



Removal of toxic metals from sewage sludge by EDTA and hydrodynamic cavitation and use of the sludge as fertilizer

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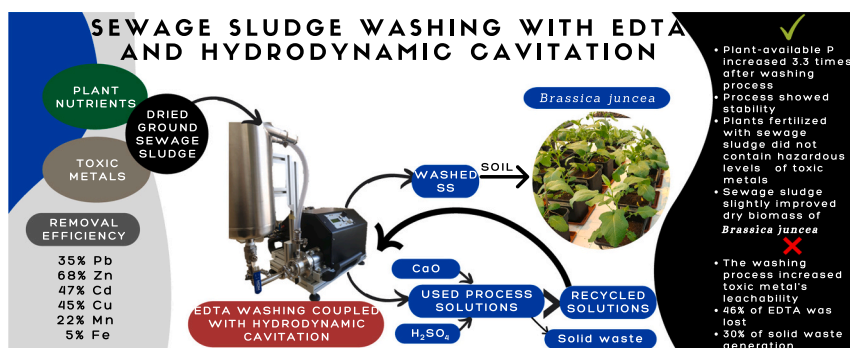
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HIGHLIGHTS

- EDTA washing & hydraulic cavitation removed toxic metals from sewage sludge.
- EDTA and process water were recycled in a closed process loop.
- The nutrient pool of P, N and K was preserved in the washed sewage sludge.
- Washed sewage sludge was tested as soil fertilizer.
- The application of washed sludge to the soil posed no risk to the environment.

GRAPHICAL ABSTRACT



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ABSTRACT

Sewage sludge (SS) is rich in plant nutrients, including P, N, and organic C, but often contains toxic metals (TMs), which hinders its potential use in agriculture. The efficiency of removal of TMs by washing with ethylenediamine tetraacetate (EDTA), in combination with hydrodynamic cavitation (HC) and the usability of washed sewage sludge as fertilizer were investigated. The environmental risk was assessed. During 8 wash batches an average 35, 68, 47 and 45 % of Pb, Zn, Cd and Cu, respectively, as well as 22 and 5 % Mn and Fe were removed from the SS. The process solutions and EDTA were recycled at a pH gradient of 12.5–2, which was achieved by adding quicklime (CaO) and then acidification by H₂SO₄, so that no wastewater was produced, only solid waste (ReSoil® method). The quality of the recycled process solutions (they remained unsaturated with salts) and the efficiency of the washing process were maintained across all batches. On average, 46 % of the EDTA was lost during the process and was replenished. The initial leachability of EDTA-mobilized Pb, Zn, Cu, Cr and Fe remaining in the washed SS increased 6-, 17-, 3-, 11- and 11-fold, respectively, but not to hazardous levels except for Zn. After washing, P and K remained in the SS, plant-available P increased 3.3-fold, while total N and C were reduced by 20.28 and 2.44 %, respectively. Washed SS was used as fertilizer in the pot experiment. The yield of *Brassica*

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juncea did not improve, the uptake of TMs by the plants and the leaching of TMs from the soil were minimal. Our study highlighted the drawbacks and potential feasibility of the new SS washing method.

1. Introduction

The escalating global volume of sewage sludge (SS) underscores the critical need for its sustainable management (Przydatek and Wota, 2020). The high content of organic matter, macro- and micronutrients in SS makes it a viable resource for use as a fertilizer (Sabia et al., 2021). Phosphorus, the main nutrient in SS, is present in the form of a combination of inorganic P bound with Al, Fe or Ca and organic P (Houben et al., 2019). The P content in SS ranges from 1 % to 5 % in dry matter (Liu et al., 2019). Soil erosion has resulted in P deficiency in agricultural soils, particularly in tropical and subtropical regions. Moreover, the remaining accessible reserves of phosphate rock are projected to be depleted within 50 years if the demand for fertilizers continues to grow at a rate of 3 % per year (Jing et al., 2019). However, the presence of toxic metals (TMs) including Zn, Ni, Cu, Pb, Co, Cd, Hg, and Cr (Olujimi et al., 2012) hinders the use of SS as fertilizer in agriculture (Towers and Paterson, 1997). In addition, the disposal of untreated SS in soil is prohibited in most EU countries (Kelessidis and Stasinakis, 2012).

Anaerobic digestion and composting are common methods of treating SS that involve mineralization of organic matter and, in the case of composting, stabilization of TMs through complexation with humic substances (Ingelmo et al., 2012; Hoang et al., 2022; Yaser et al., 2022). Chemical immobilization is an effective alternative to biological stabilization when an affordable and easy-to-use means of passivating TMs is available. Common additives include lime, aluminosilicate, phosphorus-containing materials and sulfides (Zhang et al., 2017). However, stabilization and immobilization processes do not reduce the TMs concentration. In addition, soil properties such as pH, redox potential and cation exchange capacity can affect the mobility of TMs when SS is used in agriculture (Zhang et al., 2017).

In recent years, thermal processing of SS has gained importance. Incineration and pyrolysis of SS offer advantages such as volume reduction, odor removal and destruction of organic compounds (Chanaka Udayanga et al., 2018). Nevertheless, the decomposition of the organic fraction of SS leads to an increase in the concentration of TMs (Samolada and Zabaniotou, 2014) and high temperatures to the loss of nutrients (Sichler et al., 2022).

Chemical washing of SS to remove TMs using inorganic acids (H_2SO_4 , H_3PO_4 , HNO_3), organic acids (citric acid, oxalic acid) and chelators (e.g. ethylenediamine tetraacetate, EDTA; tetrasodium glutamate, GLDA) have been extensively studied (Babel and del Mundo Dacera, 2006; Gaber et al., 2011; Li et al., 2017; Shen et al., 2001; Suanon et al., 2016; Zaleckas et al., 2009; Kaurin et al., 2022). The use of organic and inorganic acids has been reported to lead to P losses that occur during the dissolution and leaching of TMs (Camargo et al., 2016; Pathak et al., 2009; Strasser et al., 1995). Kou et al. (2020) found that the combination of EDTA with citric acid, glutamic acid or aspartic acid removed the TMs from the SS while retaining the nutrients.

During chemical washing, both acid dissolution and chelation mobilize TMs, which are then transferred from the SS into the process solutions (i.e., the washing solution). There is a knowledge gap in the efficient treatment or better recycling of contaminated process solutions prior to their safe disposal or reuse. The separation of solid SS and liquid phase with filter presses and decanter centrifuges only results in partially dewatered SS (Zhang et al., 2022a, 2022b). The leaching of acid/chelate-mobilized TMs remaining in the liquid phase of the dewatered SS poses a potential environmental threat. There is a lack of research on the scalability, economic and environmental sustainability of chemical SS washing and the efficiency of the treated SS as a soil fertilizer (Geng et al., 2020).

In our previous study (Morales Arteaga et al., 2022), a process for

closed-loop recycling of EDTA and process solutions (ReSoil®), originally developed for soil remediation (Lestan, 2017), was adapted for the washing of SS. The method uses low-cost quicklime (CaO) and H_2SO_4 , which create a strong pH gradient and recover EDTA and process solutions for further use. The method produces only solid waste and no wastewater.

