

# Soil for life

report 1323

**Advances in micronutrient  
fertilization recommendations**



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## **Advances in micronutrient fertilization recommendations**

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## Samenvatting en aanbevelingen

In de afgelopen decennia is het belang van micronutriënten voor de landbouw steeds duidelijker geworden. Daarnaast is het ook duidelijk geworden dat bemesting met micronutriënten een delicate kwestie is waarin er slechts een kleine marge bestaat tussen een tekort en toxiciteit. Er is nog steeds een wereld te ontdekken in termen van een beter begrip, ontwikkeling van meetmethodieken en voorspellen van de beschikbaarheid van micronutriënten in de bodem en de vertaling naar bemestingsadviezen.

In 2010 is een STW-project gestart onder de titel: "Micronutriënten management voor een Duurzame Landbouw en Milieu: Een nieuwe innovatieve benadering". Twee promovendi waren betrokken van de afdeling Bodemkwaliteit van Wageningen Universiteit. Het onderzoek is in de zomer van 2016 afgerond en had als doel om vanuit een beter inzicht in het gedrag van de elementen borium (B), koper (Cu), selenium (Se) en mangaan (Mn) in de bodem, de Nederlandse micronutriënten bemestingsadviezen te verbeteren. NMI was betrokken bij het toezicht op dit AIO onderzoek en de benutting van de resultaten. De NMI-activiteiten zijn uitgevoerd in opdracht van Productschap Zuivel.

Voorliggende rapport bevat aanbevelingen om de huidige Nederlandse micronutriënten bemestingsadviezen te verbeteren voor de elementen B, Cu, Se, Mn. De onderbouwing van deze aanbevelingen is afgeleid uit een compilatie van wetenschappelijke literatuur, de belangrijkste bevindingen van het STW-project in combinatie met enkele empirische gegevens uit de recente pot en veldproeven (WUR en NMI) die zijn uitgevoerd voor de gewassen gras, maïs en tarwe..

### Borium

Borium (B) stimuleert in planten de celdeling en groei, bloei en vruchtzetting. Voor dieren speelt B geen rol van betekenis. Anders dan andere micronutriënten is B slechts zwak geadsorbeerd aan bodemdeeltjes. Bij toenemende pH van de bodem wordt B in de bodem minder beschikbaar door een toename in de binding van B aan organisch materiaal (OM), klei en het oppervlak van Al- en Fe-oxide en. De volgende gronden zijn gevoelig voor boriumgebrek:

- bodems die lijden aan droogte;
- bodems met een pH lager dan 4 of hoger dan 6; en
- bodems met een laag gehalte aan organische stof en klei.

Bodems met een grove textuur (dominant in zand) zijn het meest gevoelig voor een B-tekort als gevolg van, over het algemeen, lagere organische stofgehalten en oppervlak waar B aan kan adsorberen en een lage(re) pH in vergelijking met bodems met een fijne textuur (dominant in klei). Bovendien hebben bodems met een grove textuur een laag watervasthoudend vermogen en spoelt B gemakkelijk uit tijdens natte omstandigheden.

De hoeveelheid B dat met verschillende bodem extractie methodes wordt geëxtraheerd is gerelateerd aan een combinatie van algemene bodemeigenschappen die samen de potentiële adsorptiecapaciteit bepalen; organische stofgehalte, kleigehalte en pH. Deze algemene bodem kenmerken kunnen allemaal nauwkeurig worden voorspeld met behulp van sensortechnieken op basis van NIR (nabij infrarood) en MIR (mid infrarood). Dit geeft een goed perspectief om ook B-beschikbaarheid te voorspellen met deze sensortechnieken.

De volgende conclusies en aanbevelingen worden gedaan om de B bemestingsadviezen te verbeteren:

- Het uitvoeren van grondonderzoek om te bepalen of de B-status te laag is, is in het algemeen niet relevant voor kleigronden met een organische stofgehalte van meer dan 5%. Voor deze gronden is het B-gehalte (meer dan) hoog genoeg, en door de lage bindingssterkte ook voldoende beschikbaar.

- Het uitvoeren van grondonderzoek kan echter wel nuttig zijn op klei- en zandgronden met een organisch stofgehalte hoger dan 5% voor gewassen die gevoelig zijn voor B-toxiciteit; bijv. maïs.
- Bekalken heeft een negatief effect op de beschikbaarheid van B. Voor de meest betrouwbare resultaten zou bij voorkeur B-bodemonderzoek moeten worden uitgevoerd in het voorjaar voor het groeiseizoen, maar na kalken.
- Een potexperiment met tarwe toont een duidelijke relatie tussen de B-concentratie in bladweefsel en de parameters pH en B-CaCl<sub>2</sub>. In veldexperimenten kon deze relatie niet verder worden onderbouwd. Het belang van kleigehalte, organische stof en de pH op de B-beschikbaarheid geeft echter wel aan dat het de moeite waard is om te onderzoeken of deze parameters ingebed moeten worden in de B-bemestingsadviezen.
- De reactie van gras op B-bemesting kan worden voorspeld op basis van het B-gehalte van de bodem (B-CaCl<sub>2</sub> of B-ox) of op basis van de potentiële adsorptiecapaciteit voor B in de bodem.
- De huidige bemestingsadviezen kunnen worden verbeterd door onderscheid te maken tussen gewassen op basis van hun B-behoefte. Het wordt aanbevolen om een advies voor peulvruchten te ontwikkelen als gevolg van de relatief hoge B-behoefte en de rol van B bij de vorming van wortelknollen.
- In theorie, is de levering van B uit de bodem in combinatie met de aanwending van organische mest toereikend om gewassen te voorzien in hun B-behoefte. Het effect van steeds strikter geworden gebruiksnormen en de veronderstelling dat B in organische mest direct beschikbaar is voor het gewas moeten echter nader worden onderzocht.
- Het wordt aanbevolen om de weersomstandigheden op te nemen in de adviezen voor droogte en uitspoeling gevoelige zandgronden. Een weermodule kan worden ontwikkeld die zowel feitelijke gegevens over het weer en een eenvoudig wortel-ontwikkelingsmodel bevat. Adviezen zouden zich in situaties van droogte moeten richten op irrigatie in plaats van op bemesting. Het effect van klimaatverandering op B-deficiëntie als gevolg van extreem natte en droge perioden verdient meer aandacht.
- Het voorkomen van een achteruitgang van het organisch stofgehalte en het onderhouden van de bodem pH op een voldoende en zeker niet te hoog niveau is belangrijk voor het behoud van de B-beschikbaarheid.

### **Koper**

In de bodem wordt koper (Cu) vooral geassocieerd met organische stof. De bodem pH speelt ook een belangrijke rol omdat de Cu beschikbaarheid afneemt met toenemende pH door een sterkere adsorptie aan OM, maar ook een sterkere adsorptie aan de andere bodem adsorptie oppervlakken zoals klei en Al- en Fe (hydr-) oxides. Cu-deficiëntie komt voor in gronden waar te weinig Cu aanwezig is, bijvoorbeeld door een laag organisch stofgehalte. Cu-deficiëntie komt echter ook voor in gronden waar veel Cu aanwezig is, maar vanwege de sterke binding dit niet beschikbaar is voor gewasopname, bijvoorbeeld in veen en andere organische bodems. De Cu-behoefte is hoog voor suikerbieten, spinazie, tarwe, haver, lucerne, sla en ui. De Cu-behoefte is voor koeien van dezelfde orde van grootte als van hun voedergewassen. Bij koeien kan concurrentie met molybdeen en zwavel, Cu-deficiëntie veroorzaken.

In de routinematige grondlaboratoria (Eurofins-agro) wordt Cu momenteel gemeten in een 0,01 M CaCl<sub>2</sub> extract. De methode waar het bemestingsadvies op is gebaseerd, Cu-HNO<sub>3</sub>, wordt sinds 2004 niet meer op commerciële schaal uitgevoerd. Cu-CaCl<sub>2</sub> extraheert slechts een kleine fractie (+/- 1%) van de hoeveelheid reactief Cu dat wordt geëxtraheerd met de traditionele methode (Cu-HNO<sub>3</sub>). Cu-HNO<sub>3</sub> extraheert het reactieve Cu dat vooral wordt geassocieerd met het bodem organische stofgehalte. Cu-CaCl<sub>2</sub> benadert de Cu-concentratie in de bodemoplossing, waarvan 99% is gebonden aan opgeloste

organische stof en slechts 1% aanwezig is in de vorm die ook gemakkelijk kan worden opgenomen door plantenwortels ( $\text{Cu}^{2+}$ ).  $\text{Cu-HNO}_3$  is een slechte voorspeller van Cu beschikbaarheid. Data uit een monitoringsexperiment laat zien dat  $\text{Cu-CaCl}_2$  een betere voorspeller is van het Cu-gehalte in gras dan  $\text{Cu-HNO}_3$ . Voor een redelijke voorspelling zijn echter ook co-variabelen nodig. Het AIO-project toonde contrasterende resultaten met betrekking tot de relatie tussen Cu-opname en  $\text{Cu-CaCl}_2$ .

Het is opmerkelijk dat in de drie veldproeven met gras en maïs het Cu-gehalte in het gewas op alle percelen lager was dan het streefniveau voor voedergewassen voor koeien. Het is een punt van discussie of het streefniveau van  $12 \text{ mg Cu kg}^{-1} \text{ DM}$  gras te hoog is en of deze niet verlaagd zou moeten worden. Dit vereist echter meer onderzoek.

Het (te) lage Cu-gehalte in de veldproeven kon niet worden gerelateerd aan het Cu-gehalte in de bodem. De relatie tussen Cu-gehalte in het gewas en de verschillende bodem Cu-extractiemethodes tonen tegenstrijdige resultaten; variërend van een negatieve trend (maïs, veldproef), geen relatie (gras veldproef 2011), een positieve trend (gras veldproef 2012) tot een positieve relatie (tarwe potproef). Het potexperiment met tarwe dat werd uitgevoerd onder meer gecontroleerde omstandigheden suggereert een maximum Cu-gehalte in het gewas bij een  $\text{Cu-CaCl}_2$  hoger dan  $50 \mu\text{g/kg}$ . Het werd verwacht dat de bodemfactoren organische stof en pH een rol zouden spelen in de Cu-beschikbaarheid, maar daarvoor werd in de pot- en veldproeven geen bewijs gevonden.

In de veldproef was er een duidelijke positieve reactie van het gras op bemesting met Cu-sulfaat. Ondanks het grote bereik in organische stofgehalte (1-22%) en een factor 10 verschil in beschikbare Cu in de bodem, was de respons in Cu-opname op de Cu-bemesting ongeveer gelijk voor alle percelen. Dit is een indicatie dat nog het Cu-gehalte nog het organische stofgehalte de gewasrespons op Cu-bemesting lijken te beïnvloeden. De effectiviteit van de Cu-bemesting was echter zeer laag (0,6%), wat overeenkomt met oudere studies. Dit voedt de discussie of Cu-bemesting met Cu-zouten wel duurzaam is en of bemesting met Cu-chelaten niet te prefereren is. Momenteel zijn Cu-chelaten echter niet beschikbaar voor grasland en maïs.

Voor voedergewassen wordt een laag Cu-gehalte in het ruwvoer in het algemeen (meer dan) gecompenseerd door het gebruik van krachtvoer. Alleen jongvee en droge koeien lopen het risico op een Cu-tekort tijdens de zomer, omdat deze diergroepen dan weinig of geen krachtvoer krijgen (Den Boer et al., 2011). Gegevens tot 2009 tonen een continue daling van het Cu-gehalte in organische mest (CBS). In combinatie met afnemende mestgiften en de lage Cu-gehalten in de gewassen in de veldproeven (2011 - 2012) betekent dit dat de huidige streefniveaus voor Cu in het gewas moeilijk kunnen worden gehandhaafd. Dit vergt nader onderzoek.

De volgende aanbevelingen worden gedaan om de Cu-bemestingsadviezen te verbeteren:

- Een hoge pH heeft een negatief effect op Cu beschikbaarheid in de bodem. Het is dus belangrijk om de bodem pH op een goed niveau te houden maar niet te hoog.
- Bekalken heeft een negatief effect op de beschikbaarheid van Cu. Voor de meest betrouwbare resultaten zou bij voorkeur het Cu-bodemonderzoek moeten worden uitgevoerd in het voorjaar voor het groeiseizoen, maar na kalken.
- De aanbevolen zwavel (S) bemesting handhaven bij voedergewassen omdat S een negatieve invloed heeft op Cu-opname in de koe. Te hoge S-giften, ook in de vorm van gips, moeten worden voorkomen.

### **Mangaan**

Mangaan (Mn) is een essentieel element voor zowel gewassen als voor de gezondheid van dieren. De beschikbaarheid van Mn in de bodem wordt grotendeels bepaald door het oplossen en neerslaan van Mn-mineralen. Dit is in tegenstelling tot andere (micro) nutriënten waar voornamelijk adsorptie / desorptie processen de beschikbaarheid bepalen. De oplosbaarheid van Mn-mineralen, en dus ook de

beschikbaarheid van Mn voor de plant, wordt bepaald door de pH en de redoxpotential van de bodem. Mn-deficiëntie treedt meestal op op kalkhoudende bodems omdat bij hoge pH en redox omstandigheden Mn oxideert tot Mn (IV) en precipiteert in de vorm van zeer onoplosbare oxiden of carbonaten. Op zandgronden treedt geen Mn-deficiëntie op bij een pH-KCl lager dan 5,4. Daarentegen kwam Mn-deficiëntie voor op alle percelen met een pH-waarde hoger dan 6,2. Bij een tussenliggende pH-KCl kwamen zowel niet als wel deficiënte percelen voor (De Vries en Dechering, 1960).

Aanhoudend droog weer verhoogt de kans op Mn-deficiëntie, zelfs in bodems waar voldoende Mn potentieel beschikbaar is. Dit komt omdat Mn hoofdzakelijk door de plantenwortel door 'mass flow' wordt opgenomen en de oplosbare fractie met het water naar het worteloppervlak wordt getransporteerd. De Nederlandse bemestingsadviezen zijn gebaseerd op het meten van reduceerbaar Mn (Mn-red). Deze meting wordt sinds 2004 niet meer op commerciële schaal uitgevoerd. Sinds 2004 wordt in de routinematige grondlaboratoria (Eurofins-agro) Mn gemeten in een 0,01 M CaCl<sub>2</sub> extract.

Tegen de verwachting in geeft Mn-CaCl<sub>2</sub> een goede voorspelling van reduceerbaar Mn. De hypothese is dat herbevochtiging van een gedroogde grond leidt tot de oxidatie van organische stof in de bodem waarbij Mn vrijkomt als gevolg van de reductie van Mn-oxiden (Terrones en Supriatin 2015).

Mn-CaCl<sub>2</sub> blijkt direct gerelateerd aan de pH van de bodem. In overeenstemming hiermee was het Mn-gehalte in tarwe gerelateerd aan de pH en aan Mn-CaCl<sub>2</sub>. De Mn-opname werd geremd bij een pH-CaCl<sub>2</sub> hoger dan 6 en Cu-CaCl<sub>2</sub> lager dan 1. Dit werd bevestigd in de gras veldproeven. Bladbemesting is meestal effectief in het voorkomen en verlichten van een Mn-tekort. De effectiviteit van de bladbemesting is afhankelijk van weersomstandigheden (Mulder et al., 2013). Als het duidelijk is dat aanhoudend droge weer de oorzaak is van een waargenomen niet-ernstig tekort is het raadzaam om te irrigeren of te wachten op regen.

De volgende conclusies en aanbevelingen worden gedaan om de Mn-bemestingsadviezen te verbeteren:

- Bij een bodem pH hoger dan 6, is de Mn-beschikbaarheid zo laag dat een bodemanalyse van het Mn-gehalte (Mn-CaCl<sub>2</sub>) niet zinvol is. Bij pH niveaus onder 6, is Mn-CaCl<sub>2</sub> een goede indicator voor beschikbaarheid van Mn in de bodem voor gewasopname.
- De relatie tussen Mn-CaCl<sub>2</sub> en pH zou ertoe kunnen leiden dat Mn-CaCl<sub>2</sub> goed voorspeld kan worden met bodemsensoren op basis van NIR / MIR.
- In de experimenten met verschillende extractiemethoden, een pot experiment met tarwe en veldproeven met gras bleek geen enkele andere parameter, zoals klei- of organisch stofgehalte, van invloed voor het verklaren van de Mn-opname of Mn-CaCl<sub>2</sub> of de relatie tussen pH en Mn-CaCl<sub>2</sub>. In de huidige bemestingsadviezen vindt een indeling van het Mn-gehalte van de bodem plaats op basis van het organische stofgehalte. Dit lijkt echter willekeurig en kan niet worden onderbouwd door de hier gepresenteerde gegevens.
- Een bodemonderzoek voorspelt alleen dat een potentieel tekort kan optreden. Het is wenselijk om een potentieel optredend tekort zo vroeg mogelijk aan te tonen zodat bladbemesting op tijd kan worden toegediend. Hiervoor zijn, en worden, verschillende niet-destructieve diagnostische hulpmiddelen ontwikkeld (Maarschalkerweerd en Husted 2015 Pedas et al., 2014). Dit voorkomt ook een verspilling van meststoffen als Mn-deficiëntie niet optreedt. De aanbevolen dosis is afhankelijk van het gewas en het type meststof.
- Het wordt aanbevolen om een weermodule en een beregeningsadvies te integreren in de bemestingsadviezen. De te ontwikkelen weermodule bevat zowel actuele weersgegevens als een eenvoudige wortelontwikkelingsmodel. In geval van droogte kunnen adviezen zich dan meer richten op beregening in plaats van op bemesting.



## Seleen

Seleen (Se) is een essentieel micronutriënt voor de mens, voor diergezondheid en voor de kwaliteit van dierlijke producten. Er bestaan aanwijzingen dat Se ook gunstig is voor gewasgroei en kwaliteit (Haug et al., 2007). Bemestingsadviezen richten zich uitsluitend op het dier. De optimale Se eis van koeien is ongeveer  $0,15 \text{ mg Se kg}^{-1}$  droge stof in het voer. Zonder Se-bemesting bevat ruwvoer (gras en maïs) in het algemeen onvoldoende Se voor een adequate Se-voorziening van het vee.

Selenium kan in verschillende vormen in de bodem aanwezig zijn, afhankelijk van de pH en de redox-omstandigheden. Selenaat is meestal de meest gangbare vorm onder aerobe en neutrale tot alkalische omstandigheden, terwijl seleniet de belangrijkste vorm is in zure gronden. Selenaat is heel mobiel in de bodem en als gevolg gevoelig voor uitspoeling. Seleniet adsorbeert veel sterker aan de bodem dan selenaat.

Zandgronden bevatten over het algemeen weinig Se. De Se dat in zandgronden aanwezig is wordt geassocieerd met het organisch materiaal (OM, Johnsson, 1992). In landbouwgronden met een laag Se-gehalte is Se voornamelijk geassocieerd met organische stof, zowel in de vaste bodem als in de bodemoplossing (Supriatin, 2016). Kleigronden bevatten relatief meer Se dan zandgrond door de aanwezigheid van Se-verrijkte mineralen zoals biotiet enerzijds, en hogere gehalten aan aluminium en ijzer oxides waar Se aan kan adsorberen anderzijds. Se beschikbaarheid neemt toe met toenemende pH. Bekalken van de bodem is dan ook een bekende strategie om de Se beschikbaarheid in de bodem te verhogen.

In Nederland wordt Se-deficiëntie het vaakst gemeld tijdens het weideseizoen (zomer) op zand- en veengronden. Op de zandgrond ontstaat het tekort door het ontbreken van Se en op veen en organische bodems ontstaat het tekort als gevolg van de lage beschikbaarheid van het aanwezige Se.

In Nederland is er vanuit de bemestingsadviezen geen 'historische' methode om het Se-gehalte in de bodem vast te stellen. Sinds 2004 wordt Se gemeten in een  $\text{CaCl}_2$ -extract. Onder geconditioneerde omstandigheden in een potproef kan de variatie in Se-gehalte van tarwe bladeren het beste worden verklaard door de verhouding tussen Se en opgelost organische stof (DOC) in een  $0,01\text{M CaCl}_2$  extract (Supriatin et al., 2016). Andere parameters die gerelateerd bleken te zijn aan de Se-opname waren pH (hogere opname bij een hogere pH) en de kwaliteit van organische stof (benaderd door de C:N verhouding).

Zowel Se-rijk labiel organische stof als Se-rijk opgelost organische stof is potentieel beschikbaar in de bodem voor opname door planten. In het algemeen hebben zowel klei als pH een positief effect op de potentiële beschikbaarheid van Se. De verhouding tussen C:N in het bodem organische stof – als maat voor de kwaliteit van het organische stof, is negatief gerelateerd aan de potentiële beschikbaarheid van Se.

De relatie tussen Se-opname en Se- $\text{CaCl}_2$  kon in de gras veldproeven (2011 en 2012) niet worden bevestigd. Deze veldproeven toonden wel aan dat de Se-concentraties in het gras zonder Se-bemesting (veel) lager is dan de aanbevolen concentratie in gras voor graasdieren.

Door het toedienen van Se-meststoffen in de vorm van natriumselenaat ( $4,4 \text{ g Se / ha}$ ) nam het Se-gehalte van het gras sterk toe tot een voldoende niveau. In het algemeen bestaat er een hoge respons van gewassen op Se-bemesting (meta-analyses van Ros et al., 2016). De lokale situatie heeft grote invloed op de gewasreactie. Uit de meta-analyses bleek dat kleigehalte een significant effect heeft op de Se-opname respons op Se-bemesting (gedefinieerd door de verhouding tussen Se-gehalte van het gewas met en zonder Se-bemesting). Met toenemend kleigehalte, was er een lagere respons, vermoedelijk door adsorptie van de toegevoegde Se aan de bodem. Dezelfde meta-analyse laat een variabel effect zien van OS-gehalte en pH op Se-opname respons. Bij een Se-gehalte in de bodem hoger dan  $0,2 \text{ mg kg}^{-1}$  was de respons significant groter dan bij hogere Se-gehalten. Het aanpassen van

de bemestingsstrategie aan de plaatselijke agronomische situatie en bodemeigenschappen kan het gewas opname-efficiëntie van 10% (veel voorkomende situatie) tot 50% te verhogen (Ros et al., 2014). In Nederland is het optreden van Se-deficiëntie het meest waarschijnlijk op zandgronden met een laag organisch stofgehalte. Vanwege het lage klei- en Se-gehalte is op deze gronden echter ook de grootste respons op Se-bemesting te verwachten.

De volgende aanbevelingen worden gedaan om de Se-bemestingsadviezen te verbeteren:

- Bij gebruik van Se-meststoffen dient rekening te houden met de Se-levering en beschikbaarheid in de bodem en eventueel resterende effecten van de voormalige Se bemesting.
- Bemesting met selenaat is te prefereren boven seleniet; Selenaat is ongeveer 8 keer meer effectief op de korte termijn dan seleniet.
- Zowel blad- als bodem meststoffen zijn in staat om de Se-opname te verhogen, maar bladbemesting is efficiënter. Zaadcoating kan een alternatief zijn maar de opname-efficiëntie is meestal minder dan 10%.
- Gedeelde bemestingsgiften tijdens het groeiseizoen resulteren in een hogere Se-niveaus in het gewas ten opzichte van één enkele gift vóór het groeiseizoen. Een meer frequente bemesting bij een lagere dosis wordt aanbevolen om een voldoende hoog Se-gehalte tijdens het groeiseizoen te waarborgen, alsmede de verliezen door uitspoeling en het risico van toxische niveaus te minimaliseren. De frequentie is afhankelijk van de gekozen meststof.

## Summary and recommendations

In the past decades the importance of micronutrients for agriculture has become increasingly clear. In addition, it has also become clear that fertilization management of micronutrients is a delicate issue in which there is a fine line between deficiency and toxicity. There still is a world to be explored in terms of understanding, measuring and predicting availability of micronutrients in the soil and the translation into fertilization recommendations.

In 2010 a STW project "Micronutrient Management for Sustainable Agriculture and Environment: A new innovative approach" was started at the Department of Soil Quality of Wageningen University. Two PhD students were involved. The study was finalized in the summer of 2016. The aim was to get a better understanding of the behavior of the elements boron (B), copper (Cu), selenium (Se), and manganese (Mn) in order to improve the Dutch micronutrient fertilizer recommendations. NMI was involved in supervising the PhD work and the utilization of the results. The NMI activities were commissioned by the Dutch Dairy Board.

This report aims at recommending possible improvements of the current Dutch micronutrient fertilizer recommendations. These recommendations for the elements B, Cu, Se, Mn are derived from a compilation of the scientific literature, the main findings of the STW project in combination with some empirical data from recent pot and field trials (WUR and NMI) for grass, maize and wheat.

