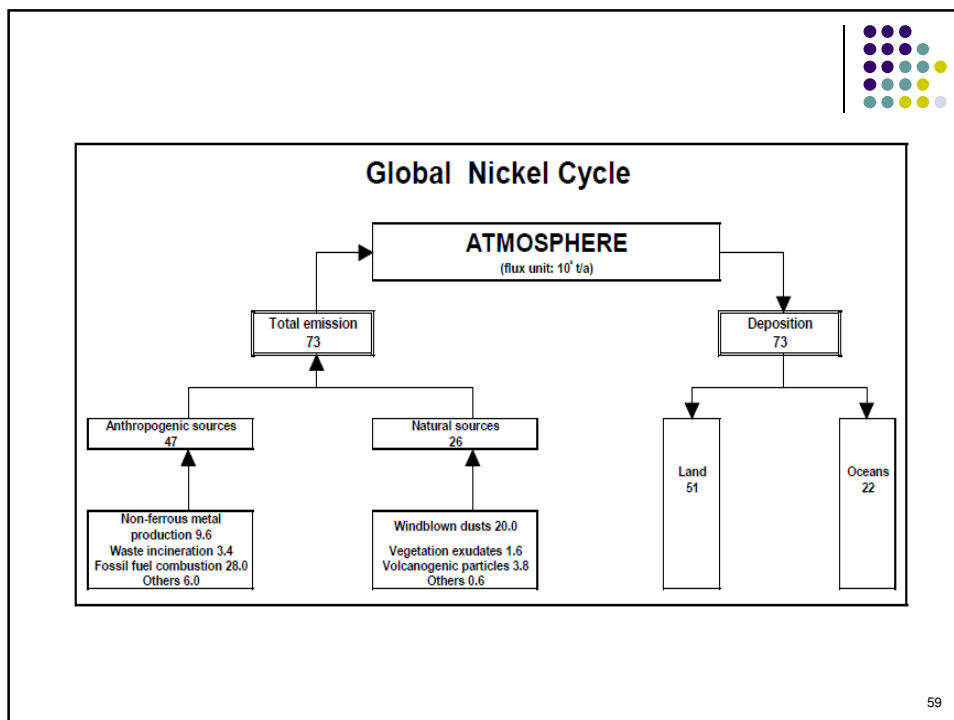
Anthropogenic Sources of Nickel in the Atmosphere



1. Combustion of oil for heat or power generation (1 to 80 mg/kg)
2. Coal combustion for heat or power generation (~ 100 mg/kg)
3. Incineration of waste and sewage sludge
4. Steel manufacture, electroplating, and nickel mining and primary production.



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Reference value for ambient air

- EU Ambient Air Guidance value (Directive 2004-107 EC): **20 ng/m³**.
 - Based on carcinogenic and non-carcinogenic respiratory effects
 - Reporting measured data. Not an Env Quality Standard
- 2010 EU REACH: long term inhalation DNEL for general public (man via the environment): **20 ng/m³**.
 - Based on Ambient Air guidance
 - Compliance based on measured/modeled at nearest population
 - Tiered approach with increasingly refined models

Reference value for ambient air



- 2009 Ontario (Canada)
 - A half-hour standard of $0.3 \mu\text{g}/\text{m}^3$ for nickel and nickel compounds (PM10), based on carcinogenic and non-carcinogenic effects
 - A 24-hour average AAQC (Ambient Air Quality Criterion) and standard of $0.1 \mu\text{g}/\text{m}^3$ (PM10); based on carcinogenic and non-carcinogenic effects
 - Annual average AAQC of $20 \text{ ng}/\text{m}^3$, (PM10) based on the carcinogenic and non-carcinogenic effects
 - Compliance mandates only modeled data at plant's border!

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Reference value for ambient air



- California OEHHA (Office of Environmental Health Hazard Assessment)
 - 1-hour REL (inhalation reference exposure level) = $1.1 \mu\text{g}/\text{m}^3$; bronchial constriction in human volunteers with asthma
 - 8-hour REL = $80 \text{ ng}/\text{m}^3$; immunotoxicity in rats
 - chronic REL = $14 \text{ ng}/\text{m}^3$; chronic respiratory toxicity in rats

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Reference value for ambient air



- Texas CEQ
 - Acute ESL = 330 ng/m³; bronchial constriction in human volunteers with asthma
 - Chronic ESL (24h) = 70 ng/m³; based on chronic active inflammation and associated lesions in rats
 - Chronic ESL(linear) (24h) = **60 ng/m³**; lung cancer in workers based on unit risk 1.7×10^{-4} per ug/m³ and tolerable risk of excess 1/10⁵

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Representative Annual Mean Total Nickel Concentrations in Ambient Air