To improve the removal of TMs, SS washing with chelators can be supported by chemical and physical SS (pre)treatment. For example, washing SS with EDTA solution removed 61, 24, 4, 31, and 12 % of Pb, Zn, Cu, Mn, and Fe, respectively. Cr was not removed. The integrated EDTA wash and microwave-assisted acid hydrolysis improved the removal of Pb, Zn, Mn, and Fe by 1.28, 3.25, 2.16 and 6.08 times, respectively, and enabled the removal of 17 % of Cr. However, the process led to a depletion of nutrients. Specifically, 70 % of P, 23 % of N, and 92 % of plant-available K were lost (Morales Arteaga et al., 2023).

In the current study, we combined EDTA washing with hydrodynamic cavitation (HC). In hydrodynamic cavitation, vapor bubbles are formed and collapsed in a liquid, releasing considerable energy in a localized area. This generates mechanical forces, high temperatures, microjets, shear forces and shock waves (Benito et al., 2005). TMs are primarily bound to organic matter and residual fractions (Xu et al., 2022; Zhang et al., 2022a, 2022b), and our previous research has shown that HC effectively reduces TMs concentration by oxidizing organic matter (Repinc et al., 2022).

We hypothesized that the combination of EDTA chelation and HC increases the efficiency of TMs removal from SS while preserving the pool of (available) nutrients (N, P, K). The feasibility of the novel combined approach was tested by several consecutive SS wash batches at pilot scale, using ReSoil® “liferesoil.envit.si” to recover EDTA and process solutions. Further objectives were to evaluate the feasibility of washed SS as a soil fertilizer in a pot trial and to assess the risk of leaching of TMs and plant uptake.

2. Materials and methods

2.1. Chemical analyses of soil and SS

SS from a Slovenian wastewater treatment plant (WWTP) was air-dried at 40 °C to constant weight and then ground to a particle size of <2 mm.

The pH value and the electrical conductivity (EC) of the SS were measured in a suspension with deionized water in a ratio of 1:10 (w:V) (DIN 38414-S4, 1984). Soil pH was measured in an aqueous $CaCl_2$ suspension (ISO 10390, 2005). In both soil and SS, the organic C content, total C content and total N content were measured with the elemental analyzer (Vario MAX CNS, Elementar Analysen Systeme GmbH, Hanau, Germany) following a process of dry incineration. The total P content of the digestate was measured after *aqua regia* extraction using a microwave oven (Mars Xpress, MDS-2000, CM GmbH, Kamp-Lintfort, Germany). After extraction, the resulting digestate underwent filtration using a 0.45 μm cellulose acetate membrane. Subsequently, the filtered digestate was diluted with deionized water (ISO 54321, 2020) and measured by inductively coupled plasma optical emission spectroscopy (ICP-OES) (Agilent 5900, Agilent Technologies, Inc., Santa Clara, CA). The analysis accuracy of trace elements was assessed by employing certified reference material, LGC6181 sewage sludge from LGC Standards (UK).

Plant-available P and K in soil and SS were extracted using ammonium lactate solution. The samples were shaken, filtered, and subsequently measured by ICP-OES, while carbonates were determined volumetrically (ÖNORM L1087, 1993; SIST EN 15936, 2012; SIST EN

16168, 2012).

2.2. Preliminary washing of SS in small-scale experiments

In a first step, laboratory-scale experiments were performed to optimize the concentration of reagents (EDTA ($C_{10}H_{12}CaN_2Na_2O_8 \cdot xH_2O$) with a molar weight of $374,27 \text{ g mol}^{-1}$, H_2SO_4 , oxalic acid, dithionite, citric acid) needed in the washing solution (WS), as well as the extraction time and solid-liquid ratio for washing SS without the HC process. For the experiments, 5 g of SS dry weight were washed in an overhead rotator at a speed of 20 rpm (rpm). The SS thus washed was separated by centrifugation at $3430 \times g$ for 10 min and then rinsed three times with tap water at the same solid:liquid ratio (1:7, w:V) to remove mobile EDTA and TMs from the SS. After rinsing, the washed SS was dried at $105 \text{ }^\circ\text{C}$, ground in an agate mortar, sieved through a $250\text{-}\mu\text{m}$ sieve, and then stored for TMs removal efficiency analysis.

2.3. SS washing by coupled HC and EDTA chelation on a pilot scale

The HC used in the experiments was both a cavitator and a pump. Its stator-rotor configuration resembles a Venturi channel, which creates

favourable hydrodynamic conditions for cavitation. This is achieved by positioning the rotor teeth at an angle to the stator teeth, as explained in Fig. S1 in Supplementary material.

To optimize the time of HC treatment, 429 g of dried and ground SS was mixed in a 5 L plastic beaker with a 3 L washing solution (WS) containing 50 mmol L^{-1} EDTA and 50 mmol L^{-1} citric acid. The slurry was then slowly added to the HC working at approximately 6000 rpm. Once all the slurry was in the HC, the speed of the HC was increased to 10,000 rpm and the duration of cavitation (10, 20, 30, 40, 50 and 60 min) was tested. After the cavitation process, the slurry was centrifuged in 500-mL centrifuge bottles (10 min at $3430 \times g$) and rinsed three times with tap water (solid:liquid ratio 1:5 (w:V)). A homogeneous sample from the washed SS was dried at $105 \text{ }^\circ\text{C}$ for 24 h, ground, sieved through a $250\text{-}\mu\text{m}$ sieve, and stored for TMs removal analysis.

For pilot-scale coupled HC and EDTA washing of SS in a closed-loop system, 429 g of dry SS in each batch was suspended in WS containing 50 mmol L^{-1} EDTA and 50 mmol L^{-1} citric acid at a solid:liquid ratio of 1:7 (w:V). EDTA, WS and process solutions were recycled using the modified ReSoil® method (see Fig. 1). In the first step, the suspension was cavitated at 10,000 rpm for 30 min. In step 2, the washed SS was separated from the used WS (uWS) by centrifugation (10 min at 3430