### Boron

In plants B stimulates cell division and growth, flowering and fruit set. For animals B does not play a role of importance. Contrary to the other micronutrients B is adsorbed only weakly to soil particles. With increasing soil pH, B adsorption on to organic matter (OM), clay and Al and Fe-oxide surfaces increases and B in the soil becomes less available. The following soils are sensitive to boron deficiency:

- soils suffering from drought,
- soils with a pH lower than 4 or higher than 6, and
- soils with a low content of organic matter and clay.

B deficiency is most prone to occur on coarse textured soils (dominance of sand) due to generally lower organic matter (OM) levels a low(er) pH and adsorption surface area compared to fine textured soils (dominance of clay). In addition, moisture content and water holding capacity are both low on coarse textured soils and B is easily leached during wet conditions. Under dry circumstances there is the risk of a drought- hindered mass flow.

The amount of B extracted with different soil extraction methods is related to a combination of general soil properties that together determine the potential adsorption capacity; organic matter content, clay content and pH. These general soil characteristics can all be accurately predicted using sensor techniques based on NIR and MIR. This provides a good perspective to also predict B availability.

The following conclusions and recommendations are made to improve the B fertilization recommendation:

- Soil analysis is generally not relevant for clay soils with organic matter content greater than 5% as B content is (more than) sufficient, and due to the low binding strength also sufficiently available.
- However, soil analyses may be useful in terms of toxicity on clay soils and sandy soils with an organic matter content higher than 5% for crops that are sensitive for B toxicity; e.g. maize.
- It should be taken into account that liming decreases B availability in the soil. To get the most reliable results B soil analysis should ideally be performed in spring before the growing season but after liming.

- A greenhouse experiment with wheat shows a clear relationship between B concentration in leaf tissue and the combined parameters pH and B-CaCl<sub>2</sub>. In field experiments this relationship could not be further substantiated. The importance of texture, organic matter and pH on B availability indicates that is worthwhile to investigate if these parameters need incorporating in the B recommendations.
- The response of grass to B-fertilization could be predicted based on the B level of the soil (B-CaCl<sub>2</sub> or B-ox) or on the potential adsorption capacity for B in soil.
- Current fertilizer recommendations can be improved by distinguishing between crops in terms of B requirement. It is recommended to develop a fertilizer recommendations for legumes due to relatively high B requirements and the role of B in the formation of root nodules.
- In theory, the combination of an adequate B level of the soil together with the application of organic manure results in sufficient B to equal removal rates by the crop. The effect of stricter application rates and the assumption that the B in organic manure is readily available for the crop should be investigated.
- It is recommended to incorporate weather conditions in the recommendations for coarse textured drought and leaching sensitive soils. A weather module may be developed that includes both actual weather data and a simple root development model. Recommendations in situations of drought must focus on irrigation rather than on fertilization. B deficiency as a result of extreme wet and dry periods due to climate change deserves further attention.
- Preventing the deterioration of soil OM content and maintaining soil pH at an adequate and not too high level is important for maintaining B availability.

### Copper

In soil Cu is mainly associated with soil organic matter (OM). The soil pH also plays an important role as Cu availability decreases with increasing pH due to increasingly strong adsorption to OM but also to the other soil constituents clay and Al- and Fe-(hydr-)oxides to which Cu adsorbs. Cu deficiency occurs in soils where too little Cu is present, e.g. soils low in OM. Cu deficiency also occurs in soils where a lot of Cu is present but due to strong binding it is not available for crop uptake, e.g. in peat and organic rich soils. Cu requirement is high for sugar beets, spinach, wheat, oats, alfalfa, lettuce, onion. The Cu requirement for cows is of the same order of magnitude as for the fodder crops on which they feed. In cows competition with Molybdenum and Sulphur is known to induce Cu deficiency.

In Dutch routine soil laboratories (Eurofins-agro) Cu is currently measured in a 0.01M CaCl<sub>2</sub> extract. The method on which the fertilizer recommendations are based, Cu-HNO<sub>3</sub>, has not been used on a commercial scale since 2004. Cu-CaCl<sub>2</sub> extracts only a small fraction (+/-1%) of the amount of reactive Cu that is extracted using the traditional method (Cu-HNO<sub>3</sub>). Cu-HNO<sub>3</sub> extracts reactive Cu that is mainly associated with soil organic matter content. Cu-CaCl<sub>2</sub> approximates the Cu concentration in solution of which 99% is bound to dissolved organic matter and only 1% is in the form that can readily be taken up (Cu<sup>2+</sup>). Cu-HNO<sub>3</sub> is a poor predictor of Cu availability. Cu-CaCl<sub>2</sub> is better predictor of grass content, based on monitoring data. However co-variables are involved. The PhD project showed contrasting results regarding the relation between Cu content and Cu-CaCl<sub>2</sub>.

It is remarkable that of the three field trials that were conducted with grass and maize all fields showed a Cu content in the crop that was lower than the required levels for crops and cows. This raises the discussion if the target level of 12 mg Cu kg<sup>-1</sup> DM grass is not too high and whether or not it should be decreased. This needs further investigation.

The (too) low Cu-levels in the field trials could not be related to soil test results. The relationship between Cu content in the crop and the different soil Cu extraction methods show contradicting results; varying

from a negative trend (maize field), to no relationship (grass field 2011), to a positive trend (grass field 2012), to a positive relationship (wheat pot). The wheat pot experiment that was performed under more controlled conditions suggests a maximum Cu uptake by wheat at a Cu-CaCl<sub>2</sub> higher than 50 µg/kg. The soil factors organic matter and pH were expected to play a role in Cu availability but no evidence was found in the pot and field trials.

There was a clear positive response of grass in the field to fertilization with Cu-sulfate. Despite the large range in OM level (1-22%) and a factor 10 difference in available Cu, the response in Cu-uptake to Cu-fertilization was approximately equal for all fields. Thus indicating that Cu-levels or soil organic matter content do not influence the crop response to fertilization. The efficiency of the Cu fertilizer was however very low (0.6%), which is in line with older trials. This opens the discussion if Cu fertilization with Cu salts is sustainable and if Cu chelates are not a better option. Currently Cu chelates are however not available for grassland and maize.

For the fodder crops a low Cu content of the roughage is generally (more than) compensated for by the use of concentrates. Only young stock and dry cows are at risk for Cu deficiency during the summer because these groups then receive little or no concentrates (Den Boer et al., 2011). Data until 2009 show a continuous decrease in Cu-content in organic manure (CBS). In combination with decreasing fertilizer dose and the low Cu-levels in the crops in the field trials (2011 – 2012) this implies that maintaining current Cu levels in the crop may become difficult. This needs further investigation.

Aspects that need taking into account in the fertilization recommendation are:

- A high pH has a negative effect on Cu availability in the soil. It is thus important to keep soil pH at a good, and not too high level.
- When applying lime (CaCO<sub>3</sub>) it is recommended to perform soil analyses after liming;
- The recommendation levels for S fertilization of fodder crops should be followed as S has a negative influence on Cu uptake in the cow. Too high an application of S, including gypsum, should be prevented.

## Manganese

Manganese (Mn) is an essential element for crops as well as for animal health. The availability of Mn in soil is to a large extent determined by the dissolution and precipitation of Mn-minerals. This is in contrast to other (micro) nutrients where adsorption/desorption processes generally determine the availability. The solubility of Mn-minerals and thus the availability of Mn for the plant is determined by the pH and redox potential of the soil. Mn deficiency generally occurs on calcareous soils because at high pH and redox conditions Mn is oxidized to Mn (IV) which precipitates in the form of highly insoluble oxides or carbonates. On sandy soils no Mn deficiency is found to occur at pH-KCl lower than 5.4 while deficiency was found to occur on all fields with a pH higher than 6.2. At an intermediate pH-KCl both healthy and deficient fields occurred (De Vries and Dechering, 1960).

Persistently dry weather increases the chance of Mn deficiency, even in soils where sufficient Mn is potentially available. This is because Mn is mainly transported in the liquid phase, via mass flow from the soil to the root surface.

The Dutch fertilizer recommendations are based on measuring reducible Mn (Mn-red). From 2004 onwards Mn-red is no longer measured at the routine soil laboratories but a switch was made to Mn-CaCl<sub>2</sub>. Contrary to expectations Mn-CaCl<sub>2</sub> gives a good prediction of reducible Mn. The hypothesis is that rewetting a dried soil induces the oxidation of organic matter in the soil which liberates Mn due to reduction of Mn-oxides (Terrones and Supriatin, 2015).

Mn-CaCl<sub>2</sub> is found to be directly related to the soil pH. In agreement herewith the Mn content in wheat leaves was closely related to pH and Mn-CaCl<sub>2</sub> and showed a clear inhibition in Mn uptake at pH-CaCl<sub>2</sub> higher than 6 and Cu-CaCl<sub>2</sub> lower than 1. This was confirmed in the grass field trials.

Foliar applications are generally found to be effective in preventing and alleviating Mn deficiency. The effectiveness of foliar Mn-application was found to depend on the weather (Mulder et al., 2013). When it is clear that continuous dry weather is the cause of an observed non-severe deficiency it is advised to irrigate or to wait for rain rather than to spray foliar fertilizer.

The following conclusions and recommendations are made to improve the Mn fertilization recommendation:

- At pH levels higher than 6, Mn availability is so low that further analyses of the soil Mn level (Mn-CaCl<sub>2</sub>) is not useful. At pH levels below 6, Mn-CaCl<sub>2</sub> is a good indicator for Mn availability in soil for crop uptake.
- The relationship between Mn-CaCl<sub>2</sub> and pH may lead to a good prediction of Mn-CaCl<sub>2</sub> using pH predicted with soil sensors based on NIR/MIR.
- In experiments with different extraction methods, the pot experiment with wheat, and the field trials with grass no other factors than pH, e.g. organic matter content or clay content were found to affect Mn uptake or Mn-CaCl<sub>2</sub> or the relationship between pH and Mn-CaCl<sub>2</sub>. The division based on organic matter content in the currently used classification of Mn-levels in soil for the fertilizer recommendations seems arbitrary and cannot be substantiated by the data presented here.
- The soil testing only predicts that a deficiency might potentially occur. It is desirable to diagnose potentially occurring deficiency as early as possible in order to apply foliar Mn fertilizer on time. To this means different non-destructive diagnostic tools have been and are being developed (Maarschalkerweerd and Husted 2015, Pedas et al., 2014). This also prevents wasting Mn fertilizer in the case Mn deficiency does not occur. The recommended dose is crop and foliar fertilizer brand specific.
- It is recommended to integrate a weather module and irrigation advice in the fertilizer recommendation. A weather module may be developed that includes both actual weather data and a simple root development model. Recommendations in situations of drought may focus more on irrigation rather than on fertilization.

## Selenium

Selenium (Se) is an essential micronutrient to humans, for animal health and animal product quality, with indications that it might also be beneficial for crop growth and quality (Haug et al., 2007). Recommendations focus solely on cattle. The optimum Se requirement of cows is around 0.15 mg Se kg<sup>-1</sup> dry matter in the feed. Without Se fertilization roughage (grass and maize) generally contains insufficient Se to ensure adequate supply of cattle.

Selenium may occur in different forms in the soil depending on the pH and the redox conditions.

Selenate tends to be the predominant form in aerobic and neutral to alkaline environments, whereas selenite is the major form present in acid soils. Selenate is quite mobile in soil and as a consequence is prone to leaching. Selenite is adsorbed much stronger to the soil than selenate.

Sandy soils generally contain little Se. The Se present in sandy soils is associated with organic matter (OM, Johnsson, 1992). In agricultural soils low in Se, Se is also mainly associated with OM, both in the solid phase sand in the soil solution (Supriatin et al., 2015). Clayey soils contain relatively more Se than sandy soils due to the presence of Se-enriched minerals such as biotite on the one hand, and higher

levels of aluminum and iron oxides for Se sorption on the other. Se availability increases with increasing pH. Liming of soils is therefore a well-known strategy to increase Se availability in the soil solution.

In The Netherlands the risk of Se deficiency has been reported to be greatest during the grazing season (summer) in sandy and peaty soils. In the sandy soils the deficiency will be due to the lack of Se and in the peat and organic rich soils due to the low availability of the Se that is present.

In the Netherlands, there is no 'historic' method to determine available Se in soil. Since 2004 Se is measured in the CaCl<sub>2</sub> extract. Under controlled conditions of a pot trial the variability in Se content in the wheat shoots is largely explained (88%) by the Se to DOC ratio both measured in a 0.01 M CaCl<sub>2</sub> extract (Supriatin et al., 2016). Other parameters that were found to be related to Se-uptake were soil pH (higher uptake at higher pH), and soil organic matter quality (i.e. soil C:N ratio).

Both Se-rich labile organic matter and Se-rich DOM are potential bioavailable Se in soils for plant uptake. In general, both clay content and soil pH have a positive effect on the potential bioavailable Se. The soil C:N ratio, as a parameter for organic matter quality, is inversely related to potential bioavailable Se.

The relationship between Se-uptake and Se-CaCl<sub>2</sub> could not be reproduced under field conditions in grass trials (2011 and 2012). These grass field trials did underpin that the Se concentration in grass without micronutrient fertilization is (far) below the recommended concentrations for ruminants.

Application of Se fertilizer in the form of sodium selenate (4.4 g Se/ha) significantly increased the Se content of the grass to a sufficient level. Crops are generally found to have a high response to applied Se fertilizer (meta-analyses by Ros et al., 2016). The local situation greatly affects crop response. The meta-analyses showed that clay content has a significant effect on the crop uptake response to Se-fertilization (defined by the ratio of Se content of the crop with and without Se fertilization). With increasing clay content, response decreased presumably due to sorption of the added Se. The same meta-analyses did not show an effect of OM content and the response to pH was variable. At a Se content of the soil lower than 0.2 mg kg<sup>-1</sup> the response was significantly larger than at higher Se levels. Adapting fertilizer strategies to the local agronomic situation and soil properties can increase the crop uptake efficiency from 10% (common situation) up to 50% (Ros et al., 2014). In the Netherlands Se deficiency is most likely to occur on sandy soils with low OM content. Due to the low clay content and low Se content, these soils are thus expected to show the largest response to Se-fertilization.

The following recommendations are made to improve the Se fertilization recommendation:

- Se fertilizer dose should account for the Se supply and availability in the soil and any residual effects of former Se fertilizer applications.
- Fertilization with selenate is preferred above selenite. Selenate is about 8 times more effective on the short term than selenite but the residual effect over time is lower.
- Both foliar and soil applied fertilizers are able to enhance Se uptake but foliar application is more resource efficient. Seed coating can be an alternative but crop uptake efficiency is usually less than 10%.
- Split fertilizer application during the growing season results in higher Se levels in the crop compared to a single application before the growing season. A more frequent fertilization at a lower dose is recommended to ensure a good Se content during the growing season, to minimize losses by leaching and to minimize the risk of toxic levels. The frequency depends on the chosen fertilizer.

## 1 Introduction

In the past decades the importance of micronutrients for agriculture has become increasingly clear (Mortvedt et al., 1991; Bell and Dell, 2008; Voortman, 2012; Udo de Haes et al., 2012; Chardon and Oenema, 2013). In addition, it has also become clear that fertilization management of micronutrients is a delicate issue in which there is a fine line between deficiency and toxicity and that there still is a world to be explored in terms of understanding, measuring and predicting availability of micronutrients in the soil and the translation into defining fertilization recommendations.

Diminishing worldwide reserves of soil nutrients, micronutrient deficiency in crops, animals, and humans in combination with a growing world population make it our obligation to use micronutrients as efficiently as possible. For this a robust and reliable fertilization strategy is needed that takes the spatial and temporal variability in climatic conditions, soil properties, and cropping systems into consideration.

Fertilizer dose must comply with crop demand in which the capacity of soils to supply or retain micronutrients during the growing season must also be taken into account. The main soil properties controlling availability of the micronutrients in the soils and of the applied micronutrients include acidity, redox potential, clay- and organic matter content, biological activity and cation- exchange capacity (e.g. Fagaria et al., 2002, Wattel-Koekoek and Bussink, 2003). Agronomic practices such as liming, irrigation and basic fertilization (nitrogen, phosphorus and sulphur) additionally affect the crop uptake efficiency of applied micronutrients.

The capacity of the soil to supply nutrients is generally determined using selective extraction methods.

Total micronutrient content in the soil is generally a poor indicator of the amount of a specific micronutrient that is available in the soil solution for uptake by the plant roots. The reason is that the nutrients are also part of the soil matrix in the form of minerals and stable organic matter. For plant uptake the nutrients that are adsorbed to the surface of soil particles or are part of the organic matter that mineralizes during a growing season are important. There is not necessarily a relationship between the total and the available nutrient content in the soil.

The reason that traditionally relatively strong extractants are used to measure the micronutrient content in a soil generally has an analytical background. When the methods were developed the apparatus that was used generally had high detection limits for the micronutrients. The strong extractants, extract relatively large quantities and thus high concentrations that could be measured. The last decades have shown a trend towards weaker extraction methods that can be used to predict the availability of more than 1 element (multi nutrient). Currently it is becoming common practice to assess nutrient availability using sensors based on near or mid infrared spectroscopy. These are especially interesting because, after an appropriate validation, they are fast and cheap and the soil does not need to be processed using chemicals but it stays in tact during the analysis. Appendix1 describes the theoretical background of assessing micronutrient levels in soil using different soil extraction methods and the prospects of using spectroscopy based sensors.

In 2010 a STW project "Micronutrient Management for Sustainable Agriculture and Environment: A new innovative approach" was started at the Department of Soil Quality of Wageningen University. Two PhD students were involved. The study was finalized in summer 2016. The aim was to get a better understanding of the behavior of the elements B, Cu, Se, Mn in order to improve the Dutch micronutrient fertilizer recommendations. NMI was involved in supervising the PhD work and the utilization of the results. The NMI activities were commissioned by the Dutch Dairy Board.

This report aims at recommending possible improvements of the current Dutch micronutrient fertilizer recommendations. These recommendations are derived from a compilation of the scientific literature, the main findings of the STW project in combination with some empirical data from recent pot and field trials (WUR and NMI). The elements B, Cu, Se, Mn will be discussed in this order in the following chapters.



## 2 Approach

The need to improve the current Dutch fertilization recommendations resulted in a STW funded project at Department of Soil Quality of Wageningen University, in which two PhD students were involved. One focused on Boron (B) and Manganese (Mn), the other on Copper (Cu) and Selenium (Se). The project aimed at a more mechanistic understanding of micronutrient dynamics in the soil and the relationship with crop uptake. It involved lab experiments and pot trials in combination with soil chemical modeling. Background is that in 2004 the largest routine agricultural soil laboratory changed from using different element specific selective extraction methods to measuring all micro- and macronutrients in one extract of 0.01M CaCl<sub>2</sub>. The fertilization recommendations were not adapted. The only change was that the old indices are no longer measured but are now calculated from the new CaCl<sub>2</sub> based indices. Improvement of the micronutrient recommendation based on CaCl<sub>2</sub> is needed to use micronutrients more efficiently, to prevent unnecessary losses to the environment and to maintain an optimal crop quality.

For the macronutrients different studies have been performed to ensure a correct interpretation of the new CaCl<sub>2</sub> based indices in terms of availability for crop uptake and response to fertilization. For the micronutrients the results from the STW project are important to derive direct relationships between the CaCl<sub>2</sub> based indices, and crop and fertilizer response.

In this report the following steps are taken for each of the four micronutrients In order to incorporate the gained knowledge in the current fertilization recommendations:

1. Theoretical background to understand and predict micronutrient availability
  - a. Literature review
  - b. Results from Ph.D. research C. Torrones and S. Supriatin
2. Synthesis of the results and new insights into adapting the fertilization recommendation. If necessary:
  - a. incorporating other parameters that influence micronutrient availability, e.g. soil parameters, weather
  - b. adapting the fertilizer dose for sensitive crops and current crop yields
  - c. suggestions concerning timing of soil sampling
  - d. interaction with other elements

The Ph.D. research contains different elements. These are:

1. How does sample preparation and storage affect the amount of micronutrients that are extracted with 0.01M CaCl<sub>2</sub>?
2. What fractions are extracted with different extraction methods, based on soil chemical modelling?
3. How do the new micronutrient availability indices relate to crop uptake? Different datasets are used from pot- and field trials.
4. How can micronutrient availability be best predicted based on the standard methods used in routine soil laboratories to characterize the soil and available nutrients? Coupling the gained knowledge concerning micronutrient speciation in soil to easy to measure soil parameters. This will lead to a prediction of micronutrient availability that includes factors such as, for example, soil organic matter content, clay content and pH.

This study uses data from different experiments that were conducted between 2011 and 2013 by the PhD students in close cooperation with NMI:

1. 2013; Wheat pot experiment in the greenhouse using 19 widely varying soils from grass fields;
2. 2011; Grass from 21 field plots without fertilizer application (1st cut, 3 plots were fertilised with N)

3. 2012; Grass (1st cut) from 26 fields with 2 plots per field. Both plots received NPK (per ha: 120 kg N; 31 kg P<sub>2</sub>O<sub>5</sub>; 60 kg K<sub>2</sub>O) and one plot also received micronutrients (per ha: 4.4 g Se (Na<sub>2</sub>SeO<sub>4</sub>); 3.5 kg Cu; 1 kg soluble B; 1 kg soluble Mn);
4. 2011; Maize plant from 15 fields on farms in Friesland, with NPK;
5. 2011 and 2012; Potato from a B/S fertilization experiment (1 kg B/Ha +B/+S, +B/-S, -B/+S, -B/-S).

In all these experiments the general soil parameters pH, organic matter content, clay content and cation exchange capacity were measured in combination with different selective extraction methods and crop yield and micronutrient uptake in the aboveground biomass.

### 3 Boron

#### 3.1 *Boron in crops and animals*

In plants B stimulates cell division and growth, flowering and fruit set. B influences the sugar transport in the plant and thereby has indirect influence on carbohydrate metabolism, protein synthesis and the active uptake of ions. In legumes B is necessary for the formation of root nodules. B deficiency is first visible in the dying of growth points of root and stem, which stimulates the sprouting of side buds. Root vegetables are susceptible to B deficiency; in The Netherlands B deficiency in sugarbeets is known as “hartrot” of the beets.

The critical levels range from 5-10 mg B kg<sup>-1</sup> dry weight (DM) in grasses and cereals to 20-70 mg B kg<sup>-1</sup> DM in dicots. Sugar beet is at risk of hartrot if the B-levels in the leaves are higher than 35-40 mg kg<sup>-1</sup> B (literature review by Wattel-Koekoek and Bussink 2003).

For animals B does not play a role of importance.

#### 3.2 *Boron in soil*

Concerning the availability of nutrients for crops the concentration in solution determines what the roots can directly take up. The amount in solution is generally much lower than what a crop takes up due to replenishment from Boron (B) that is adsorbed to or part of soil particles. To define the bioavailability of B both the concentration in solution and the capacity of the soil to buffer this concentration by desorption and dissolution are important.

Boron in the soil solution is primarily present as the uncharged boric acid molecule (B(OH)<sub>3</sub>) and when the soil pH exceeds 7, B is also partly present in the anionic form B(OH)<sub>4</sub><sup>-</sup>. Only a charged ion is adsorbed to the oppositely charged surface of soil particles. This explains why B is only weakly adsorbed to soil particles and adsorption increases with increasing pH due to the formation of the negatively charged borate. With increasing pH B becomes less available due to B adsorption on to clay and Al and Fe-hydroxyl surfaces (Keren and Bingham, 1985).

The concentration in solution is dictated by B adsorbed to soil particles. B is adsorbed to a range of soil particles; Fe- and Al (hydr)oxides, edges of clay minerals, calcium carbonate and organic matter (OM). Organic matter is especially important for B adsorption. Anion competition increases in the order phosphate (P) > Molybdate (Mo) > sulphate (S) but the competitive effect is low, indicating that B adsorption sites are generally specific for B (Fageria et al., 2002).

The potential of a soil to buffer the B concentration in solution is generally low because the amount adsorbed is relatively small compared to the amount in solution. This has the following consequences:

- B is easily leached from the soil profile. B fertilization may thus not be too high due to toxicity problems and leaching.
- Main plant uptake mechanism is mass flow. As a result B availability is very sensitive to drought.

The risk of B deficiency is especially present on light textured soils (sandy soils) and not or hardly present on medium and coarse textured soils (marine and river clay soils). In a study of maize fields (2009 and 2010, Ros and Den Boer (2011) found that B levels were too low on 25% of the alluvial sandy soils compared to only 1% on the clay soils. On the clay soils 85% had a sufficient B level (>0.35 mg kg<sup>-1</sup>).

