1. Introduction

1.1 Demand for alternative solutions to pollution by trace contaminants

Due to high cost of technical mining water treatment plants, phytoremediation likewise phytostabilisation are increasingly being proposed as alternative low cost technology. Despite the capacities of chemical technologies to eliminate high contamination loads of high concentrations from point sources, e.g. mine and tailings water from flooded mines and abandoned tailings (Gatzweiler et al. 2000; Jakubick and Kahnt 2002), there is a principal and growing demand for alternative solutions to treat:

- low concentrations (traces), but with high annual load due to many diffusive input into the catchments, which is typical in old mining areas in Germany;
- low input point sources, but with relatively high concentrated loads, which is characteristic of leaching or seepage water springs below mining dumps and ore processing tailing; and
- point sources with decreasing concentration of contaminants but with annual loads that are expected to last for decades or even centuries.

The loads include effluents from mining water treatment plants, discharges from closed wastewater treatment plant and mine flooding. The last scenario, above, is especially relevant in the decommissioned uranium mining areas in Germany and most eastern European countries where
mining was realised without regard for the environment (Hurst 2002). In addition to current strict German and international regulations and standards (e.g. EC 1998) for radionuclides mainly $^{226}$Ra, chemical toxicity of uranium is increasingly being considered (Gilman et al. 1998, Meinrath et al. 2003). Some national and European guidelines have already been introduced, or are in progress to introduce the limit levels of uranium concentration in drinking water to extremely low mg l$^{-1}$-range. The WHO provisional guide line recommends a uranium content limit in drinking water as $0.6 \ \mu g \ \text{uranium kg}^{-1} \ \text{body weight per day}$ as Tolerable Daily Intake (TDI) (Gilman et al. 1998). The U.S. Environmental protection agency (EPA) set the uranium content limits at $20 \ \mu g \ l^{-1}$ (ATSDR 1999). However, EPA considers a standard of $30 \ \mu g \ l^{-1}$ as within safe limits. Currently, the German States Commission on Water Issues (LAWA, Länderarbeitsgemeinschaft Wasser) is considering introducing very low limits levels of $1-2 \ \mu g \ l^{-1}$ for natural uranium in surface and ground water. Unfortunately, such uranium concentration levels can be found in natural surface and ground water of most anomalies regions worldwide, even without anthropogenic influences like mining and tailings. Further, higher values have been observed in natural water downstream of uranium ore surface mineralisation spots (Dudel et al. 2001, Meinrath et al. 2003, Salas and Ayora 2004).

Considering the limits and the contaminant loading in the decommissioned uranium mines, technical mining water treatment plants cannot be very costly and unable to meet all no source pollutions. Hence, passive treatment technologies, which have been well proven, and already established to eliminate organics and inorganic contaminants from different sources using plant communities, seems to be an alternative. Most organic contaminants originate from municipal and industrial waste piles, while most inorganic originates from ore processing. Generally, these passive technologies target heavy elements, radionuclides, arsenic, sulphate, and acidity (H$^+$) contamination.

Following the introduction of constructed wetlands to treat municipal wastewater, similar technologies have been adapted for inorganic contaminations (Wieder 1989; Younger 2001). Currently, there are technologies specifically designed for water impacted by acidity and sulphate load e.g. the Acid Mine Drainage (AMD) technology and the landfill leachates treatment (e.g. Dvorak et al. 1992, Kadlec 1999). The well-investigated efficient natural process of microbial sulphate reduction is used to reduce the acidity, decrease the sulphate load and fix the heavy metals in the course of microbial sulphate reduction. This availability of organic matter acts as electron donators (energy source) to microbes. For treatment of water, mainly organic acids provide the energy that act as the driving force. Thus,
it is necessary to supplement mine water, which are poor in degradable organic matter to facilitate the microbial activity. This empirical improvement and detailed knowledge of the processes involved are attributed to success of the AMD technological approach. Consequently, they are currently being proposed for application in uranium contaminated mine water. It has been found that some micro-organisms like autotrophic Fe (III) and sulphate reducing bacteria are capable of reducing uranium from soluble U(VI) to insoluble U(IV) states (Ganesh et al. 1997, Gorby & Lovley 1992, Lovley & Philips 1992). This is relatively well proven passive technology, and has already been adapted for the elimination of U and Ra from water pathway (e.g. Kunze et al. 2002, Kalin et al. 2002).