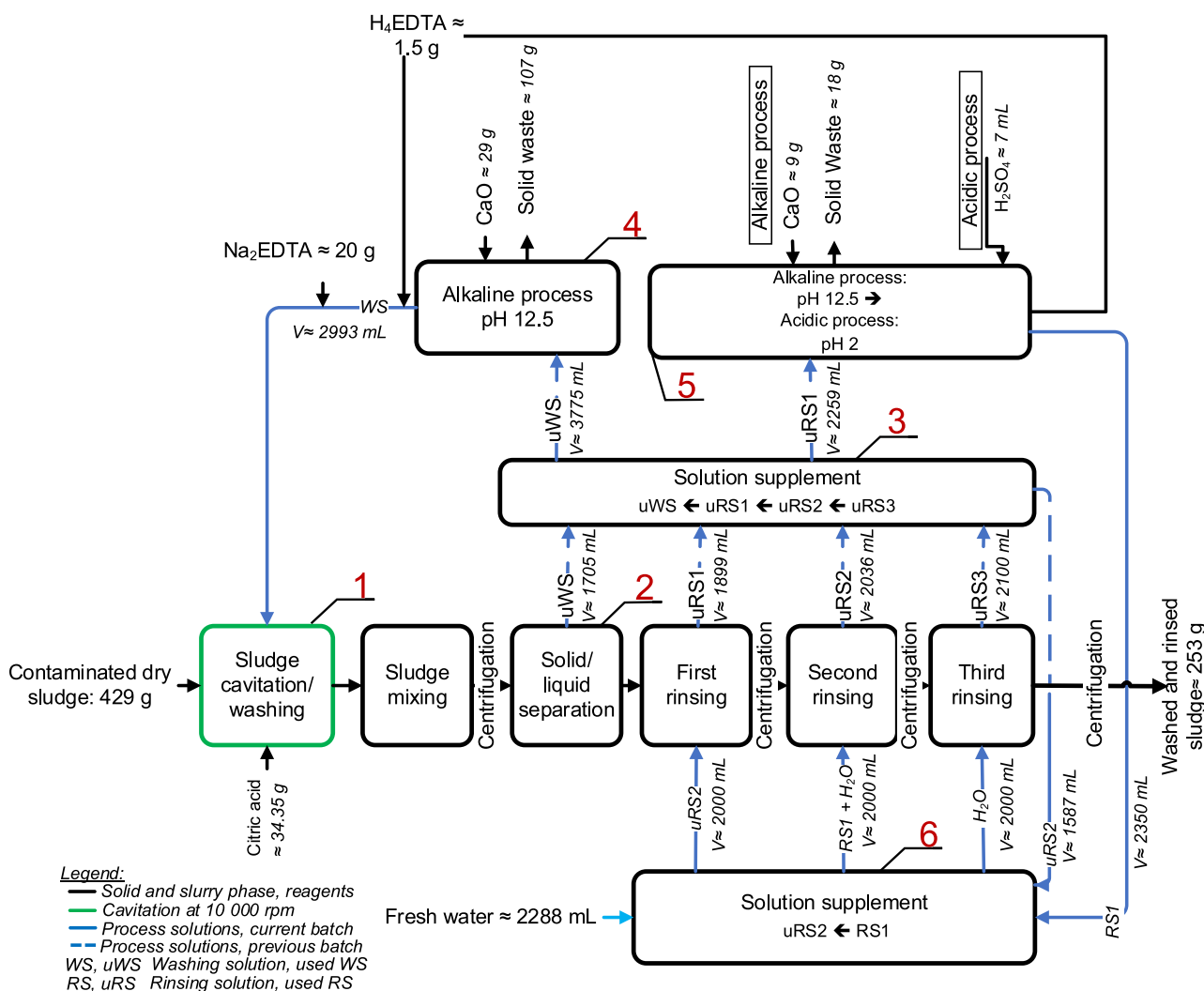


Fig. 1. The process flow diagram with material balance illustrates the following steps: (1) EDTA coupled with hydrodynamic cavitation washing of the sewage sludge (MWF), (2) separation of solid and liquid phase and rinsing of the MWF, (3) solution replenishment for water losses, (4) alkalization of used washing solution (uWS), (5) alkalization or Acidification of the first used rinsing solution (uRS1), (6) Upstream supplementation of the process solutions with fresh water to achieve the desired final volume. The flow of solutions is represented by blue lines, dashed blue lines indicate the flow of solutions from the previous batch, and black lines represent the flow of solids.

×g). Subsequently, the SS in the first batch was rinsed three times with tap water (solid:liquid ratio 1:5 (w:V)) by vortex mixing for 10 s in 500-mL centrifuge bottles and then separated by centrifugation (10 min at 3430 ×g). Subsequent washed SS batches were rinsed three times under the same conditions as the first batch, but with two rinsing solutions (RS) obtained from the previous batch in the following order: second used RS (uRS2) and first treated RS (RS1). For the third rinsing step, additional tap water was added to the system. The uRS1, uRS2 and uRS3 were separated from the washed SS by centrifugation.

During the process, some water was lost due to the treatment of the process solutions with CaO and hydration to Ca(OH)₂ (see below) and the high temperature (61 °C) during HC. To maintain a solid:liquid ratio of 1:7 (w:V) in a series of batches, uWS was supplemented with uRS1, uRS1 with uRS2 and uRS2 with uRS3 and tap water (Fig. 1, step 3).

As shown in Fig. 1, uRS2 was used untreated for the next batch due to the low concentration of TMs and EDTA. CaO was added to the uWS to raise the pH to 12.5 and precipitate the TMs as hydroxides, which were removed by centrifugation with excess hydrated lime (Ca(OH)₂) (Fig. 1, steps 4 and 5). EDTA in WS was recovered as Ca-EDTA. uRS1 was alkalized to pH 12.5 to remove TMs and then acidified with H₂SO₄ to pH 2 to recycle the remaining EDTA in the solid acid form (H₄EDTA). H₄EDTA was recovered by centrifugation and recycled back to WS with fresh disodium ethylenediaminetetraacetate dihydrate (Na₂EDTA) C₁₀H₁₄N₂Na₂O₈•2H₂O, molar weight: 372,24 g mol⁻¹ to replenish the portion of EDTA lost during the process. Step 6 shows the replenishment of WS and RS due to water loss as described above. The washed SS was dried to constant weight at 40 °C, ground to pass through a 2-mm sieve, and stored cold for the pot experiment and further analysis.

2.4. Pot experiment

10 kg of soil with 11 % moisture, 1137 mg kg⁻¹ of total P, 7078 mg kg⁻¹ K and an N concentration of 0.2 % were amended with dried, ground original (unwashed) and washed SS at different doses to a final amount of 26, 52, 104 and 208 kg ha⁻¹ of total P, respectively. Mineral fertilizers 5Ca(NO₃)₂•NH₄NO₃•10H₂O, and K₂SO₄ were also used to balance the total concentration of N and K in the soil with the P dosage (due to the lack of N and K in SS). In the control treatment (NPK), (NH₄)₂PO₄ was used along with the aforementioned mineral fertilizers. The amounts of added SS and mineral fertilizers are listed in Table S1. Non-fertilized soil was used as a negative control. Each treatment was repeated six times.

The fertilized and unfertilized soils were placed in 10-liter plastic pots and kept in a greenhouse. Meanwhile, the seeds of *Brassica juncea*, var. Scala, were sown in a seedling tray on February 22, 2023. The seedlings were planted in the pots on March 29, 2023 and the pots were distributed randomly (random block design) in the greenhouse. The plants were watered regularly every 3 to 4 days at 9 am to keep the humidity at about 75 % of the maximum water holding capacity.

Before the flowering stage on April 25, 2023, the plants' physiological status and fitness were evaluated conducting gas-exchange and chlorophyll fluorescence measurements. Gas-exchange measurements were taken from 9 till 12 am with LI-6400 XT portable photosynthesis system (LI – COR, Lincoln, USA) on the middle, fully developed leaf the plant (4th leaf from the top; one leaf per plant) at a constant reference CO₂ concentration (400 μmol mol⁻¹) and photon flux density (PFD; 1500 μmol m⁻² s⁻¹). Temperature (T) and water vapor deficit (VPD) were controlled at the ambient level targeting to average for measuring time respecting daily weather conditions which was 20° for T and 0.9 kPa for VPD). Net photosynthesis (A), transpiration (E), stomatal conductance (g_s), intercellular leaf CO₂ concentration (C_i) and effective photochemical efficiency (F_v'/f_m'; yield) were recorded when steady state conditions were reached. Photosynthetic water use efficiency (WUE = A/E; μmol CO₂ mmol H₂O⁻¹) was derived from gas exchange measurements.