Adsorption to light textured soils is generally lower due to a low(er) pH and adsorption surface area compared to coarse textured soils. In addition, moisture content and water holding capacity are both low on light textured soils and B is easily leached. As a consequence the risk of low amounts of available B

for plant uptake is high, and under dry circumstances there is also the risk of a drought- hindered mass flow.

Recent (2010 – 2016) work by C. Torres (Ph.D. Wageningen University) shows that, depending on the soil type and pH, certain B fractions in the soil determine the B availability for the crop. In an adsorption study B was added to soil and the distribution of this added B over the soil solution and adsorption to soil particles was measured and modelled. The model was a multisurface model combining the NICA-Donnan and CD-music model (o.a. Weng et al., 2002) Input parameters were: 0.01M CaCl<sub>2</sub> extractable B, total phosphate (ammonium oxalate, P-ox), pH, organic matter content (OM; HA), total clay content, and total Fe-, Al-, Mn(hydr)oxides. It was shown to be possible to model B adsorption with one single set of adsorption affinity constants and some easy to measure soil parameters.

It is striking that in a sandy soil only 15% of the added B is adsorbed and in a clay soil only 35%, the rest of the B remains in the soil solution. Both soils thus have a low B buffer capacity although the buffer capacity is higher of the clay soil compared to the sandy soil. The concentration in solution ranged between 5 and 100 µM and was comparable to the general range for agricultural soils reported by Barber (1995). The modelling results confirm that OM is the most important surface for B adsorption. In a sandy soil with 6% OM, clay and oxide surfaces did not contribute to B sorption, B was adsorbed only to OM. In clay soils (38.5% clay and 17% OM) the mineral surfaces were found only to be important at pH levels below 5. The increase in B adsorption with increasing pH was thus mainly due to adsorption to OM. Modelling B adsorption to a soil thus showed that OM and pH were the most important parameters determining B adsorption.

In summary, the following soils are sensitive to boron deficiency:

- soils suffering from drought,
- soils with a pH lower than 4 or higher than 6, and
- soils with a low content of organic matter and clay.

B deficiency may thus occur in freshly limed soils as B availability is reduced due to adsorption to the CaCO<sub>3</sub> and an increased adsorption to OM and clay due to the increase in soil pH (Fleming, 1980).

### 3.3 *Measuring B in soil*

Worldwide the most commonly used extraction method is hot water. In the Netherlands B is mostly measured in 0.01 M CaCl<sub>2</sub>

Soil OM content is the dominant soil parameter that determines the amount of B extracted with hot water when distinguishing between sandy and clay soils (Figure 3-1, based on a Eurofins Agro dataset from 2003 of 409 soil samples). For clay soils this relationship is slightly affected by pH. For sandy soils pH does not influence this relationship. For clay soils the B-content per percentage OM is lower at low pH (pH<6) than at high pH (pH>6). This is in agreement with the increase in B adsorption with increasing pH. The parameters OM, clay and pH together determine the amount potential adsorption capacity of the soil and thereby the amount of B that is extracted with B-HW. The same is found for B that is extracted with an acid ammonium oxalate extraction where B-ox could also be predicted based on the combination CEC and pH.

In the Dutch fertilizer recommendations no distinction is made between soil types. All soils with a B content greater than 0.35 mg kg<sup>-1</sup> are categorized as having a high B content. From Figure 3-1 **Error! Reference source not found.** it is clear that clay soils with an OM content greater than 5% will have a B content that is more than sufficient. In addition moisture content and water holding capacity are relatively high and buffering is also relatively high on clay soils. It is recommended that for these soils, a soil B analyses is not necessary.

It is questionable if a soil B analyses is necessary for clay soils at all. In the dataset of Eurofins Agro (409 soils) only 2% of the clay soils have a B content classified as being low or very low ( $<0.3 \text{ mg kg}^{-1}$ ). For sand and reclaimed peat soils 45% have a B content that can be classified as being (very) low. The same results were found by Velthof & Van Erp (1998) when analysing the B-state of Dutch soils based on the soil survey of Eurofins Agro for the season 1995/1996. They also found the B-content of clay soils to be generally good. About 20% of the surveyed reclaimed peat soils and 50% of the diluvial sandy soils had a low or very low B-status. When investigating the Eurofins Agro data of the period 1987 – 1996 Velthof and Van Erp (1998) did not find a trend in the B status of arable land. Extrapolating this to the Eurofins dataset of 2003 this indicates that also in the period 1996 – 2003 the B status of arable land (based on the available datasets) does not seem to have deteriorated.

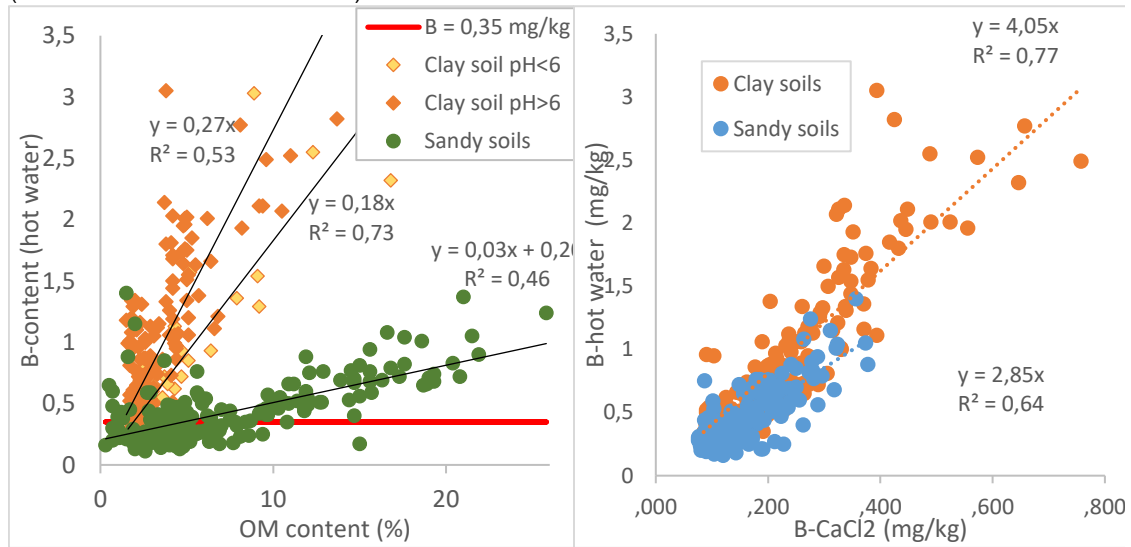


Figure 3-1 Left figure shows the relationship between B-content measured using a hot water extraction and organic matter content when a distinction is made between clay (N=180) and sandy soils (N=229). Right figure shows the relationship between B-content measured using 0.01M CaCl<sub>2</sub> and a hot water extraction when a distinction is made between clay (code 20, 40, 45, 71, N=159) and sandy soils (code 0, 10, 50, N=166). Data from Eurofins.

The relation between the 0.01M CaCl<sub>2</sub> multi-nutrient extraction method and B-HW appears to be different for sandy and clay soils: B-CaCl<sub>2</sub> is approximately a factor 2.8 (sandy soils) and 4.1 (clay soils) lower than B-HW (Figure 3-1).

B-CaCl<sub>2</sub> was found to be significantly correlated with organic matter content (OM), clay content and pH indicating that adsorption-desorption are the important processes that control B-solubility during the extraction procedure. This is confirmed in a dataset containing 1000 soil samples. B-CaCl<sub>2</sub> could be predicted (77% explained variance, Figure 3-2) based on pH, OM, and clay content using the following equation:

$$B\text{-CaCl}_2 (\mu\text{g/kg}) = -463 + 88.9 \times \text{pH} + 22 \times \text{OM} (\%) + 5.58 \times \text{Clay} (\%) \quad \text{Equation 1}$$

Fe- and/or Al oxides (measured in an ammonium oxalate extract) did not improve the explained variance when clay content was included in the model. Organic matter and clay content were equally important in explaining B-CaCl<sub>2</sub> (explained variance +/-41%). CEC was a better single predictor (70% explained variance, Figure 3-2). The explained variance of 77% is high when the analytical error is taken into account. The prediction is based on the sum of the three parameters and as a result the error of these parameters must also be added.

The good prediction of B-CaCl<sub>2</sub> based on the combination of clay, organic matter and pH implies that B-CaCl<sub>2</sub> is determined by the adsorption capacity of the soil and that it is hardly affected by fertilization and

other factors. In the dataset of 1000 samples there are only two clear outliers (red dots in Figure 3-2) which may be due to fertilization or to an analytical error.

Measuring B in the laboratory in an accurate and reproducible way is a challenge. The main reason is that B tends to 'stick' to glassware and tubing. That is also a reason why there is not a perfect relation between 0.01 M CaCl<sub>2</sub> and B-HW. Since B-HW measurement values are 3-4 times higher, these 'stick' problems are less prominent compared to B-CaCl<sub>2</sub>. Recent developments in implementing sensors to predict general soil characteristics and nutrient availability show promising results for B (pers. Comm. Soil Cares Research based on Mid Infrared spectroscopy, data not shown). The reason for this is most probably the close relationship between the availability parameters for B (B-CaCl<sub>2</sub> and B-HW) and the general soil characteristics that determine the adsorption (equation 1, Figure 3-1, Figure 3-2).

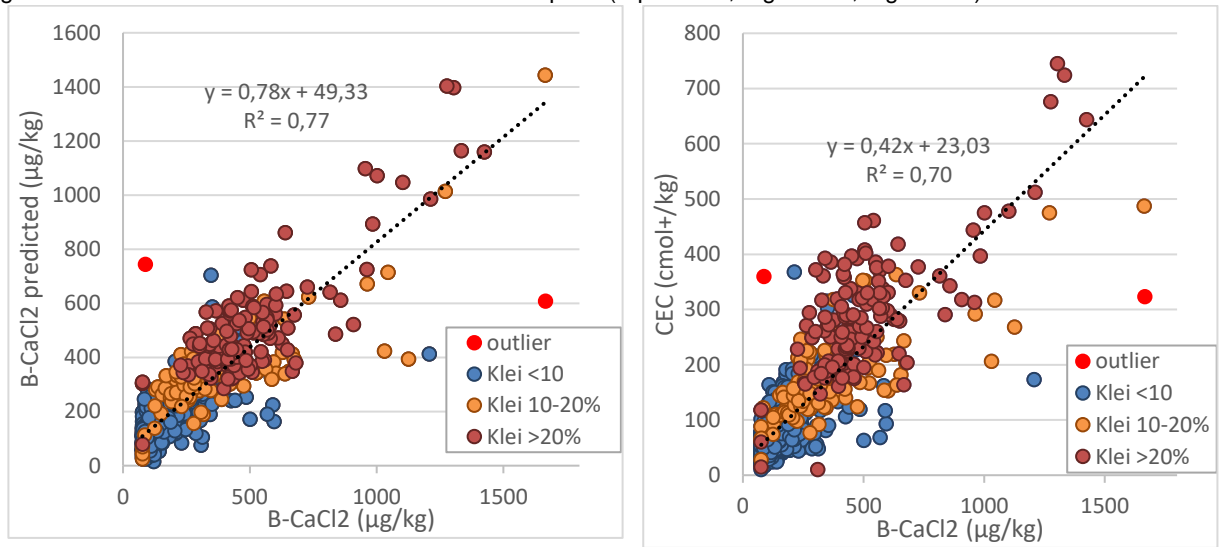


Figure 3-2 Relationship between predicted B-CaCl<sub>2</sub> based on standard soil parameters (pH, OM and clay) and B-CaCl<sub>2</sub> (left figure) and CEC and B-CaCl<sub>2</sub> (right figure).

### 3.4 Predicting B bioavailability for the crop

The relationship between B-CaCl<sub>2</sub> and some easy to measure readily available soil parameters raises the question what the agronomic significance is of B-CaCl<sub>2</sub> in terms of B availability for the crop. This is presented using the data from different field experiments that were conducted in 2011 (grass, maize, potato), 2012 (grass with and without micronutrients fertilization (1 kg B) and 2013 (Potato fertilized with B and S). In addition to these field trials, a wheat experiment was performed in the greenhouse by C. Torrones at Wageningen University. In all these experiments the relationship between B concentration in the crop and the measured soil parameters to approximate B availability in the soil (combination of B-CaCl<sub>2</sub> and other soil characteristics) are investigated. The relationship between B content in the plant tissue and B-CaCl<sub>2</sub> in the different field and greenhouse experiments is shown in Figure 3-3.

There seems to be a good relationship ( $r^2=0.71$ ) between B concentration in the wheat leaves and B-CaCl<sub>2</sub> in the wheat experiment that was conducted under standardized conditions in the greenhouse (Figure 3-3). However, the relationship is mainly based on the soils that have a high B-CaCl<sub>2</sub> and on which the uptake is high. When excluding these two pots the relationship is no longer significant ( $r^2 = 0.14$ ). This relationship becomes more significant (increase to  $r^2 = 0.87$ , Figure 3-4) when pH and B-CaCl<sub>2</sub> are combined in a linear model. Again 2 of the 20 soils are important but excluding them still results in a significant relationship ( $r^2=0.65$ ).

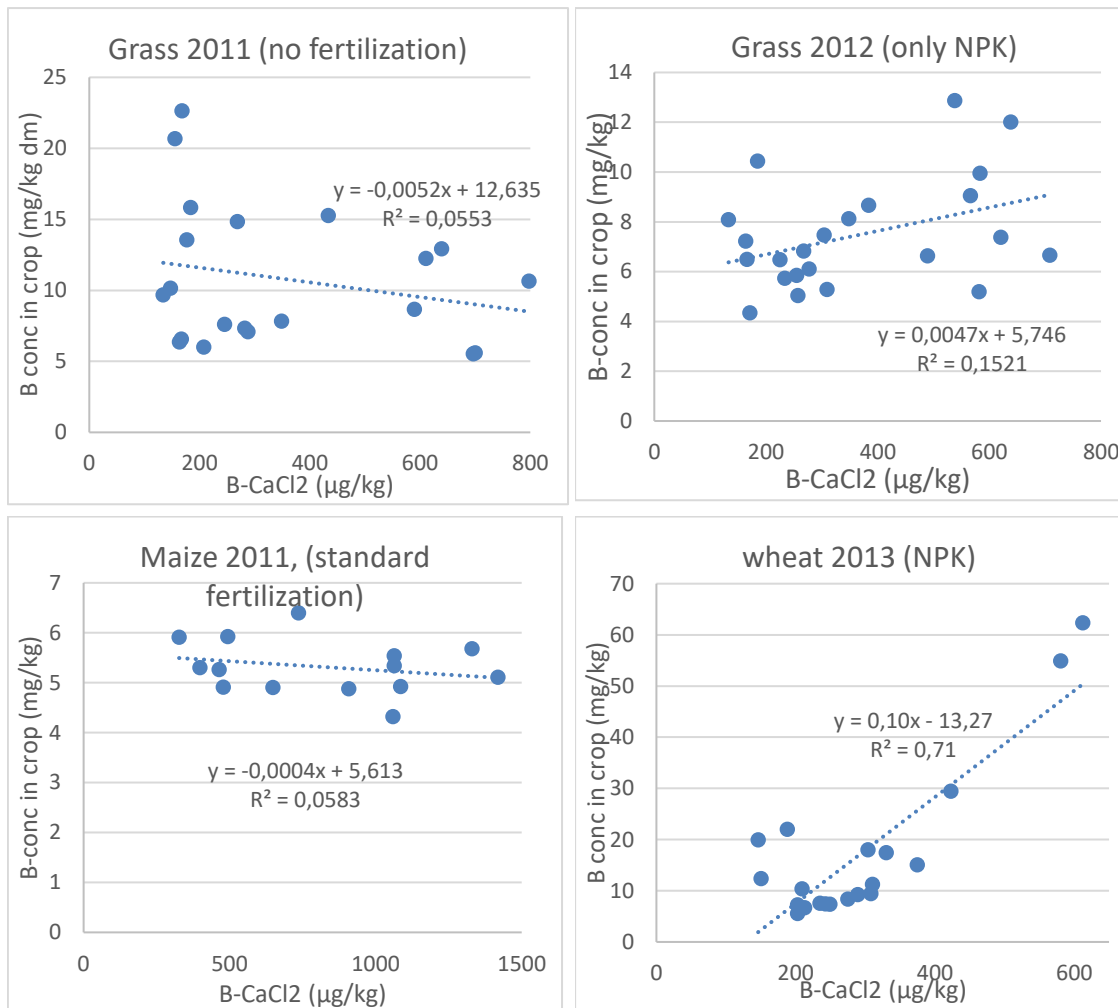


Figure 3-3 relationship between B content of the crop and B-CaCl<sub>2</sub> in different field (grass and maize) and pot (wheat) trials. In none of the shown experiments B fertilizer was applied. In the maize experiment 2011 the fields were fertilized according to common practice.

Of the two parameters pH is more important than B-CaCl<sub>2</sub> in explaining the B concentration in wheat leaves (Figure 3-4). At a pH below 6 there is a clear increase in B concentration of the wheat with decreasing pH ( $r^2=0.75$ ). Above pH 6 the availability seems to be restricted and B levels in the plant tissue are consistently low. This is in accordance with literature where availability and uptake of B are known to decrease dramatically at pH higher than 6 (Fageria et al., 2002).

When considering data from 4 field experiments (grass 2x, maize and potato) there seems to be no relationship between B availability in the soil as measured with B-CaCl<sub>2</sub> and the B concentration in the crop (Figure 3-3, data not shown for the potato trial). In first instance B-CaCl<sub>2</sub> thus does not seem to have a predictive value for B-availability in soil for crop uptake in the field. However, different general soil parameters are related to the B concentration in the crop (as pointed out before B availability is largely driven by adsorption processes and pH). Which parameter differs per experiment / crop. Contrary to the results from the wheat experiment in the greenhouse pH was not a main factor controlling B concentration in the crop in the field trials. For the grass experiments similar results were found in 2011 and in 2012; an increase in B concentration in the grass with increasing OM level of the soil when distinguishing between soils with OM levels lower and higher than 8% (Figure 3-5). In the previous paragraphs the indices for B availability in soil (B-HW and B-CaCl<sub>2</sub>) were also found to be closely related to the combination of organic matter, texture and pH. For the grass experiment only OM was a significant

factor. For soils with OM level higher than 8% there is a clear increase in B concentration in grass with increase in OM level. For soils with less than 8% OM B uptake shows a large variation. Including clay and pH did not improve the explained variance.

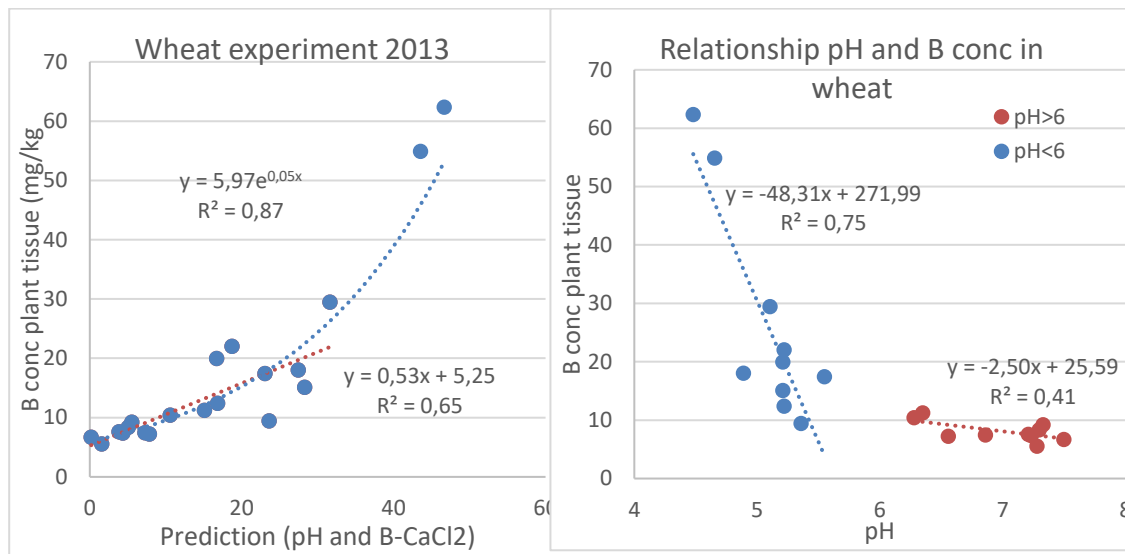


Figure 3-4 Relationship between B concentration in wheat leaves and the combination of pH and B-CaCl<sub>2</sub> (left figure) and with only pH (right figure).

In the field trial with maize, clay is the only soil parameter with which a significant relationship with B concentration in the crop is found ( $r^2=0.4$ , Figure 3-5). The maize trial was fertilized according to common practice and with the application of organic manure some B will also have been added. However, the B concentration in the maize is low and the differences between fields are small (4.2 to 6.4 mg B kg<sup>-1</sup> DM). The level is on average comparable to the critical level of 5 mg B kg<sup>-1</sup> below which deficiency is known to occur. This cannot be related to low B levels in the soils as B-CaCl<sub>2</sub> ranges up to 1.4 mg kg<sup>-1</sup> (Figure 3-3). In the maize experiment another (unknown) factor seems to be suppressing B uptake. Clay content seems related to some extent in suppressing B uptake. If clay content was related to B availability it would be positively related to B concentration in maize and not negatively related.

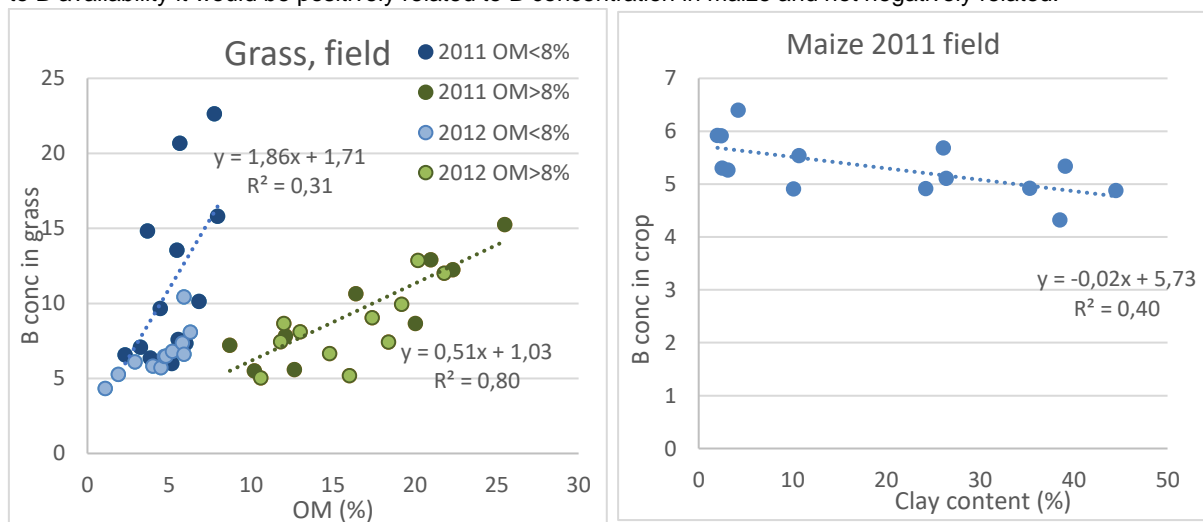


Figure 3-5 Relationship between B concentration in grass and soil organic matter content (left figure) and the relationship between maize and clay content (right figure).



### 3.5 Response to fertilizer applications

B fertilization at a rate of 0.99-1.01 kg B/ha did not increase the amount of B in the potato samples. In the potato trial fertilization with B or with B and S did not affect B-uptake compared to no fertilization. This was contrary to expectations because the trial was performed on a sandy soil with low pH (4.8) and relatively low OM content (3.8%).

For grass fertilization with B (1 kg B/ha) increased B uptake (data field experiment 2012). This change in B concentration in the grass with fertilization compared to without fertilization (ratio) was related to the B content in the soil as approximated using B-CaCl<sub>2</sub> ( $r^2=0.50$ , Figure 3-6). The relationship is even slightly better ( $r^2=0.58$ ) when using the predicted B-CaCl<sub>2</sub> based on equation 1 (OM, Clay, pH). These three general soil characteristics that determine the adsorption potential for B can thus also be used to get an indication whether grass is expected to respond to B fertilization. With increasing adsorption potential for B in the soil or with increasing B-CaCl<sub>2</sub> the response in B concentration in the grass to B fertilization is lower. At a B-CaCl<sub>2</sub> of approximately 550  $\mu\text{g kg}^{-1}$  there is on average no response to B fertilization. This would on average correspond with B-HW of 1.4  $\text{mg kg}^{-1}$  on sandy soils and 2  $\text{mg kg}^{-1}$  on clay soils (Figure 3-1). Above this level the response is slightly negative and below this level the response is increasingly positive. On average the maximum increase in B concentration in the grass is approximately a factor 1.6.

