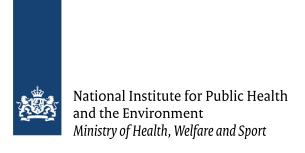


## Update of ecological risk limits of nickel in soil

RIVM Letter report 2015-0137 A.J. Verschoor



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## Colophon

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This investigation has been performed by order and for the account of the Ministry of Infrastructure and the Environment, within the framework of the project M607711 Soil quality and Risk assessment.

This is a publication of:

National Institute for Public Health
and the Environment
P.O. Box 1 | 3720 BA Bilthoven
The Netherlands
www.rivm.nl/en

## Publiekssamenvatting

## Actualisatie van risicogrenzen voor nikkel in bodem

Het RIVM doet een voorstel voor nieuwe Nederlandse risicogrenzen voor nikkel in de bodem. Deze grenzen geven aan bij welke concentraties nikkel negatieve effecten op het ecosysteem in de bodem kan veroorzaken. Het voorstel is gebaseerd op de meest recente data en inzichten in Europa.

Met deze grenzen worden de risico's van een nikkelverontreiniging realistischer ingeschat. Als gevolg daarvan zullen de normen strenger zijn voor enkele typen bodems, zoals bodems waar meer nikkel uit vrijkomt. Voor het merendeel van de bodems zullen de normen soepeler uitvallen.

Nieuw is dat bij deze normen rekening wordt gehouden met de mate waarin bodemorganismen worden blootgesteld aan de vervuilende stof, de zogeheten biobeschikbaarheid. Uit de bodem komt namelijk niet de totale concentratie vrij, omdat een deel aan bodemdeeltjes 'vast blijft zitten'. In welke mate dat gebeurt, is afhankelijk van de samenstelling van de bodem en verschilt daarom per bodemtype. In het onderzoek is ook een correctiemethode ontwikkeld waarmee de totale concentratie van een stof in de bodem kan worden omgerekend naar de concentratie die vrijkomt.

De huidige bodemnormen voor nikkel dateren van 2001 en zijn gebaseerd op drie testgegevens met regenwormen. De laatste 15 jaar is er binnen de Europese Unie veel experimenteel onderzoek gedaan naar de mate waarin nikkel giftig is voor bodemorganismen. Het RIVM heeft op basis van deze 184 testresultaten, verdeeld over 43 verschillende soorten organismen of bodemprocessen, nieuwe maximaal toelaatbare risico- (MTR) en ernstige risicoconcentraties (ER) in de bodem afgeleid.

Kernwoorden: nikkel, biobeschikbaarheid, veroudering, bodem, normstelling

#### **Synopsis**

#### Update of ecological risk limits of nickel in soil

RIVM proposes new Dutch risk limits for nickel in soil. These limits indicate the concentrations above which nickel can have negative effects on the soil ecosystem. The proposed risk limits are derived based on the latest insights in Europe on risk assessment.

By using these new risk limits, the risks of nickel pollutions are predicted more realistically. As a result, the standards will be more stringent for some types of soils, such as soils that release more nickel but for the majority of the soils, the risk limits are lower than the current values.

New to these standards is that bioavailability of the contaminant is taken into account. Part of the total contaminant concentration is trapped in the soil. The soil therefore does not release all of the contaminant. To what extent this occurs is dependent on the composition of the soil, and varies per type of soil. This study therefore also developed a correction method with which the total concentration of a substance can be converted into the a bioavailable concentration.

The current soil standards for nickel date from 2001 and are based on three tests with earthworms. The last 15 years there has been much experimental research on the nickel toxicity on soil organisms within the European Union. Based on 184 test results, spread over 43 different species of organisms or soil processes, RIVM derived new maximum permissible risk (MPR) values and serious risk concentrations (ER) for soil.

Keywords: nickel, bioavailability, aging, soil, risk limits

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## Summary

This report updates the scientific background of maximum permissible added concentrations (MPC $_{added}$ ), maximum permissible total aged concentrations (MPC $_{total}$ ), serious risk addition (SRC $_{added}$ ) and serious risk concentrations (SRC $_{total}$ ) of nickel in soil. The last update dated back to 2001, when only 3 terrestrial ecotoxicity studies were used. In 2008 the EU Risk Assessment Report of nickel was finalized, which evaluated an overwhelming number of recent terrestrial ecotoxicity studies of nickel. This information formed the basis of the derivation of updated MPC and SRC values. Besides updating the generic ecological risk limits, we adopted the method proposed in the EU-RAR, to calculated soil-specific risk limits. Based on a total of 184 terrestrial soil toxicity data, the MPC and SRC values are computed for added concentrations, and soil specific ecological risk limits expressed as total aged concentrations as a function of the cation exchange capacity.

The  $MPC_{added}$  and  $SRC_{added}$  are respectively 10 and 144 mg Ni/kg. Because they reflect bioavailable fractions, they should be compared with concentrations that are extracted with 0.43 M HNO<sub>3</sub> or another mild extractant.

Nickel concentrations added in toxicity tests are corrected for ageing in order to reflect field conditions. Ageing is dependent of the pH in the test and a correction is applied for tests with a exposure time of less than 120 days, to convert the added NOEC or EC10 value to a value that is relevant for the chemical availability under field conditions. The bioavailability of nickel is further determined by the cation exchange capacity of the soil. If measured CEC values are not available, the CEC may eventually be computed by an appropriate transfer function. Seven species-specific regression models for bioavailability were present: for plants (2), soft-bodied (1) and hard-bodied invertebrates (1) and enzymatic and microbial processes (3). These regression functions were used to calculate soil-specific NOEC or EC10 values for each species and each test. By read-across (looking at similar uptake pathways) these models are assigned to species for which no specific model was present. In order to enable routine calculation of soil-specific risk limits, we derived simple soil-type corrections for MPC and SRC:

$$MPC_{total} = 2.7 \times CEC$$
 and  $SRC_{total} = 22 \times CEC$ .

The calculated risk limits reflect the total aged nickel concentrations, therefore it should be compared with concentrations that are extracted with aqua regia or a comparable "total" extraction method. For a Dutch standard soil with 10% organic matter and 25% clay a MPC<sub>total</sub> of 81 mg Ni/kg soil was proposed, and a SRC<sub>total</sub> of 660 mg Ni/kg soil.

The relation is valid for soils with CEC between 1.8 and 52.8 cmol/kg. This is the CEC-range in soils used for the BLM-derivation. For higher CEC values a cut-off value of 52.8 cmol/kg could be used, or MPC and SRC should be maximized to values of respectively 150 and 1200 mg Ni/kg.

#### 1 Introduction

#### 1.1 Background of the report

Ecological risk limits play an important role in the Dutch soil protection policy. Together with human health related risk limits, they are used for assessment of soil quality in the context of decision making on remediation, re-use of soil and risk management in case of chemical spills or other emergency situations.

The derivation of most risk limits was performed in 2001 [1], mostly based on data from ecotoxicity tests that had been evaluated previously [2-6], but using an adapted methodology. Since then, risk limits for some (groups of) compounds have been updated, by adding new data to the already available datasets and taking into account methodological developments [7, 8], but the majority of the currently used ecological risk limits originates from the 2001-report. Upon request of the Dutch Ministry of Infrastructure and the Environment, it was investigated to what extent the existing ecological risk limits for soil can (should) be improved to meet new scientific developments and to solve practical problems that arise when using those risk limits in practice [9]. As a follow-up, a scoring method was developed to rank the existing ecological risk limits with respect to uncertainty related to data quality and changes in the methodology [10]. Based on this evaluation arsenic, nickel, drins and DDT/DDE/DDD were selected for a closer review in 2014.

This report deals with the evaluation of new data and methodologies in order to derive up-to-date ecological risk limits for nickel in soil. Arsenic, drins and DDT/DDE/DDD are evaluated in separate reports. Before focusing on nickel, the following sections provide some background information on the risk limits considered in this report and the aspects that are considered most important when discussing the scientific validity of the previously derived risk limits.

#### 1.2 Relevant risk limits

The relevant ecological risk limits in the context of this report are the Maximum Permissible Concentration (MPC) and the Serious Risk Concentration (SRC).