Potential photochemical efficiency (F_v/F_m) and the electron

transport rate (ETR) of Photosystem II were measured with a portable pulse amplitude modulated chlorophyll fluorometer (PAM 2500; Walz, Effeltrich, Germany). After a 30 min dark acclimation fluorescence was excited with a saturating irradiance of a 'white light' pulse (PPFD; 8000 μmol m⁻² s⁻¹; 0.8 s). On the same leaves the chlorophyll content was determined by Soil Plant Analysis Development (SPAD) meter (Konica-Minolta Sensing Inc., Osaka, Japan).

After these measurements were completed, the plants were harvested. The shoots were washed under running tap water to remove dirt and cut into small pieces. They were then weighed and dried at 70 °C for approximately 72 h until the weight stabilized, allowing the measurement of the dry biomass. After harvesting the plants, the pots were soaked in water and the leachates were collected to measure TMs.

2.5. Leachability of TMs

The leachability of TMs from SS was determined according to (DIN 38414-S4, 1984). This test has been reported to give reliable, consistent results (Kasselman and Snyman, 2004). In brief, SS was mixed with deionized water at a ratio of 1:10 in an overhead rotator for 24 h, the liquid phase – the leachate – was separated by centrifugation and analyzed for TMs as described below.

2.6. Determination of TMs

In laboratory-scale experiments, TMs in SS were determined using an X-ray fluorescence spectrophotometer (XRF, Delta DS -4000, Olympus) after drying, grinding and sieving of SS up to <250 μm. The limit of quantification (LQ) of Pb, Zn, Cd, Cu, Cr, Mn, and Fe was 6, 5, 10, 10, 50, 10, and 50 mg kg⁻¹, respectively.

The TMs content in the soil, in the plants and in the SS was determined after dissolution in *aqua regia*, using the same procedure as for the analysis of P and K (see above). The filtrate was analyzed by ICP-OES (Agilent 5900, Agilent Technologies, Inc., Santa Clara, CA). The LQs for Pb, Zn, Cd, Cu, Cr, Mn and Fe were 0.2, 0.02, 0.02, 0.1, 0.1, 0.05 and 2 mg L⁻¹, respectively. The TMs in the process solutions were measured directly by ICP-OES.

Low Cd concentrations in SS, plants and process solutions were measured with graphite cuvette AAS (GF-AAS 240Z, Agilent Technologies, Santa Clara, CA, USA). The LQ for Cd was 0.01 μg L⁻¹.

2.7. Determination of EDTA

The concentration of EDTA in the process solutions was determined spectrophotometrically according to the method of Wang et al. (2013). The LQ for EDTA was 0.15 mmol L⁻¹.

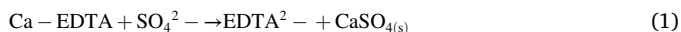
2.8. Statistical analysis

Statistical analyses were performed to evaluate the differences between treatments using ANOVA, Welch's two independent sample *t*-test, Kruskal-Wallis, and Dunn's test with Bonferroni correction based on data distribution characteristics. The R Studio program was used to perform all statistical analyses (R Core Team, 2020).

3. Results and discussion

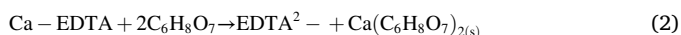
SS contained 61, 1166, 0.62, 408, 216, 282, 47,056 mg kg⁻¹ Pb, Zn, Cd, Cu, Cr, Mn and Fe, respectively, and had a moisture content of about 72 %. According to the concentration, Zn was the main contaminant. However, according to Slovenian legislation (Ur. l. RS, št.62), Cu and Cr exceeded the established safe limits for toxic metals when using sewage sludge as fertilizer by 1.36 and 1.08 times, respectively. In the first set of SS washing experiments with EDTA, we evaluated the use of auxiliary reagents, optimised the wash time and the solid to liquid ratio on a small laboratory scale. EDTA was used as Ca-EDTA salt, which is the main

form of chelator recycled from the ReSoil® process (Lestan, 2017). Washing with 50 mmol L⁻¹ EDTA for 1 h at a solid:liquid ratio of 1:7 (w:V) removed 9, 39, 16, 4, 23, and 1 % of Pb, Zn, Cu, Cr, Mn, and Fe, respectively, from SS (treatment No. 1 in Table S2). The Cd concentration in SS was below the LQ of XRF in all laboratory-scale experiments. Increasing the EDTA concentration to 100 mmol L⁻¹ increased the removal of Pb and Cu by 1.8- and 1.4-fold, respectively. The removal of Zn, Cr, Mn and Fe did not change significantly (treatment No. 2). The chelation of TMs with Ca-EDTA is kinetically hampered. In our previous study, we introduced activation of the chelator. By adding H₂SO₄ and polycarboxylic acids, Ca was captured out of EDTA and insoluble Ca salts were formed (Eq. (1), Morales Arteaga et al., 2021). Activation of EDTA with 50 mmol L⁻¹ H₂SO₄ in treatment No. 3 improved the removal of Pb, Zn, Cu, Mn and Fe by 2.8-, 1.3-, 1.7-, 1.4- and 12-fold compared to treatment No. 1.



Fe is regularly added to improve coagulation and dewatering in wastewater treatment plants (Wang et al., 2022). Coagulation mechanisms include the formation of Fe oxides and hydroxides and the colloid entrapment. A secondary reaction is the binding of TMs to Fe oxides and hydroxides (Lack et al., 2002). The TMs were released by reductive dissolution of Fe oxides and hydroxides after addition of the strong reducing agent Na-dithionite to the SS washing solution (WS). Washing conditions: 25 mmol L⁻¹ Na-dithionite, 50 mmol L⁻¹ EDTA, 50 mmol L⁻¹ H₂SO₄, solid:liquid ratio 1:7 (w:V) (treatment No. 4) increased Fe removal by 1.6-fold, while the removal of Pb, Zn, Mn and Cr did not change significantly compared to treatment No. 3.

In treatment No. 5, oxalic and citric acid were used to activate Ca-EDTA and as additional chelators. Washing with 50 mmol L⁻¹ EDTA and 50 mmol L⁻¹ oxalic acid for 1 h and at a solid:liquid ratio of 1:7 (w:V) removed 24, 53, 24, 33 and 12 % of Pb, Zn, Cu, Mn and Fe, respectively. This was not a significant improvement compared to treatment No. 3 where H₂SO₄ was used to activate the chelator. Ma et al. (2020) reported that citric acid in combination with electroosmosis significantly improved the removal of TMs from SS. Washing with 50 mmol L⁻¹ EDTA, 50 mmol L⁻¹ citric acid for 1 h at a solid:liquid ratio of 1:7 (w:V) removed 35, 58, 27, 36 and 12 % of Pb, Zn, Cu, Mn and Fe, respectively (treatment No. 6). Oxalic and citric acids presumably activated Ca-EDTA in a reaction similar to reaction by H₂SO₄ (Eq. (1)), capturing Ca²⁺ from EDTA by chelation of Ca on carboxyl groups and forming sparingly soluble Ca oxalate/citrate, which was removed from the wash system by precipitation (Eq. (2)).