In the grass experiment B was also measured in an ammonium oxalate extraction. B-CaCl<sub>2</sub> is closely related to B-ox ( $r^2=0.80$ , data not shown). This parameter for B availability in soils also shows a close relationship with the grass response to fertilization (Figure 3-6).

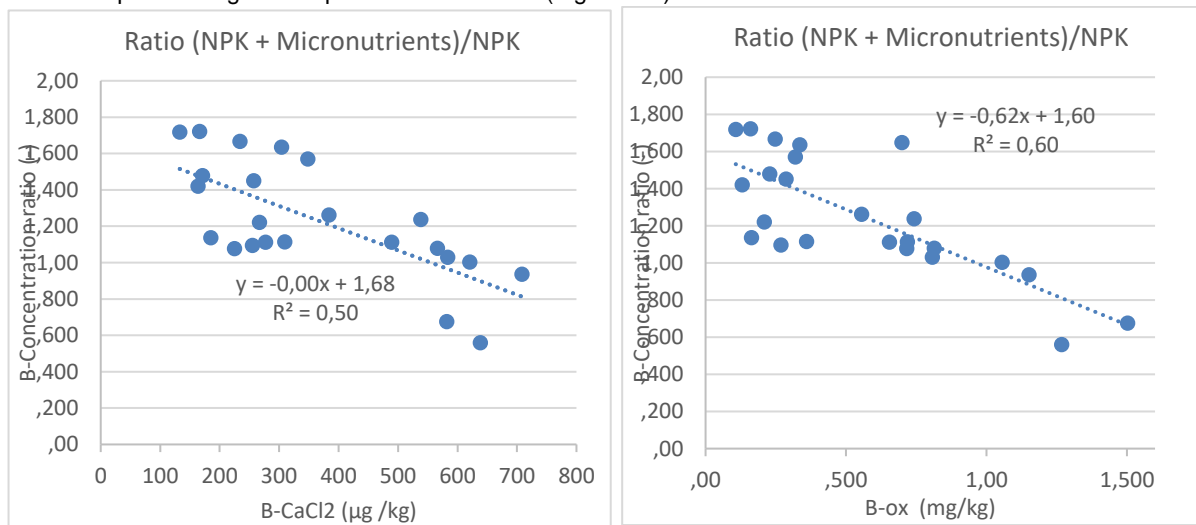


Figure 3-6 Relationship between the ratio B concentration in grass of with and without B fertilization and B-CaCl<sub>2</sub> (left figure) and B in an acid ammonium oxalate extract (B-ox, right figure).

### 3.6 Improving fertilization recommendations

#### 3.6.1 Current fertilizer recommendations

The Dutch fertilizer recommendation is based on the hot water extraction and consists of a recommendation for both specific arable crops and the fodder crop maize. The specific arable crops are sugarbeet, alfalfa, turnip, celery, maize and carrots.

The recommendation distinguishes four classes (Table 3-1). Above a B-content of 0.35  $\text{mg kg}^{-1}$  the

classification is high and the advice is that no fertilizer is needed. A B content lower than  $0.2 \text{ mg kg}^{-1}$  is classified as being very low. The current fertilizer recommendation does not distinguish between soil types. This is despite the fact that deficiencies are found primarily to occur on sand. The recommended fertilizer dose is for 1 year because B is prone to leaching from the topsoil and toxicity (Van Dijk and Van Geel, 2010).

When using organic manure this can be subtracted from the recommended dose. Organic manure contains approximately 4 g B per ton. When applying 40 tons this coincides with  $160 \text{ g B ha}^{-1}$ . This application rate is equal to the average removal rate of maize (Table 3-2). When the soil B level is (moderately) high sufficient B is applied. For soils with (very) low B level additional b should be applied in the form of a liquid fertilizer (for maize applied foliar or in the row) or as a blend containing B (Ros and Den Boer 2011). For arable crops the higher dose of the solid fertilizer results from the poor spatial distribution during application.

The B content of organic manure is based on relatively old analyses (Van Dijk and Zwanepol, 1993 and Smilde and Van Luit, 1985) and the availability of the B applied with the manure has not properly been tested (Ros and Van Dijk, 2011). It is recommended to further investigate both these aspects.

Table 3-1 Boron content of the soil with associated categories and corresponding fertilizer recommendations based on hot water extraction (from Adviesbasis bemesting arable crops and grassland and fodder crops 2012).

Classification	B-content soil mg/kg	Fertilizer advice (kg B/ha)	
		Liquid fertilizer, Foliar	Solid fertilizer
Very low	<0.2	0.4*	1.5
Low	0.2-0.29	0.3*	1
Mod. high	0.3-0.35	0.2*	0.5
High	>0.35	0*	0

\* the only recommendation for (silage) maize

The Dutch classification and recommendation is based on numerous field trials in the 50s and 60s and practical experience. Van Driel and Smilde (1990) concluded that in The Netherlands B deficiency mainly occurs in B sensitive crops such as maize and sugar beets on Pleistocene sandy soils with a water-soluble B content of less than  $0.3 \text{ mg kg}^{-1}$ .

The Dutch classification is low compared to critical B concentrations used internationally for some field crops (alfalfa, sugar beet, cotton, maize, peanut) which ranged between  $0.1 - 2 \text{ mg kg}^{-1}$  (mean  $0.8 \text{ mg kg}^{-1}$ , review by Fageria et al., 2002). According to old Swedish fertilizer recommendations the B content in soil should be higher than  $1 \text{ mg kg}^{-1}$  (Stoltz en Wallenhammar 2009).

### 3.6.2 Adapting and including additional crops and conditions

For some crops, e.g. maize, differences between B sufficiency and toxicity are small. In addition, the differences in B requirement between crops is large. The current recommendation only differentiates between the fodder crops maize, alfalfa and fodder beets and the specific arable crops sugar beet, turnip, celery, and carrots. In Table 3-2 a comparison is made between the level at which B is deficient, sufficient and toxic for sugar beet, potato, and maize (Reuter & Robinson, 1986 in Wattel-Koekoek and Bussink, 2003). From this the B removal from the soil by the crop is calculated in g B per hectare and expressed in mg B per kg soil (based on a root depth of 30 cm and a soil bulk density of  $1.15 \text{ kg/dm}^3$ ).

Table 3-2 Levels at which B is deficient, sufficient and toxic for sugar beet, potato, and maize (Reuter & Robinson, 1986 in Wattel-Koekoek and Bussink, 2003). From this B removal by the crop is calculated in g B per hectare (based on a root depth of 30 cm) and expressed in mg B per kg soil (based on a soil bulk density of 1.15 kg dm<sup>-3</sup>).

	<u>B- content crop</u>			<u>Yield</u> ton	<u>B removal by crop</u> (g B ha <sup>-1</sup> )			<u>Expressed in mg B kg<sup>-1</sup> soil (based on 30 cm soi)</u>		
	Deficient	Sufficient t	Toxic		DM ha <sup>-1</sup>	Deficient	Sufficient	Toxic	Deficient	Sufficient
Sugar beet	<40	40 – 200	>500?	18	<720	720 – 3600	9000	<0,21	0.2 – 1.0	2.61
Potato	<15	20 – 60	>180	15	<225	300 – 900	2700	<0,07	0.09 – 0.26	0.78
maize	<5	5 – 25	>30	15	<75	75 - 375	450	<0.02	0.02 – 0.11	0.13

#### Maize

For the fodder crop maize only the low recommended fertilizer dose is valid (Table 3-2). This recommendation is in use since 2012 after an update by the CBGV based on a report of (Ros & Den Boer, 2011). Banded B application together with N is the common way of application. It matches roughly the B removal rate of maize (0.075 – 0.38 kg ha<sup>-1</sup>).

Differences between B sufficiency and toxicity are narrow (Marschner, 1995). This is especially clear for maize (Table 3-2). When B uptake exceeds 0.45 kg ha<sup>-1</sup> this can cause B cause toxicity. When expressed in B removal from the soil this equals only 0.13 mg kg<sup>-1</sup> (based on a root zone of 30 cm).

When direct B availability / uptake is higher than 0.13 mg kg<sup>-1</sup> maize may already have problems with B toxicity. In addition, calculation show that with a common application rate of 40 m<sup>3</sup> dairy manure in combination with 150 kg of the commonly used 'Maize-fertilizer' the application of B is 0.45 kg ha<sup>-1</sup> (Ros and Den Boer, 2011). Therewith the B application is equal to the level above which toxicity may occur.

This was also the case for other common fertilizer practices, indicating that generally sufficient B seems to be applied to maize to prevent deficiency and that seldom B fertilizer is required. However, in most cases the amount in manure is neglected. B fertilization is mainly restricted to sandy soils.

The results of B supply with common fertilizer application levels imply that it might be necessary to execute soil testing to make sure that there is no problem with toxicity. High B levels are generally found to occur on clay soils and sandy soils with OM content greater than 10% (Figure 3-1, Table 3-2). Apart from further research in the effectiveness of the B in organic manure it is recommended to measure the B content of these soils to determine the level of B toxicity for maize.

#### Potato

B requirement and removal by Potato is intermediate between that of sugar beet and maize. Despite the relatively large amounts of B removed by the crop, there is currently no B fertilizer recommendation for potato.

In a potato field experiment B content was not found to be affected by B application (results potato trial 2011, 2012, C. Torres), most probably due to drought overruling the effect of B fertilizer application (Mulder et al., 2013). In 2012 weather was generally cool with sufficient rain. However, again 1 (Festien) of the 2 investigated potato species showed B deficiency symptoms, regardless of whether or not B was applied to the soil. The other potato species (Dartiest) only showed deficiency symptoms when no B was applied to the soil. Potato is thus sensitive to low B availability induced by drought. Deficiency can also occur in wet weather conditions when B is leached out of the rooting zone of the soil. The degree of deficiency symptoms and the response to fertilizer application seems potato species specific. The deficiency symptoms reported by Mulder et al. (2013) and the relatively high B requirements (Table 3-2) suggest that it is deserves recommending to develop a fertilizer recommendation for potatoes.

### Clover and legumes

B appears to be important for the symbiosis between rhizobia and legumes (Redondo-Nieto et al, 2007). In addition, legumes have a relatively high B requirement. Fertilization with boron (B) was found to promote the number of root nodules of white clover (Marschall et al, 1991). Sufficient B supply also influences clover yield (Marschall et al., 1991; Sherrell, 1983). B in clover is important for seed production (Stoltz and Wallenhammar 2014). Stoltz and Wallenhammar (2014) also found that in organic red clover B application had no effect on shoot B concentration and in only one experiment increased seed B concentration at soil B-content  $<1 \text{ mg kg}^{-1}$ . However, B application did affect seed yield and quality. It is thus possible that B application does not result in higher B concentrations in the shoot but does result in better seed yield and quality.

Important side remark is that research on only clover cannot be used for grass clover because the interaction between grass and clover is complex in terms of the absorption of nutrients. When considering these different studies and the increasing interest in the cultivation of legumes for feed in The Netherlands, it appears worthwhile to develop a B fertilizer recommendation for clover and other legumes.

### Sugar beet

For sugar beet under sufficient conditions B removal is 0.7 – 3.6 kg B per hectare. For soils that would just qualify as having a B content classified as being high ( $>0.35 \text{ mg kg}^{-1}$  which corresponds with  $\sim 1.4 \text{ kg ha}^{-1}$ ) the B content of the soil may not be sufficient and will also deteriorate quite rapidly. This calculation of  $1.4 \text{ kg ha}^{-1}$  is based on the assumption that the roots take up nutrients from the top 30cm. Sugar beet will however take up nutrients from a deeper soil layer. It is still recommended to increase the B-level of the four classes for sugar beets as a result more soils will be classified as low in B and to increase the fertilizer dose accordingly.

### Drought

Despite that a soil may be classified as having 'sufficient' or 'high' B-content, deficiency may occur in the field during periods of drought. For example, turnip was B deficient when grown in soil with hot water-extractable B  $<0.3 \text{ mg kg}^{-1}$ , but also became B deficient when grown in the field at B-levels of 0.5 - 0.6  $\text{mg kg}^{-1}$  during a dry summer (In Fageria et al., 2002; Batey, 1971). A recent study (Mulder et al., 2013) also shows that potatoes became B deficient as a result of drought (field study 2011). Deficiency was observed in both unfertilized and fertilized ( $1 \text{ kg B ha}^{-1}$ ) fields. Due to prolonged drought at the beginning of the growing season (2011), the absorption of B by the roots was greatly limited. After irrigating, no further deficiency symptoms were observed. Irrigation seems more effective to overcome B deficiency caused by drought than the application of B fertilizer.

### 3.6.3 Conclusion on crops and other factors

The current fertilizer recommendation need to differentiate more between different crops. For sugar beet (and fodder beet) B requirements is approximately 10 times higher than for maize. Without adequate B fertilization B may become easily depleted after sugar beet in the rotation. This is especially true for sandy soils with poor buffer capacity for B. In addition, the fertilizer recommendations should be extended to include levels above which B toxicity may give rise to problems. Potato should be added to the list of crops for which a B soil test is advisable. It is also advised to develop a B-recommendation for grass-clover.

When B deficiency is signaled in the field drought must also be considered as the cause, in which case irrigation is more effective than fertilization.

### 3.7 Conclusions for the B fertilization recommendation

The following conclusions and recommendations are made to improve the B fertilization recommendation:

- The amount of B extracted with different soil extraction methods is related to a combination of general soil properties that together determine the potential adsorption capacity; organic matter content, clay content and pH. This provides a good perspective to predict B availability using sensor techniques based on NIR and MIR;
- For fertilizer recommendations, soil analysis does not seem relevant for clay soils with organic matter content greater than 5% as B content is (more than) sufficient for these soils to grow maize. On the other hand, B levels are generally low in sandy soils with low organic matter content due to the low capacity to bind B, the sensitivity to drought and leaching.
- For crops that are sensitive for B toxicity; e.g. maize, soil analyses may be useful on clayey soils and sandy soils with an organic matter content higher than 5%.
- It should be taken into account that liming decreases B availability in the soil. To get the most reliable results B soil analysis should ideally be performed in spring before the growing season but after liming.
- A greenhouse experiment with wheat shows a clear relationship between B concentration in leaf tissue and the combination of pH and B-CaCl<sub>2</sub>. In field experiments this relationship could not be further substantiated. The importance of texture, organic matter and pH on B availability indicates that is worthwhile to investigate if these parameters need incorporating in the B recommendations.
- The response of grass to fertilization with B could be predicted based on the B level of the soil (B-CaCl<sub>2</sub> or B-ox) or on the potential adsorption capacity for B in soil.
- Current fertilizer recommendations can be improved by distinguishing between crops in terms of B requirement. It is recommended to develop a fertilizer recommendations for legumes due to relatively high B requirements and the role of B in the formation of root nodules.
- In theory the application of organic manure results in a B application that together with an adequate B level of the soil is generally equal to removal rates by the crop. The increasingly strict application rates of organic manure and the assumption that B in organic manure is readily available for the crop should be investigated.
- It is recommended to incorporate weather conditions in the recommendations for coarse textured drought and leaching sensitive soils. A weather module may be developed that includes both actual weather data and a simple root development model. Recommendations in situations of drought must focus on irrigation rather than on fertilization. B deficiency as a result of extreme wet and dry periods due to climate change deserves further attention
- Preventing the deterioration of soil OM content and maintaining soil pH at an adequate and not too high level is important for maintaining B availability.

## 4 Copper

### 4.1 *Copper in crops and animals*

For crops Copper (Cu) is essential because Cu is part of enzymes in the chloroplasts which are involved in the photosynthesis. Further, Cu is a component of several enzymes that are involved in the oxidation of organic compounds. Cu deficiency is especially visible on the generative plant parts and results in poor fruit set and waste of young fruits. The critical levels of Cu vary from 1 to 5 mg kg<sup>-1</sup> Cu dry matter depending on the crop type. Cu has a very low mobility in the plant since 98% of it is organically bound. Remobilization is therefore limited and symptoms occur on the youngest leaves. Cereals are sensitive to Cu deficiency, especially wheat and oats but also spinach.

In the animal Cu is a component of several enzymes that play an important role in the metabolism. Cu deficiency may include diarrhea, deterioration of health, growth and / or milk production, rough and dull hair, block feet (ref). The Cu requirement is of the same order of magnitude for animals as for plants. The Cu content needed for a crop growth and quality is relatively low; 5-30 mg kg<sup>-1</sup>. For cows the requirement of the feed is between 12 mg kg<sup>-1</sup> for lactating cows and 25 mg kg<sup>-1</sup> for dry cows (CBV, 2012). In crops Cu deficiency can be latently present for a longtime before symptoms can be visually recognized on the plants. Symptoms of Cu deficiency may thus be apparent in grazing animals before the crop exhibits symptoms of deficiency. It is therefore important that there is sufficient Cu present in crops used for roughage (grass and maize) for a good animal performance.

On the one hand, Cu deficiency in animals can occur due to a too low Cu content in the grass. On the other hand, Cu-deficiency may be caused in the animal when the Cu from the consumed grass is insufficiently absorbed. This can occur by the formation of poorly soluble salts in the rumen between Mo, S, and Cu. Especially S in the rumen contributes to poor absorption of Cu. The utilization of Cu by the animal is thus not only determined by Cu, but also by the availability of S- and Mo (CBGV, 2012). In a study in the province of Drenthe Cu deficiency was only found to occur in young animals older than 1 year in the summer because they have a high demand but do not receive Cu in the form of concentrates during the grazing period. In all the other life stages of the cows excessive amounts of Cu were given due to the composition of the concentrates (Den Boer et al., 2011).

### 4.2 *Copper in soil*

In the soil solution most Cu (99%, Terrones and Supriatin, 2015) is associated with dissolved organic molecules (DOC). Only relatively little is in the Cu<sup>2+</sup>-form that can readily be taken up by the plant roots. The strong reaction of Cu with DOC has a twofold effect; on the one hand, more Cu is in the soil solution making it more mobile as the organic ligands act as carriers of Cu to plant roots (Lindsay, 1979). On the other hand, the availability of organically complexed Cu is lower than of Cu<sup>2+</sup> for uptake by the root because the Cu must be separated from the ligand before it is taken up. The availability of the Cu in the Cu-complex in solution depends on the amount and stability of the complex. The amount of Cu in the soil solution is mainly determined by the Cu which is bound to soil constituents (Barber, 1995).

Organic matter (OM) is the primary constituent for Cu adsorption in soil and readily complexes Cu. Modelling the reactive Cu in soil (extracted with 0.43M HNO<sub>3</sub>) shows that up to 95 percent is related to OM in the soil, around 4% is adsorbed to Fe-(hydr)-oxides and little Cu is adsorbed to clay (Terrones and Supriatin 2015). Apart from adsorbed to OM, Cu is also found in silt and clay fractions of soil and usually present in carbonate fractions in alkaline soils and in Fe oxide fractions in acid soils (Shuman, 1991 in Fageria et al., 2002). Available concentrations of Cu have been found to increase with increased

amounts of clay (Lee et al., 1997 in Fageria et al., 2002).

Solubility of Cu is very soil pH dependent and decreases 100-fold for each unit increase in pH. Plant uptake also decreases in accordance (Fageria et al., 2002). As the pH increases, the number of sorption sites on the organic matter increases thus increasing the Cu adsorption (Geering and Hodgson, 1969 in Fageria et al., 2002). Adsorption to clay becomes important when the pH of the soil is above 6 and adsorption to metal (hydr-)oxides between pH 4 and 7 (Wattel-Koekoek and Bussink, 2003).

Other elements affect Cu availability. For instance, competition takes place between Ca cations and the Cu adsorption of organic matter. In principle, Cu adsorbs much more strongly to the organic matter, but the concentration of Ca-cations is much higher so that competition becomes relevant. At the root surface competition for Cu uptake is of little relevance except for the competition with  $Zn^{2+}$  (Mengel and Kirkby 1987).

Opposing results have been found on the effect of moisture content on the uptake of Cu. Nevertheless, the general assumption is that drought reduces the Cu availability.

Cu deficiency can occur for several reasons. The first is that the soil does not contain enough Cu, e.g. when the organic matter content of a soil is very low. The second reason is that the soil does contain enough Cu but that the availability is too low, e.g. due to the strong adsorption in organic matter rich soils. Cu deficiency can also be caused by management measures like liming and P fertilization or a high S content in the ration of the cow.

#### 4.3 *Measuring Cu in soil*

To assess the distribution and availability of Cu in soil, Supriatin investigated extractability and speciation of Cu using 83 widely varying Dutch soils (Terrones and Supriatin 2015). In general, the total Cu content is relatively low in the sandy soils and high in the clay-peat soils. The total Cu content extracted with a strong extraction method, Aqua Regia, ranged from 5 to 76 mg kg<sup>-1</sup>. Efficiency of Cu extraction follows the order of Aqua Regia (taken as 100%) > ammonium oxalate ( $59 \pm 12\%$ )  $\approx$  0.43 M HNO<sub>3</sub> ( $52 \pm 12\%$ ) > 0.01 M CaCl<sub>2</sub> ( $0.4 \pm 0.1\%$ ). Amounts of Cu extracted in different extractions are correlated. Cu-HNO<sub>3</sub> and Cu-am.ox are best related ( $r^2 > 0.9$ ). About 50% of total Cu extracted with Aqua Regia cannot be extracted with 0.43 M HNO<sub>3</sub> or ammonium oxalate and is considered as non-reactive. Of the other 50%, i.e. reactive Cu as extracted in 0.43 M HNO<sub>3</sub>.

Organic matter is the most important soil component associated with both the reactive Cu (Cu-HNO<sub>3</sub> and Cu-ox) and with the directly available Cu (Cu-CaCl<sub>2</sub>). About 96% of reactive Cu in the soil is adsorbed to soil OM and 4% is adsorbed to Fe-(hydr)oxide based on soil chemical modelling Cu extracted with 0.43M HNO<sub>3</sub> (Terrones and Supriatin, 2015). Little Cu is adsorbed to clay. Only 0.4% Cu is extracted as Cu-CaCl<sub>2</sub>, of which 99% is bound to DOC, and only 1% present as free Cu<sup>2+</sup> ion. The results indicate that 0.01 M CaCl<sub>2</sub> extractable Cu can be used to estimate mobile Cu concentration in soils, whereas 0.43 M HNO<sub>3</sub> extraction can be used to estimate the quantity of Cu that can buffer Cu in the soil solution. The amount of Cu extracted with 0.01M CaCl<sub>2</sub> increases by a factor 2 when drying the soil sampling prior to extraction. This increase is related to an increase in dissolved organic carbon (DOC). Also when a soil sample is stored at room temperature Cu-CaCl<sub>2</sub> can increase over time. This increase is also related to an increase in DOC over time.

The Dutch fertilizer recommendations are based on measuring Cu after extracting the soil with 0.43N HNO<sub>3</sub>. This extract approximates the total amount of reactive Cu. Since 2004 this extract has been changed to the weak multi-nutrient extraction with 0.01M CaCl<sub>2</sub>. A statistical relationship was developed by Eurofins to relate CaCl<sub>2</sub> extraction results to HNO<sub>3</sub> extraction results. This relationship includes a correction for pH and is used for the CaCl<sub>2</sub> based fertilizer recommendation.

Based on 87 samples Supriatin (2015) found a good direct correlation between Cu-CaCl<sub>2</sub> and Cu-HNO<sub>3</sub>

with an explained variance of 79%. For predictive purposes and to extrapolate Cu-CaCl<sub>2</sub> to the fertilization recommendations based on Cu-HNO<sub>3</sub> an explained variance of 79% is not very helpful. All the more so because Cu-HNO<sub>3</sub> is poorly related to crop uptake. It is better to derive a direct relationship between Cu-CaCl<sub>2</sub> and crop response.

Increasing the pH of acidic soils can reduce the Cu-availability. It is therefore suggested to perform soil analysis after liming and not prior to liming.

Concluding:

- 0.43M HNO<sub>3</sub> and ammonium oxalate extract reactive Cu that is mainly associated with soil organic matter content. This method was used until 2004 to measure Cu in soil as a base for Dutch fertilizer recommendations;
- 0.01M CaCl<sub>2</sub> only extracts about 1% of this reactive Cu. this method is used since 2004 to measure Cu in soil as a base for Dutch fertilizer recommendations;
- Of the Cu in solution 99% is bound to dissolved organic matter and only 1% is in the form that can readily be taken up (Cu<sup>2+</sup>)
- A direct relationship between Cu-CaCl<sub>2</sub> and crop response is needed.