The MPC $_{soil}$  is defined as the concentration in soil at which no negative effect on ecosystems is expected [11, 12]. The MPC $_{soil}$  is derived considering direct ecotoxicity to soil organisms and/or bacterial or enzymatic processes and then indicated as MPC $_{soil, eco}$ . If deemed necessary in view of compound characteristics, secondary poisoning of predatory birds and mammals may also be considered by deriving an MPC $_{soil, secpois}$ . Considering the protection level and methodology, the MPC $_{soil, eco}$  is comparable to a Predicted No Effect Concentration (PNEC) as derived in various international frameworks [13, 14].

The  $SRC_{soil, eco}$  is the environmental concentration at which possibly serious ecotoxicological effects on soil organisms and/or processes are to be expected, meaning that 50% of the species or processes is potentially affected. The  $SRC_{soil}$  is usually derived for direct ecotoxicity to soil organisms and/or processes only. In some cases, secondary poisoning was taken into consideration for derivation of the SRC [7, 15].

For Nickel secondary poisoning is deemed not relevant and is therefore not included in de SRC [17]. Detailed guidance for the derivation of the MPC and SRC for soil is given in [16].

#### 1.3 Using ecotoxicity data: data quality and treatment of results

The derivation of ecological risk limits basically follows a four step approach: collection of literature, evaluation of the scientific reliability, selection of relevant endpoints and using the endpoints to derive the risk limits. It can be imagined that if new data were generated since the last evaluation, this may potentially lead to a different result. This is the case for nickel, for which a huge amount of terrestrial ecotoxicity data has become available since 2001.

However, even if this is not the case and the same literature data would be used, newly derived risk limits will differ from those derived in 2001. Re-evaluation of the literature according to current insights may lead to different conclusions regarding the quality of the data, and the way risk limits are derived given a certain dataset has been adapted in several ways during the past years.

#### 1.3.1 Changes in data treatment

Once reliable and relevant ecotoxicity endpoints are selected, the available data can be used in different ways to derive risk limits. If the number of data is limited, an assessment factor is put on the lowest endpoint. If more data are available, statistical extrapolation using Species Sensitivity Distributions (SSDs) can be applied. Changes in the requirements for using the latter were identified as an important factor when considering the uncertainty related to the previously derived risk limits [10]. An SSD displays the fraction of species potentially affected as a function of the exposure concentration. The Hazardous Concentration for 5% and 50% of the species (HC5 and HC50), are used as input for the MPC<sub>soil, eco</sub> and SRC<sub>soil, eco</sub>, respectively. Application of SSDs for terrestrial species is possible in rare cases only. In 2001, SSDs were applied when data for at least four taxonomic groups were available<sup>1</sup>, regardless of the trophic levels represented in the dataset. The HC5 and HC50 were used without any additional assessment factors. With the implementation of the European Technical Guidance Document (TGD) for risk assessment of new and existing substances in 2003 [13], the requirements for performing SSDs have been extended. At present, SSDs can only be performed when at least 10 (preferably 15) values are available for at least eight different taxonomic groups, representing primary producers, and primary and secondary consumers. For the aquatic compartment, it is specified in detail which are the required taxonomic groups. This not the case for soil, but the requirements with respect to the number of data and the inclusion of at least three trophic levels are considered to be the same. For the SRC<sub>soil, eco</sub>, whether or not performing an SSD is not a major change if No Observed Effect Concentrations (NOECs) are present for at least two trophic levels. The 50th percentile of the SSD that was used previously, is equal to the geometric mean of the NOECs that will be used now.

<sup>&</sup>lt;sup>1</sup> e.g. bacteria, fungi, insects and earthworms

#### European risk assessment of nickel

The European Risk Assessment Report of Nickel (EU-RAR) will form the basis for the update of ecological risk limits in the Netherlands [17]. In the late 1990s, nickel metal, nickel sulphate, nickel chloride, nickel nitrate and nickel carbonate were prioritized and selected for extensive hazard and risk assessment by the European Union under the former European Existing Substances Regulation (EEC 793/93). Denmark acted as Rapporteur Member State for the Nickel Risk Assessment (NiRA) for the EU. The process, in which other European Member States were enabled to comment, took more than eight years and included a hazard evaluation and classification, a risk data set generation, a risk assessment (RA), and finally a risk reduction strategy (RRS). The Risk Assessment Report (EU-RAR) was finalized in 2008 and is considered the most thorough, up-to-date risk assessment, with wide agreement amongst Member States and the nickel industries.

### 1.4 Readers guide

In the present report, a closer look is taken at the underlying data and methodology of the nickel RAR of 2008. High quality soil toxicity data were adopted from the RAR (Chapter 2). After the Dutch derivation of nickel risk limits in 2001, a lot of new research was published about the (bio)availability of nickel in soil. This resulted in a methodology to calculate soil specific risk limits, which is described in Chapter 3. Chapter 4 shows the calculated generic risk limits (MPC $_{\rm soil,\ eco}$ ) and soil specific functions that were derived using the RAR methodology. A glossary of abbreviations is provided in appendix 1.

## 2 Ecotoxicity of nickel to soil organisms

#### 2.1 Previous assessment

At the time of the previous RIVM assessment in 2001 [1] only 3 reliable terrestrial data were available, and 15 aquatic toxicity data. The HC5 and HC50 values that were derived from aquatic ecotoxicity test (n=15) when using equilibrium partitioning, were higher than the NOEC and EC50 values from the terrestrial tests. A logKp value of 3.3 was employed.

Table 1 HC5 and HC50 values derived in 2001, based on aquatic and terrestrial toxicity tests.

	Number of tests	HC5 (mg	HC50 (mg Ni/kg)
		Ni/kg)	
Terrestrial tests	3	0.26	65
Aquatic tests	15	38	990

The ultimate set of soil quality criteria is based on policy decision. The current maximum permissible concentration (MPC) in soil were set to the background-concentration in soil (35 mg Ni/kg) [18]. The current Serious Risk Concentration was set to 100 mg/kg.

#### 2.2 Selection criteria for toxicity data in EU-RAR

In the EU-RAR reliable chronic terrestrial ecotoxicity test results are selected. Sufficient terrestrial data were thus available for the derivation of nickel risk limits in soil and it was not necessary to include aquatic toxicity data. Selected tests include 184 standard and non-standard tests comprising 43 species: 11 plants, 6 invertebrates, 19 microbial species or processes and 7 different enzymatic processes. We adopted all these data for revision of the new Dutch risk limits. Here a summary is given of the selection criteria that were used in the EU-RAR [17]. An overview of species and number of records is provided in Appendix 2. The complete list with toxicity data is provided in Appendix 3.

#### Test compound

 The toxicity of nickel depends on the type of nickel-compound used in the test, mainly caused by differences in solubility. Only test results with soluble nickel salts (NiCl<sub>2</sub> and NiSO<sub>4</sub>) are selected. Test results of poorly soluble nickel compounds (NiO and metallic Ni) were excluded.

### Soil types

- Only test results with natural or artificial soils were selected. Tests with other substrates, for instance agar agar, nutrient solutions, pure quartz sand or manure were excluded.
- Tests with soils from deeper soil layers were excluded due to low organic matter content.
- Tests with soil from sub-tropical and tropical areas outside Europe, were excluded because they are considered to be not representative for the European situation.

 Only soils with cation exchange capacity (CEC) between 1.8 en 52.8 cmol/kg, pH between 3.3 and 7.7 and clay content between 0.4 and 55.5% were selected, because these were within the applicability domain of the developed bioavailability models.

#### Chemical analyses

- Tests with measured (with strong acid extraction) as well as nominal (total added) nickel concentrations were selected. Elimination of tests with nominal concentrations only would have resulted in an unacceptable reduction of the number of tests, while for nickel differences between nominal and actual total concentrations due to e.g. biodegradation are not relevant. In case a study did not report whether concentrations are measured or not, it was assumed that it concerned nominal concentrations.
- Background concentrations that were estimated based on regression models were not taken into account. Instead, the median background concentration for European topsoils from the FOREGS database was used (that is 14 mg Ni/kg dry soil) [17].
- For OECD standard soil (artificial soil) there is no background concentration. In that case it was assumed that the reported nominal concentration was equal to the total concentration.

