# THE APPLICATION OF THERMAL HYDROLYSIS BIOSOLIDS ON THE SOILS OF WESTERN SYDNEY

A report undertaken on behalf of Sydney Water

# WESTERN SYDNEY UNIVERSITY





# The application of thermal hydrolysis biosolids on the soils of Western Sydney

A report undertaken on behalf of Sydney Water

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### **Executive Summary**

This report evaluates the application of Thermal Hydrolysis Biosolids (THB) produced by Sydney Water to improve soil condition across five representative sites in the Western Sydney region. The study assessed THB performance in terms of soil structure, nutrient status, microbial diversity, and compliance with regulatory thresholds including nitrogen-limited biosolids application rates (NLBAR), contaminant limits (CLBAR), and PFAS risk management. The data show that THB offers substantial potential to restore degraded or lowfertility soils but requires management to mitigate the risks associated with nutrient loading, metal bioavailability, and salinity.

### Scope and Methodology

THB was applied to topsoil and subsoil materials from Penrith Lakes, Picton Farm, Sydney Science Park, Western Sydney Parklands, and WSU Richmond. These locations span a range of soil landscape units, fertility levels, and land use histories. A 12-week incubation program was conducted under controlled laboratory conditions to simulate THB-soil interactions, using a dry equivalent application rate of 115 to 127 t/ha. This application rate is in in line with the raw NLBAR 130 t/ha but greater than field adjust values ranging from 19 to 121 t/ha across the five sites. The raw NLBAR application rate was selected to induce immediate measurable physicochemical and microbiological changes. Key soil parameters were monitored before and after incubation to determine the impact of THB on soil function.

### Soil Improvement and Structural Benefits

The addition of THB improved soil aggregation, reduced dispersion, increased cation exchange capacity, and enhanced overall structural stability, particularly at sites with sodic or erodible subsoils. At Penrith Lakes and Western Sydney Parklands, dispersion indices decreased significantly following THB incubation, and effective cation exchange capacity increased despite elevated sodium additions. These results indicate that THB can offer substantial physical benefits to degraded soils when the organic matter and cationic composition of the material is appropriately balanced.

### Nutrient Enrichment and Chemical Change

The THB incubation resulted in large increases in total nitrogen, phosphorus, sulfur, and carbon across all five sites. The mean topsoil total nitrogen increased by 0.38%, total carbon by 2.6%, phosphorus by 1,497 mg/kg, and sulfur by 803 mg/kg. While these increases can benefit plant growth and microbial activity, they must be carefully managed in the field. The THB has a carbon-to-nitrogen (C:N) ratio of 7:1 and a nitrogen-to-phosphorus (N:P) ratio of 1:1, both lower than agronomic targets. Following THB addition, the soils generally shifted to an average C:N ratio of 8:1 and N:P ratio of 2:1, potentially creating a nitrogen limitation unless additional inputs are provided. Phosphorus levels were significantly elevated but remain largely recalcitrant, with Bray 1 and Colwell extractable phosphorus indicating most of the pool not being immediately available for immediate loss.

### Contaminant Risks and PFAS

The THB material met regulatory thresholds for most contaminants, with copper concentration (494 mg/kg) exceeded the Grade B threshold and resulted in an overall Contaminant Grade C classification under the NSW EPA (2000) framework. The THB zinc concentration (698 mg/kg) approached the Grade B limit and may exceed thresholds under routine variability. Calculated CLBAR values ranged from 105 to 711 t/ha across the five sites, indicating CLBAR was not placing an additional restriction and the adjusted NLBAR was the limiting factor for biosolids application. Only one PFAS compound, perfluorooctanesulfonic acid (PFOS), was detected at 0.03  $\mu$ g/kg. This value is well below the 0.22  $\mu$ g/kg threshold for unrestricted use under the PFAS NEMP 3.0 guidelines and does not currently present a limitation to reuse. Nonetheless, the persistence and cumulative behaviour of PFAS compounds support a recommendation for long-term monitoring.