1.2 Search for applicable mechanisms and process control

When searching for solutions to the water contamination problem in abandoned uranium mines, there is need to understand specific principles and mechanism in a wetland ecosystems without artificial input of organic amendments. The knowledge would help to overcome the empirical approaches based only on effects, and to achieved a sustainable function of the nature, and to develop alternative technologies based on primary production for:

- Elimination of trace contamination of heavy elements with special biogeochemical behaviour, such as solubility differences of the element’s species under anaerobic or aerobic conditions;
- Immobilisation of soluble species of the contaminants under oxidised conditions (uranium carbonate species) and radioactive decay products ($^{226}$Ra); and
- Fixation of toxic arsenic species, or change them to chemical species with lower toxicity

In this study, a few selected examples are used to demonstrate how processes in plants and associated microbes, and in entire wetland structures can be used to eliminate low uranium concentration loading from the water pathway. The processes involve the employment of plant physiological and community functions like primary production and detrical system (White et al. 1989) for uranium immobilisation. Thus, the key role of “sun-driven” carbon assimilation and carbon sequestration is demonstrated. The current work has not considered the aeration effect and anaerobiosis on uranium speciation and immobilisation in the root zone due to the structure and physiological activity of plant community, e.g. thermosmosis (Armstrong et al. 1988), interstitial water residence time, and oxygen consumption by bacteria. This paper presents mainly the bulk ef-
fects in selected compartments, and the consequences that are important for wetland constructed or modified for uranium removal from water pathway.

2. Samples and analytical methods

The field studies were conducted in two abandoned uranium mining and milling sites of Lengenfeld and Neuensalz-Mechelgrün in the state of Saxony in Germany. Details of site location and sampling procedures in the field are described elsewhere (Dudel et al. 2001; Mkandawire et al. 2003a, 2004b). A comprehensive overview of the sites is also given by Mkandawire et al. in this book. Intact sediments cores were sampled with 6 cm PE tubes from the sedimentation area of the ponds and wetlands below the tailing dam and seepage water spring at the site Neuensalz-Mechelgrün. The sediment were sliced and handled according to Kleeberg and Dudel (1997). A few plant species, dominant in the abandoned uranium mine sites, were isolated for laboratory microcosms and microcosm investigations. Laboratory microcosms investigation used *Lemna sp.* and was set in semicontinuous culture mode as described in Mkandawire and Dudel (2002), Mkandawire et al. (2003b, 2004a). The laboratory experiments were conducted in synthetic mine tailing water based on Hutner nutrient solution but, adapted to mine water properties of the study sites. For instance, the synthetic mine tailing water contained about 10% of element (e.g. N, P) concentrations of standard Hutner nutrient. To ensure that the synthetic tailing waters had composition of uranium species like in the mine waters where uranyl carbonate \( UO_2(CO_3)^{2-} \) species dominates, chemical speciation were simulated using PhreeqC geochemical modelling package (Parkhurst and Appelo, 1999). All experiments started in mineral water without any dissolve organic matter. The samples were handled according to the German standards (DIN). All plant and sediment samples were digested, and analysed with ICP-MS as described elsewhere (Meinrath et al. 1999, Brackhage et al. 1999, Brackhage, Dudel 2002).
3. Results and discussion

3.1 Uranium fixation in photoautotrophs: special emphasis on rhizofiltration of vascular plants

3.1.1 Surface processes

The immobilisation of soluble uranium species, usually dominated by uranyl carbonates, in the surface water takes place through biosorption on the surfaces of living or dead organic matter, or through biomineralisation in which uranium co-precipitation with secondary minerals. Biosorption processes include adsorption, electrostatic attraction, precipitation, and complexation. These processes are rapid, and are completed within a few minutes e.g. maximum 30 minutes of uranium biosorption on *Lemna gibba* dry biomass (Guibal et al. 1992, Mkandawire et al. 2003). Biomineralisation include calcification processes due to photosynthetic inorganic carbon uptake.