#### 4.4 *Cu in soil in relation to crop uptake*

A first indication that Cu-CaCl<sub>2</sub> might be a better direct indicator for Cu uptake by grass than the traditional Cu-HNO<sub>3</sub> method was shown by Bussink (unpublished) based on the dataset from the project "Koeien en Kansen". In addition, the Cu content of the crop was found to be strongly correlated with crude protein content, soil clay content and K –CaCl<sub>2</sub>. The explained variance using this combination of parameters was about 65%. Temminghoff et al. (1998) also found a better correlation of Cu content in grass with Cu-CaCl<sub>2</sub> than with Cu-HNO<sub>3</sub>.

Cu-HNO<sub>3</sub> is a poor predictor of Cu availability (explained variance 25%), based on the results of a pot experiment with grass on 72 different sandy and reclaimed peat soils (Van Luit & Henkens, 1967). No distinction could be made between the sandy and reclaimed peat soils. When combining the results from the pot experiment with data from various field trials in the North of The Netherlands, Van Luit and Henkens (1967) showed that the Cu-content in grass is associated with Cu-HNO<sub>3</sub>, N content of the grass (crude protein content) and pH-KCl of the soil (explained variance increased from 25% to 40%).

In the wheat pot experiment (2013, N=19) there is a relatively small variation in Cu content measured in the wheat shoots; between 3.4 and 6.9 mg Cu kg<sup>-1</sup>dw. Of the two investigated extraction methods the correlation of Cu concentration in the wheat leaves was better correlated to Cu-CaCl<sub>2</sub> ( $r^2=0.42$ ) than to Cu-ox ( $r^2=0.32$ ). It seems that at Cu-CaCl<sub>2</sub> levels in the soil higher than 50 µg kg<sup>-1</sup> the Cu content of the crop reaches a maximum (Figure 4-1). This would imply that above 50 µg/kg the Cu content in the soil is more than sufficient to meet wheat crop needs (critical level young wheat leaves is 1.5 mg kg<sup>-1</sup>). When incorporating a measure for amorphous Fe-(hydr-)oxides as extracted with ammonium oxalate (Fe-ox) the explained variance increases to 62%. From a mechanistic point of view it is not clear why amorphous Fe-(hydr-)oxides would have such a dominant effect on Cu uptake by wheat. It is therefore suggested to only focus on the direct relationship with Cu-CaCl<sub>2</sub>. Despite an expected effect and a large variation in both soil pH (4.5 to 7.5) and in OM content (1.6 – 14.5) they do not seem to affect Cu content in the wheat shoots.



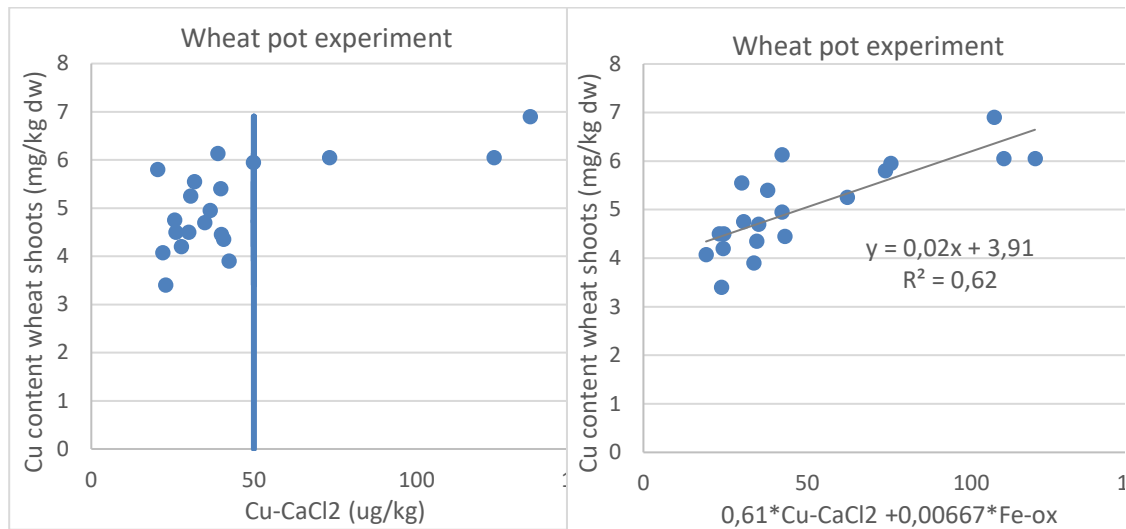
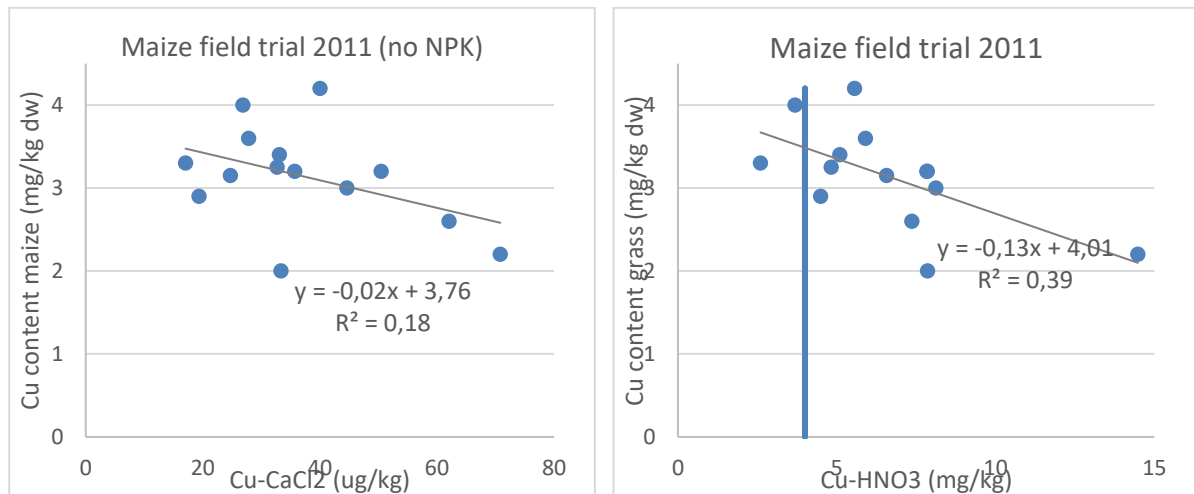


Figure 4-1 Relationship between Cu content of wheat shoots and Cu-CaCl<sub>2</sub> (left) and the combination of Cu-CaCl<sub>2</sub> and amorphous Fe (hydr-)oxides (right).

In the maize field trial the Cu content in the maize varies between 2 and 4.2 mg Cu kg<sup>-1</sup>dw. This is within the range of 1-5 mg Cu kg<sup>-1</sup> DM where Cu is deemed to be deficient for crops (Kabata-Pendias and Pendias, 2001). Despite the low levels of Cu in the maize the Cu-HNO<sub>3</sub> content of 12 of the 14 soils is higher than the 4 mg kg<sup>-1</sup> below which the soil Cu content is defined as being low according to Dutch fertilizer recommendations. There seems to be a poor negative trend between Cu content and the soil parameters Cu-CaCl<sub>2</sub> and Cu-HNO<sub>3</sub> when considering all soils. Astonishingly the trend is negative and the Cu content seems to decrease with increasing Cu-CaCl<sub>2</sub> content in the soil. The negative trend is also visible with Cu-HNO<sub>3</sub> and Cu-ox. This is against expectations and contrary to results found in literature.



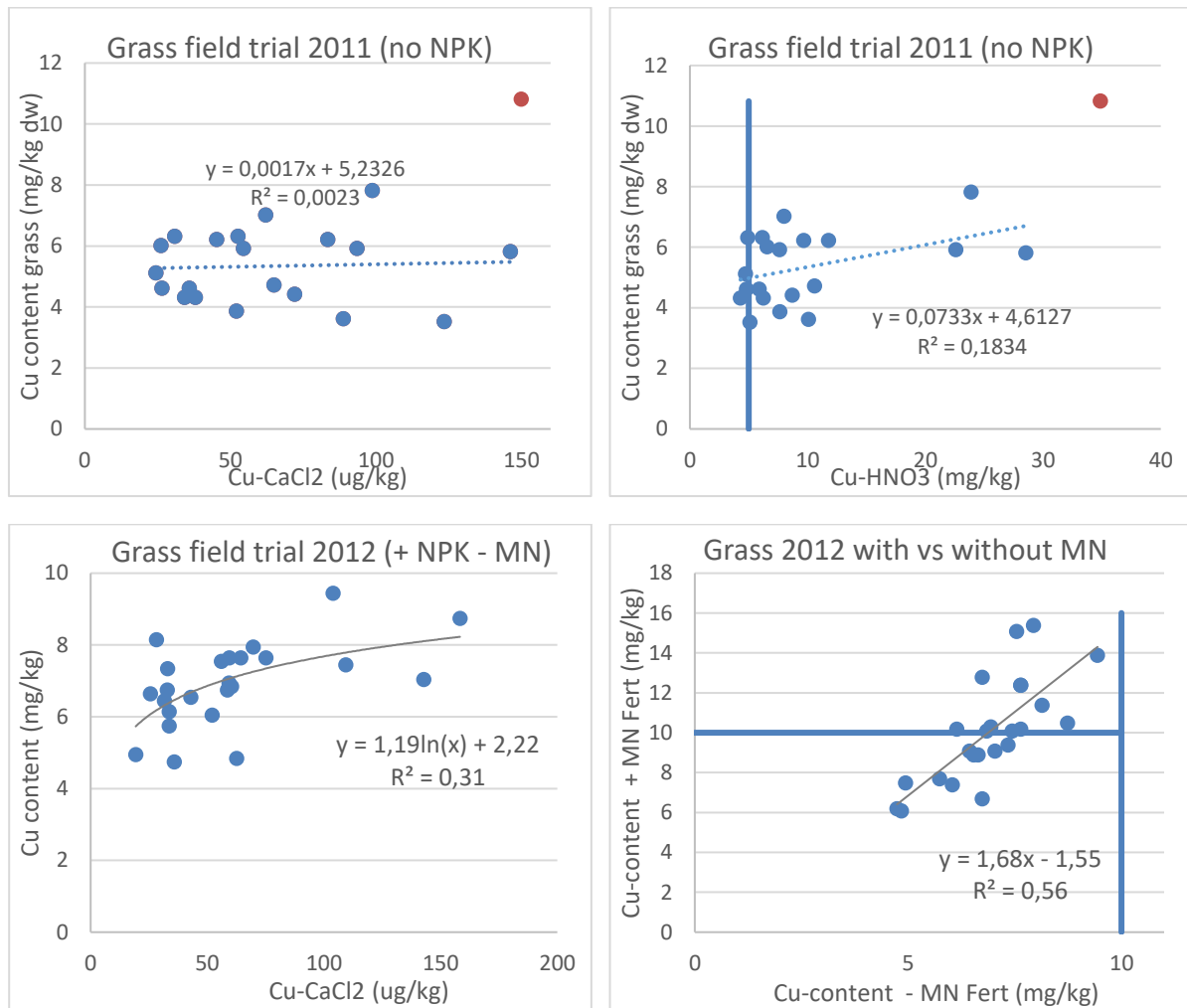


Figure 4-2 Relationship between Cu content of grass and Cu-CaCl<sub>2</sub> (left) and Cu-HNO<sub>3</sub> (right) in the maize experiment (top figures), the grass field trial 2011 (central figures) and the grass field trial in 2012 (bottom figures). The field with a Cu-content in grass in the 2011 trial of 11 mg/kg DM is considered an outlier (red dot).

In 2011 field trial the Cu content of the grass is lower than the 12 mg kg<sup>-1</sup> DM required for feed grass for cattle despite the wide range in Cu levels in the soils. That year (2011) the grass was not fertilized, with the exception of three soils where N was applied. In the current recommendations a minimum level of 5 mg kg<sup>-1</sup> Cu-HNO<sub>3</sub> is required in the soil for an adequate availability for grass. Despite the fact that only three soils are just below this level the Cu content of the grass was below the required amount in grass for cattle. This further substantiates the poor result that was found in different studies between Cu content in the crop and Cu-HNO<sub>3</sub> (o.a. Van Luit and Henkens, 1967).

The grass field trial in 2012 (+ NPK, +/- micronutrients) shows a poor relationship between Cu content in grass and Cu-CaCl<sub>2</sub> ( $r^2=0.31$ ,  $n=24$ , Figure 4-2). The relationship with Cu-ox is even a little poorer ( $r^2=0.27$ ). In 2012 mineral NPK was applied and the Cu uptake ranged between 4.7 and 9.4 mg Cu kg<sup>-1</sup> DM and was thus in all cases also below the required level of 10 mg kg<sup>-1</sup>. Again the Cu level of the soil could not prevent a too low Cu content in the grass to provide cattle sufficient Cu.

For the grass grown on sandy and clay soils the Cu uptake was higher in 2012 with NPK application compared to 2011 without NPK (16 fields were used in both 2011 and 2012). However, on the clay-peat and peat soils the Cu content was the same in 2012 with NPK compared to 2011 without NPK fertilization (Terrones and Supriatin, 2015). A tentative conclusion may be that NPK fertilization increases Cu uptake in sandy and clay soils but not on clay-peat and peat soils. This hypothesis is based on very little number of soil samples ( $n=5$ ) and should be further investigated.

In both 2011 (-NPK) and in 2012 (+NPK) all fields except one showed a too low Cu content in the grass for feed requirements.

Conclusions concerning Cu uptake in the different pot- and field experiments:

- Cu uptake is generally (too) low, especially for grass and maize compared to the current required levels set-out by cattle animal nutrition; The low Cu uptake in the maize and grass field trials could not be related to soil test results
- The relationship between Cu uptake and the different soil Cu extraction methods show contradicting results; varying from a negative trend (maize field) to no relationship (grass field 2011) to a positive trend (grass field 2012) to a positive relationship (wheat pot)
- The soil factors organic matter and pH were expected to play a role in Cu availability but no evidence was found in the pot and field trials;
- Previous studies found N content of the grass to be of influence on Cu-content;
- The wheat pot experiment suggests a maximum Cu uptake at Cu-CaCl<sub>2</sub> higher than 50 µg kg<sup>-1</sup>, this could not be reproduce in the field trials;

#### 4.5 *Response to fertilizer applications*

In the 2012 grass field trial the application of 3.5 kg CuO ha<sup>-1</sup> as copper sulphate results in a structural increase in Cu content on all fields; average 1.5 times higher Cu content (range of 6 to 15 mg Cu kg<sup>-1</sup>, Figure 4-2). This dose corresponds to the recommended dose for grass when the Cu-status of the soil is low (Table 4-1). For only 13 of the 24 fields this resulted in sufficient (>12 mg kg<sup>-1</sup>) Cu in grass. A hypothesis may be that the level of 12 mg Cu kg<sup>-1</sup> DM grass is too high and that the target level should be decreased to for instance 6 mg kg<sup>-1</sup>dw. This needs further investigation.

The response in Cu content of the crop to Cu fertilization was similar in all fields. The response could not be related to any of the soil parameters.

#### 4.6 *Improving fertilization recommendations*

##### 4.6.1 *Current fertilizer recommendations*

Problems with Cu deficiency have most commonly been reported for cereals on humic podzolic soils, reclaimed peat soils (dalgronden), peat soils and other soils with high organic matter content with less than 3-4 mg kg<sup>-1</sup> Cu-HNO<sub>3</sub> (based on numerous field trials in the 50s and 60s, and practical experience Van Driel and Smilde, 1990). In grass fields no response was found to occur above a Cu-HNO<sub>3</sub> of 5 mg kg<sup>-1</sup> (Van Luit and Henkens, 1967)

The Dutch Fertilization recommendation for arable crops distinguishes four classes of Cu in the soil based on Cu-HNO<sub>3</sub> but it does not distinguish between different soil types (Table 4-1). The recommendation for arable land is based on analyzing data from field trials with oats and wheat from 1954 and 1955. In addition, a pot trial was performed with 70 different soils with wheat in 1957. Both the field trial and pot experiment show that no deficiency occurs in wheat at Cu-HNO<sub>3</sub> above 4 mg kg<sup>-1</sup>. Oats were found to be a little less sensitive and Cu-HNO<sub>3</sub> of 3 mg kg<sup>-1</sup> was found to be sufficient (De Vries and Dechering 1960).

The recommendation for grassland is based on the study by Van Luit and Henkens (1967) based on a pot experiment with 72 soils and several field trials in the North of The Netherlands. After correction for the influence of the N content and the influence of the pH, they concluded that Cu content in grass reached a maximum at Cu-HNO<sub>3</sub> higher than 5 mg kg<sup>-1</sup>. The current fertilizer recommendation that no

fertilizer is recommended at a Cu-HNO level above 5 mg Cu kg<sup>-1</sup> soil is based on this conclusion. The current recommendation does not consider N-content of the grass or pH-KCl.

The fertilizer recommendations in arable farming were thus developed for oats and wheat. Other crops, including rye and barley, are less sensitive. For the fodder crops maize, cereals for GPS, fodder beets and alfalfa the same recommendations are used as for the arable crops. For grassland the minimum Cu content of the soil is 5 mg kg<sup>-1</sup>. The Cu content of grass on peat and poorly drained sand soils is sometimes insufficient to meet crop demand. Generally it is too low to meet animal requirement. In the fertilizer trials of Van Luit en Henkens (1969) and the 2012 grass field trials the maximum Cu content was respectively 13 and 15.4 mg kg<sup>-1</sup> DM. With Cu fertilization an increase of at maximum 16 mg kg<sup>-1</sup> DM seems possible. Different strategies have been identified (concentrates, lick blocs, bolus etc.) to prevent Cu deficiencies (Bussink et al., 2007).

Special attention has to be paid to young stock and dry cows. They are at risk for Cu deficiency during the summer because these groups then receive little or no concentrates (Haas et al., 2013). A good Cu soil status in combination with Cu fertilization is than a possibility. In all other stages of the cow's life it receives a surplus of Cu due to the application of concentrates.

Table 4-1 Classification of the copper status of soils based on Cu-HNO<sub>3</sub> and the advised fertilizer dose for the arable crops oats and wheat (1968, De Haan and Van Geel 2013) and the fodder crops maize, cereals for GPS, fodder beets and alfalfa (Commissie Bemesting Grasland en Voedergewassen, 2012). The recommended Cu fertilization is sufficient for a period of 4 years.

Classification	<u>Arable land</u>		<u>Grassland</u>	
	Cu-content soil, mg kg <sup>-1</sup>	Advised dose, kg Cu ha <sup>-1</sup>	Cu-content soil, mg kg <sup>-1</sup>	Advised dose, kg Cu ha <sup>-1</sup>
Very low	< 3.0	6	< 2.0	6
Low	3.0 – 3.9	2.5	2.0 – 4.9	3.5
Neutral	4.0 – 9.9	0	5.0 – 9.7	0
High	>10	0	≥ 9.8	0

Due to a possible decrease in Cu availability in the cow by the formation of insoluble salts with S (and Mo) it is important to fertilize grassland with S in accordance with the recommendation and not apply more.

The availability of Cu is kept at an appropriate level by applying organic and mineral fertilizers and, if possible, by maintaining a slightly acidic soil pH. When the crop shows symptoms of deficiency foliar application in the form of Cu-sulphate or a Cu-chelate may be applied. Comparing different foliar Cu fertiliser types for wheat shows that chelated-Cu is the most effective foliar form of Cu for increasing grain yield (Brennan, 1990). A balance approach showed that when pig manure is applied, no deficiency occurs in a normal arable crop rotation (Velthof and Van Erp 1998).

On a national scale there is a net surplus of Cu in Dutch agricultural soils. This surplus has seen a steep decrease between 1980 and 2005 since which it is approximately stable (

Table 4-2). This trend is mainly due to decreased Cu-levels in organic manure. The monitoring stopped in 2009.

Table 4-2 Loading of agricultural soils with Cu (data CBS).

	1980	1990	2000	2005	2007	2008	2009
Gross load of Cu in 1 000 kg	1360	970	780	515	485	480	465
Of which: Organic manure	1050	750	700	435	420	420	405
Mineral fertilizer	150	120	50	40	30	25	25
Wet and dry deposition	80	50	20	20	20	20	20
Other sources	80	50	10	20	15	15	15
Removal with the crop	140	130	100	95	95	100	100
Netto Cu surplus	1220	840	680	420	390	380	365

#### 4.6.2 Predicting Cu availability

A prerequisite of fertilizer recommendations is possibility to predict if Cu availability in the soil will be sufficient and if not how the soil will react to a fertilizer dose. For this a reliable indicator to determine soil and/ or crop Cu status is needed.

In Dutch routine soil laboratories (Eurofins) Cu is currently measured in a 0.01M CaCl<sub>2</sub> extract. The method on which the fertilizer recommendations are based, Cu-HNO<sub>3</sub>, has not been used on a commercial scale since 2004. Cu-CaCl<sub>2</sub> extracts only a small fraction (+/-1%) of the amount of reactive Cu that is extracted using the traditional method (Cu-HNO<sub>3</sub>). Cu-HNO<sub>3</sub> extracts reactive Cu that is mainly associated with soil organic matter content. Cu-CaCl<sub>2</sub> approximates the Cu concentration in solution of which 99% is bound to dissolved organic matter and only 1% is in the form that can readily be taken up (Cu<sup>2+</sup>). The correlation between Cu-CaCl<sub>2</sub> and Cu-HNO<sub>3</sub> is not good enough to accurately predict the one from the other and it is better to derive a direct relationship between Cu-CaCl<sub>2</sub> and crop response.

None of the field trials nor the wheat pot trial showed proof of a reliable relation between Cu content in the crop and the soil test results. The relationship between Cu content in the crop and the different soil Cu extraction methods show contradicting results with a positive relationship in the wheat pot trial. However these were relatively small datasets. It is in contrast with monitoring data 10-15 years ago in which there was clear relation between CaCl<sub>2</sub> extractable Cu and herbage content (Bussink, unpublished).

Furthermore, contradictory to theoretical knowledge and imposed expectations, the soil factors organic matter and pH did not play a role in affecting Cu availability in the pot and field trials. Further research is necessary to investigate the relationship between soil parameters and Cu plant availability and uptake.

#### 4.6.3 Fertilizer efficiency

Besides a clear relationship between a soil parameter and crop content it is also important that the Cu fertilizer is effective in order to prevent Cu loading of the soils. In the grass field trial (2012) only 0.6% of the applied Cu sulphate is recovered. This is very low but in accordance with older trials. If it is possible to increase Cu content by 10 mg kg<sup>-1</sup> DM, then this equals an amount of 150 g ha<sup>-1</sup> for herbage production levels of 15 tons DM per ha. The fertilizer recommendation is valid for 5 years. Over a period of 5 years at maximum an uptake of 750 g Cu ha<sup>-1</sup> might be expected. At application levels of 4 or 6.5 kg ha<sup>-1</sup> this means an efficiency of at maximum 13-18%. Mostly the efficiency will be (much) lower because of the increase in Cu-content is less than 10 mg kg<sup>-1</sup> DM. This means that the recovery is lower than 10%. The conclusion might be that using Cu salts for fertilization is a very ineffective way to improve Cu content of herbage. This opens the discussion if Cu fertilization with salts should still be recommendation

for Dutch agriculture. From literature it is known that Cu-chelates are far more effective, so that much lower amounts are needed. The strategy could be to shift to these products. Currently these products are not available on the Dutch market.

#### 4.7 *Conclusions for the Cu fertilization recommendation*

Results of the grass and maize trials help to answer the question whether current Cu fertilizer recommendation for grass will lead to a feed quality covering Cu needs of cattle livestock. In none of the trials performed in the PhD project grass contained the from animal nutritionists recommended 12 mg kg<sup>-1</sup> DM. However, for the fodder crops low Cu content in the feed is generally (more than) compensated for by the use of concentrates. Only young stock and dry cows are at risk for Cu deficiency during the summer because these groups then receive little or no concentrates (Den Boer et al., 2011). Data until 2009 shows a continuous decrease in Cu-content in organic manure. In combination with decreasing fertilizer dose and the low Cu-levels in the crops in the field trials (2011 – 2012) this implies that Cu deficiency may be a more generally occurring problem than expected. This needs further investigation. There was a clear positive response of grass in the field to fertilization with Cu-sulphate. The response in Cu-uptake to Cu-fertilization was approximately equal for all fields, indicating that Cu-levels does not influence the crop response to fertilization. The efficiency of the applied Cu sulphate fertilizer was however very low (0.6%) which is in agreement with other trails. This opens the discussion if Cu fertilization in a salt form is sustainable.

Currently, the following general advice concerning Cu management in fertilization can be added to current recommendations:

Aspects that need taking into account are:

- A high pH has a negative effect on Cu availability in the soil. It is thus important to keep soil pH at a good, and not too high level.
- When applying lime (CaCO<sub>3</sub>) it is recommended to perform soil analyses after liming;
- The recommendation levels for S fertilization of fodder crops should be followed as S has a negative influence on Cu uptake in the cow.