### Metal Mobility and Bioavailability

Post-incubation analysis revealed that THB significantly increased the levels of bioavailable metals in the soil, particularly iron (309 mg/kg), manganese (93 mg/kg), zinc (13 mg/kg), and copper (7 mg/kg). Although total metal concentrations remained within acceptable limits, the elevated bioavailable fractions suggest potential mobility under field conditions. These effects are most pronounced in low pH or sandy soils, where retention capacity is limited as presents within the five studied soils. In addition, THB contributed high levels of bioavailable sulfur (411 mg/kg), which may impose a further limitation on application rates.

### Microbial Community Response

THB introduced a diverse suite of microbial taxa dominated by Firmicutes, Bacteroidota, Chloroflexi, Caldatribacterota, Desulfobacteria, and Euryarchaeota. These groups persisted in low-fertility soils such as Penrith Lakes, Western Sydney Parklands, and WSU Richmond, where existing microbial communities were limited. In contrast, the THB had little effect on microbial composition at Picton Farm and the Sydney Science Park topsoil, both of which maintained their original diversity throughout incubation. This suggests that THB may implant microbial diversity in nutrient-depleted soils but has minimal impact where soil biodiversity is already high. The biological origin of THB means that the presence of viable pathogens cannot be entirely excluded, and this possibility should be further assessed when considering application in sensitive or high-risk environments.

### Implications and Recommendations

THB provides a strong candidate material for soil improvement, particularly in disturbed or degraded soils of Western Sydney. Its ability to enhance soil structure, nutrient availability, and microbial function supports its role in land rehabilitation. However, careful application is required to manage nutrient imbalances, particularly low N:P ratios and elevated sulfur. The elevated bioavailability of several trace metals and nutrients necessitates long-term monitoring for mobility and leaching. Site-specific management plans should include microbial and chemical assessments before and after application, with particular focus on sodicity, sulfur, PFAS, trace metal accumulation, and potential pathogenic organisms, especially where soils are used for food production or located in environmentally sensitive areas.

In summary, THB presents low contaminant risk under current conditions, provides measurable soil quality benefits, and improves soil structure and biological function. Its use in Western Sydney soils should be targeted to areas of low fertility and poor structure, with regulated application rates, soil-specific blending strategies, and a monitoring program that includes nutrient ratios, bioavailable fractions, and microbial community tracking.

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### Introduction

Sydney Water Corporation engaged Western Sydney University (WSU) to investigate the application of thermal hydrolysis treated biosolids (THB) to the common soils of the western Sydney region. The soils of the western Sydney region are characterised by low fertility attributable to historic land management practices. Broadly, the soil types in the western Sydney region are classified by the NSW *Land and Soil Capability Mapping* as being poor due to issues around acidity, salinity, water logging, and water erosion. The application of THB materials may assist in improving the overall soil function and mitigate some of the identified limitations. This report provides data on potential soil improvements using THB as produced by Sydney Water. Previous reporting on the application of recycled water by Sydney Water provides complimentary data on soil types and classification (see Reynolds *et al.*, 2024).

The geographic areas under investigation as part of this study are the Penrith Lakes precinct, the Picton Farm, which exists as part of the Picton Water Resource Recovery Facility site, the Sydney Science Park Urban Living Lab site, Western Sydney Parklands, and the WSU Richmond Campus. These sites present a diversity of landscapes that overall represent the soils of the Western Sydney region. The study areas are located on the Richmond (9030ri), Blacktown (9030bt and 9029bt), South Creek (9030sc), and Berkshire Park (9030bp) soillandscape units respectively (Figure 1).