The capacity of uranium biosorption depends on the growth increment, which increases the biomass and sorption surface, and the yield during the development of the population and the community. Under close to steady state conditions in nature, submerse and emerse floating macrophytes reach specific growth rates in the range of 0,02 and 0,35 d\(^{-1}\) (Mkandawire et al. 2003b, Vymazal 1994). Some micro- and macrophytic algae of different taxonomic groups, mainly *Characeae*, have been described to efficiently immobilise U and Ra up to some grams per kg dry matter (Franklin et al. 2000, Kalin et al. 2002, Dienemann et al. 2002). However, it was observed that close to 80% of biosorbed uranium can be removed from algae and submerse vascular plant after washing with distilled water, 0,1 M EDTA or citric acid solution (Mkandawire et al. 2003b). Unfortunately, there is no information on the distribution of adsorbed contaminant in sedimanted algae or plant detritus between the stable and permanently fixed in the course of sedimentation as well as humification, and the unstable that could be remobilised in the course of microbial mineralisation.

3.1.2 Incorporation into emerse wetland plants (helophytes)

The incorporation of uranium into a plant depends on one hand, its solubility and the influence of physicochemical conditions on its speciation in an aquatic system, and on the other hand, on the influence of plant and asso-
ciated microbes metabolic activities on milieu e.g. exudation of complex-building organic matter, oxygenation of the environment and oxygen consumption. The interaction processes and the uptake of uranium are accelerated by the flow of water from the interstitial water into the root cells and vascular systems (tracheary elements) up to the shoots and leaves into the atmosphere. The flow is usually plant mediated but also, regulated by water pressure in Sediment (Soil)-Plant-Atmosphere-Continuum (SPAC). Consequently, uranium is transported with the flow of water to above ground (emerge) plant parts, likewise inorganic ions (White, Broadley, 2003). The accumulated of U in the shoots and leaves, which is usually small fraction, depends on the plant species and the environment (Brackhage at al. 2002). The largest fractions of U accumulate in the roots of emerging plants (Fig. 1). Others studies have also observed high accumulation of other heavy metals like Pb, Cd and Cu in the roots (Brackhage et al. 1999, Schierup, Larsen 1981, St-Cyr, Crowder 1990).

![Uranium content in selected compartments of different wetland plants in the field (site Neuensalz-Mechelgrün in Saxony, East Germany). The average U concentration in milieu water in the fields is presented in Mkandawire et al. in this book. Values are mean of four repeated sampling, and n=4, and all are significant at p<0,01 with Wilcoxon signed rank.](image)

In detailed layer-wise fractionation of outer and inner root cell layers Brackhage et al. (2004) showed that the great amount of uranium is concentrated in the rhizodermis (“root wall” including the Casparian strip). The observations suggested that inside contaminants fixed on refractory ligno-cellulose and associated compounds (e.g. amino acids) are more sta-
ble immobilized than fast surface adsorbed, especially when compact root litter is fast deposited into the sediment (Lewis and Yamamoto 1990). Considering the fast fine roots turnover which is within a few weeks (Berg and McClaugherty, 2003), and the large root-shoot-ratio in helophytes and related carbon flow below ground e.g. reed (Ulrich and Burton 1985) the availability of electron acceptors for complete respiration (oxygen, nitrate) of this slow degradable or even recalcitrant organic C to CO₂ and/or transport limitation of microbial decay processes (e.g. due to sedimentation in the course of burial) should determine the mobilisation of fixed U back to the water body.

3.2 The impact of primary production

The efficiency of carbon assimilation on contaminant immobilization from the water pathway has two aspects:
- Organic matter provide surface for sorption, and for incorporation into biomass; and
- Organic carbon in form of litter form durable fixation which depend on biomass increment and quality i.e. stability against decay.

Hence, biosorption and incorporation is directly related to productivity (rate) and litter quality.