The double concentration of EDTA and citric acid was also tested. Washing of SS with 100 mmol L⁻¹ EDTA and 100 mmol L⁻¹ citric acid, solid:liquid ratio 1:7 (w:V) for 1 h did not result in a significant increase in TMs removal efficiency (treatment No. 7). Washing with citric acid as the sole chelating agent (50 mmol L⁻¹ citric acid, solid:liquid ratio 1:7 (w:V) for 1 h) removed only 3, 7 and 6 % of the Zn, Mn and Fe, respectively, while Pb and Cu were not removed (treatment No. 8). Washing the SS with only 50 mmol L⁻¹ EDTA in the following combinations: (i) 25 mmol L⁻¹ oxalic acid and citric acid (treatment No. 9), (ii) 25 mmol L⁻¹ oxalic acid and H₂SO₄ (treatment No. 10) and (iii) 25 mmol L⁻¹ citric acid and H₂SO₄ (treatment No. 11) did not significantly improve the efficiency of TMs removal compared to treatment No. 6 with 50 mmol L⁻¹ EDTA and citric acid. SS washing with 50 mmol L⁻¹ EDTA and citric acid and increasing the wash time to 16 h improved the

removal efficiency for Zn, Cu, Mn and Fe by 1.15-, 1.22-, 1.19- and 1.92-fold, respectively (treatment No. 12). However, a long washing time may not be a viable option in large-scale operation. Overall, the SS wash with 50 mmol L⁻¹ EDTA and 50 mmol L⁻¹ citric acid at a solid:liquid ratio of 1:7 (w:V) for 1 h (treatment No. 6) was the most favourable and was selected for further pilot-scale trials.

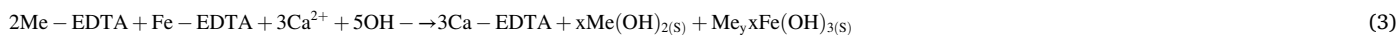
No or very little Cr was removed from the SS by small-scale treatments. The Cr was probably present in the SS as Cr³⁺, which is due to reduction by excess Fe. EDTA chelation of trivalent cations is generally much slower than the almost instantaneous chelation of divalent metal ions. For Cr³⁺, the main reason for the slow chelation kinetics (and the lack of removal from the SS) is the constant of water displacement from the inner sphere of the highly hydrated Cr(H₂O)₆³⁺, which is very low at 5.8 × 10⁻⁷ s⁻¹. Similarly, the constant of water displacement is faster for chelation of divalent Co²⁺ with a rate constant of ~10⁶ s⁻¹, while the corresponding value for trivalent Co³⁺ is ~10¹ s⁻¹ (Byegaard et al., 1999).

3.1. SS washing tests on a pilot scale

In pilot-scale experiments, the EDTA wash (50 mmol L⁻¹ EDTA, 50 mmol L⁻¹ citric acid in an solid:liquid ratio of 1:7 (w:V) for 1 h) was coupled with HC. First, the time of cavitation: 0-, 20-, 30-, 40-, 50- and 60-min was tested. As shown in Table S3, the TMs removal from SS increased with the time of HC and reached a plateau after 40 min. However, the temperature of the cavitated suspension also increased rapidly (Table S3). To prevent damage to the HC apparatus due to operation at high temperature, we limited the cavitation time to 30 min, after which 42, 68, 54, 37, and 17 % of Pb, Zn, Cu, Mn, and Fe were removed from SS, respectively (Table S3). This corresponds to 1.2-, 1.2-, 2.0- and 1.4-fold more efficient removal of Pb, Zn, Cu and Fe, respectively, compared to the small-scale treatment without cavitation (treatment No. 6, Table S2). The results indicate a modest positive effect of HC on the removal of Pb and Zn and a significant effect on the removal of Cu. The Cd concentration was below the LQ of the XRF in all samples.

The removal of Cr from SS was not observed during laboratory-scale washing (Table S2). Interestingly, the concentration of Cr in washed SS increased after the combination of EDTA washing and HC (negative removal efficiency in Table S3). Sequential extraction of SS showed that Cr is strongly bound to the organic matter and residual fraction of SS (Repinc et al., 2022). In addition, Hao et al. (2022) showed that oxygen-containing carboxyl, hydroxyl and phosphate functional groups coordinate with Cr to form polymers. Most of the Cr-enriched polymerized material presumably resisted fragmentation by HC, while less polymerized SS fragments with low Cr concentration were lost after rinsing three times and solid-liquid separation (Fig. 1). This resulted in a significant mass loss of SS in a series of 8 wash batches, on average 41 % (mass balance in Fig. 1). The data in Table S3 show that the Cr concentration in the washed SS increased with the duration of cavitation.

The feasibility of the novel SS wash method was further tested in a series of 8 wash batches with recycling of EDTA and process solutions in the ReSoil® process (Fig. 1). The used washing and rinsing solutions (uWS, uRS) were treated in a pH gradient. The addition of CaO alkalinized pH of the process solutions to pH > 12. This high pH destabilized Me-EDTA (Me = TMs, Mn), while Fe precipitated from Fe-EDTA at pH > 9 as hydroxide. The Me was replaced by Ca, and the EDTA was recycled as Ca-EDTA. The Me precipitated as hydroxides and co-precipitated with highly absorbing Fe hydroxide. The (co-)precipitation shifts the equilibrium of the displacement and precipitation reaction (Eq. (3)) to the right (Morales Arteaga et al., 2022):



The mass balance shown in Fig. 1 shows that most of the EDTA was recycled by the alkaline process. Addition of citric acid activated Ca-EDTA in WS, as shown in Eq. (2). The remainder of the EDTA precipitated from the process solutions in acidic form after RS1 (Fig. 1) was acidified with H_2SO_4 to a pH of 2 (Eq. (4)). Excess Ca^{2+} from the alkaline phase and SO_4^{2-} from the acidic phase of ReSoil® was removed as insoluble gypsum (Eq. (5)). This prevented the saturation of process solutions (Lestan, 2017).



Consistent quality of the process solutions is crucial for a functional closed-loop process (Fig. 1). As shown in Fig. 2, the pH, conductivity and Na^+ concentration in uWS, uRS1 and uRS2 fluctuated without an upward trend. Na^+ (the amount of Na_2EDTA addition is indicated in Fig. 1) presumably left the system by adsorption at the SS solid phase. There was no increasing trend in the concentrations of Pb, Zn, Mn and Fe. The Cu concentration reached a plateau after the fourth batch (Fig. 2). The Cr concentration in uWS was very low and fluctuated in a U-shaped time axis over eight batches.