#### **Effects**

- For plants and invertebrates only single species tests with endpoints survival, growth or reproduction were selected.
- Microbial tests were included when involving litter breakdown, respiration, microbial growth, carbon- or nitrogen-mineralization. These are multispecies tests, which are considered representative for the activity of terrestrial microbial ecosystems.
- Soil enzymatic tests were only included when they were performed in buffered soil suspensions at a pH-value that is equal (± 0.5 pH-unit) to that of undisturbed soil.
- Effects on cell metabolism, chlorosis and cell membrane stability were excluded because the results are difficult to interpret at the population level.
- Concentrations with 10% effect (lethal LC<sub>10</sub> or sublethal, EC10) were considered to be similar to No Observed Effect
  Concentrations (NOECs). When both were reported, the EC<sub>10</sub> was selected.

#### Reliability

The following criteria were considered to assess the reliability of a test:

- Negligible mortality or loss of body weight in controls
- Appropriate controls
- Random distribution of organisms in test containers
- Sufficient replica's for a sound statistical analysis
- No multi-metal exposure
- Effect concentrations were excluded when based on visual estimation or when the statistical method was not described
- At least one control and two test concentrations were required for a reliable estimation of effect-concentrations.

#### 3 Method for derivation of risk limits

## 3.1 Risk assessment options

For a proper risk assessment it is important that the measured nickel concentration and soil risk limits refer to the same metal pool. Changing or updating the risk limits requires a critical evaluation of extraction methods too.

Since decades, risk limits for soil are expressed as total concentrations, which are compared with soil concentrations measured by *aqua regia* extraction (a near-total extraction). This way of risk assessment is perceived to be a rather conservative, worst case method, because risk limits are mostly based on freshly contaminated soils whereas many studies have shown that ageing and soil properties lower the actual toxicity. In current legislation, these phenomena are corrected by background concentrations and soil type corrections.

An alternative, more direct, way to deal with bioavailability would be to employ a mild extraction to soil samples (extracting the bioavailable nickel fraction) and compare the measured bioavailable fraction with a soil risk limit that also reflects the bioavailable fraction. Experts agree [19] that an extraction with 0.43 M HNO $_3$  reflects the potentially bioavailable fraction. It extracts the actual bioavailable fraction, present in the pore water plus the fraction that is weakly bound to the soil matrix. Concentrations measured by 0.43 M HNO $_3$  are therefore considered appropriate for comparison with soil risk limits that account for added test concentrations.

We will provide two options for ecological risk limits of nickel:

- 1. Risk limits expressed as added concentrations, for comparison with  $0.43\ HNO_3$  extracted nickel concentrations,
- Risk limits expressed as total concentrations in a standard soil for comparison with aqua regia extracted nickel concentrations. Risk limits are subject to a soil-type correction that account for the influence of ageing and bioavailability on nickel toxicity.

In principle these options aims at the same protection level; i.e protection of 95% of the species/processes for the MPC<sub>soil</sub> and 50% of the species/processes in an ecosystem for the  $SRC_{soil}$ .

#### 3.2 Derivation of HC5 and HC50

HC5 and HC50 values were derived by statistical extrapolation using a species sensitivity distribution (SSD). In the SSD the toxicity dataset was aggregated to 43 data points, each test species or tested process was represented by its most sensitive endpoint.

The SSD comprises plants, invertebrates, microbial processes and enzyme activities.<sup>2</sup> If several data were available for the same endpoint, the geometric mean was calculated.

The aggregated toxicity data were fitted with a log-normal distribution model, according to recommendations in the EU-RAR. Other mathematical fits were tested in the RAR and (i.e. gamma distribution, log-logistic, Weibull) resulted in HC5 and HC50 values that differed a factor 1.2 at the most. Statistical uncertainty is one of the elements that determine the choice of an assessment factor (see paragraph 3.4). All calculations were implemented in R statistical software. The HC5 and HC50 values were computed with the R-package fitdistrplus [20].

#### 3.3 Calculation of soil-specific risk limits

The calculation of soil-specific risk limits contains a correction for fixation in the toxicity test (expressed by a leaching/ageing factor) and a correction for bioavailability taking into account differences in soil properties between test and soil of interest. The whole procedure for calculation of a soil-specific HC5 is shown in Figure 1.

<sup>&</sup>lt;sup>2</sup> The ecological relevance of enzymatic assays can be discussed for several reasons. The enzymatic activities are measured at conditions that are not representative for *in situ* conditions. Several assays are conducted in pH buffered soil suspensions (some tests even at pH>10) and since the metal-enzyme interaction is pH dependent, this might obscure the relationship with effects in the soil. Almost all assays use saturating substrate concentrations (typically several mM), a condition that is unlikely to occur *in situ*. The *in situ* effect of metals on an enzymatic reaction may be rather insensitive to the enzyme capacity (as measured with the enzyme assays) if substrate supply is the rate-limiting step. Not considering the incorporation of the enzymatic toxicity data in the terrestrial database resulted, for the scenarios considered, in higher HC<sub>5</sub> values. The factor of difference is factor of 1.2, which is considered negligible (EU-RAR, 2008).

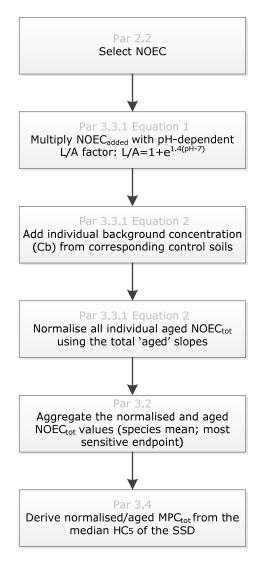


Figure 1 Calculation steps for normalization of nickel toxicity data. Corresponding paragraphs (grey) contain more detailed information about the procedure.

#### 3.3.1 Correction for leaching/ageing and bioavailability

The bioavailability of nickel in soil is dependent of soil type and the duration between nickel application and measurement of effects (leaching/ageing).

Effect concentrations observed in toxicity tests are normalized for the duration between nickel application and measurement of effects. Leaching is an effect that occurs at longer time periods when macro-ions are leached, causing a change in ion strength. Lower ion strength leads to higher bioavailability because there are less competitive ions that compete with nickel for binding to cell membranes. Ageing is a factor that indicates stronger binding and potential incorporation of nickel in the soil particles. Ageing reduces the nickel concentrations in pore water and leads to a lower observed toxicity. Both effects are combined in a leaching/ageing factor.

The EU-RAR described the following procedure for calculation of the leaching/ageing (L/A) factor. The L/A factor was studied on 16 soil types, and appeared to be dependent on the pH:

$$L/A = 1 + e^{1.4 \times (pH-7.0)} \tag{1}$$

Where the pH is pH-CaCl<sub>2</sub>. If pH was measured in KCl is was transformed to pH(CaCl<sub>2</sub>) by: pH-CaCl<sub>2</sub> = 0.795+0.894\*pH-KCl

The L/A factor is only applied on added concentrations, with an equilibration time of  $\leq$  120 days.

$$EC_{x,aged} = EC_{x,added} \times L/A \tag{2}$$

After correction for leaching/ageing, the background concentration is added. The remaining differences in nickel toxicity between soil types can be best explained by differences in CEC (compared to pH, organic matter and clay). A high CEC, implies that high concentrations of cations such as Ca<sup>2+</sup>, Mg<sup>2+</sup>, Na<sup>+</sup> or K<sup>+</sup> are available in the soil solution. These cations suppress the toxicity of Ni<sup>2+</sup>, because they hinder the binding of Ni<sup>2+</sup> to biological targets, and they supplement potential deficiencies of these macro-ions caused by the toxic effects of Ni<sup>2+</sup>. Regression functions for the relation between CEC and NOEC or EC10 values were derived for seven different soil organisms.

$$log(EC_{x,aged} + Cb) = a + b \times log(CEC)$$
(3)

The applicability range of the regression functions is pH 3.3-7.7, organic matter content 0.4-56.8%, CEC 1.8-52.8 cmol/kg and 1-113 mg Ni/kg. The applicability range was determined by the range of soil properties of the 16 soils used for derivation of the regression functions.

The overall correction is described by the following formula:

$$EC_{x ref,} = \left(EC_{x, test} \times L/A + Cb_{test}\right) \times \left[\frac{CEC_{ref}}{CEC_{test}}\right]^{b} \tag{4}$$

For organisms with no experimental regression functions, read-across was applied according to EU-RAR recommendations. The values for the parameter b and the read-across rationale is included in Appendix 4.

The effect of the bioavailability correction on the nickel HC5 and HC50 is demonstrated for 2 Dutch soil types that were also selected in the EU-RAR. These data are used to check if we implemented the bioavailability calculations correctly.