Cu requirement is high for sugar beets, spinach, wheat, oats, alfalfa, lettuce, onion. Therefore it should be considered to spend research effort on developing crop-specific Cu fertilizer recommendations for these crops. Currently, the recommendations derived from oat and wheat trials are applied.

Results of the grass field trial suggest an interaction with NPK fertilization. However, further research is needed to be able to conclude on this. From theoretical knowledge it seems logical since a large fraction of the Cu in the plant is organically bound. As these compounds are built-up of in particular N and P should the N and P status of the plant be expected to influence the mobility of Cu in the plant.

## 5 Manganese

### 5.1 Manganese in crops and animals

Manganese (Mn) is an essential element for crops as well as for animal health. In crops Mn plays an important role in the production of oxygen during the photosynthesis. Symptoms of Mn deficiency are yellow coloration between the veins, followed by chlorosis and withering of the leaves. Mn deficiency is first visible in the youngest leaves. In more severe cases, there is often a kink in the leaves. The Mn requirement varies greatly between different plant species and even between varieties. Mn sensitive crops are: potatoes, beets, beans, wheat and oats, but also spinach and lettuce require relatively large quantities of Mn. Danish research shows that barley varieties differ in susceptibility to Mn deficiency (Pedas et al, 2008). Dutch research confirms this difference between varieties for potatoes (Wattel-Koekkoek & Postma, 2003) and beet (Anonymous, 2003d).

For animals Mn plays an important role, e.g. in the formation of cartilage and bone and carbohydrate metabolism. For bovine animals, grass should contain at least 25 mg Mn kg<sup>-1</sup> DM, and a maximum of 500 mg kg<sup>-1</sup> DM. This is consistent with the majority of plants in which the minimum Mn-concentration lies between 10 and 25 mg kg<sup>-1</sup> DM, depending on the type of crop, growth stage and ecosystem (Mengel & Kirkby, 1987). Mn toxicity occurs in the majority of plants when the Mn concentration is greater than 500 mg kg<sup>-1</sup> DM.

### 5.2 Manganese in soil

Mn availability is to a large extent determined by the dissolution and precipitation of Mn-minerals such as Mn-oxides (pyrolusite and birnessite (MnO<sub>2</sub> (IV)) manganite (MnOOH (III)) and Mn-carbonates (MnCO<sub>3</sub> (II)). This is in contrast to other (micro) nutrients where the dissolution of the mineral form is not or hardly relevant to agricultural availability. The solubility of Mn-minerals and thus the availability of Mn for the plant is determined by the pH and redox potential of the soil. Where pH is a measure for the number of free acid particles (protons), redox is a measure of the number of free electrons. The presence of free electrons is closely associated with the moisture and oxygen content in the soil. When a soil is waterlogged the number of free electrons increases and the redox potential decreases. This is because the microbial processes to mineralize soil organic matter (OM) in the soil continue, oxygen quickly becomes depleted (anaerobic) and subsequently Mn minerals are dissolved. The soluble Mn<sup>2+</sup> that is released is available to the plant. When a soil becomes water logged due to restricted drainage or flooding, anaerobic conditions may thus lead to a large increase in Mn solubility and toxicity problems. At higher pH and redox conditions Mn is oxidized to Mn (IV) which precipitates in the form of oxides or carbonates. Mn-carbonates are highly insoluble and the direct availability of Mn is too low for Mn sensitive crops. Mn deficiency therefore often occurs on calcareous soils.

In solution the principal ionic Mn species is Mn<sup>2+</sup> which can be adsorbed to metal (hydr) oxides or OM, and clay particles. With increasing soil pH the adsorption of Mn<sup>2+</sup> strongly increases. At high pH Mn is thus not only precipitated as Mn-oxide or Mn-carbonate, but it is also strongly sorbed to the surface of various soil particles. The Mn<sup>2+</sup> concentration in the soil solution decreases a 100-fold for each unit increase in soil pH. As a result Mn in plants decreases extensively with increasing soil pH. Mn deficiency occurs mainly in soils with a high pH. On sandy soils no Mn deficiency is found to occur at pH-KCl lower than 5.4 while deficiency was found to occur on all fields with a pH higher than 6.2. At an intermediate pH-KCl both healthy and deficient fields occurred (De Vries and Dechering, 1960). At the other end of the spectrum, Mn solubility can be sufficiently high to induce toxicity problems in sensitive crop species in extremely acid soils.



Organic matter in the soil can also promote the occurrence of Mn deficiency because it strongly binds  $Mn^{2+}$ . Mn deficiency frequently occurs in limed and drained peat soils. In the Netherlands, Mn deficiency was first known as the “peat oats disease” (veenkoloniale haverziekte). This was described by Sjollema and Hudig in 1909 in which they connected the occurrence of grey specks with prolonged alkaline fertilizer applications. They suggested acid dressings and the application of Mn sulphate as the most effective means of curing the affected plants (Gerretsen, 1937).

A high availability of other divalent cations (such as  $Ca^{2+}$  and  $Mg^{2+}$ ) has an adverse effect on the availability of Mn. The size of the  $Mn^{2+}$  ion is similar to that of  $Mg^{2+}$  and  $Ca^{2+}$  and, therefore, competition between these nutrients occurs at the root surface. Mn uptake is further hampered by fertilization with a large dose of Potassium, Zinc and / or Molybdenum. Phosphate Fertilizer can also decrease Mn availability by the precipitation of Mn-phosphate. An interaction between Mn and Fe on the root surface is the result of a similar crop response mechanisms for Mn and Fe. Corn Plants with Mn deficiency are more sensitive to herbicides. It is therefore advised to first carry out a Mn fertilization and then to wait for 2 to 3 days with weed control (Anonymus, 2003c).

Weather conditions can have a major effect on Mn availability. Persistently dry weather increases the chance of Mn deficiency, even in soils where sufficient Mn is potentially available. This is because Mn is mainly transported in the liquid phase, via mass flow from the soil to the root surface.

Manganese deficiency has been reported for plants grown in in peaty (Netherlands, England and Denmark), coarse-textured (Sweden and Denmark), coarse/fine-textured (Netherlands), and podzolic and brown forest (Scotland) soils (Welch et al., 1991). In the Netherlands Mn deficiency mainly occurs in cereals and potatoes due to persistent dry weather on soils with a high pH. Mn deficiency occurs especially in rye, but also in barley in the north-eastern sandy soils.

### 5.3 *Measuring Mn in soils*

The Dutch fertilizer recommendations are based on measuring reducible Mn by extracting the soil with a solution of 1N-ammonium acetate to which a weak reducing agent (0.2% hydrochinon) is added. From 2004 onwards Mn-red is no longer measured at the routine soil laboratories but a switch was made to Mn- $CaCl_2$ . Mn availability is thus currently approximated using the weak multi-nutrient extractant 0.01M  $CaCl_2$  in which Mn is measured using an ICP-MS.

Mn-reducible extracts more Mn than does Mn- $CaCl_2$ , in addition a correction must be made for pH when converting the one into the other. For the recommendation this implies that a distinction must be made based on pH. This is in agreement with the common practice in the north-western part of Germany that borders The Netherlands (Niedersachsen).

In theory, 0.01M  $CaCl_2$  is expected to extract very low Mn concentrations. When fresh soil is analyzed indeed the Mn concentration is very low. Drying and rewetting has a large impact on the amount of Mn extracted which is related to dissolved organic carbon (DOC) dynamics (data C. Torrões). Rewetting a dried soil induces a DOC flux by oxidation of OM which liberates Mn due to reduction of Mn-oxides. This hypothesis explains why Mn- $CaCl_2$  is a relatively good indicator for Mn availability as the dissolution of Mn in the  $CaCl_2$  extract is directly linked to the nature and lability of the OM in the soil (Terrones and Supriatin 2015).

Another important aspect is the increasing accuracy of the measurements and decreasing detection limits over the years. In a dataset from 2002 containing Mn- $CaCl_2$  of 1236 samples there is a very poor relationship with pH due to a large variation in the data and a relatively high detection limit (Figure 5-1 ). Ten years later the relationship between Log Mn- $CaCl_2$  and pH is considerably better which is assumed to be (partly) due to a higher accuracy of the method as a whole and a lower detection limit. Data from the soils that were used for the wheat experiment indicate that with a lower detection limit the

relationship may be further improved.

With increasing accuracy and decreasing detection limits there is reason to believe that in the future Mn availability can be predicted based solely on pH. The sensors based on Near Infra Red (NIR) and Mid Infra red (MIR) spectroscopy are well capable of predicting soil pH. For this reason it is expected to lead to a (reasonably) good prediction of Mn-CaCl<sub>2</sub> using pH predicted with NIR/MIR.

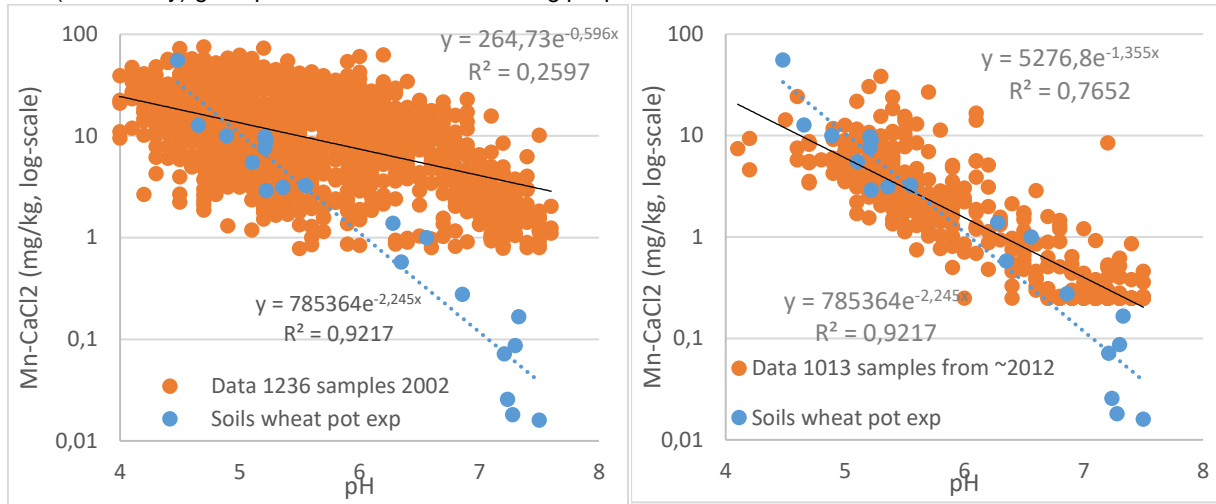


Figure 5-1 Relationship between Mn-CaCl<sub>2</sub> and pH in 1236 soil samples analyzed in 2002 (left Figure) and in 1013 samples analyzed in 2012 (right figure). In both figure the data from a recent wheat trial is also shown.

#### 5.4 Mn in soil in relation to crop uptake

In the wheat trial in the greenhouse 20 widely varying Dutch soils were used which show a large range in Mn-CaCl<sub>2</sub> (0.02 to 56 mg kg<sup>-1</sup>) and in pH (4.5 to 7.5). Both at low pH as well as at high pH there were soils containing high and low clay and OM levels. Mn-CaCl<sub>2</sub> was related to pH (Figure 5-1). In accordance with theory, the indices for Mn availability was low at pH greater than 6 and increased exponentially with decreasing pH. The Mn content in the wheat followed this pattern. Below pH 6, the Mn content of the wheat increased linearly with increasing LOG Mn-CaCl<sub>2</sub> ( $r^2=0.85$ , **Error! Reference source not found.**) but also inversely with pH ( $r^2=0.70$ , **Error! Reference source not found.**). At pH values greater than 6

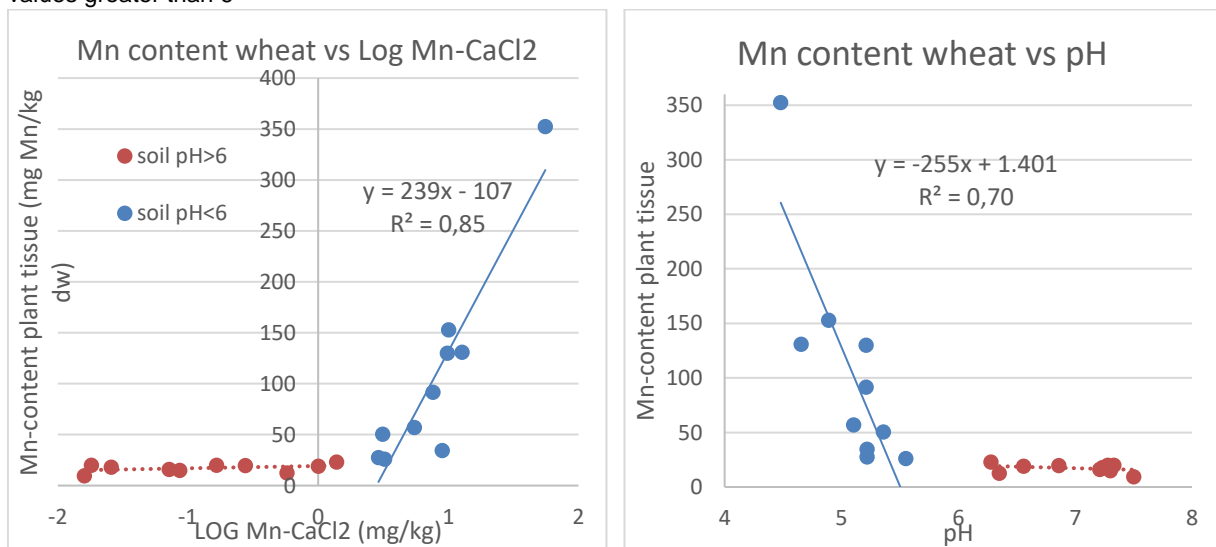


Figure 5-2 Relationship between Mn-CaCl<sub>2</sub> and pH in 1236 soil samples analyzed in 2002 (left Figure)

and in 1013 samples analyzed in 2012 (right figure). In both figure the data from a recent wheat trial is also shown.

The Mn content of the wheat was consistently low ( $17 \pm 4 \text{ mg kg}^{-1}$ ). The relationship between Mn uptake and Mn-CaCl<sub>2</sub> or pH was not affected by either clay content (range 1-33%) or OM content (range 1.4-15%) of the soils.

Different datasets from field trials (grass 2011, 2012 and maize 2011) were also analyzed with the aim to understand and predict Mn uptake. For the grass trials (N=21 in 2011 and N=24 in 2012) the Mn content of the crop is inversely related to pH and linearly related to Log Mn-CaCl<sub>2</sub>. Based on all data points the relationship with Log Mn-CaCl<sub>2</sub> is generally better than with pH. The Mn-CaCl<sub>2</sub> indicator for Mn availability seems independent of soil type as the grass experiment comprised of sand, peat, marine- and river clay.

In the maize experiment (n=14) the relationship between Mn uptake and pH was very poor ( $r^2=0.39$ ) as was the relationship with Mn-CaCl<sub>2</sub> ( $R^2=0.42$ , data not shown). Despite the poor correlation the trend is similar to the wheat trial.

The pot- and field experiments clearly show a consistently low Mn content in both the grass and wheat trial at pH higher than 6. At pH lower than 6 the Mn content of the crop linearly increases with increasing Log Mn-CaCl<sub>2</sub>. For the maize trial this trend is confirmed but the data shows too much variation and the number of data points is limited.

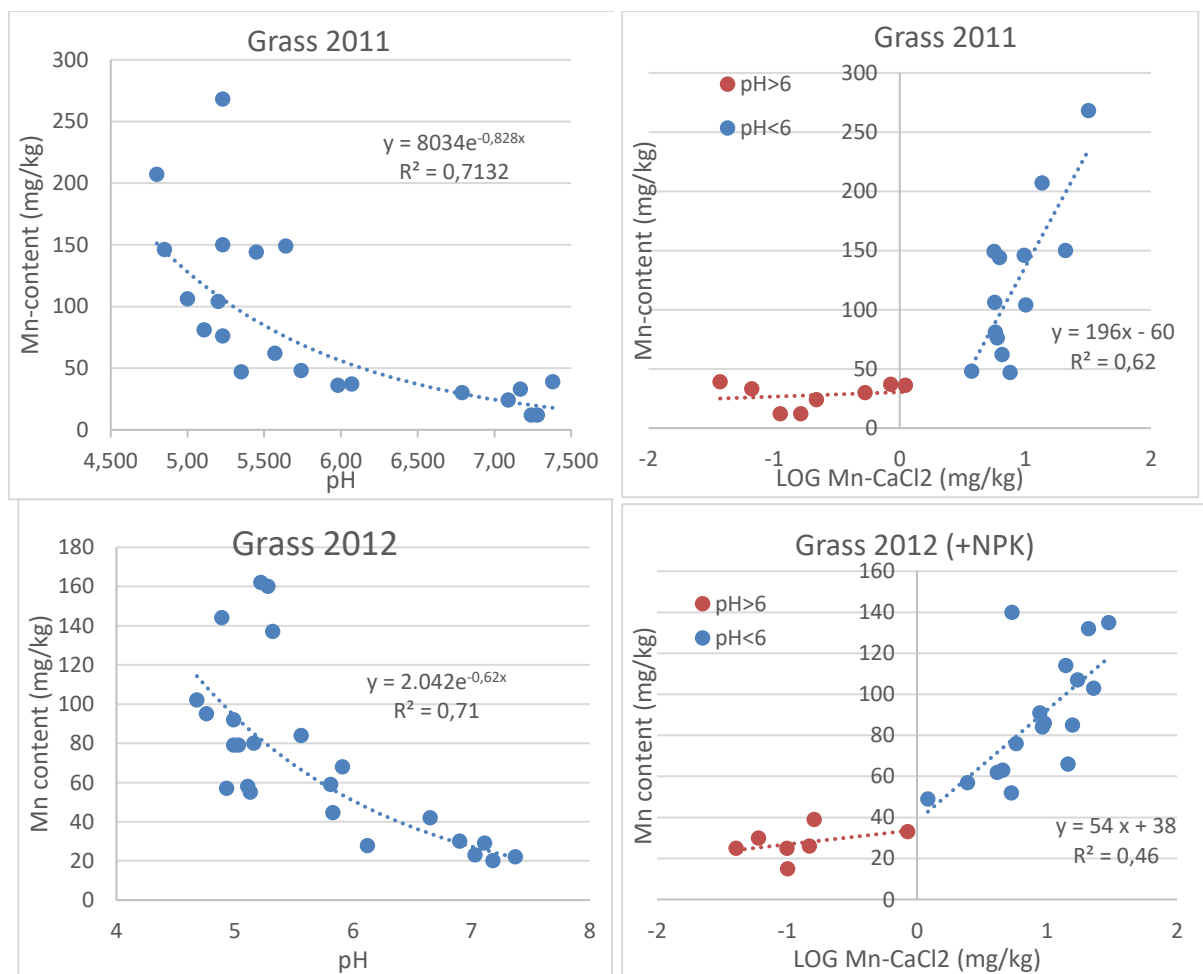


Figure 5-2 Relationship between Mn-content in grass and pH (left figure) and Mn-CaCl<sub>2</sub> (right figure) in field trial 2011 (top) and 2012 (bottom).

## 5.5 *Response to fertilizer applications*

The fertilization recommendation only recommends foliar application of Mn because it is much more effective than applying Mn to the soil. In The Netherlands deficiency is generally not caused by the lack of Mn but by the low availability of the Mn that is present due to the soil chemical processes at high pH. The same soil chemical processes are the reason that the availability of applied fertilizer Mn has a low effectiveness. In the 2012 grass field trial no response was found in Mn content of the grass when  $\text{MnSO}_4$  was added at a dose of only 1 kg Mn per ha. This dose is very low compared to the dose of 50 kg Mn  $\text{ha}^{-1}$  that was found to be effective to prevent Mn deficiency in oats (Smilde, 1968). In this field trial both MnO and  $\text{MnSO}_4$  were found to be comparably effective and at a dose of 50 kg Mn  $\text{ha}^{-1}$  soil application was as effective as foliar application (dose not mentioned). In the same study (Smilde 1968) a pot experiment with soil from the Biesbosch was conducted and application levels of up to 176 kg Mn  $\text{ha}^{-1}$  was not effective. In the fertilization recommendations the Biesbosch soil is treated differently because in these soils deficiency was found to occur at high reducible Mn levels. The C/N coefficient was found to be helpful to identify where deficiency might occur. However, it is unclear what prevents Mn uptake in these soils when it is clearly present in a potentially available form. This deserves further attention as the underlying mechanism might also be applicable in other regions.

Foliar applications are generally found to be effective in preventing and alleviating Mn deficiency. Different commercial foliar Mn fertilizers are available including Mn-sulphate, Mn-chelate, Mn-nitrate, Mn-carbonate. The effectiveness of the latter has not been proven (Wattel-Koekoek and Bussink, 2003). It is advised to apply the Mn fertilizer to young leaves during weather conditions of high humidity (Anonymous 2003). Mn is very little mobile in a plant which makes it necessary to repeat the application several times.

Continued dry weather increases the risk of Mn deficiency. When it is clear that continuous dry weather is the cause of an observed non-severe deficiency it is advised to irrigate or to wait for rain. In a field trial with potato the effectiveness of foliar Mn-application (in combination with B and S fertilization) was also found to depend on the weather (Mulder et al., 2013). In the dry, hot growing season of 2011 apart from a positive effect of irrigation, there was also a clear positive effect of applying Mn to both investigated potato varieties (Festien and Seresta). In the humid and cooler growing season of 2012, no effect of foliar Mn application was observed on yield. No symptoms of Mn deficiency were observed in either 2011 or 2012. In 2011 a timely irrigation and in 2012 the damp weather allowed transport of the applied Mn to the plant thereby preventing deficiency.

Balance calculations show that Mn supply to arable land decreases due to lower application rates of animal manure and the decreased use of Mn-containing pesticides. It is not known what the effect is on the availability of soil Mn for crop uptake of reduced Mn supply to potentially Mn deficient soils, e.g. calcareous soils (Velthof en Van Erp 1998). This deserves further investigation.

## 5.6 *Improving fertilization recommendations*

### 5.6.1 Current fertilizer recommendations

In The Netherlands the problem of Mn deficiency is not related to a lack of Mn in soils but to a too low plant availability of the Mn that is present in the soil (De Vries and Dechering, 1960). Due to the observation that Mn deficiency only occurs when the pH of the soil is above a certain pH, De Vries and Dechering (1960) distinguish two categories: 1) Silt- and clay soils that contain lime ( $\text{CaCO}_3$ ) and 2) Dilluvial sandy soils that originally have a low pH but where Mn deficiency may occur locally due to liming.

The current recommendations are principally based on two studies: De Groot, 1956 for arable crops and

maize and De Vries and Dechering, 1960 for grassland. In the study by De Groot (1956) beets and grains were grown on marine silt and clay soils with a high pH and the crop was classified as being healthy or deficient (Figure 5-3). This study resulted in the current classification for arable crops (including maize) on clay soils based on the reducible manganese level and OM content (Table 5-1). Three areas form an exception; de Biesbosch, Noordoostpolder and Kreekrakpolder where deficiency was found to occur at very high reducible Mn levels in the soil. In the Biesbosch the C/N coefficient was found to be the factor with which a distinction can be made between deficient ( $C/N > 11$ ) and no deficiency symptoms ( $C/N < 11$ ). As mentioned before it is recommended to further investigate the Mn limiting processes in these areas so that this knowledge may also be used to establish if it is prone to occur in other areas.

Table 5-1 Classification of silt and clay soils based on reducible Mn levels

	Boundary Mn-reducible mg/kg		Remark
	≤2.5% OM	>2.5% OM	
Low	≤60	<100	Deficiency possible
sufficient	>60	>100	No deficiency

After changing from measuring Mn-red to Mn-CaCl<sub>2</sub> Bussink et al. (2002) suggested to adapt the classification based on the combination pH-KCl and Mn-CaCl<sub>2</sub> (Table 5-2). This suggestion has not been implemented in the current recommendations.

For sandy soils soil testing is not recommended, the recommendation is based solely on pH-KCl. For diluvial sandy soils with a pH-KCl lower than 5.4 deficiency does not occur. When the pH-KCl of the soil is greater than 5.4 it is recommended to check the crop for Mn-deficiency.

When a deficiency is expected to occur, based either on Mn-red or pH-KCl, a foliar application with one of the commercial foliar fertilizers is recommended. Foliar application may be applied when deficiency occurs or to prevent deficiency. The application may be repeated after a couple of weeks. The recommendation refers to the dose that is recommended for that specific crop by the producer of the fertilizer of choice.