Figure 1 – Study site location and soil types. Soil mapping units of Richmond (9030ri) map unit (green shadow), Blacktown (9030/9029bt) map unit (orange shadow), South Creek (9030sc) map unit (blue shadow), and Berkshire Park (9030bp) map unit (yellow shadow). Map unit data from NSW DPIE (2021a)

In its simplest definition, the *fertility* of soil is an interaction between the soil structure, water availability and retention capacity, and soil nutrient levels. Measures of soil fertility are traditionally focused on nutrient availability (carbon, nitrogen, and phosphorus) within the soil profile with limited recognition of the restrictive factors of sodicity and dispersion on soil performance. The overall soil *health* is ultimately a measure of resilience to perturbations (e.g. droughts) and is subsequently linked to soil physical and chemical properties. Previous reporting has focused on applying recycled water to soil health improvement (Reynolds *et al.* 2024). The work reported here identifies several limitations and highlights how the use of THB materials may interact with the limitations of these same soil materials.

### **Scope of Works**

The investigation covered the soils across a broad geographic area to provide data on their interactions with THB sourced from Sydney Water. The geographic regions included the Penrith Lakes precinct, the Picton Water Resource Recovery Facility site, the Sydney Science Park Urban Living Lab site, Western Sydney Parklands, and the WSU Richmond Campus.

The works undertaken include:

- 1. An investigation into the nature and properties of THB material as supplied by Sydney Water.
- 2. The collection of soil cores and samples from each site and use in lab-based incubation experiments with THB materials.
- 3. The identification of key changes to soil physicochemical and microbiological properties following incubation of soils with THB materials.

Site investigations, material collection, and lab-based incubations occurred between April 2022 and September 2023.

### The study sites within Western Sydney

The pre-existing soil materials were characterised and mapped using soil and landscape data from the NSW Department of Planning, Industry and Environment (DPIE, 2020a, DPIE, 2021a). The five nominated sites present the typical soil landscapes of the Western Sydney region (Table 1). These are broadly characterised as possessing; the absence of organic horizon, high permeability, low water holding capacity, low nutrient levels, erodibility, and high bulk density. The exceptions to these broad characterisations are at the Picton Farm as part of the Picton Water Resource Recovery Facility site, where liquid injected biosolids have been added historically (circa 10 years) with recycled water irrigation and the WSU Richmond site, which is a sandy alluvial soil also receiving recycled water irrigation and agriculturally relevant soil amendments. Detailed reviews of each site are provided in Reynolds *et al.* 2024.

Study areas	Soil type	Soil map unit	ASC map (DPIE, 2021a)	Limiting factors
Penrith Lakes (Bund wall at Hadley House)	Alluvial brown mottled stiff medium clay (ri4)	9030ri / 9030xx (Richmond / disturbed)	Dermosol into Kurosol into heavily disturbed	High erodibility, shrink swell
Picton Water Resource Recovery Facility (Picton Farm paddock S8)	Residual brown clay loam (bt2)	9029bt (Blacktown)	Dermosol into Kurosol	Hardsetting, acidity.
Sydney Science Park (Urban Living Lab)	Residual brown clay loam (bt4)	9030bt (Blacktown)	Kurosol into Kurosol (natric)	Low permeability, low water holding capacity, stoniness
Western Sydney Parklands (Pikes Lane)	Clay loam overlies whole- coloured medium to heavy clay (sc2)	9030sc (South Creek)	Kurosol into Kurosol (natric)	High erodibility (sodic). Hard setting. Low fertility.
WSU Richmond Campus (WSU Farm D2)	Alluvial dark brown sandy loam (bp1)	9030bp (Berkshire Park)	Kurosol (natric)	Erodibility, low nutrient capacity, low CEC

Table 1 – Sampling locations, soil type, and limiting (source data: DPIE, 2021a)

### Thermal Hydrolysis Biosolids (THB)

The thermal hydrolysis biosolids (THB) collected for this study are internally identified by Sydney Water as "*St Marys WWTP Composited Dewatered Biosolid*" and are produced using a thermal hydrolysis protocol by Sydney Water. The THB materials were collected on 'as is' basis in bulk from freshly processed materials directly from the end point of processing at the Sydney Water St Marys facility. Bulk samples (30kg) were collected, sealed, and stored at <5°C in the dark. Samples for pure THB characterisation were immediately taken and frozen at -80°C until analysis. Additional pure THB samples were taken at pre-homogenisation with soil, and lastly as a third pure sample at the end of the experimental period to investigate potential changes to the pure THB over time.