The treated process solutions were reused in the next SS wash batch (Fig. 1). Pb, Zn, Cd and Fe were efficiently removed: their concentrations in WS and RS1 were significantly lower compared to uWS and uRS1 (Figs. 2, 3). The Cu concentration was comparable in the used and treated solutions. The increasing trend in concentration indicates that the equilibrium between Cu displacement from Cu-EDTA and precipitation as Cu hydroxide was at a higher concentration than was achieved

in the series of eight batches. Nevertheless, Cu was removed from SS at a constant rate in all series of batches (Table 1). The concentrations of Cr and Mn in the treated solutions were below the LQ, as were the concentrations of TMs in RS2 (Fig. 3). The concentrations of EDTA, Na^+ and conductivity in the treated solutions were stable over series of batches. The EDTA concentration in WS was lower than the original 50 mmol L^{-1} . The mass balance shown in Fig. 1 showed an average EDTA loss of 46 %. The lost EDTA was probably adsorbed onto the solid phases of the washed SS. The lost chelator was replenished by adding Na_2EDTA at the beginning of each batch together with H_4 -EDTA recycled from the acidic phase of ReSoil® (Eq. (4)). The material balance of SS, EDTA, solid waste (hydrated lime mixed with metal hydroxides, Eq. (3)) and other materials used in ReSoil® is shown in Fig. 1. On average, 0.3 g of solid waste was generated per g of SS treated. In general, the properties of the process solutions indicate the stability and robustness of the SS washing process.

The removal efficiency of Pb, Zn, Cd and Cu from SS was quite constant in eight batches, averaging 35, 68, 47 and 45 %, respectively (Table 1). The removal of Mn and Fe was lower and varied between 12–29 % and 0–10 %, respectively. Washed SS contained 40, 365, 0.32, 219, 262, 216, 45,079 mg kg^{-1} of Pb, Zn, Cd, Cu, Cr, Mn and Fe, respectively (Table S4). The Cr accumulated in the washed SS, presumably for the reasons mentioned above.

SS is a hygroscopic, gelatinous material. After solid-liquid separation by centrifugation, a large proportion of the TMs enriched WS remained entrenched in the washed SS with a moisture content of approx. 70 %. To remove the TMs, the washed SS was rinsed three times with (recycled) RS (Fig. 1). The cleanliness of the RS was important for efficient rinsing of the washed SS. The TMs concentration in RS1 (Fig. 3) was significantly lower compared to the TMs concentration in uWS (Fig. 2); the TMs concentration in RS2 was below the LQ.

Despite rinsing, the leachability of Pb, Zn, Cu, Cr and Fe from washed SS remained 6-, 17-, 3-, 11- and 11-fold higher compared to the original SS (Table 2). No statistical differences were found for Mn. No or only a minimal Cd concentration was detected in the leachates of original and washed SS. Despite the increased leachability, the Pb, Cd, Cu and Cr concentrations remained 12.5, 9, 2 and 1.9 times below the hazardous limit values (DIN 38414-S4, Council Decision 2003/33/EC, Table 2). However, the leachability of Zn from washed SS exceeded the hazardous limit by a factor of 2.2. In a small-scale laboratory experiment, Kou et al. (2020) washed the SS with EDTA and citric acid and reported that the leachability of Cr, Cd and Cu decreased after the washing process. A more efficient solid-liquid separation was probably possible on a small scale than on the pilot scale used here.

3.2. Properties of washed SS as fertilizer

On a global scale, SS contains a much higher P content compared to any other type of biomass (Cordell et al., 2011). As shown in Table 3 the total P concentration in washed SS did not change. Moreover, plant-available P in washed SS, measured as P_2O_5 , increased by a factor of 3.3. Kou et al. (2020) reported a decrease in total P after washing SS with EDTA and citric acid-SS. Similarly, we reported a depletion of P after combined washing of SS with EDTA and acid digestion (Morales Arteaga et al., 2023).

For the other nutrients, washing with SS slightly reduced the total N content by 1.25-fold (Table 3). The total K content was not affected, while the concentration of plant-available K (measured as K_2O) was reduced by 60 %. Presumably, the high Na^+ concentration in WS and RS (Figs. 2 and 3) exchanged and washed K^+ from the SS solid phase. The C concentration was not affected by washing. Of the other properties, pH and electrical conductivity (EC) decreased slightly in the washed SS, while $CaCO_3$ content remained at the same level (Table 3). Kou et al. (2020) reported that washing with EDTA and citric acid had no effect on the N concentration in SS.

Washed SS was tested as a soil fertilizer in a pot experiment with

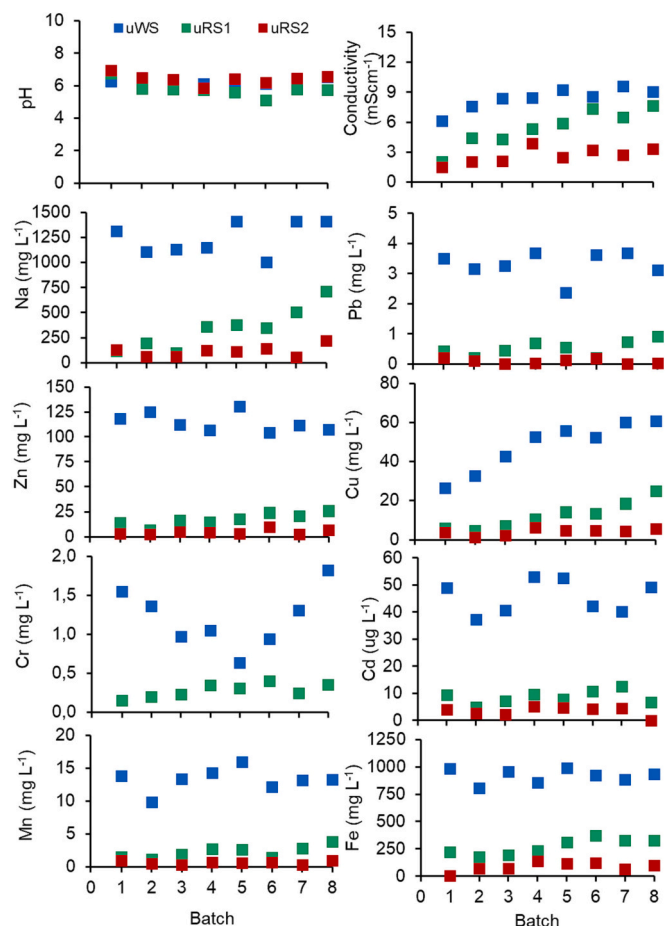


Fig. 2. pH, conductivity, Na and metal concentrations of the used washing solution and the used first and second rinsing solutions (uWS, uRS1 and uRS2 respectively) over the 8 consecutive wash batches.