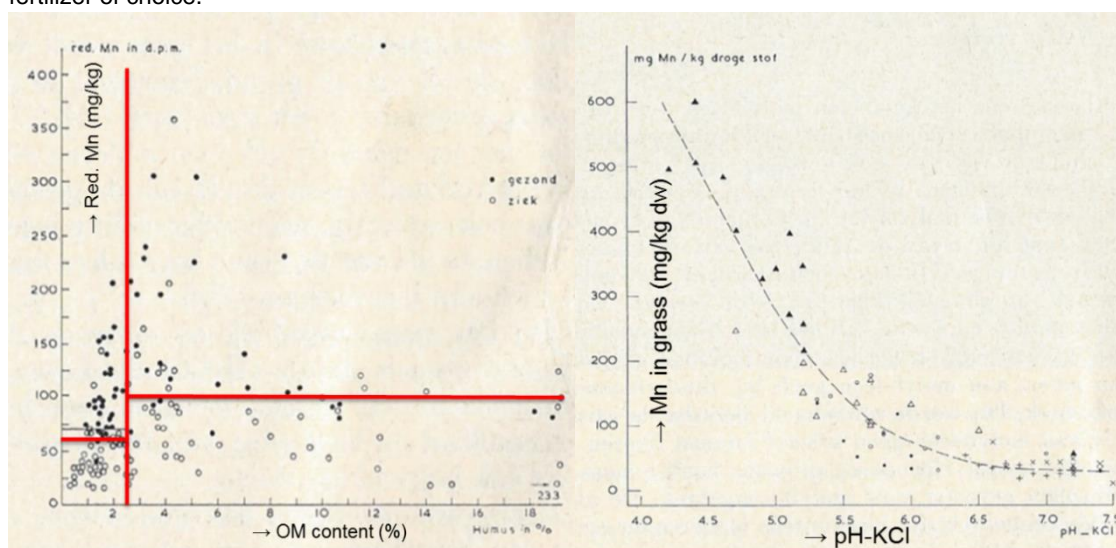


Figure 5-3 Healthy (closed symbols) and deficient beets and grains (open symbols) in relation to reducible Mn and OM levels on marine silt and clay soils (left figure, De Groot 1956). Relationship between Mn content in grass and pH-KCl (right Figure, De Vries and Dechering, 1960).

Table 5-2 Limits on the basis of Mn-CaCl<sub>2</sub> and reducible Mn, depending on pH and organic matter content.

pH	boundary Mn-CaCl <sub>2</sub> (mg/kg)		boundary Mn-reducible (mg/kg)	
	≤ 2,5% OM	> 2,5% OM	≤ 2,5% OM	> 2,5% OM
< 5,5	5,8	8,0	60	100
5,5-6,0	3,2	5,0	60	100
6,0-6,5	1,2	2,7	60	100
> 6,5	0	0,9	69	100

For grassland the recommendation is based on the study by De Vries and Dechering (1960) in which Mn levels in grass were related to pH (Figure 5-3). It is recommended to maintain the pH of the soil at the right level (pH between 4.8 and 5.6) for optimal grass growth as Mn availability will then be sufficient for both crop and cow. For grass soil testing is not advised as pH is a sufficient indicator and Mn requirement of grass is relatively low. Apart from maintaining the soil at the right pH no further recommendations concerning fertilization are given.

#### 5.6.2 Predicting Mn availability

Recent pot- and field studies show that at pH levels lower than 6 Mn-CaCl<sub>2</sub> is a good indicator for Mn uptake by a crop (wheat and grass). Below pH 6, pH itself may also be used as an indicator for Mn availability. At pH higher than 6 the availability of soil Mn for crop uptake is very low and also amounts of Mn extractable with CaCl<sub>2</sub> are that low that sampling and analysis do not seem to be worthwhile. With increasing accuracy and decreasing detection limits the relationship between Mn-CaCl<sub>2</sub> and pH has improved considerably over the past years.

Sensor techniques have, and are being developed rapidly. Techniques based on Near Infra Red (NIR) and Mid Infra Red (MIR) spectroscopy give good predictions of soil pH. This gives confidence that based on the relationship with pH, Mn availability will also be predicted using sensors based on NIR/MIR.

At pH higher than 6, both Mn-CaCl<sub>2</sub> and Mn uptake is consistently low. This is consistent with previous studies which underlie the current Dutch fertilization recommendations and it is the general consensus on this topic in literature. In the current recommendation a foliar application is advised to prevent deficiency or to cure a detected deficiency. The soil testing only predicts that a deficiency might potentially occur. It is desirable to diagnose potentially occurring deficiency as early as possible. To this means different non-destructive diagnostic tool have been and are being developed (Maarschalkerweerd and Husted 2015). A non-destructive diagnostic tool that has successfully been developed is based on chlorophyll a fluorescence (Pedas et al., 2014). It monitors photosynthetic performance and thereby specifically diagnoses manganese deficiency in plants.

The advantage of these handheld, simple to use, non-destructive methods that can be used in the field is that deficiency can be diagnosed before the symptoms become visible and Mn- fertilization can be applied on time. The preventive application of Mn can thus be prevented and Mn fertilizer does not have to be wasted.

#### 5.6.3 Adapting and including additional crops and conditions

Plants exhibit a wide variation in the uptake of Mn (Table 5-3). Worldwide research shows that meadow grasses, for example, may have a Mn-content ranging from 17 to 334 mg kg<sup>-1</sup> DM. A Mn content in the

plant of 20 mg kg<sup>-1</sup> DM is usually deemed sufficient. At lower levels Mn deficiency may occur.

The Dutch fertilizer recommendation does not distinguish between crops other than between grass and arable crops. The recommendations do not include a fertilizer dose as the farmer should apply the dose that is recommended for that specific crop by the producer of the fertilizer of choice.

Table 5-3 Average Mn content of different crops, expressed in mg kg<sup>-1</sup> of dry matter (Kabata-Pendias & Pendias, 2001).

Crop	Mn content, mg kg <sup>-1</sup> DM
cabbage (leaves)	28
salad (leaves)	29
Carrots (root)	14
Onion (bulb)	17
Potato (root)	15
Wheat (grain)	15-80
Grass	17-334

### 5.7 Conclusions for the Mn fertilization recommendation

At pH levels higher than 6, Mn availability is so low that further analyses of the soil Mn level (Mn-CaCl<sub>2</sub>) is not useful. At pH levels below 6 Mn-CaCl<sub>2</sub> is a good indicator for Mn availability in soil for crop uptake. Mn-CaCl<sub>2</sub> seems a better indicator than pH. However with increasing accuracy and decreasing detection limits there is reason to believe that in the future Mn availability can be predicted based solely on pH. This may also lead to a (reasonably) good prediction of Mn-CaCl<sub>2</sub> using pH predicted with NIR/MIR. Mn-CaCl<sub>2</sub> was found to be a good indicator for Mn availability in soil. This was not expected in first instance because it is a very weak extraction method. Extracting moist soil samples indeed shows very low Mn levels. However, when the soil is first dried, which is common practice, Mn levels are much higher and correlate well with crop uptake (when pH<6). The rewetting of the soil during the extraction procedure induces a dissolved organic carbon (DOC) flux by oxidation of OM. This liberates Mn due to the reduction of Mn-oxides. This hypothesis explains why Mn-CaCl<sub>2</sub> is a relatively good indicator for Mn availability as the dissolution of Mn in the CaCl<sub>2</sub> extract is directly linked to the processes that determine Mn availability in the soil (Terrones and Supriatin 2015).

In experiments with different extraction methods, the pot experiment with wheat, and the field trials with grass no other factors than pH, e.g. organic matter content or clay content were found to affect Mn uptake or Mn-CaCl<sub>2</sub> or the relationship between pH and Mn-CaCl<sub>2</sub>. The division based on organic matter content in the currently used classification of Mn-levels in soil for the fertilizer recommendations seems arbitrary and cannot be substantiated by the data presented here.

Soil testing only predicts that a deficiency might potentially occur. It is expected that in the future more hand-held easy to use non-destructive diagnostic tools will be used to diagnose deficiency in the crop before the symptoms become visible and Mn- fertilization can be applied on time. In the case Mn deficiency does not occur, the “preventive” Mn fertilizer application does not have to be wasted.

In the current recommendations weather conditions such as drought that might limit transport of Mn in the soil to the plant are not considered. A weather module may be developed that includes both actual weather data and a simple root development model. Recommendations in situations of drought may focus more on irrigation rather than on fertilization. It is recommended to integrate a weather module and irrigation advice in the fertilizer recommendation.

## 6 Selenium

### 6.1 Selenium in crops and animals

Selenium (Se) is an essential micronutrient to humans, for animal health and animal product quality, with indications that it is also beneficial for crop growth and quality (Haug et al., 2007). Without Se fertilization roughage (grass and maize) generally contain insufficient Se to ensure adequate supply of cattle (Den Boer, 2008). For grazing dairy cows the Se-intake can be lower than the animal requirement due to limited supply of concentrates. In winter supplies are sufficient when roughage due to its supplementation with concentrates or . The dry cows and young stock (< 2yr) are the high-risk groups with regard to the supply of Se, especially in rations which predominantly consist of grass (without additional concentrates, Den Boer et al., 2011). In The Netherlands the optimum Se requirement of dairy cows is around 0.15 mg Se kg<sup>-1</sup> dry matter. International studies report Se deficiency in animals when fed rations and/or feed crops containing less than 0.1 mg Se kg<sup>-1</sup> (Gupta and Gupta, 2000).

When dairy products contain too low amounts of selenium this may also negatively affect human intake. Worldwide the variation of Se status in humans largely depends on their diet, which is strongly related to the geographical variation in soil's Se level (data FAO). The wheat grown on arable land soils in the Netherlands also contains low selenium for human intake (Supriatin et al., 2016). The range of intake between which Se deficiency and toxicity occurs is relatively narrow, with current estimates suggesting that intakes below 40 µg day<sup>-1</sup> are inadequate and those exceeding 900 µg day<sup>-1</sup> are potentially harmful (WHO, 1996). A prolonged excess of Se (> 2 mg Se kg<sup>-1</sup> DM) can lead to blindness, excessive salivation, paralysis, abnormal development hoof and hair loss.

### 6.2 Selenium in soil

Sandy soils generally contain little Se. The Se present in sandy soils is associated with organic matter (OM). Clayey soils contain relatively more Se than sandy soils due to the presence of Se-enriched minerals such as biotite on the one hand, and higher levels of aluminum and iron oxides forming main components for Se sorption on the other. In organic rich soils, the native Se content varies depending on the origin of the soil (Hartikainen, 2005). When peaty soils contain large quantities of Se the availability may be low due to the strong binding of Se to the OM. According to Johnsson (1992) organic matter contains 80 times more Se than clay. In agricultural soils low in Se, Se is also mainly associated with OM, both in the solid phase and in the soil solution (Supriatin et al., 2015).

Selenium may occur in different forms in the soil depending on the pH and the redox conditions. The Se speciation is basically controlled by three transformation mechanisms: oxidation vs. reduction, mineralization vs. immobilization, and volatilization. Schematic Se cycle in soil is shown in Figure 6-1 (Ros et al., 2014). The different forms of Se include selenate (SeO<sub>4</sub><sup>2-</sup>), selenite (SeO<sub>3</sub><sup>2-</sup>), elemental Se (Se<sup>0</sup>) and selenide (Se<sup>2-</sup>). In addition, it can also occur as volatile diselenides (Hartikainen, 2005).

Plants acquire Se predominantly as selenate, but may also take up Se as selenite and small organic compounds. Selenate tends to be the predominant form in aerobic and neutral to alkaline environments, whereas selenite is the major form present in acid soils (Mayland et al., 1991). Selenate is quite mobile in soil and as a consequence is prone to leaching. Selenite is adsorbed much stronger to the soil than selenate. The predominance of selenate in neutral and alkaline soils is mainly due to a higher affinity of selenite for sorption to iron oxides, clay minerals and soil organic matter. The binding to these soil constituents decreases at high pH. At high pH Se is usually present in the more available selenate form. Liming of soils is therefore a well-known strategy to increase Se availability in the soil solution.



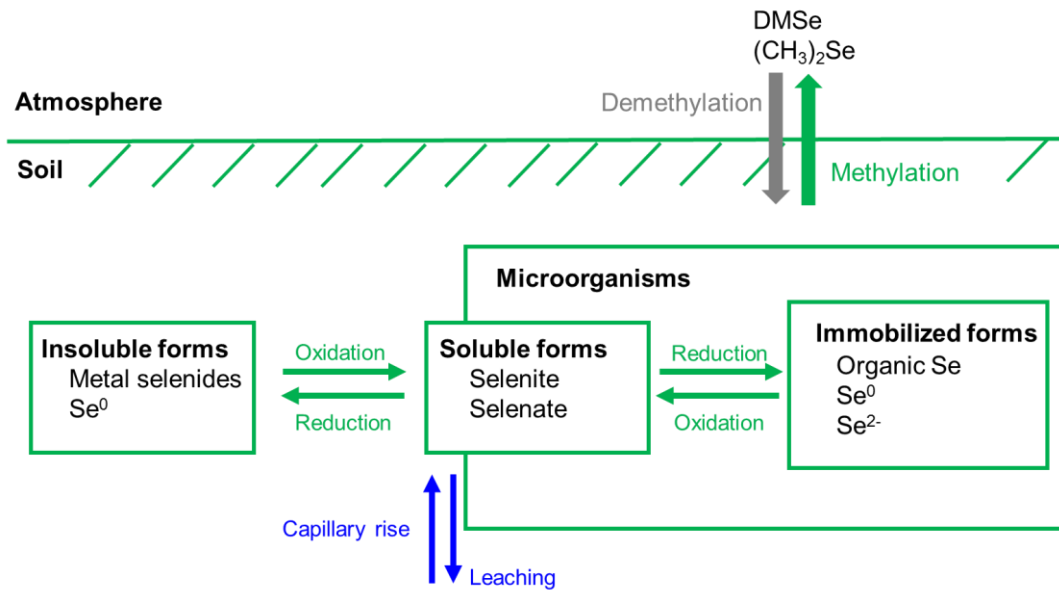


Figure 6-1 Schematic Se cycle in soil (in Ros et al, 2014, adapted from Flury et al., 1997).

Selenium in Dutch agricultural soils is mainly (+/-80%) associated with organic matter (Supriatin et al., 2015). Relatively little Se is in the soil solution (approximately 1.5%). Of the Se in solution only 13-34% is inorganic Se or Se in organic nanoparticles. In solution Se is also mainly (>85%) associated with colloidal organic Se (Weng et al., 2011).

A study of Se content in Dutch soils Figure 6-2 shows that as described above, Se content is in general low in sandy soils and high(er) in clay, and especially peat soils. A recent study showed that around 75% of 83 soil samples taken from grasslands in the Netherlands are mainly in the low range of total selenium content (i.e. selenium deficient, Supriatin, 2016). The low total selenium content in the soils causes the low selenium content in grass. However, total Se may not be the best indicator as not the total amount but the available amount of Se is relevant for Se uptake. In The Netherlands the risk of Se deficiency has been reported to be greatest during the grazing season (summer) on sandy and peaty soils. On the sandy soils the deficiency will be due to the lack of Se and on the peat and organic rich soils due to the low availability of the Se that is present. On peat soils the low Se availability is associated with the adsorption of Se due to the high OM level and the combination of relatively large quantities of Fe- and Al(hydr-) oxides and a low pH. Se deficiency has also been associated with acid soils high in Fe- and Al(hydr-) oxides (Gupta and Gupta 2000).

The results regarding the effect of sulfate (S) and phosphate (P) on the availability of Se are contradictory (Wattel-Koekoek and Bussink, 2006). Se uptake by the crop was adversely affected by high S and P concentrations. On the other hand competition with both S and P in the soil can result in more Se becoming available. Extensive research in Finland Ylärinta (1983, 1990, 1993) showed a strong reduction in Se uptake of selenite and selenate when sulfate and phosphate were applied in pot experiments with Italian ryegrass.

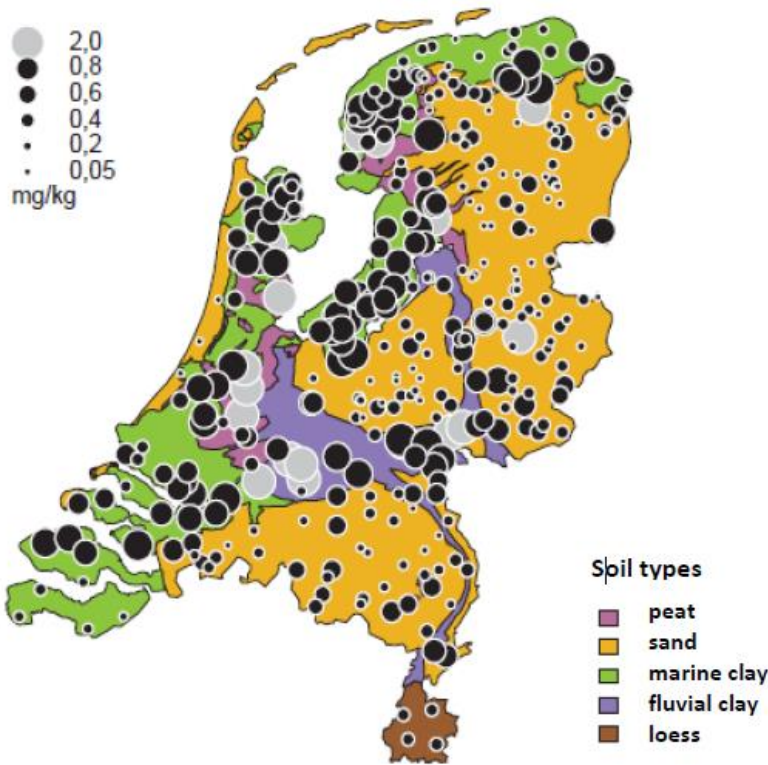


Figure 6-2 Map of Se content in soils in the Netherlands. Source: Mol et al. (2012).

### 6.3 Measuring Se in soil

In the Netherlands, there is no 'historic' method to determine available Se in soil. In other countries, the methods to determine Se levels in soil are based on the total Se-content or on a wet destruction (Kubota & Cary, 1982). Total Se is often not a good indicator of the Se-availability. By including soil parameters such as pH, clay and organic matter increased the explained variance between total Se in soil and Se in crop to above 60% (Johnsson, 1992). With modern techniques (ICP-MS) very low Se concentrations can now also be determined in weak extraction solvents. Since 2004 Se is measured in the  $\text{CaCl}_2$  extract.

### 6.4 Se in soil in relation to crop uptake

In a pot experiment with wheat using 19 widely varying soils low in Se from arable fields, the variability in Se content in the wheat shoots is largely explained (88%) by the Se to DOC ratio both measured in a 0.01 M  $\text{CaCl}_2$  extract (Figure 6-3, Supriatin et al., 2016). Other parameters that were found to be related to Se-uptake were soil pH (higher uptake at higher pH), and soil organic matter quality (i.e. soil C:N ratio). Johnsson, (1992), found that by including the soil parameters pH, clay and OM the explained variance between Se-total in the soil and Se-uptake increased to above 60%.

The wheat trial results indicate that in soils that are low in Se the bioavailability of Se is dominated by organic Se and that it is related to the solubility and quality of Se present in dissolved organic matter. The Se uptake in the wheat shoots was highest for clay soils with clay content  $\geq 20\%$  and  $\text{pH} > 6$ . This was primarily related to the high Se to DOC ratio in 0.01 M  $\text{CaCl}_2$  extraction of these soils (Surpiatin et al., 2016).

In contrast with the results from the trials in the greenhouse, where conditions could be kept optimal, results from field trials with grass (2011 and 2012) did not show a relationship between Se-uptake and Se- $\text{CaCl}_2$ . Incorporating additional parameters also did not increase the explained variance. The Se

content in the grass field trials did show a positive trend with Se-CaCl<sub>2</sub> in 2011 ( $r^2=0.38$ , no NPK) but there was no relationship or trend with Se-CaCl<sub>2</sub> in 2012 (with NPK, Figure 6-3). Se in grass could not be predicted to any satisfactory extent by Se-CaCl<sub>2</sub> or any other Se fraction. The different results between pot and field trial are related to the much larger variability in moisture – temperature and related redox conditions in the field trial.

Weng et al., 2011 also did not find a direct relationship between Se in grass and Se-CaCl<sub>2</sub>. However, in combination with soluble S (sulphur) and/or P (phosphor), and the concentration of small DOC particles was found to explain most of the variability of Se content in grass. The results indicate that mineralization of organic Se is the most important factor that controls Se availability in soils. Competition with sulfate and phosphate needs to be taken into account.

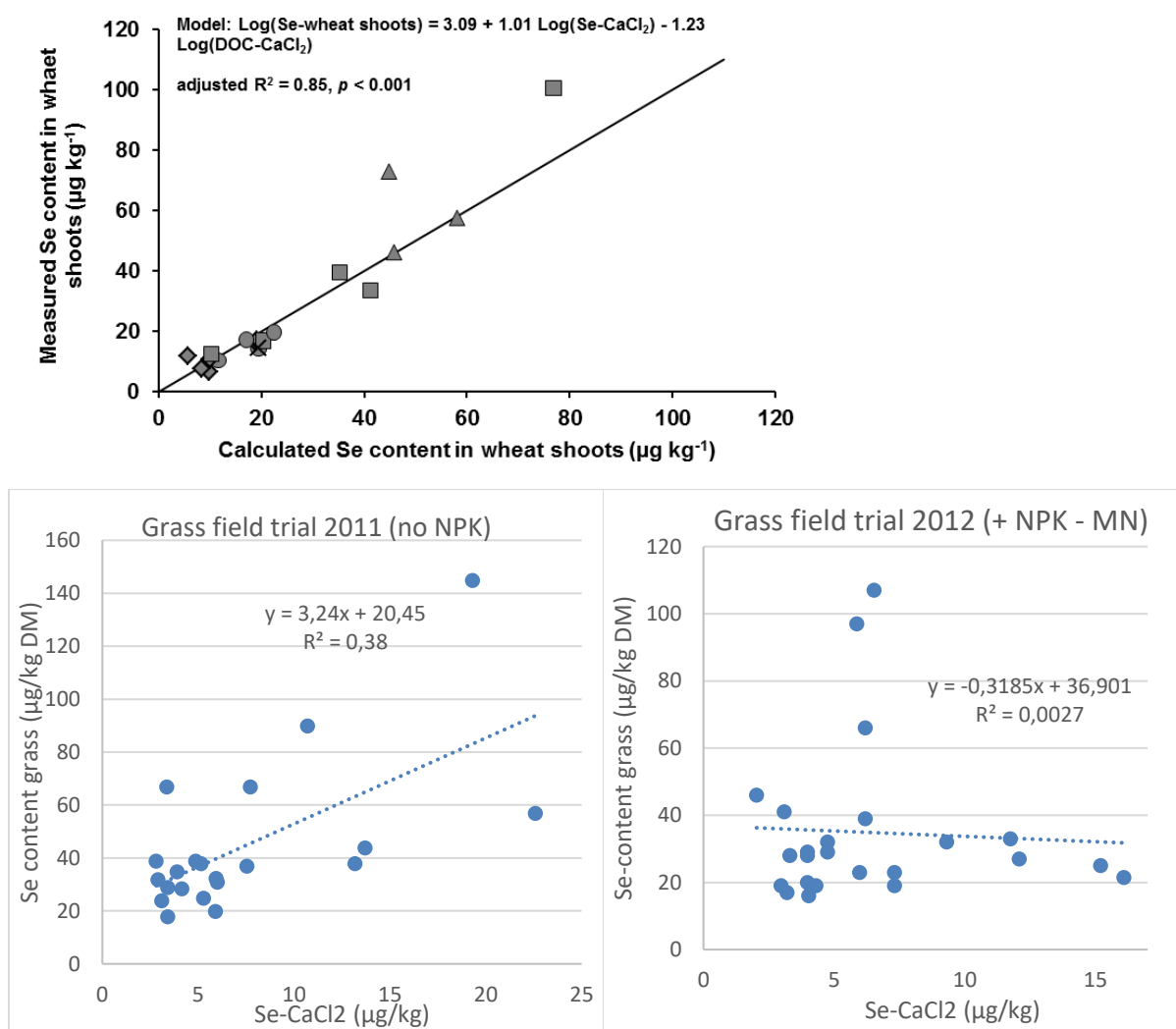


Figure 6-3 Relationship between Se-CaCl<sub>2</sub> and Se content in wheat in a pot trial (top figure, Surpiatin et al., 2016) and grass in the field trial in 2011 (bottom left figure) and 2012 (bottom right figure)

### 6.5 Response to fertilizer applications

Data from the grass field trials show that the Se concentrations in grass without micronutrient fertilization are (far) below the required 150  $\mu\text{g kg}^{-1}$  for grass for dairy cattle intake. The Se content of the grass ranged between 16 and 145  $\mu\text{g kg}^{-1}$ ; only 2 fields had a Se content higher than 100  $\mu\text{g kg}^{-1}$ . Application

of Se fertilizer in the form of sodium selenate (4.4 g Se/ha) significantly increased the Se content of the grass to a sufficient level (Figure 6-4). Crops are generally found to have a high response to applied Se fertilizer (meta-analyses by Ros et al., 2016). The local situation greatly affects crop response. Based on the grass field trial (2012) it is suggested that on clay-peat and peat soils the response to Se-fertilizer is lower than on the sand and clay soils but this was not found to be significant (Supriatin, 2016, Figure 6-4). Response is defined as the ratio of the Se-content in grass with Se-fertilization versus without fertilization. After Se-fertilization there was no significant difference in Se-uptake between soil types. Without fertilization Se-uptake was highest on the clay soils with pH>6. In a meta-analyses of worldwide data, Ros et al. (2016) found that clay content has a significant effect on crop uptake response to Se-fertilization (also defined by the ratio of Se content of the crop with and without Se fertilization). At low (<5%) clay content the response was higher (average response of 1100%) than at high (>35%) clay content (average response of 400%). With increasing clay content, response decreased presumably due to sorption of the added Se. The same meta-analyses did not show an effect of OM content and the response to pH was variable. At a Se content of the soil lower than 0.2 mg kg<sup>-1</sup> the response was significantly larger than at higher Se levels. In the Netherlands Se deficiency is most likely to occur on sandy soils. Due to the low clay content and low Se content, these soils are thus expected to show the largest response to Se-fertilization.