Table 2 Properties of 2 soils in The Netherlands used in the EU-RAR to calculate normalized HC5 and HC50 values

normalized rico and ricovalues.					
	рН	OM%	Clay%	CEC	
Loam	7.5	2.2	26	20	
Peat	4.7	40	24	35	

OM=Organic matter content (%)

3.3.2 Derivation of a relation between HC5, HC50 and CEC A simplification of the whole leaching ageing and bioavailability procedure is very useful for routine risk assessments. The full procedure described in the RAR comprises a non-linear correction of each individual NOEC or EC10 value in the SSD (see equation 4). The exponential factor b is species-dependent and varies from 0.95 to 1.34. As a consequence the magnitude of the correction  $\left[\frac{CEC_{ref}}{CEC_{test}}\right]^b$  is dependent of the species and the test conditions (CEC<sub>test</sub>). This implies that the ranking of species in the SSD is not always the same and may change between reference soil types.

For application of nickel bioavailability in routine assessments a relation between soil properties (for instance CEC) and HC5 or HC50 would be very more useful and straightforward, because risk assessors may skip the procedure of normalisation for individual species, constructing SSDs and fitting distributions. In addition to the RAR procedure, we investigated the potential of a relation between nickel toxicity and CEC. The HC5 and HC50 were computed for 17 hypothetical soils with CEC between 1.8 and 52.8 cmol/kg, with intervals of 3 cmol/kg. Individual corrected NOEC and EC10 values were calculated for these 17 hypothetical soils, SSDs were constructed and HC5 and HC50 were derived. Subsequently, linear regression was applied to derive the transfer function between HC5, HC50 and CEC. A discussion about uncertainties is given in paragraph 4.3.

#### 3.3.3 Estimation of CEC

The CEC describes the interaction between dissolved and adsorbed cation in the soil. The CEC can be estimated from measured cation concentrations in the pore water and on the soil matrix [21]. A simpler approach is to estimate the CEC from basic soil properties such as clay content, organic matter content and/or pH.

Organic matter and clay are standard parameters in routine soil analysis in the Netherlands, whereas CEC is not. Therefore a relation of HC5 and HC50 with OM and clay content would be more practical. In general the CEC is also dependent upon the pH; in most soils increasing pH lead to increasing CEC [22]:

$$CEC = (30 + 4.4 \times pH) \times Fclay + (-34.7 + 29.7 \times pH) \times Fom$$
 (5)

where CEC is expressed in meq/100 g soil=cmol/kg, Fclay=fraction of clay, Fom= fraction organic matter, and pH=pH-KCl<sup>3</sup>. This equation is used in the EU-RAR, and in (slightly different form) in several Dutch studies (i.e. [23, 24])

 $<sup>^3</sup>$  The underlying study was performed in the 1960's. Measuring pH in a KCl solution was quite common then. Nowadays pH is mostly measured in a CaCl<sub>2</sub> solution. The relation between both pH's is: pH-CaCl<sub>2</sub> = 0.795+0.894\*pH-KCl.

However, when the pH has not been measured or is not present in a database the CEC may be estimated from an equation without pH [25]:

$$CEC = 1.5 \times \%OM + 0.6 \times \%Clay \tag{6}$$

where CEC is expressed in meq/100 g soil = cmol/kg.

A thorough discussion about the validity of each equation to estimate CEC from soil properties is beyond the scope of our report. The message is that a relation of  $HC5_{total}$  or  $HC50_{total}$  with CEC may be transformed to a relation of  $HC5_{total}$  or  $HC50_{total}$  with clay, OM and/or pH, if measured CEC values are not available.

#### 3.4 Derivation of MPC and SRC

It is common practice in risk assessment methodology to apply assessment factors that account for different types of variations and uncertainties [26]. When MPC is derived using the SSD-method, an assessment factor between 1 and 5 is put on the HC5, to be justified on a case by case basis. The following aspects are considered:

- 1. Overall quality of the database and the endpoints
- 2. Diversity and representativity of the taxonomic groups
- 3. Mode of action data
- 4. Statistical uncertainties around the HC5 estimate
- 5. Effects data from the field.

After setting an appropriate assessment factor (AF), the maximum permissible concentration and the serious risk concentrations are computed as follows:

$$MPC = HC5/AF \tag{7}$$

The SRA or SRC is based on the geometric mean of the available NOEC/EC10-values, which is equivalent to using the HC50 without an additional assessment factor.

#### 4 Results

#### 4.1 Added concentrations

The SSD based on added nickel concentrations is provided in Figure 2.

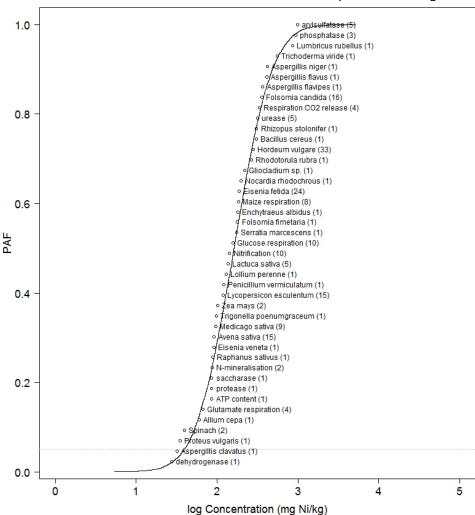


Figure 2 Species sensitivity distribution based on added nickel NOEC or EC10 values. Between brackets the number of underlying data. PAF = Potentially Affection Fraction of species.

The graph is illustrative because it shows all the species for which nickel-toxicity data are available, and their relative sensitivity. The NOECs or EC10 values in this graph are not corrected for bioavailability. The HC5 and HC50 values are therefore not reported because they cannot be related to a reference soil.

#### 4.2 Correction for soil-type specific bioavailability

The variability of NOECs could best be explained by variation of the CEC. The effect of bioavailability on the nickel HC5 and HC50 is demonstrated for two soil types that were also selected in the EU-RAR. This is done to verify our implementation of the bioavailability correction. The estimated

HC5 and HC50 values are listed in Table 3. The agreement between the EU-RAR and our own estimates is good. The minor differences in HC5 may be caused by using different software to fit the log-normal distribution.

Table 3 Normalized HC5 and HC50 values (mg Ni/kg soil) for 2 soil types in The Netherlands. Between brackets the values that were estimated in the EU-RAR.

	HC5 (mg Ni/kg soil)	HC50
Loam	99 (100)	408
Peat	186 (189)	746

The relation between CEC and HC5 and HC50 for soil types with CEC between 1.8 and 52.8 cmol/kg is shown in Figure 3 and can be described by linear functions:

$$HC5_{total} = 5.40 \times CEC$$
 [8]

$$HC50_{total} = 21.6 \times CEC$$
 [9]

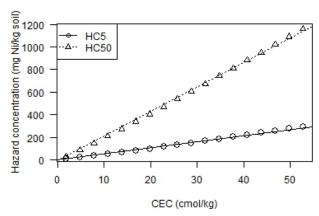


Figure 3 Relation between HC5, HC50 and the CEC of the soil.

The applicability domain of the bioavailability regression functions is restricted to CEC between 1.8 and 52.8 cmol/kg. If higher CEC values are measured or predicted, cut-off values are proposed: maximum HC5 = 293 mg Ni/kg and maximum HC50 = 1162 mg Ni/kg.

#### 4.3 MPC and SRC values and soil-type corrections

Acknowledging the large number of high quality terrestrial ecotoxicity studies, covering 8 taxonomic groups, the EU-RAR recommended an assessment factor (AF) of 2. In addition to the EU-RAR reasoning we considered the fact that in the aquatic ecosystem snails and molluscs belong to the most sensitive organisms as far as nickel is concerned. In the terrestrial toxicity database there are no snails or molluscs included. A summary of the uncertainties indicated by the EU-RAR is given in Appendix 5.

Given the AF of 2 the following ecological risk limits are proposed (seeTable 4).

Table 4 Summary of proposed MPC and SRC values and relations with soil properties, compared with current legal criteria. Rounded to 2 significant digits.