The THB results are compared to previous published research on biosolids for St Marys (Oliver *et al.* 2004) which did not undergo pyrolysis, North Head (Hossain *et al.*, 2011) which did undergo various pyrolysis protocols and with recent work from Victoria (Reichman *et al.*, 2025) on materials that did not undergo pyrolysis and were repeatedly applied to an agricultural land. The biosolids from the St Marys site in the Oliver *et al.* (2004) study were sourced from the same wastewater treatment plant in New South Wales and consisted of aerobically digested sewage sludge. These biosolids underwent mechanical dewatering using a belt press and were air-dried to field moist prior to collection and testing. Comparisons between these various biosolids, along with THB material at the time of collection (initial), at the time of the experiment (pre-mixing), and for the control material left untouched for the duration of the experiment (post-experiment) are provided in Table 2.

Properties	THB (initial) (n = 3)	THB (pre- mixing) (n = 2)	THB (post- exp) (n = 2)	Oliver <i>et</i> <i>al.</i> (2004) St Marys	Reichman <i>et al.</i> (2025) Victoria	Hossain <i>et al.</i> (2011) 300 °C North Head
рН	6.47	6.45	6.46	5.76	6.7	5.32
EC (mS/m)	1.31	1.32	1.31	5.41	2.8	412*
Organic Matter %	49	49	49	31	53.4	55.7
Soluble Magnesium	1,286	1,267	1,274	-	-	-
Soluble Calcium	3,021	3,036	3,046	-	-	-
Soluble Potassium	301	298	299	-	-	-
Total coliform bacteria (MPN)	<2	<2	<2	-	<2	-

 Table 2 - Summary data of THB as received (initial) at the commencement of sampling for mixing (premixing) with soil and a pure THB sample left sealed and untouched for the duration of the experiment (post-exp). Concentrations are in mg/kg unless otherwise noted. Note: \* reported value in paper which may be inaccurate.

The biosolids used in the study by Hossain *et al.* (2011) were sourced from the North Head Sewage Treatment Plant in Sydney (as known) and were generated through anaerobic digestion. After digestion, the biosolids underwent mechanical dewatering and thermal treatment at a range of temperatures ranging to 700°C (the closest relevant temperature to this

study is 300°C). The biosolids used in the Reichman *et al.* (2025) study were sourced from four Melbourne Water treatment facilities and consisted of anaerobically digested sewage sludge that had been air-dried and stockpiled.

The THB material presents as low-density aggregates (clumps) with a 10YR 2/2 colour. The mean pH was 6.46 and would be suitable for use as a bulk soil. Coliforms were determined using IDEXX colisure kits with mean values <2 MPN. The mean electrical conductivity (EC) of the pure THB was 1.3 ds/m and would be considered elevated if used as bulk soil material. The THB material itself was stable, showing minimal change throughout the experiment when tested as received (initial), at the time of mixing with the soil materials (pre-experiment), and as tested after the experiment, having sat in storage for the duration (control).

The nutrient load of the pure THB material is associated with organics having high concentrations of carbon, nitrogen, phosphorus, and sulfur (Table 3). The total organic carbon value is 15%, and the C/N ratio is 7.2 which is below that used in field experiments of Reichman *et al.* (2005). The total sulfur loads are lower than those of Oliver *et al.* (2004), and ammonium loads are greater than those found in Hossain *et al.* (2011). The total nitrogen values of the THB constrain broadscale application rates and dosage rates used in the incubation program are calculated to the upper limit of these constraints.