3.2.1 Growth rate and yield dependent contaminant removal

Based on the above and other prerequisites such as light limitation in submerse growth, it is expected that high productive emerse vascular plants which containing ligno-cellulose should be more useful in uranium immobilisation than submerse macrophytes. In order to quantify the efficiency we measured the growth rate and the yield of two Lemnaceae, *Lemna gibba* and *Lemna minor*, under laboratory and field conditions. Figure 2 presents the growth rates and yields of *Lemna gibba* with and without uranium influence. From the field investigations, it was found that *Lemna* sp. in the field attained mean growth rate of 0.27 and yield of 20 g m⁻² d⁻¹. Table 1 shows that *Lemna* sp are capable to removed a relevant amount of the uranium and arsenic from the water pathway under laboratory conditions characterised with nutrient limitation. Because the individual *Lemna* unit (frond = shoot and leaf, and root) can sediment or be removed from the water, the entire *Lemna* plant was considered. Field investigation revealed results within the range observed in the laboratory experiments (Mkindawire et al. 2003b). The contaminant elimination rate that depended on growth rate and yield was influenced by the temperature and resource
availability (e.g. photosynthetic active radiation and nutrients), as well as the growth inhibition due to toxicity of the contaminants.

![Graph](image)

**Fig. 2.** Influence of 1 mg U l⁻¹ and on the steady state growth rate and yield of *Lemna gibba* in laboratory culture; error bars: standard deviation. n=4

**Table 1.** *Lemna gibba* extraction potential of U and As estimated from measured productivity data (specific growth rate and yield) under optimal conditions in the laboratory. The values are mean of four replications ± standard deviation (Phyto-Extraction potential is defined in Mkandawire et al. 2004b)

<table>
<thead>
<tr>
<th>Contaminant elimination (kg ha⁻² a⁻¹)</th>
<th>Extraction potential (%)</th>
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<tr>
<td>Arsenic 751,9 ± 250</td>
<td>48,3 ± 15,1</td>
</tr>
<tr>
<td>Uranium 662,7 ± 203</td>
<td>41,4 ± 11,9</td>
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It was observed that the U had negligible effect on investigated *Lemna* species. In a separate study, the effects of increasing U or As load were examined in the laboratory. It was found that U in concentrations lower than 1000 µg l⁻¹ affects the specific growth rate and yield slightly. However, uranium toxicity to *Lemna gibba* was observed more under extremely P-limiting conditions. The effects of arsenic were relatively sharp. It was observed that under aerobic conditions, toxicity of arsenic salts to *Lemna gibba*, were related to P availability (Mkandawire et al. 2004a).
3.2.2 Screening for efficient contaminant extracting species in communities

Comparison of growth of a few helophyte species in the study sites showed that despite associated algae, emergent vegetation is the most productive. However, Veronica beccabunga L. and Cardamine amara are adapted to changing wetness. Scirpus sylvaticus and Equisetum fluviatile have typical limited productivity in comparison to the other species like Phalaris arundinacea, Glyceria fluitans and Phragmites australis. Further, investigation should be concentrate on these last three plant species, especially Phragmites australis. Typha latifolia and Phalaris arundinacea have a broad ecological valency. Consequently, they easily establish monospecies stands, and replace each other under changing environmental conditions e.g. eutrophication, natural succession, grazing pressure by muskrats (compare with Mitsch et al. 2004).

According to the literature, cattail (Typha latifolia and T. angustifolia) is more productive than common reed (Phragmites australis) under natural conditions (Kvet and Husak 1978, Grace and Wetzel 1982). However, cattail seems to suffer from nutrient deficiency under the conditions of mine water of the study sites. This suggested that under more mesotrophic conditions Typha is out-competed by Phragmites. The productivity of the reed stands of 0.8-6 kg dry matter m\(^{-2}\) a\(^{-1}\) is higher than that of duckweed and under nutrient limitation (Kvet & Husak 1978, Ondok and Kvet 1978, Ostendorp 1988, Ulrich and Burton 1985, 1988).