Option	so, compared wen can one regar enterial it	MPC (mg	SRC (mg
'		Ni/kg soil)	Ni/kg soil)
1	Added concentration	10	140
2	Soil type correction	$2.7 \times CEC$ includes an AF of 2	22 × CEC
	Total concentration in a standard soil (10% OM, 25% clay, pH 6.4)	81	660
3	Maximum total concentration <sup>1</sup>	150	1200
	Minimum total concentration <sup>2</sup>	11	90

<sup>&</sup>lt;sup>1</sup> Determined by the applicability domain of the bioavailability correction <sup>2</sup> Calculated for a standard soil with 2% OM and 2% clay

The MPC $_{added}$  of 10 mg Ni/kg reflects the chemically available fraction, which can be extracted with 0.43 M HNO $_{3}$ . The MPC $_{added}$  is independent of soil type, but it ignores the effect of other cations on the bioavailability of nickel. The same reasoning holds for SRC $_{added}$ .

To account for bioavailability, relations were derived between MPC $_{\text{total}}$  and SRC $_{\text{total}}$  and the CEC. If the CEC is not measured, it may be estimated from transfer functions. This introduces an additional uncertainty to the correction of MPC and SRC values.

To facilitate comparison with current legal ecological risk limits for nickel in soil, MPC $_{total}$  and SRC $_{total}$  were calculated for a standard soil with 25% clay and 10% organic matter. The standard soil would have an estimated CEC of 30 cmol/kg according Equation 6. According to Equation 5, this standard soil would have a pH of 6.4. Taking into account ageing and bioavailability (option 3), the MPC $_{total}$  for such a standard soil is 81 mg Ni/kg. The SRC $_{total}$  for a standard soil is 657 mg Ni/kg soil.

For soils and sediments with very low clay and organic matter contents the current legislation recommends to use minimum contents of 2% in the soil type correction [27]. This would imply a minimum MPC $_{total}$  of 11 mg Ni/kg soil and a minimum SRC $_{total}$  of 90 mg Ni/kg soil.

#### 5 Discussion and conclusions

The current legal MPC<sub>total</sub> (35 mg Ni/kg) and SRC<sub>total</sub> (100 mg Ni/kg) of nickel are defined for a standard soil of 25% clay and are independent of organic matter content. These current legal ecological risk limits ignore the effect of organic matter and pH. Beside this uncertainty, the weight-of-evidence of the current ecological risk limits is small; only 3 toxicity tests were considered and the methodology of derivations of the environmental quality criteria was out-of-date.

It is up to policy makers to decide if and how the current ecological risk limits for nickel in soil will be replaced. This will also depend on choices on how to deal with bioavailability of metals in general, and what extraction methods are to be prescribed. These issues are outside the scope of this report. This report contains ecological risk limits for the two extraction methods that are currently considered: i.e. the 0.43 M HNO<sub>3</sub> extraction and the *agua regia* extraction.

The proposed new ecological risk limits are based on a state-of-the-art assessment of 184 terrestrial soil toxicity data, distributed over 43 species or soil processes together with application of the most up-to-date guidelines for derivation of environmental quality criteria.

The proposed  $MPC_{added}$  is 10 mg Ni/kg and the  $SRC_{added}$  is 140 mg Ni/kg. They reflect the potentially available fraction, which can be extracted with 0.43 M HNO<sub>3</sub>. The MPC<sub>added</sub> is independent of soil type, but it ignores the competitive effect of other cations on the bioavailability of nickel. The same reasoning holds for  $SRC_{added}$ .

The soil-type corrections of the ecological risk limits with CEC are:

$$MPC_{total} = 2.7 \times CEC$$
 and  $SRC_{total} = 22 \times CEC$ .

Where CEC is the cation exchange capacity in cmol/kg. In these equations the ageing and bioavailability of nickel are accounted for. The resulting MPC- and SRC-values should be compared with *aqua regia* extracted nickel concentrations. If measured CEC values are not available, the CEC may eventually be computed by an appropriate transfer function.

For a standard soil with 10% organic matter, 25% clay a CEC of 30 cmol/kg is estimated. This results in  $MPC_{total} = 81 \text{ mg Ni/kg and } SRC_{total} = 660 \text{ mg Ni/kg}$  for a standard soil with 10% OM and 25% clay.

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## Appendix 1 Glossary

Abbreviation	Explanation
AF	Assessment Factor
CEC	Cation Exchange capacity; property of soil to reflect the potential to hold and exchange cations.
EC10	Effect Concentration where 10% effect is observed compared to untreated control. This statistically derived concentration of a substance in an environmental medium expected to produce a 10% effect compared to the control.
ER	Dutch synonym for SRC: "Ernstig Risico Concentratie"
EU-RAR	European Union Risk Assessment Report
HC5	Hazard Concentration 5%. This concentrations is statistically derived from a SSD and reflects a concentration at which 5% of species in an ecosystem may be adversely affected.
HC50	Hazard Concentration 50%. This concentrations is statistically derived from a SSD and reflects a concentration at which 50% of species in an ecosystem may be adversely affected.
MPC	Maximum Permissible Concentration. This is a legal risk limit reflecting a concentration that protects the ecosystem. It is derived from the HC5, eventually divided by an assessment factor. English synonym for MTR.
MTR	Dutch synonym for MPC: "Maximaal Toelaatbare Risico-concentratie"
NOEC	No Observed Effect Concentration. This parameter represents the concentration of a pollutant that will not harm the species involved, with respect to the effect that is studied.
ОС	Organic Carbon
ОМ	Organic Matter

Abbreviation	Explanation
SRC	Serious Risk Concentrations. This is a legal risk limit, used in the Netherlands, reflecting a concentration that induces additional investigation and possible risk management (intervention). It is derived from the HC50.
SSD	Species Sensitivity Distribution. A SSD is an important probabilistic tool for environmental risk assessment (ERA) and accounts for differences in species sensitivity to a chemical.

## Appendix 2 Species and records EU-RAR

 $EC10_{added}$  for the most sensitive endpoint, total number of underlying data (n). Between brackets the original RAR data, when different.

Species or proces	n	EC10 <sub>added</sub> (mg Ni/kg)
<b>Enzymatic processes</b>	18	
arylsulfatase	5	993
dehydrogenase	1	7.9
N-mineralisation	2	72
phosphatase	3	875
protease	1	77
saccharase	1	77
urease	5	281
invertebrates	44	
Eisenia fetida	24 (17)	149 (179)
Eisenia veneta	1	85
Enchytraeus albidus	1	180
Folsomia candida	16	275
Folsomia fimetaria	1	173
Lumbricus rubellus	1	842
microbial growth or processes	50	
Aspergillis clavatus	1	13
Aspergillis flavipes	1	347
Aspergillis flavus	1	393
Aspergillis niger	1	400
ATP content	1	77
Bacillus cereus	1	285
Gliocladium sp.	1	200
Glucose respiration	10	127
Glutamate respiration	4	55
Maize respiration	8	152
Nitrification	10	116
Nocardia rhodochrous	1	177
Penicillium vermiculatum	1	102
Proteus vulgaris	1	15
Respiration CO2 release	4	299
Rhizopus stolonifer	1	288
Rhodotorula rubra	1	247
Serratia marcescens	1	155
Trichoderma viride	1	530
plants	85	
Allium cepa	1	46
Avena sativa	15	76
Hordeum vulgare	33 (16)	235 (296)
Lactuca sativa	5	104
Lollium perenne	1	110
Lycopersicon esculentum	15	103

Species or proces	n	EC10 <sub>added</sub> (mg Ni/kg)
Medicago sativa	9	78
Raphanus sativus	1	80
Spinach	2	32
Trigonella poenumgraceum	1	84
Zea mays	2	48

Appendix 3 EU-RAR terrestrial toxicity data used for derivation of ecological risk limits of nickel