Properties	THB (initial) (n = 3)	THB (pre-mixing) (n = 2)	THB (control) (n = 2)	Oliver <i>et al.</i> (2004) St Marys	Reichman <i>et al.</i> (2025) Victoria	Hossain <i>et al.</i> (2011) 300 °C
Sulfur (mg/kg S)	4,921	4,901	4,905	9,587	-	-
Chloride (eqv mg/kg)	844	845	845	-	-	-
Total Organic Carbon (%)	15	15	14	18	11	25.6
Total Carbon (%)	28	28	28	25	25	
Nitrate (mg/kg N)	3.8	3.7	3.6	-	-	< 0.2
Ammonium Nitrogen (mg/kg N)	2,761	2,755	2,780	-	-	1,175
Total Nitrogen (%)	3.9	3.9	3.9	-	-	3.27
Total Kjeldahl Nitrogen (TKN)	3.7	3.6	3.6	-	1.2	-
C/N ratio	7.2	7.2	7.2	-	20.8	-
Bray 1 Phosphorus	434	451	448	-	-	-
Colwell Phosphorus	1,447	1,451	1,450	-	-	492
Total Phosphorus	38,152	38,100	38,134	-	52,658	-

 

 Table 3 - Summary data of THB as received (initial) at the commencement of sampling for mixing (premixing) with soil and a pure THB sample left sealed and untouched for the duration of the experiment (control). Concentrations are in mg/kg unless otherwise noted

The main concerns for soils of western Sydney are acidity, sodicity, erodibility, and fertility. The THB materials may induce a greater response when higher application rates are applied. The exchangeable sodium percentage (ESP) is a measure of soil sodicity using the proportion of sodium on the soil exchange sites. Sodic soils may become dispersive, resulting in structural collapse and erosion upon wetting. The ESP for pure THB material is 4.6 (Table 4), which places the material as *mildly* sodic if considered as a soil material. However, the Dispersion Index of the THB is 0, indicating a stable material not prone to erosion. The THB effective cation exchange capacity (ECEC) is high (relative to the soils of western Sydney), which can be considered as the capacity of the soil to maintain and hold nutrients. The higher ECEC is attributed to the amount of organic matter in the soil, which may degrade/decompose over time, resulting in a lower ECEC and a potential loss of soil fertility if not adequately managed.

Exchangeable components	THB (initial) (n = 3)	THB (pre-mixing) (n = 2)	THB (post-exp) (n = 2)
Ex-sodium percentage (ESP)	4.6	4.6	4.6
Dispersion Index	0	0	0
Ex-Calcium (mg/kg)	6,232	6,228	6,232
Ex-Magnesium (mg/kg)	1,508	1,470	1,490
Ex-Potassium (mg/kg)	750	755	761
Ex-Sodium (mg/kg)	503	510	499
Ex-Aluminium (mg/kg)	28	28	28
Ex-Hydrogen (mg/kg)	<1	<1	<1
Ca/Al ratio	40	<b>51</b>	16
ECEC (cmol+/kg)	48	51	46

Table 4 – Effective cation exchange capacity (ECEC) and exchangeable cation distribution in pure THB. Data for as received 'initial', at the time of mixing with the soil 'pre-mixing' and for control material not exposed to soils but left for 12 weeks 'post-exp'. No data available from Oliver (2004), Hossain (2011) or Reichman (2025)

A potential risk is the amount of 'metals' and 'heavy metals' that might be stored within the THB materials, noting that biosolids have been reported as having higher 'heavy metal' loads. The total elemental pool is the most resistant to alteration and change. The determination is undertaken by complete acid digestion of the THB material (Table 5). The total concentration of elements is within expectations, with values generally below those of Hossain *et al.* (2011) and above those of Reichman *et al.* (2025). The THB is very close to the values determined in Oliver *et al.* (2004), noting that both use the St Mary's facility.