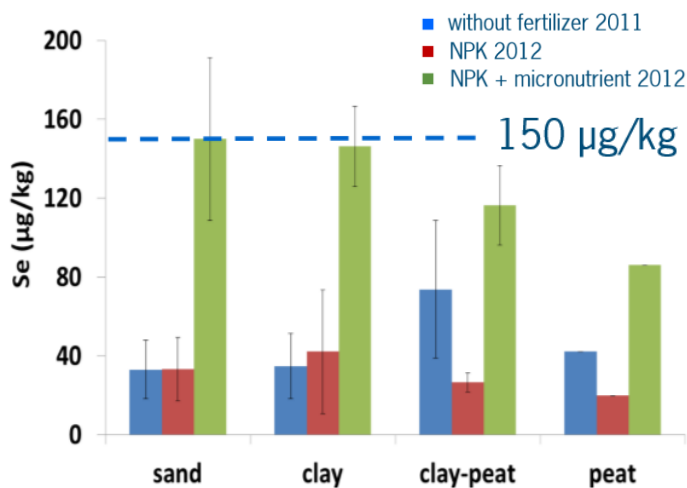


Figure 6-4 Se uptake in grass in the 2011 and 2012 field trials.

The efficiency of the applied Sodium Selenate was on average 12% in the grass field trial in 2012. In older field trials recoveries of above 50% were found (Bussink, unpublished) when Selenate was applied before the first cut. Adapting fertilizer strategies to the local agronomic situation and soil properties can increase the crop uptake efficiency from 10% (common situation) up to 50% (Ros et al., 2014). Important fertilizer strategies include

- The use of a site specific fertilizer dose: Se fertilizer use should account for the Se supply and availability in the soil and any residual effects of former Se fertilizer applications.
- The choice for a specific Se fertilizer: selenate is about 8 times more effective on the short term than selenite but the residual effects over time are lower.
- Application technique: both foliar and soil applied fertilizers are able to enhance Se uptake but foliar application is more resource efficient. Seed coating can be an alternative but the crop uptake efficiency is usually less than 10%.
- Application timing: fertilizer application during the growing season results in higher Se levels in the crop in comparison with fertilizer applications before the growing season.

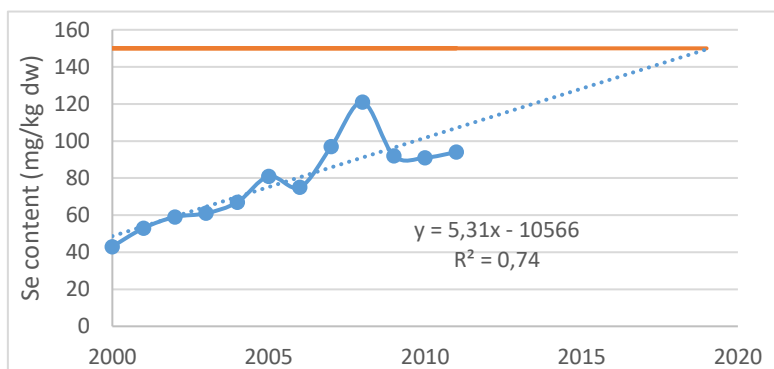


Figure 6-5 Trend in Se levels in grass silage since 2000 (data Eurofins, <http://www.melkvee.nl/nieuws/1719/>)

Both grass and maize naturally contain less Se than the target of  $150 \mu\text{g kg}^{-1}$  dry matter. Since 2000, farmers have started to apply more Se fertilizer. This has resulted in an increase in the averaged Se levels in grass silage (Figure 6-5). However, the increase that was seen between 2000 and 2006 seems to be lower between 2009 and 2011 (data Eurofins published by OCI agro). The average content of  $95 \mu\text{g Se kg}^{-1}\text{DM}$  in 2011 is still significantly lower than the recommended  $150 \mu\text{g kg}^{-1}$  and thus needs attention.

### 6.6 Improving fertilization recommendations

Quantities of  $150 \mu\text{g Se kg}^{-1}$  dry matter in the ration are desirable. Grass often contains only one-third of this amount. Research shows that a dose of 8-10 g Se per hectare is sufficient to keep the Se levels in the grass above the target value of  $150 \mu\text{g kg}^{-1}$  DM for several months. For all the grass (silage) it is therefore recommended to fertilize the grasslands annually with up to 10 grams of Se per hectare to ensure a sufficient Se content. This advice applies to all soil types because a literature review did not show an effect of soil type on Se-response (Den Boer et al., 2008).

It is advised to apply Se fertilizer at least once a year. A more frequent fertilization at a lower dose is however recommended to ensure a good distribution of the Se content in the grass during the growing season (several cuts), and to minimize losses by leaching. Bussink (2000) concluded that in order to obtain  $150 \mu\text{g kg}^{-1}$  grass DM, in theory 2-3 g Se  $\text{ha}^{-1} \text{yr}^{-1}$  (applied as selenate) is required if this Se is evenly distributed over all cuts. The frequency depends on the chosen fertilizer. In practice it is often only one or two applications. Therefore in total 10 g Se  $\text{ha}^{-1} \text{yr}^{-1}$  is recommended. Grass has a quick response to applied Se. The risk of toxic levels is large when applying a high single dose or by foliar application the grass. At a level of 2-3 mg Se  $\text{kg}^{-1}$  DM in grass, Se is toxic for the cow. The best time of application of Se fertilizer is in spring before the first cut but no later than a few weeks after the first cut (Bussink, 2000). Selenate is about 8 times more effective on the short term than selenite. However, the residual effect over time of the applied fertilizer is lower for selenate compared to selenite. Positive effects of selenate rarely last longer than 1 year. Positive effects of selenite may, however, last for periods up to 3 years. In view of the low recommended dose, application takes place with the aid of carrier fertilizers. The most commonly used carriers are sodium and nitrogen fertilizers.

Because high sulfur levels in the feed ration reduce the Se absorption by the cattle it is important to keep to the recommended S fertilization, and not over-apply S.

### 6.7 Conclusions for the Se fertilization recommendation

Estimates indicate that worldwide there is currently only sufficient Se for approximately 39-47 years (Udo de Haes et al., 2012). This results in the necessity to only use Se where really needed and to use the

most effective form. For this a robust and reliable fertilizer strategy is needed that takes the spatial and temporal variability in climatic conditions, soil properties, and cropping systems into consideration.

In The Netherlands the Se-content in roughage without fertilization is too low to meet the cattle demand. The risk of Se deficiency has been reported to be greatest during the grazing season (summer) in sandy and peaty soils. In the sandy soils the deficiency will be due to the lack of Se and in the peat and organic rich soils due to the low availability of the Se that is present.

In the Netherlands, there is no 'historic' method to determine available Se in soil. Since 2004 Se is measured in the CaCl<sub>2</sub> extract. Under controlled conditions of a pot trial the variability in Se content in the wheat shoots is largely explained (88%) by the Se to DOC ratio both measured in a 0.01 M CaCl<sub>2</sub> extract (Supriatin et al., 2016). Other parameters that were found to be related to Se-uptake were soil pH (higher uptake at higher pH), and soil organic matter quality (i.e. soil C:N ratio. In general, both clay content and soil pH have a positive effect on the potential bioavailable Se.

The relationship between Se-uptake and Se-CaCl<sub>2</sub> could not be reproduced under field conditions in grass trials (2011 and 2012). These grass field trials did underpin that the Se concentration in grass without micronutrient fertilization is (far) below the recommended concentrations for ruminants.

Application of Se fertilizer in the form of sodium selenate (4.4 g Se/ha) significantly increased the Se content of the grass to a sufficient level. Crops are generally found to have a high response to applied Se fertilizer (meta-analyses by Ros et al., 2016). The local situation greatly affects crop response. The meta-analyses showed that clay content has a significant effect on crop uptake to Se-fertilization (defined by the ratio of Se content of the crop with and without Se fertilization). With increasing clay content, response decreased presumably due to sorption of the added Se. The same meta-analyses did not show an effect of OM content and the response to pH was variable. At a Se content of the soil lower than 0.2 mg kg<sup>-1</sup> the response was significantly larger than at higher Se levels. In the Netherlands Se deficiency is most likely to occur on sandy soils. Due to the low clay content and low Se content, these soils are thus expected to show the largest response to Se-fertilization. Adapting fertilizer strategies to the local agronomic situation and soil properties can increase the crop uptake efficiency from 10% (common situation) up to 50% (Ros et al., 2014).

The following recommendations are made to improve the Se fertilization recommendation:

- The use of a site specific fertilizer dose: Se fertilizer use should account for the Se supply and availability in the soil and any residual effects of former Se fertilizer applications.
- Se fertilizer dose should account for the Se supply and availability in the soil and any residual effects of former Se fertilizer applications.
- Fertilization with selenate is preferred above selenite. Selenate is about 8 times more effective on the short term than selenite but the residual effects over time are lower.
- Both foliar and soil applied fertilizers are able to enhance Se uptake but foliar application is more resource efficient. Seed coating can be an alternative but crop uptake efficiency is usually less than 10%.
- Split fertilizer application during the growing season results in higher Se levels in the crop compared to a single application before the growing season. A more frequent fertilization at a lower dose is recommended to ensure a good Se content during the growing season, to minimize losses by leaching and to minimize the risk of toxic levels. The frequency depends on the chosen fertilizer.

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## Appendix 1 Background extraction methods to assess micronutrient levels in soil

### *General*

Until recently many soil extractants were based on “trial and error” research. Extracting solutions of different composition were tested which were aggressive enough to detect differences in nutrient contents between soils. These differences were related to yield and fertilizer response trials. This correlative approach has led to the situation that numerous methods (variation in shaking time, extractant type and soil to soil extractant ratio) are used for the different regions all over the world (Lindsay and Cox, 1985; Sims and Johnson, 1991). The extractants vary from mild to aggressive. Especially in the past the more aggressive extracts (Figure 6, and Table 3) had to be used compared to macronutrients in order to detect the low amounts available in soil.

For optimum plant growth nutrient concentration in soil solution should be maintained above a certain level. A growing crop will first use what is directly available (intensity). The concentration in the soil solution will decrease and then the quantity that can readily come into solution (capacity or quantity), determines if the concentration can be maintained above the required minimum level for optimum plant growth. If not, fertilization is required. This concept is well explained by Mengel and Kirkby (1987). When we use only one extractant for soil it is however difficult or even impossible to make a clear distinction between intensity and capacity. Experimental correlation of the extraction result with crop response becomes then a necessity. The result may still be that the correlation coefficient between yield response and the soil analysis result is often fairly low. Besides difficulties in distinguishing what is directly available and what can be delivered, the extractant may also overestimate the deliver capacity because of release of micronutrients from components in the soil matrix, which are normally not accessible by plant roots.

Two types of extraction, one for intensity and one for capacity, is in principle a better approach but more expensive and therefore not common.

### *The extraction process*

Figure 1 schematically shows in which forms micronutrients can be available in soil. Roughly speaking the amount of nutrients present in a form decrease in the order of mineral >> adsorbed > chelated >> free cation/anion. The free cation/anion in solution is the form directly taken up by roots (Marschner, 1995). The release by (natural) chelates, the desorption rate and/or the weathering rate (usually very small) determine the deliver capacity of the soil. On the right hand side of Figure 1 extractant groups are presented. The amount extracted with these groups roughly decreases going from strong acids towards water. Water extraction or dilute neutral salt extraction gives an impression of what is available or what can be exchanged. Both may have a very good relationship with crop response. However, the relatively high analytical detection was a serious problem in the past and therefore a major reason why weak extractants were seldom or not measurable and feasible. Much effort was put in extractants based on weak and strong acids, salt solutions, complexing agents or mixtures of these to reach the high detection limits (see also Table 3).

From the late forties onwards many experiments were performed to relate extractants with crop response. This was also the case in the Netherlands for the Cu and Co content in grass. Extractions were performed with 0.43 M HNO<sub>3</sub> and 0.4 M acetic acid, respectively and at present these extractions are still in use without much change in the basic recommendations (Henkens 1998a and b).

Because of the trial and error approach extractants had to be correlated with crop response trials whenever the soil type, climatic region and or the crop type was changing. Correlation results with crop type often increased when soil pH, organic matter content and/or CEC were taken into account. Although this approach is laborious good results were obtained between extraction and crop response in individual experiments with correlation coefficients sometimes up to 90% and more, as is shown by Sims and Johnson (1991). Differences in soil type, climatic conditions and crop type result in a range of critical values (below values were an effect of fertilizer on yield or quality might be expected) as is shown in Table 4 for some common extractants.

The trial and error approach has led to the situation that in the Netherlands for the elements B, Co, Cu, Zn and Mn all are determined with different extractants. This is expensive. International the tendency has shifted towards the use of multinutrient extractants. Jones (1998) gives some examples of universal extractants like: Morgan, Wolf-Morgan, Mehlich 1 and 3 and AB-DTPA. Houba et al. (1990) proposed 0.01 M CaCl<sub>2</sub> as multi-nutrient extractant for macro- and micronutrients and later also for heavy metals (Novozamsky, 1993), resulting finally in a general analysis protocol (Houba et al. 2000). Van Erp et al. (1998) gives a literature overview of the quality of the relationship obtained between the amounts of nutrient extracted with 0.01 M CaCl<sub>2</sub> for macro- and micronutrients. The quality of the relationships varies from poor to very good ( $r^2 = 0.90$ ). They concluded that the 0.01 M CaCl<sub>2</sub> procedure is a promising tool in near future farm nutrient management.

#### *The analytical measurement of micronutrients*

Developments in analytical techniques enhanced the use of multi-nutrient extractants and make it also possible to use weaker extractants. During the past decades detection limits decreased by several order of magnitude and furthermore there was a shift towards multi-element analysers. In the seventies optical emission spectrometry based on inductively coupled plasma (ICP-OES or ICP-AES) came up. Later ICP-MS came up which was a big step forward to very low detection limits in multi-element analysis. ICP-MS (inductively coupled plasma mass spectrometry) about three orders of magnitude better than ICP-OES. This makes ICP-MS the preferred technique. The development of these sensitive techniques makes it in principle possible to determine the very low amount of free micronutrient ions (Cu<sup>2+</sup>, Zn<sup>2+</sup>) in solution and thereby micro nutrient speciation. This is important because it is generally believed that plant roots take up the free the micronutrient form. However, the total amount of micronutrient dissolved in the extractant is determined by ICP(-MS). This can be overcome by the Donnan Membrane Technique (DMT). It can measure the free ionic form in soil solutions in the presence of inorganic (Cl, PO<sub>4</sub>) and organic (dissolved organic matter) complexing agents (Temminghoff et al., 2000, Weng et al., 2001). The DMT consists of two parts: a donor side and an acceptor side. A negatively charged cation exchange membrane separates the two sides. The donor side contains the solution with metals and ligands in which one would like to measure the free metal concentration. The acceptor side only contains a salt used for metal exchange (e.g. Ca(NO<sub>3</sub>)<sub>2</sub>). The membrane allows free cations to pass the membrane whereas complexed cations and anions will not pass the membrane. This opens new ways to a more fundamental and less trial and error approach in determining the availability of micronutrients.

#### **Interpretation of results - Towards mild extractants –**

It is important to include the enormous knowledge gain in understanding soil (chemical) processes affecting micronutrient availability into the evaluation of the soil analysis result. This means: knowledge

about the deliver capacity of the solid phase is necessary: adsorption and desorption behaviour of clays, oxides, organic matter and the chelating properties of dissolved (in)organic ligands

Determining the composition of the solid phase (organic matter and clay content, CEC, the amount of  $\text{CaCO}_3$  and metal oxides) may be relatively expensive but is only necessary once every 5-10 years and cheap techniques are underway. Non-invasive techniques like Near (1200- 2500 nm) or Mid Infrared (2500-22.000 nm) Spectroscopy have a great potential in this respect. With these techniques spectra of soil samples are recorded. These spectra are calibrated and validated against the classical analytical technique. Chang et al. (2001) tested 33 chemical, physical, and biochemical properties of 802 soil samples from 4 areas in the USA. They showed that total C, total N, CEC, sand and silt could be predicted accurately ( $R^2 > 0.8$ ) with near infrared spectroscopy. Clay content was predicted somewhat less accurately. Near infrared spectroscopy is now embedded in routine soil analysis by Eurofins Bllg. Many parameters are analysed, including total C, clay content, CEC and  $\text{CaCO}_3$  content, with high precision ( $R^2 > 0.9$ ) (Vedder, personal communication). In Australia CSIRO even offers over 20 parameters to be analysed on a routine basis by mid infrared. Besides total C, clay content and CEC also cationic composition of the CEC is determined. Since 2004 near infrared analysis is common in the Netherlands. Eurofins offers a broad range of parameters. Nowadays total C, N and S, clay, clay, sand and silt content but also CEC, the cationic composition of the CEC and pH is determined Further research should make it clear if oxides and the surface characteristics of particles can be characterised by these spectral techniques.

For several elements like Cu (Temminghoff, 1998), Co (Mclaren et al., 1987), Ni (Weng et al) and Zn (Qian and Xie) the methodology of using weak electrolytes and determining the composition of the solid phase and multi-surface modelling looks very promising. This methodology can also be very useful in agronomical research, also to include other micronutrients. The proposed methodology could save a lot of money because a basic concept may work in the end on all types of soil, whereas up to now many different extracting procedures have been developed, which might work rather well in one region but only poorly in other region.

Furthermore, in the long-term real multi-nutrient based fertilizer recommendations may become possible. It is well known that the availability and uptake of a (micro)nutrient is affected by the level of the other (micro)nutrients (Mengel and Kirkby, 1987; Kabata-Pendias & Pendias, 2001). This is up to now only seldom taken in account (with the exception of water cultures like in glass horticulture).

At present the first step in using data of the solid phase in combination with the extraction result may still be in the way as Sillanpää did (1982); developing statistical relations between crop yield and the extraction result in combination with one other soil parameter like CEC. However such an approach should then be regarded as a transition phase towards predicting micronutrient availability based on the extraction result, the solid phase characterisation and multi-surface models.

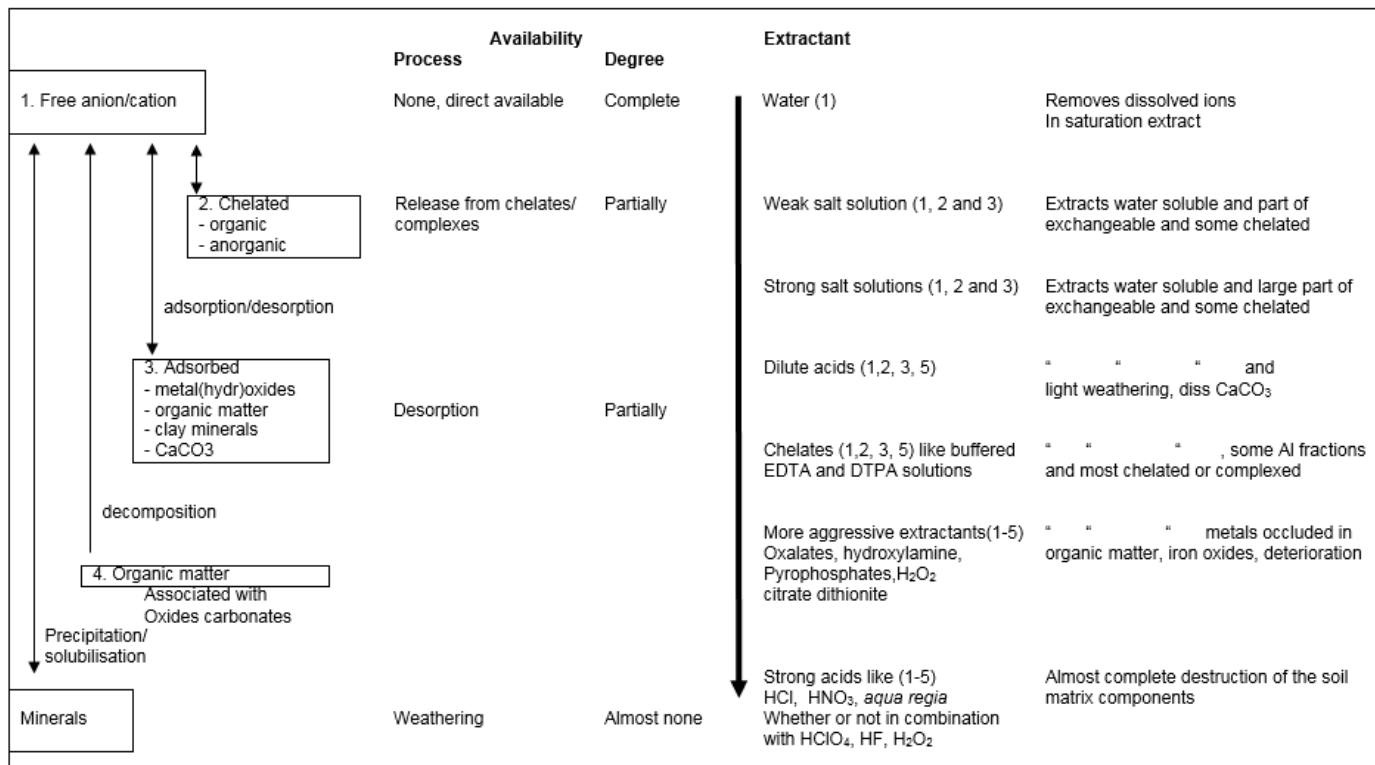
## Conclusions

- There is a great diversity in extractants and soil extraction procedures. More uniformity is required to meet today demands. Soil analysis for micronutrients started decades ago as trial and error research with rather aggressive extractants (containing all kind of combinations of acids, salts and chelating compounds) in order to make it possible to measure anything. The analytical result was related with crop response to develop relationships for fertilizer application. Protocols developed in former days were well tested, but the disadvantage is that they have all a very local and or crop

specific basis, so that results cannot be easily extrapolated to other situations. This is a great disadvantage for improving micronutrient recommendations.

- The development of multi element techniques as ICP-MS make low determination limits possible for micronutrients. This opens new opportunities for multi-nutrient extraction with weak electrolytes such as 0.01 M CaCl<sub>2</sub>, instead of using all kind of aggressive electrolytes for each micronutrient. Weak electrolytes are in principle better suited to describe soil processes on a fundamental basis. In 0.01 M CaCl<sub>2</sub> also pH can be determined and can be taken into account for model calculations.
- Prediction of micronutrient availability on a more fundamental basis is possible. For future development no new extractants should be developed but the knowledge gain in soil chemistry should be embedded in making better predictions of micronutrient availability. It is now possible to predict metal speciation in the soil solution, using multi-surface models that describe the adsorption and desorption behaviour of soil components (e.g. clay, oxides, organic matter, CaCO<sub>3</sub>) simultaneously. The free (metal) ion, the form that is taken up by plants roots, can now be measured directly by the Donnan membrane technique. It has a direct relationship with the in 0.01 M CaCl<sub>2</sub> extracted amount. In measuring properties of the solid phase mid and near infrared spectroscopy are promising tools. In this way micronutrient availability is estimated on a fundamental basis, which has in principle the advantage that it can be used for different soils types and different regions.

Figure 1. Schematic overview of the fractions containing micronutrients, the processes involved for releasing nutrients and type of extractant related to the release of micronutrient from a certain soil fraction. It is only a rough indication what are the main fractions delivering/releasing micronutrients to the extraction solution when certain types of extractants are used (Bussink & Temminghoff, 2004).





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