										_		EC-added
Species	Medium	Location	рН	OC	clay	Cb	CEC	Eq.Period	Duration	Parameter	Endpoint	(mg Ni/kg) Ref.
invertebrates												
Folsomia candida	Loamy sand	Houthalen	3.6	1.73	0.4	1			28d	EC10	reproduction	36.4 [28]
Folsomia candida	Sandy clay loam	Zegveld	4.1		34	26	52.75	7		EC10	reproduction	558
Folsomia candida	Loamy sand	Montpellier	4.1	0.25		16	8.39	7		EC10	reproduction	120
Folsomia candida	Loamy sand	Rhydtalog	4.2			3	_	7	28d	EC10	reproduction	527
Folsomia candida		Jyndevad	4.5	1.32	1.5	1	1.84	7	28d	EC10	reproduction	104
Folsomia candida	Clay	Aluminosa	5.6	0.9	46.9	19	19.26	7	28d	EC10	reproduction	101
Folsomia candida		Borris	5.6	1.33	4.3	3	4.91	7	28d	EC10	reproduction	180
Folsomia candida	Sandy clay loam	Woburn	6.1	4.3	35.3	39	28.87	7	28d	EC10	reproduction	622
Folsomia candida	Silt loam	Ter Munck	6.7	1.09	9.6	11	7.8	7	28d	EC10	reproduction	269
Folsomia candida	Clay	Souli	7	0.45	33.2	81	12.85	7	28d	EC10	reproduction	384
Folsomia candida	Clay	Marknesse	7.6	1.14	19.9	19	19.44	7	28d	EC10	reproduction	662
Folsomia candida	Clay	Brecy	7.5	1.37	49.2	113	23.57	7	28d	NOEC	reproduction	828
Folsomia candida		Cordoba 2	7.6	0.49	55.4	24	35.26	7	28d	EC10	reproduction	1100
Folsomia candida		Cordoba 1	7.6	0.53	19.8	18	13.35	7	28d	EC10	reproduction	61.7
Folsomia candida	Loam	Guadalajara	7.7	0.31	17.2	11	13.27	7	28d	EC10	reproduction	562
Eisenia fetida	Loamy sand	Houthalen	3.6	1.73	0.4	1	1.84	7	28d	EC10	reproduction	49.8
Eisenia fetida	Sandy clay loam	Zegveld	4.1	33.05	34	26	52.75	7	28d	EC10	reproduction	1110
Eisenia fetida	Loamy sand	Montpellier	4.1	0.25	25.3	16	8.39	7	28d	EC10	reproduction	54.5
Eisenia fetida	Loamy sand	Rhydtalog	4.2	12.52	12.7	3	11.91	7	28d	EC10	reproduction	362
Eisenia fetida	-	Jyndevad	4.5	1.32	1.5	1	1.84	7	28d	EC10	reproduction	46.5
Eisenia fetida	Sandy loam	Kovlinge II	5.1	2.47	3.9	2	4.31	7	28d	EC10	reproduction	182
Eisenia fetida	Clay	Aluminosa	5.6	0.9	46.9	19	19.26	7	28d	EC10	reproduction	230
Eisenia fetida	•	Borris	5.6	1.33	4.3	3	4.91	7	28d	EC10	reproduction	66.1
Eisenia fetida	Sandy clay loam	Woburn	6.1	4.3	35.3	39	28.87	7	28d	EC10	reproduction	151
Eisenia fetida	Silt loam	Ter Munck	6.7	1.09	9.6	11	7.8	7	28d	EC10	reproduction	172
Eisenia fetida	Clay	Souli	7	0.45	33.2	81	12.85	7	28d	NOEC	reproduction	297
Eisenia fetida	Clay	Marknesse	7.6	1.14	19.9	19	19.44	7	28d	EC10	reproduction	233
Eisenia fetida	Clay	Brecy	7.5	1.37	49.2	113	23.57	7	28d	EC10	reproduction	239

												EC-added	
Species	Medium	Location	рН	OC	clay	Cb	CEC	Eq.Period		Parameter		(mg Ni/kg)	
Eisenia fetida		Cordoba 2	7.6	0.49			35.26	7	_04	NOEC	reproduction	490	
Eisenia fetida		Cordoba 1	7.6	0.53			13.35	7		EC10	reproduction	186	
Eisenia fetida	Loam	Guadalajara	7.7	0.31			13.27	7	200	EC10	reproduction	198	
Eisenia fetida	OECD 207		6	5.8	10	20	14.5		21d	NOEC	reproduction	180	
Enchytraeus albidus	OECD 207		6	5.8	10	20	14.5		42d	NOEC	reproduction	180	
Folsomia candida	OECD 207		6	5.8	10	20	14.5		28d	NOEC	reproduction	320	
Eisenia veneta	Loamy sand	LUFA 2.2	5.5	2.3	5	6	7.9			EC10	reproduction	85	
Folsomia fimetaria	Loamy sand	LUFA 2.2	5.5	2.3	5	6	7.9			EC10	reproduction	173	
Lumbricus rubellus	Sandy loam	b	7.3	4.7	17	17	25.3			EC10	mortality	842	[31]
microbial growth or	processes												
N-mineralisation	Nethen		6.2	1.07	10	8	6.5		28d	EC10		257	[32]
N-mineralisation	Nethen_NH4		6.2	1.07	10	8	6.5		28d	EC10		20	
Nitrification	Sandy clay loam	Zegveld	4.1	33.05	34	26	52.75		4-28d	EC10		170	[33]
Nitrification	Loamy sand	Rhydtalog	4.2	12.52	12.7	3	11.91		4-28d	EC10		111	
Nitrification	Clay	Aluminosa	5.6	0.9	46.9	19	19.26		4-28d	EC10		44	
Nitrification		Borris	5.6	1.33	4.3	3	4.91		4-28d	EC10		137	
Nitrification	Silt loam	Ter Munck	6.7	1.09	9.6	11	7.8		4-28d	EC10		67	
Nitrification	Clay	Souli	7	0.45			12.85		4-28d	EC10		214	
Nitrification	Clay	Brecy	7.5		49.2		23.57		4-28d	EC10		439	
Nitrification		Cordoba 2	7.6	0.49			35.26		4-28d	EC10		169	
Nitrification		Cordoba 1	7.6	0.53			13.35		4-28d	EC10		53	
Nitrification	Loam	Guadalajara	7.7	0.31		11	13.27		24h	EC10		67	
Glucose respiration	Loamy sand	Montpellier	4.1	0.25		16			24h	EC10		22	
Glucose respiration	Clay	Aluminosa	5.6		46.9		19.26		24h	EC10		254	
Glucose respiration	Sandy clay loam	Woburn	6.1	4.3			28.87		24h	EC10		376	
Glucose respiration	Silt loam	Ter Munck	6.7	1.09	9.6	11	7.8		24h	EC10		45	
Glucose respiration	Clay	Souli	7	0.45			12.85		24h	EC10		242	
Glucose respiration	Clay	Marknesse	7.6	1.14			19.44		24h	EC10		116	
Glucose respiration	Clay	Brecy	7.5		49.2		23.57		24h	EC10		302	
Glucose respiration		Cordoba 2	7.6		55.4		35.26		24h	EC10		167	
Glucose respiration		Cordoba 1	7.6	0.53			13.35		24h	EC10		140	
Glucose respiration	Loam	Guadalajara	7.7	0.31			13.27		24h	EC10		56	
Maize respiration	Loamy sand	Houthalen	3.6	1.73	0.4	1	1.84		28d	EC10		42	

												EC-added	
Species	Medium	Location	рН	OC		Cb	CEC	Eq.Period	Duration	Parameter	Endpoint	(mg Ni/kg)	Ref.
Maize respiration	Sandy loam	Rhydtalog		12.52		3	11.91		28d	EC10		343	
Maize respiration		Borris	5.6	1.33	4.3	3	4.91		28d	EC10		55	
Maize respiration	Sandy clay loam	Woburn	6.1	4.3	35.3	39	28.87		28d	EC10		121	
Maize respiration	Clay	Souli	7	0.45	33.2		12.85		28d	EC10		88	
Maize respiration	Clay	Brecy	7.5	1.37	49.2	113	23.57		28d	EC10		203	
Maize respiration		Cordoba 2	7.6	0.49	55.4	24	35.26		28d	EC10		446	
Maize respiration	Loam	Guadalajara	7.7	0.31	17.2	11	13.27		28d	EC10		370	
Aspergillis flavipes		Kitchawan	4.9	3.34	9.4	20	8.15		several days	EC10	hyphal growth	347	[34]
Aspergillis flavus		Kitchawan	4.9	3.34	9.4	20	8.15		several days	EC10	hyphal growth	393	
Aspergillis clavatus		Kitchawan	4.9	3.34	9.4	20	8.15		several days	EC10	hyphal growth	13	
Aspergillis niger		Kitchawan	4.9	3.34	9.4	20	8.15		several days	EC10	hyphal growth	400	
Penicillium vermiculatu	m	Kitchawan	4.9	3.34	9.4	20	8.15		several days	EC10	hyphal growth	102	
Rhizopus stolonifer		Kitchawan	4.9	3.34	9.4	20	8.15		several days	EC10	hyphal growth	288	
Trichoderma viride		Kitchawan	4.9	3.34	9.4	20	8.15		several days	EC10	hyphal growth	530	
Gliocladium sp.		Kitchawan	4.9	3.34	9.4	20	8.15		several days	EC10	hyphal growth	200	
Serratia marcescens		Kitchawan	4.9	3.34	9.4	20	8.15		several days	EC10	colony count	155	
Proteus vulgaris		Kitchawan	4.9	3.34	9.4	20	8.15		several days	EC10	colony count	15	
Bacillus cereus		Kitchawan	4.9	3.34	9.4	20	8.15		several days	EC10	colony count	285	
Nocardia rhodochrous		Kitchawan	4.9	3.34	9.4	20	8.15		several days	EC10	colony count	177	
Rhodotorula rubra		Kitchawan	4.9	3.34	9.4	20	8.15		several days	EC10	colony count	247	
Respiration CO2													
release	Sandy loam	a	6	3.3	9	2	11		42w	NOEC		400	[35]
Respiration CO2													
release	Clay	b	7.5	1.86	60	39	30		80w	EC10		2542	
Respiration CO2													
release	Sandy peat	a	4.4	7.4	5	4	52.5		82w	EC10		291	
Respiration CO2													
release	Typic Xerochrept	a	5.2		8	14	13.1		28d	NOEC		27	[36]
ATP content	Sandy cambisol	а	6	1.2	9	9	10.3		9y	NOEC		77	[37]
Glutamate respiration	Sand	a	7	0.9	2	8	1.5		1.5y	NOEC		55	[38]
Glutamate respiration	Sandy peat	a	4.4	7.4	5	4	52.5		1.5y	NOEC		55	
Glutamate respiration	Clay	b	7.5	1.86	60	39	30		1.5y	NOEC		55	
Glutamate respiration	Sandy loam	a	6	3.31	9	2	11		1.5y	NOEC		55	