Element	THB (n=7)	Oliver <i>et al.</i> (2004) St Marys	Reichman <i>et al.</i> (2025) Victoria	Hossain et al. (2011) 300 °C
Calcium	18,428	15,502	-	34,700*
Magnesium	3,231	-	-	3,500
Potassium	992	1,836	1,100	-
Sodium	718	937	-	-
Sulfur	9,252	9,587	-	44,700*
Zinc	698	687	256	1,350
Manganese	516	-	-	-
Iron	74,976	110,512	-	78,000*
Copper	494	597	139	1,150
Boron	9.6	-	-	-
Silicon	358	-	-	-
Aluminium	15,429	11,234	-	-
Molybdenum	6.1	-	-	-
Cobalt	5.5	-	-	-
Selenium	4.6	-	-	<6.6
Cadmium	0.79	3.2	-	2.62
Lead	20	57	22	115
Arsenic	2.9	-	-	<3
Chromium	53	-	27	107
Nickel	21	-	11	182
Mercury	0.48	-	-	-
Silver	1.3	-	-	-

 Table 5 – Total elemental concentration as determined by acid digest (mg/kg). Partial data available from Oliver (2004), Hossain (2011) and Reichman (2025). \* denotes a conversion from % to mg/kg

Total elemental concentrations may be of limited concern due to solubility and bioavailability restrictions. A discernment is made between '*total*' load and '*bioavailability*'. To understand this potential risk, bioavailable metals were determined using a DTPA (diethylene triamine pentaacetic acid) extraction, which is a selective extraction aimed at estimating bioavailability and KCl (potassium chloride salt rinse) was used for bioavailable sulfur. The pure THB materials had DTPA extractable iron, manganese, zinc, and copper (Table 6). The DTPA extractable metal concentrations in the pure THB materials are higher in iron and manganese than values reported for raw (not pyrolised) and 300°C pyrolised biosolids, whilst zinc and copper are similar. Data on KCl extractable elements or even water extractable sulfur are limited in published reports.

Element	THB (initial) (n = 3)	THB (pre-mixing) (n = 2)	THB (post-exp) (n = 2)	Biosolids Raw Hossain <i>et al.</i> (2011)	Biosolids 300°C Hossain <i>et al.</i> (2011)
Zinc	120	121	121	297	143
Manganese	1,267	1,187	1,079	51	36
Iron	2,431	2,400	2,420	467	842
Copper	55	55	55	22	< 0.1
Sulfur	4,921	4,910	4,910	-	-

Table 6 – The bioavailable zinc, manganese, iron and copper (DTPA) and sulfur (KCl). Comparisondata of 'raw' and '300°C' of Hossain (2011). No data available from Oliver (2004) orReichman (2025)

#### **Guidelines for THB land use**

The THB material was assessed using the contaminant grading system outlined in NSW EPA (2000). The NSW EPA (2000) guidelines establish a contaminant grading system to evaluate biosolids quality based on the concentration of heavy metals and organic contaminants. Biosolids are assigned a contaminant grade from A to E, with Grade A representing the highest quality (lowest contaminant levels) and Grade E indicating excessive contamination or untested material. Each contaminant is individually assessed, and the lowest individual grade assigned to any contaminant determines the overall contaminant grade of the biosolids product (Appenidx A contains NSW EPA 2000 threshold values). For the THB, copper (494 mg/kg) exceeds the limits for both Grade A (100 mg/kg) and Grade B (375 mg/kg), qualifying it as Grade C. Zinc (698 mg/kg) meets the Grade B threshold (≤700 mg/kg), while all other regulated metals fall within Grade A limits (Table 7). Due to the total copper, the lowest individual contaminant grade determines the overall classification, and the THB is assigned a *Contaminant Grade C*. It is important to note that the zinc concentration of 698 mg/kg is very close to the Grade B upper limit of 700 mg/kg. This proximity to the threshold limit suggests that routine biosolid composition variability could result in Grade C classification based on zinc alone.