Because of the accumulation capacity of uranium in roots (Fig. 1) and that its litter has a higher humification potential i.e. peat building capacity (Koppisch 2001, Päivänen, Vasander 1994, Tolonen et al. 1992), common reed should be recommended for U elimination in constructed wetlands. For the other species, the knowledge on their capacity to build up recalcitrant organic carbon (peat formation) is very limited e.g. Typha sp., or is complete unknown and needs further investigation e.g. Lemna sp. Furthermore, it is unknown whether the U fixed on fine particular organic matter (FPOC) produced by the other plant species of the wetland communities sediments or remains suspended in the water. Similarly, it is not clear whether U is remobilised in the course of microbial mineralisation of organic carbon, especially under anaerobic conditions. The fate of uranium complexed on, or accumulated in organic matter in the course of organic matter deposition and sedimentation is crucial for certainty of the plant mediated U elimination from the water pathway.
Fig. 3. Distribution of uranium and organic carbon in different sediment layers of a pond below the tailing dam in Neuensalz-Mechelgrün in West-Saxony (East Germany). Data from a sediment core selected from six cores with a comparable correlation of U and organic C.

To estimate the uranium accumulation in the organic sediments, sediment cores from a shallow wetland pond below a tailing dam were investigated. The ponds are surrounded by helophyte communities dominated by cattail species, *Typha latifolia* and *Sparganium erectum* and alder, *Alnus glutinosa*. The sediments in the pond have formed in a period of about 40 years, since closure of the ore mining and processing activities in the early 60's. Figure 3 shows the relationship between uranium accumulation in the sediments and the carbon content. It was found that uranium accumulated in layers, which were rich in organic matter of sediments. However, the water pathway remains continuously loaded with U due to leaching from slug heaps, and due to flooding of the mine.

Currently, there are no evidence on the share of U that sediment together with lignocellulose and fixed during humification, or originated from microbial C mineralisation (carbon decay) which is usually accompanied by sharp decrease of redox-potential (Eh) down to negative values.
Capacity of natural attenuation of trace contaminants from uranium mine tailing waters in nature-like constructed wetlands (Abdelouas et al. 1999). The results suggested that at least for decades the flow of degradable organic carbon from the primary production of vascular plants and associated alga, and oxidation of this carbon with sulphate as electron acceptor by the sulphate reducing bacterial activities, create an efficient U sink in the sediments (Ganesh et al. 1997, Lovley & Philips 1992).

4. Conclusion and outlook

The results suggest that high productive vascular emerse plant and associated (periphytic) algae are feasible to remove U from water pathway. However, the extent (quantity) and regulation by environmental factors (flow, electron acceptors for respiration/biodegradation of organic carbon, quality of organic carbon) of remobilisation is unknown. This study showed that uranium was fixed strongly in organic sediments for some decades based on primary production.

Hence, with the knowledge acquired on U fixation in selected wetland compartments in the current study in addition to the already literature described and proven modular constructed wetland types, we propose that integrated modulate systems would be more effective in uranium removal from mine contaminated waters than a single pond or subsurface flow system (e.g. reed bed). The integrated modulate systems should have helophyte communities (e.g. reed and cattail) beds combined sequentially, and followed by sedimentation ponds. Eco-technologically, the integrated shallow sedimentation ponds could be regular cleaned, or let the primary produced organic carbon be mineralised without re-mobilisation of fixed uranium.

The concrete morphometric and hydrologic design of such a constructed wetland depends mainly on the water chemistry and the contaminant load. However, both water chemistry changes and hydro-morphologic design need gradual experimental tests and improvements in the medium scale (mesocosm experiments) and larger scale (pilot plant). Furthermore it should be considered that site (habitat) adapted species or ecotypes of dominant habitat structuring species, e.g. common reed (Phragmites australis) clones selected and tested before plantation. To start with a mixture of genotypes and species and a set of different hydrological conditions like water depth and residence time in a complex integrated wetland to be capable to accommodate “self-adjustment capacity”. This needs not so much time as self-organisation of natural communities, however a large area.
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