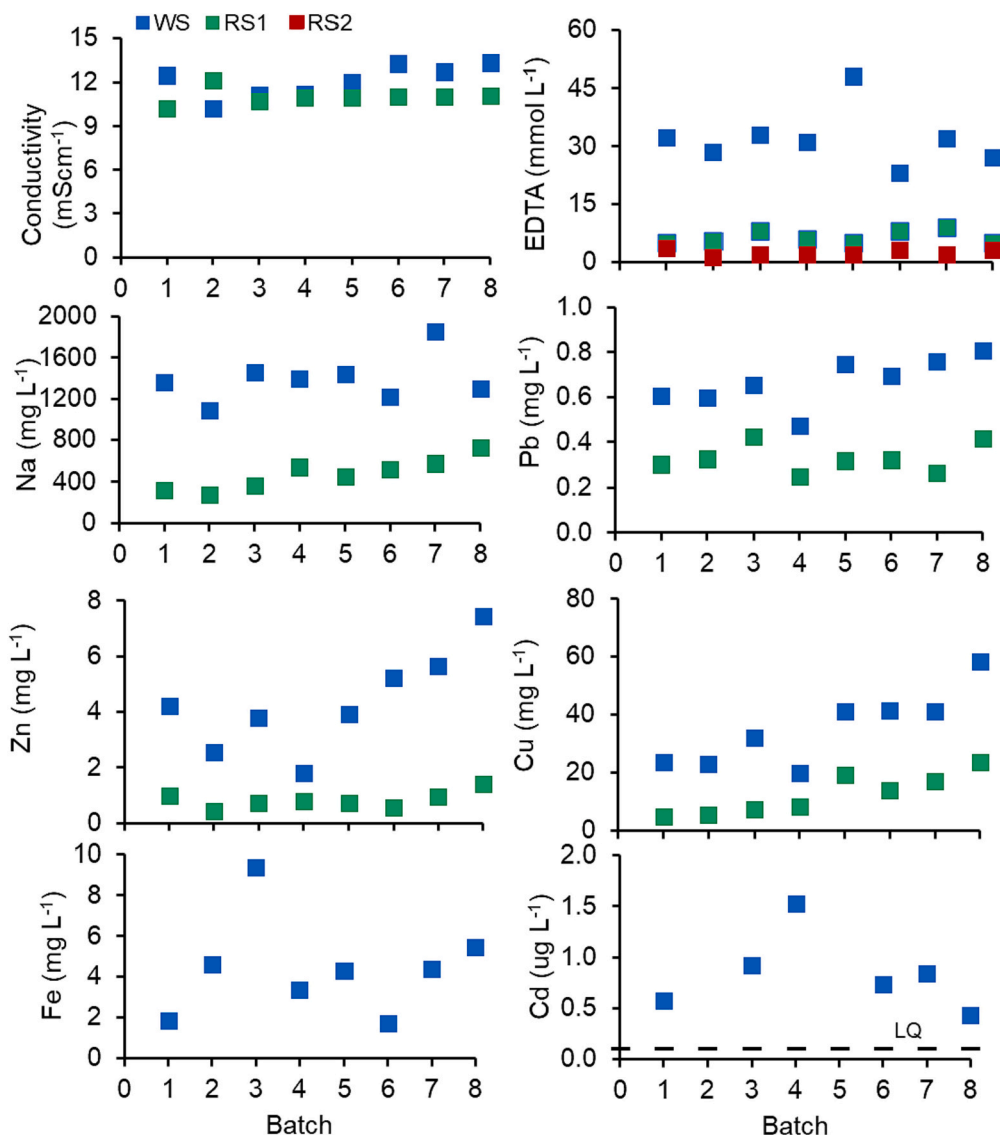


Fig. 3. pH, conductivity, Na and metal concentrations of the recycled washing, first and second rinsing solutions (WS, RS1 and RS2 respectively) over the 8 consecutive wash batches.

Table 1

The efficiency of removing metals from washed SS in a series of 8 consecutive batches.

Batch number	Removal efficiency (%)						
	Pb	Zn	Cd	Cu	Cr	Mn	Fe
1	34	62	46	41	-52	19	2
2	30	71	44	49	-30	12	0
3	32	68	49	46	-34	19	0
4	35	69	50	46	-25	19	1
5	38	65	42	41	-24	26	7
6	34	68	50	34	-16	22	10
7	37	70	49	51	-25	29	8
8	42	70	45	51	-20	29	9

Brassica juncea as a test plant (Fig. S2). *B. juncea* is an annual herbaceous plant with a short crop cycle. The average dry weight of the plants fertilized with different doses of washed SS was slightly higher than that of the control plants, but the differences were not statistically significant (Fig. 4). The content of chlorophyll in the leaves was similar in all treatments (Fig. S3). Fluorescence measurements revealed high photochemical efficiency, both potential (F_v/fm) and actual (F_v'/fm'), in all

Table 2

Leaching of metals from washed and original sewage sludge (SS). Data are given as means \pm standard error of 3 subsamples taken from the homogenized bulk of unwashed SS and from a combined sample of washed SS from batches 1–8. Different letters indicate significant differences between the mean values according to the Welch Two Sample t-test ($p < 0.05$).

Metals (mg kg ⁻¹)	Original SS	Washed SS	DIN 38414-S4*
Pb	0.13 \pm 0.03 ^a	0.80 \pm 0.00 ^b	10
Zn	6.5 \pm 0.0 ^a	110 \pm 1 ^b	50
Cd	0.0 \pm 0.0 ^a	0.11 \pm 0.01 ^b	1
Cu	7.6 \pm 0.2 ^a	24 \pm 0 ^b	50
Cr	0.47 \pm 0.03 ^a	5.2 \pm 0.0 ^b	10
Mn	1.8 \pm 0.0 ^a	1.8 \pm 0.1 ^a	/
Fe	81 \pm 2 ^a	904 \pm 7 ^b	/

* Concentrations stipulated as hazardous (DIN 38414-S4, Council Decision 2003/33/EC).

plants, regardless of treatment. Average F_v/F_m values around 0.83 indicate excellent plant fitness (Schreiber et al., 1995). There were also no treatment-related differences in the parameters derived from gas exchange measurements (net photosynthesis, stomatal conductance, transpiration, water use efficiency), despite some variability between

Table 3

Properties of the original and washed sewage sludge (SS) from all 8 batches of EDTA washing in combination with hydrodynamic cavitation (HC). Data are given as mean values \pm standard error of 3 samples. Different letters indicate significant differences between treatments according to the Welch Two Sample *t*-test ($p < 0.05$).

Properties	Original SS	Washed SS
pH H ₂ O	6.9 \pm 0.0 ^a	6.4 \pm 0.0 ^b
EC (mS cm ⁻¹)	3.1 \pm 0.1 ^a	2.7 \pm 0.1 ^b
Total P (mg kg ⁻¹)	30,352 \pm 472 ^a	30,896 \pm 1168 ^a
Available-P (mg kg ⁻¹)	2533 \pm 462	8267 \pm 945
Total N (%)	3.6 \pm 0.0 ^a	2.8 \pm 0.0 ^b
Total K (mg kg ⁻¹)	3258 \pm 112 ^a	3342 \pm 120 ^a
Available-K (mg kg ⁻¹)	130 \pm 2 ^a	322 \pm 3 ^b
Total C (%)	27 \pm 0 ^a	26 \pm 0 ^a
Total organic C (%)	26 \pm 0 ^a	25 \pm 0 ^a
CaCO ₃ (%)	12 \pm 0 ^a	12 \pm 0 ^a
C _{org} /N	7.2 \pm 0 ^a	8.8 \pm 0.0 ^b