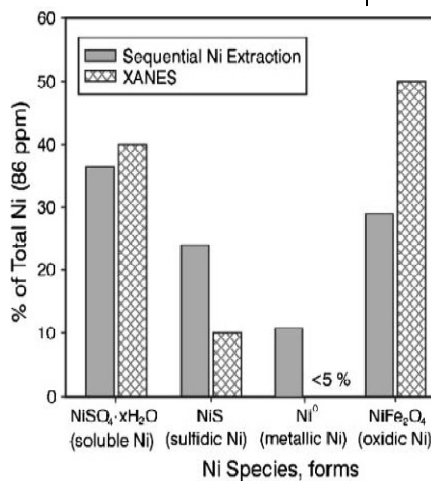
- Remote areas - 1 ng/m³ or less
- Rural areas - 0.5-2 ng/m³
- Urban areas - 2-20 ng/m³
- Heavily industrialized areas - 10 to 100 ng/m³ or higher

[It is interesting to note that cigarette smoke is an elective contribution of about 0.04-0.58 µg of nickel per cigarette]

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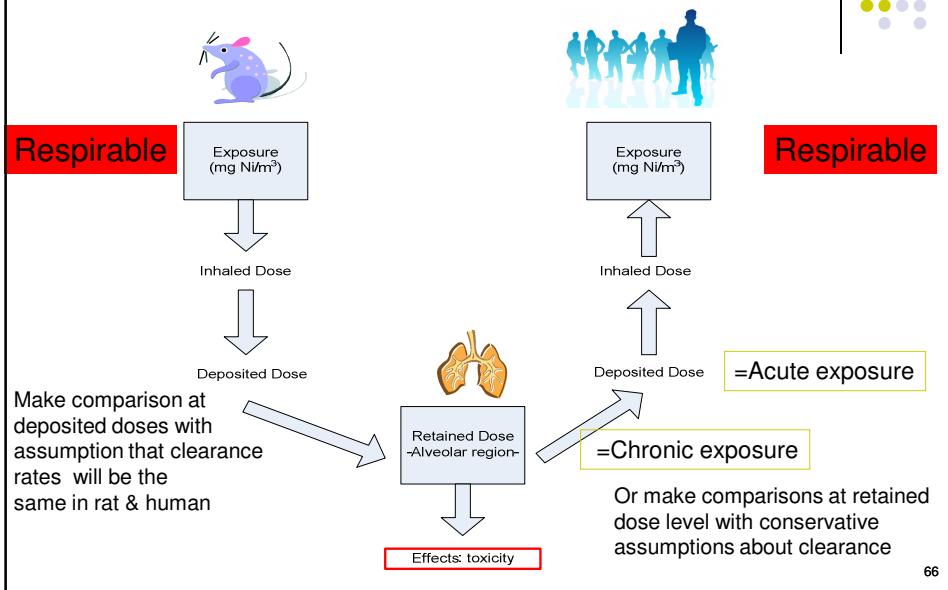
Identity of Nickel Compounds in Ambient Air

- Evaluations of the PM₁₀ particulates from non-hot spot associated urban ambient air was conducted by Galbreath et al., 2003
- XAFS demonstrated that the predominant species is hydrated nickel sulfate (NiSO₄·xH₂O) and Ni-bearing spinel such as trevorite (NiFe₂O₄)