Species	Medium	Location	рН	OC	clay	Ch	CEC	Eq.Period	Duration	Parameter	Endnoint	EC-added (mg Ni/kg)	Ref.
Species plants	Mediuiii	Location	рп	UC	Clay	CD	CLC	Lq.Periou	Duration	Parameter	Liiupoiiit	(Hig Ni/Kg)	Kei.
Hordeum vulgare	Loamy sand	Houthalen	3.6	1.73	0.4	1	1.84	7	4d	EC10	root length	31	[39]
Hordeum vulgare	Sandy clay loam	Zegveld	4.1		34	26	52.75		4d	EC10	root length	1101	
Hordeum vulgare	Loamy sand	Montpellier	4.1	0.25		16	8.39	7		EC10	root length	90	
Hordeum vulgare	Loamy sand	Rhydtalog		12.52			11.91	-	4d	EC10	root length	249	
Hordeum vulgare	Louiny Suna	Jyndevad	4.5	1.32		1	1.84		4d	EC10	root length	46	
Hordeum vulgare	Sandy Ioam	Kovlinge II	5.1	2.47	3.9	2	4.31		4d	EC10	root length	123	
Hordeum vulgare	Clay	Aluminosa	5.6	0.9			19.26		4d	EC10	root length	261	
Hordeum vulgare	City	Borris	5.6	1.33	4.3	3	4.91		4d	EC10	root length	128	
Hordeum vulgare	Sandy clay loam	Woburn	6.1	4.3			28.87		4d	EC10	root length	398	
Hordeum vulgare	Silt loam	Ter Munck	6.7	1.09	9.6	11	7.8	7		EC10	root length	106	
Hordeum vulgare	Clay	Souli	7	0.45			12.85	•	4d	EC10	root length	211	
Hordeum vulgare	Silt loam	Marknesse	7.6		19.9	19			4d	EC10	root length	268	
Hordeum vulgare	Clay	Brecy	7.5	1.37			23.57		4d	EC10	root length	289	
Hordeum vulgare	Ciay	Cordoba 2	7.6	0.49			35.26		4d	EC10	root length	587	
Hordeum vulgare		Cordoba 1	7.6		19.8		13.35		4d	EC10	root length	96	
Hordeum vulgare	Loam	Guadalajara	7.7		17.2		13.27		4d	EC10	root length	304	
Lycopersicon				0.0_				•					
esculentum	Loamy sand	Houthalen	3.6	1.73	0.4	1	1.84	7	28d	EC10	yield-shoots	21	
Lycopersicon	,										,		
esculentum	Sandy clay loam	Zegveld	4.1	33.05	34	26	52.75	7	28d	EC10	yield-shoots	599	
Lycopersicon													
esculentum	Loamy sand	Montpellier	4.1	0.25	25.3	16	8.39	7	28d	EC10	yield-shoots	16	
Lycopersicon													
esculentum	Loamy sand	Rhydtalog		12.52			11.91		28d	EC10	yield-shoots	125	
Lycopersicon esculent	cum	Jyndevad	4.5	1.32	1.5	1	1.84	7	28d	EC10	yield-shoots	10	
Lycopersicon						_		_					
esculentum	Sandy Ioam	Kovlinge II	5.1	2.47	3.9	2	4.31		28d	EC10	yield-shoots	42	
Lycopersicon esculent	um	Borris	5.6	1.33	4.3	3	4.91	7	28d	EC10	yield-shoots	52	
Lycopersicon	6 1 1 1	\A/ I	<i>-</i> 1	4.2	25.2	20	20.07	-	20.1	FC10		150	
esculentum	Sandy clay loam	Woburn	6.1	4.3	35.3	39	28.87	/	28d	EC10	yield-shoots	150	
Lycopersicon esculentum	Silt loam	Ter Munck	6.7	1.09	9.6	11	7.8	7	28d	EC10	viold shoots	118	
		Souli	6.7 7		33.2		12.85		280 28d	EC10 EC10	yield-shoots	250	
Lycopersicon	Clay	Souli	/	0.45	33.2	91	12.00	/	Zou	ECIO	yield-shoots	250	

Species	Medium	Location	рН	ОС	clay	Cb	CEC	Eq.Period	Duration	Parameter	Endpoint	EC-added (mg Ni/kg) Ref.
esculentum	ricaiaiii	Location	рп	<u> </u>	ciay	CD	CLC	Lqii cilou	Daracion	rarameter	Епароппе	(mg m/kg) Ken
Lycopersicon												
esculentum	Silt loam	Marknesse	7.6	1.14	19.9	19	19.44	7	28d	EC10	yield-shoots	200
Lycopersicon											,	
esculentum	Clay	Brecy	7.5	1.37	49.2	113	23.57	7	28d	EC10	yield-shoots	504
Lycopersicon esculen	tum	Cordoba 2	7.6	0.49	55.4	24	35.26	7	28d	EC10	yield-shoots	224
Lycopersicon esculen	tum	Cordoba 1	7.6	0.53	19.8	18	13.35	7	28d	EC10	yield-shoots	144
Lycopersicon												
esculentum	Loam	Guadalajara	7.7	0.31	17.2	11	13.27	7		EC10	yield-shoots	189
Spinach	Sand	b	4.55			3	7.4	10	30d	NOEC	yield	10 [40]
Spinach	Heavy clay		8.1			20	19.6		30d	NOEC	yield	100
Avena sativa	Sandy loam + P	Grenville	7.2	2.32		14	13	60	110d	EC10	yield-grains	453 [41]
Medicago sativa	Sandy loam + P	Grenville	7.6	2.32		14	13	60		EC10	yield-tops	371
Medicago sativa	Sandy loam + P	Grenville	7.2	2.32		14	13	60	83d	EC10	yield-tops	383
Avena sativa	Sand 3	Uplands	5.5	0.81		14	6	60	110d	EC10	yield-grains	43
Avena sativa	Sand 3+P	Uplands	5.5	0.81		14	6	60	110d	EC10	yield-grains	47
Avena sativa	Sand 3+L	Uplands	6.3	0.81		14	6	60	110d	EC10	yield-grains	64
Avena sativa	Sand 3+P+L	Uplands	6.2	0.81		14	6	60	110d	EC10	yield-grains	53
Medicago sativa	Sand 3+P	Uplands	5.5	0.81		14	6	60	83d	EC10	yield-tops	36
Medicago sativa	Sand 3+L	Uplands	6.3	0.81		14	6	60	83d	EC10	yield-tops	44
Medicago sativa	Sand 3+P+L	Uplands	6.2	0.81		14	6	60	83d	EC10	yield-tops	39
Avena sativa	Sand 4	Uplands	5.2	2.38		14	11.7	60	110d	EC10	yield-grains	49
Avena sativa	Sand 4+P	Uplands	5.1	2.38		14	11.7	60	110d	EC10	yield-grains	238
Avena sativa	Sand 4+L	Uplands	6.4	2.38		14	11.7	60	110d	EC10	yield-grains	238
Avena sativa	Sand 4+P+L	Uplands	6.1	2.38		14	11.7	60	110d	EC10	yield-grains	253
Medicago sativa	Sand 4	Uplands	5.2	2.38		14	11.7	60	83d	EC10	yield-tops	34
Medicago sativa	Sand 4+P	Uplands	5.1	2.38		14	11.7	60	83d	EC10	yield-tops	41
Medicago sativa	Sand 4+L	Uplands	6.4	2.38		14	11.7	60	83d	EC10	yield-tops	92
Medicago sativa	Sand 4+P+L	Uplands	6.1	2.38		14	11.7	60	83d	EC10	yield-tops	91
Avena sativa	Clay	a	7.5	1.34	9	8	14.6	60	to maturaty	NOEC	yield-grains	80 [42]
Raphanus sativus	Clay	a	7.5	1.34	9	8	14.6	60	30d	NOEC	yield	80
Lactuca sativa	Clay	С	8.3	1.74	45	27	34.7	60	40d	NOEC	, yield-leaves	40
Allium cepa		XX	8.3	0.28	24	14	12.6	7	56d	EC10	yield	46 [43]