the differently treated groups (the high stomatal conductance of the control plants contributes to their high net photosynthesis and transpiration) (Fig. S3). Many of the above parameters respond strongly to the addition of mineral nutrients such as N and P when the initial availability of these elements in the soil is insufficient (Flexas et al., 2012). It is important to emphasize that such a response was also absent in the NPK treatment where the experimental plants were supplied with mineral fertilizer containing readily available N and P (Fig. S3). Thus, it can be concluded that the lack of fertilizer effect and the lack of effect of SS supplementation is most likely due to a relatively rich soil, in which mineral nutrients were present in sufficient amounts to support the growth of the test plants (Table S5). Another plausible reason for the small differences between the treatments is the preplanting of *B. juncea* seedlings growing in nutrient-rich substrate. Other studies reported that the use of SS increased plant biomass. For example, Kominko et al. (2022) found that fertilization with SS increased the fresh biomass of canola by 75–138 %, maize by 96–138 % and sunflower by 23–54 %. The above-ground biomass of rice increased with the increase in the application rate of SS (Shan et al., 2021). Belhaj et al. (2016) found that the addition of SS increased root and shoot length, number of leaves, biomass and antioxidant activities of sunflower.

Direct application of SS to the soil as a fertilizer has the potential to enhance the uptake of TMs by plants. For example, Belhaj et al. (2016) showed that the addition of SS led to increased concentrations of Cr, Cu, Ni and Zn in the shoots and roots of sunflowers. We investigated the

uptake of TMs into the green plant parts (stems and leaves) only for the highest dose (206 kg ha⁻¹) of washed and original SS. The SS dosage was not sufficient to significantly change the total concentration of TMs in the soil (Table S6). Nevertheless, as mentioned above, the leachability (mobility in aqueous media) of TMs in washed SS was up to 17 times (Zn) higher than in original SS and exceeded the hazardous limit by 2.2 times (Table 2). This was not reflected in increased uptake in *B. juncea*, which is known as a TMs (hyper)accumulator (Vocciante et al., 2019). A small but statistically significant difference was only observed for Cd, where increased uptake was observed in soil amended with washed SS and decreased uptake in soil amended with original SS (Table 4). In contrast to other toxic metals, Cd has a relatively high degree of mobility in natural substrates (Zulfqar et al., 2023).

Similar to plant uptake, the increased leachability of TMs from washed SS (Table 2) was not reflected in a significant leaching of TMs from the soil. No TMs were detected in the leachates of the control soils. An average of 0.001 mg Cu per kg soil was leached from the soil fertilized with original SS (206 kg ha⁻¹ dose). Small amounts of Pb, Zn, Cd and Cu: on average 0.001, 0.012, 0.002 and 0.002 mg kg⁻¹, respectively, were leached from the soil enriched with washed SS.

4. Conclusion

The trials were mainly conducted on a pilot scale and demonstrated the technical feasibility of EDTA washing in combination with HC for the efficient removal of TMs from SS. The nutrient pool remained largely intact and plant-available. However, Cr, which is often present in

Table 4

Metal concentrations in the green parts (stems and leaves, dry weight) of *Brassica juncea* grown on soil fertilized with original sewage sludge (SS), washed SS and non-fertilized soil. Shown are the mean values of metal concentrations with standard error of six replicates. Mean values were compared using Kruskal-Wallis and Dunn tests with Bonferroni correction. Different letters indicate significant differences between the mean values ($p < 0.05$).

Soil fertilization	Toxic metals concentration (mg kg ⁻¹)			
	Zn	Cd	Cu	Cr
Original SS (206 kg P ha ⁻¹)	52 \pm 3 ^a	1.1 \pm 0.1 ^a	11 \pm 0 ^a	2.8 \pm 1.1 ^a
Washed SS (206 kg P ha ⁻¹)	67 \pm 10 ^a	1.5 \pm 0.1 ^b	12 \pm 0 ^a	1.7 \pm 0.4 ^a
Non-fertilized control	51 \pm 2 ^a	1.4 \pm 0.1 ^b	11 \pm 0 ^a	2.1 \pm 0.9 ^a

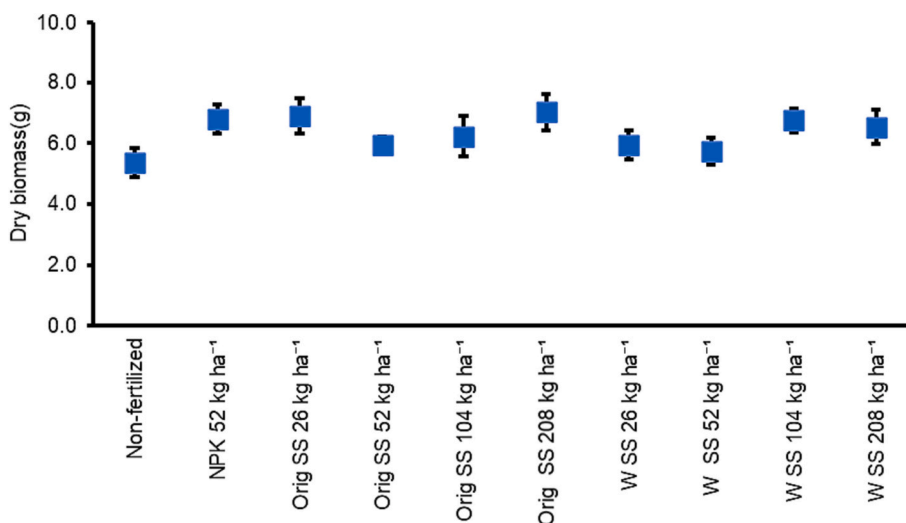


Fig. 4. Dry biomass of *Brassica juncea* on unfertilized soil, with NPK fertilized soil, and soils fertilized with different doses of original (Orig) and washed (W) sewage sludge. The mean values \pm standard error of 6 replicates are shown. No statistically significant differences were found according to one way ANOVA test.

elevated concentrations in SS, was not removed and the leachability of TMs from washed SS increased. The novel washing process also resulted in a significant loss of biomass and EDTA, which could affect economic sustainability. We could not detect a clear fertilizing effect of washed SS on the test crop. The fertilization potential would be better investigated further by using nutrient-poor soils, and the nutrient requirements of the selected test plants would need to be carefully considered. The pot trial did not reveal any evidence of potential environmental risks associated with the application of washed SS.

Further research is needed to clarify the questions that have arisen from this initial comprehensive study.

CRedit authorship contribution statement

Juan Francisco Morales Arteaga: Writing – original draft, Investigation. **Dominik Vodnik:** Investigation. **Damijana Kastelec:** Formal analysis. **Mojca Zupanc:** Investigation. **Matevž Dular:** Investigation. **Jernej Ortar:** Investigation. **Marija Đurić:** Investigation. **Anela Kaurin:** Investigation. **Rok Mihelić:** Investigation. **Domen Lestan:** Writing – review & editing, Writing – original draft, Project administration, Methodology, Funding acquisition, Conceptualization.

Declaration of competing interest

We declare that we do not have any conflict of interest.

Data availability

No data was used for the research described in the article.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.scitotenv.2024.171444>.

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