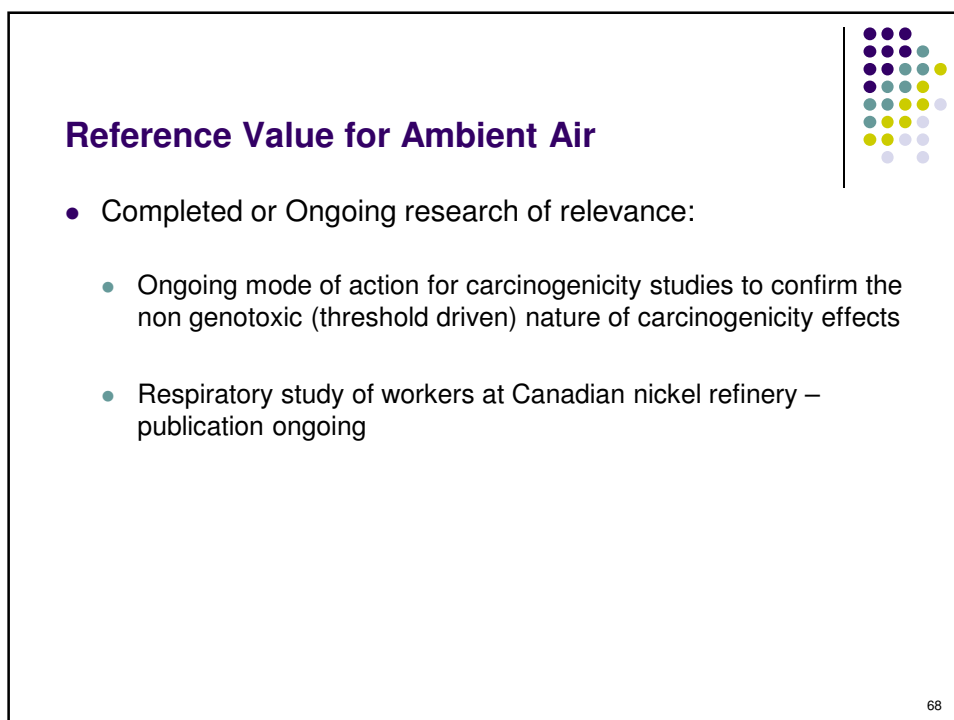
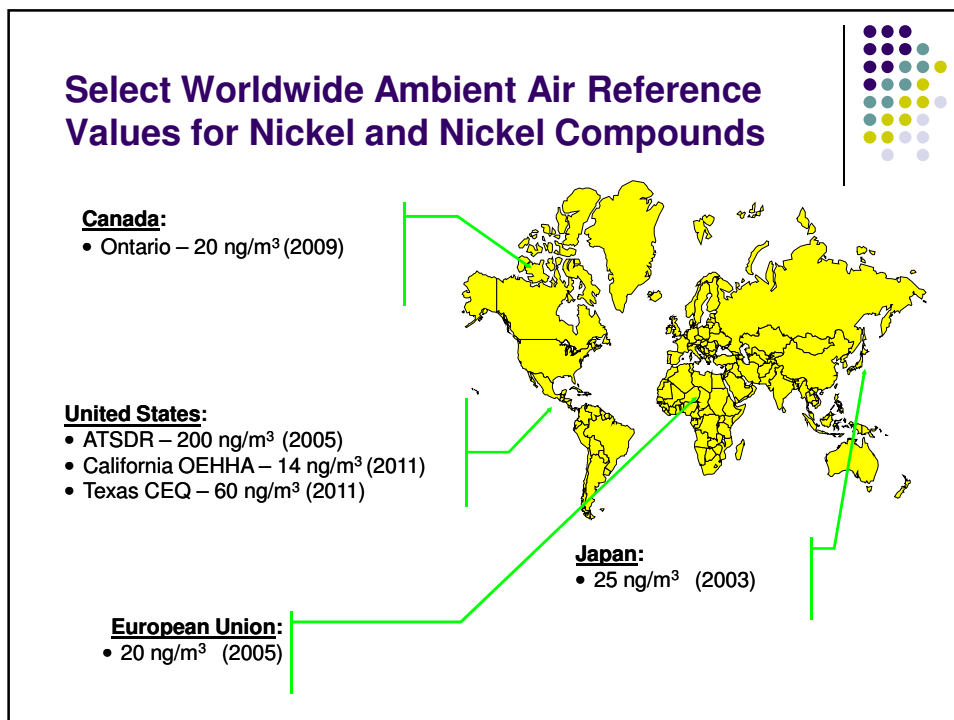


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Particle Size Extrapolation



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Reference Value for Ambient Air



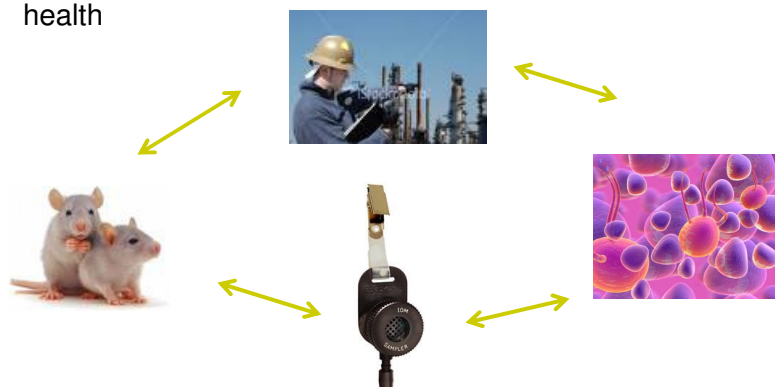
- Datagaps not addressed by ongoing NiPERA work:
 - A published solid study of dose-response for adverse respiratory effects of nickel in a human population
 - Improved speciation methods for workplace exposure
 - Clarification of cancer risks associated with lateritic operations
 - Good quality exposure data

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Summary Human Health Research



- Through NiPERA, the nickel industry continues its commitment to a broad scope, high quality research program to improve workplace exposure assessment and health monitoring, as well as refining the risk assessment for human health



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