												EC-added	
Species	Medium	Location	рН	OC	clay	Cb	CEC	Eq.Period		Parameter		(mg Ni/kg)	
Trigonella poenumgr		XX	8.3	0.28	24	14	12.6		56d	EC10	yield	84	
Lollium perenne	Sandy loam	С	6	1.7		19	31		56d-64	EC10	yield	110	
Lactuca sativa		Steinhof	4.9			16	8		63d	EC10	yield-leaves	18	[45]
Lactuca sativa		Gansemos	5.6			17	41		63d	EC10	yield-leaves	153	
Lactuca sativa		Erlach	7.7			26	10		63d	EC10	yield-leaves	257	
Lactuca sativa		Gasel	6.6			21	20		63d	EC10	yield-leaves	422	
Avena sativa		X	5.6	0.93	12	10	15		150d	EC10	yield-grains	66	[46]
Avena sativa		X	5.4	1.4	40	26	21		150d	EC10	yield-grains	45	
Avena sativa		X	5.2	1.86	58	46	33		150d	EC10	yield-grains	47	
Avena sativa		X	5	1.98	4	2	9		150d	EC10	yield-grains	16	
Avena sativa		X	5.4	3.95	5	1	19		150d	EC10	yield-grains	40	
Zea mays	Loam	Giza	7.9		26.6	54	17.3		45-50d	EC10		119	[47]
Zea mays	sandy loam	Nobaria	8.2	0.7	15.1	40	12.6		45-50d	EC10		19	
enzymatic process	es												
urease	Sand	а	7	0.9	2	8	1.5		1.5y	EC10		120	[48]
urease	Sandy loam	a	6	3.31	9	2	11		1.5y	EC10		2300	
urease	Silty loam	a	7.7	1.4	19	25	16		42d	EC10		130	
urease	Clay	b	7.5	1.86	60	39	30		1.5y	EC10		90	
urease	Sandy peat	a	4.4	7.44	5	4	52.5		1.5y	EC10		540	
phosphatase	Sandy loam	a	6	3.31	9	2	11		1.5y	EC10		7021	[49]
phosphatase	Silty loam	a	7.7	1.4	19	25	16		1.5y	EC10		251	
phosphatase	Clay	b	7.5	1.86	60	39	30		42d	EC10		380	
arylsulfatase	Sand	а	7	0.9	2	8	1.5		42d	EC10		372	[50]
arylsulfatase	Sandy loam	a	6	3.31	9	2	11		42d	EC10		610	
arylsulfatase	Silty loam	a	7.7	1.4	19	25	16		42d	EC10		2207	
arylsulfatase	Clay	b	7.5	1.86	60	39	30		1.5y	EC10		272	
arylsulfatase	Sandy peat	a	4.4	7.44	5	4	52.5		1.5y	EC10		7080	
dehydrogenase	haplic luvisol		7	1.1	15.2	19.4	12.4			EC10		7.9	[51]
saccharase	Sandy cambisol	а	6	1.2	9	9	10.3		9y	NOEC		77	
protease	Sandy cambisol	a	6	1.2	9	9	10.3		9y	NOEC		77	_

## Appendix 4 Overview of parameter b according to EU-RAR

Overview of parameter b experimentally determined for 7 test organisms and read-across choices for other organisms, in order to enable bioavailability corrections of nickel NOEC or EC10 values for all species in the toxicity database (according to EU-RAR descriptions).

Test organism	b	Read across to:	Reason
Lycopersicon esculentum (tomato)	1.27		
Hordeum vulgare (barley)	1.12	All other plants in the database: Allium cepa, Avena sativa, Lactuca sativa, Lollium perenne, Medicago sativa, Raphanus sativus, Spinach, Trigonella poenumgraceum and Zea mays	Slopes quit similar to tomato. No critical choice according to RAR uncertaintly analysis
Eisenia fetida	0.95	To all soft bodied invertebrates: <i>Eisenia</i> veneta, Lumbricus rubellus, Enchytraeus albidus	Based on dominant metal uptake route via their dermis.
Folsomia candida	1.17	To all hard bodied invertebrates: Folsomia fimetaria,	Based on dominant metal uptake route through gut wall.
Substrate induced respiration	1.34	Glutamate respiration, glucose respiration, ATP content	Indicators of biomass and ATP content
Maize induced respiration	1.22	Dehydrogenase,respiration	Natural substrates and basal soil respiration
Nitrification	1.00	N-mineralization, arylsulfatase, phosphatase, protease, saccharase, urease	Processes related to the N-cycle, and all other microbial processes not covered by substrate induced or maize induced respiration

## Appendix 5 Summary uncertainties EU-RAR

Summary of uncertainties in the derivation of ecological risk limits, as discussed in the EU-RAR.

Acknowledging the large number of high quality terrestrial ecotoxicity studies, covering 8 taxonomic groups, the EU-RAR recommended an assessment factor (AF) of 2 is suggested, based on the following considerations:

- 1. The overall quality of the database and the end-points covered, e.g., if all the data are generated from "true" chronic studies; Although part of the included studies could be considered long-term studies, very few of these studies could be considered truly chronic studies, i.e. including at least one or more generations. Inclusion of one or more generations was the case for some microbial studies, but not for the plant and invertebrate studies. Although some studies are not true chronic studies, they are often more sensitive compared to longer-term studies, e.g. microbial glucose and maize respiration.
- 2. The diversity and representativeness of the taxonomic groups covered by the database; The plants species included represent solely agricultural species and it is uncertain how wild species will be affected by Ni compared to agricultural species. Three of the 4 earthworm species tested were closely related and the 2 collembolan species were also closely related. This introduces uncertainty in how the toxicity relates to less closely related species. The species used in the soil-type models are 2 plant species, 2 invertebrate species and 3 microbial processes. Hence, in relation to the cross-species implementation it is uncertainty as to what predictive power these models have for other species.
- 3. Statistical uncertainties around the 5th percentile estimate, e.g., reflected in the goodness-of-fit or the size of confidence interval around the 5th percentile; The choice of SSD curve fitting and goodness of fit approaches impacted the derivation of the HC5 to some degree. But the impact was no greater than a factor of 1.2.
- 4. Comparisons between field/microcosm studies and the HC5(50%) to evaluate the laboratory to field extrapolation. No field data or mesocosm studies were available that allowed deriving threshold concentrations of Ni in soils at the field scale. Hence, for plants and invertebrates no food-web studies were reported, which indicate that influence of species interaction such as predation, mutualism and competition on the Ni toxicity is not covered and quantified.
- 5. Use of bioavailability models
  The limited number of Phyla, classes, species and life-strategies
  for which the models have been developed and tested, implies

- uncertainty with regard to the validity of the models for other less closely related species.
- 6. Interaction with other environmental related factors
  The impact of soil-type, ageing and leaching have been studied
  and quantified, whereas other field related factors have not been
  studied or quantified. The latter include among others
  temperature fluctuation and soil-humidity.