Contaminant	THB	Grade A	Grade B	Grade C	THB Grade
Arsenic	2.9	20	20	20	А
Cadmium	0.79	3	5	20	А
Chromium	53	100	250	500	А
Copper	494	100	375	2000	С
Lead	20	150	150	420	А
Mercury	0.48	1	4	15	А
Nickel	21	60	125	270	А
Selenium	4.6	5	8	50	А
Zinc	698	200	700	2500	В

**Table 7** – THB graded against NSW EPA (2000) biosolids criteria. Values in mg/kg.Thresholds for Grade A, B, and C provided for reference

A second determination, referred to as the stabilisation grade, is a classification system that evaluates the extent to which biosolids have been treated to reduce pathogens, odours, and the potential to attract disease vectors. The primary aim of the stabilisation grade is to ensure biosolids are microbiologically safe for land application and do not pose health or environmental risks after disposal or reuse. The THB materials achieve a *Stabilisation Grade A* based on the treatment protocol being a thermal hydrolysis approach and using the NSW EPA criteria (Appenidx A contains threshold values). The overall contaminant grading result is a classification of the THB material as *Restricted Use 2* (Table 8).

Overall Classification	Minimum Contaminant Grade	Minimum Stabilisation Grade	Allowable Uses
Unrestricted Use	А	А	Home gardens, public spaces, and all other.
Restricted Use 1	В	Α	Urban landscaping, public contact sites, and all other.
Restricted Use 2	С	В	Agriculture, forestry, land rehabilitation, landfill, surface land disposal.
Restricted Use 3	D	В	Forestry, land rehabilitation, landfill, surface land disposal.
Not Suitable for Use	E or ungraded	C or ungraded	Landfill or surface disposal only

Table 8 – The overall THB classification based on NSW EPA (2000) Biosolids grading criteria

The frequency of biosolids application to land is governed by the classification of the biosolids product and the specific site management practices employed. For the THB materials used in this report and any other biosolids classified as Restricted Use (Grades 1, 2, or 3), the guidelines stipulate that once applied to a portion of land, *no further application should occur on that same area for a minimum of five years*. This interval is designed to mitigate cumulative contaminant build-up risk and ensure safe nutrient assimilation in agricultural soils.

However, an exception to this 5-year interval is permitted where the receiving soil has been limed and maintained at a minimum pH of 5.5 (using CaCl<sub>2</sub>) for a period of at least two years following the biosolids application. This pH management strategy reduces the mobility and plant uptake of certain heavy metals, thereby lowering potential environmental and food safety risks. The THB material alone may provide a benefit by increasing pH to >5.5 but the longevity of this increase requires further investigation and addition of lime may be required at certain intervals. Biosolids classified as Unrestricted Use (those meeting both Contaminant Grade A and Stabilisation Grade A criteria) are not subject to this five-year restriction.

#### **Guideline NLBAR application rate**

The Nitrogen-Limited Biosolids Application Rate (NLBAR), based on the Environment Protection Authority (NSW EPA) (2000) *Environmental Guidelines: Use and Disposal of Biosolids Products*, is the current guideline for biosolids use and application. It ensures that applied biosolids provide nitrogen for plant growth without exceeding environmental safety thresholds. The guidelines differentiate between agricultural and non-agricultural sites with allowances for land rehabilitation, leading to different nitrogen application strategies.

The application of biosolids as a nitrogen source in agricultural systems is subject to strict regulatory controls to prevent environmental degradation. In New South Wales, the NSW Biosolids Guidelines impose a maximum nitrogen application rate of 1,200 kg N/ha for agricultural land, ensuring nitrogen inputs do not exceed plant uptake and minimising the risk of nitrate leaching into groundwater. This limit is particularly relevant in cropping systems, pasture management, and horticultural production, where excessive nitrogen accumulation can lead to soil imbalances and water contamination. The NSW EPA guidelines require that baseline soil nitrogen be accounted for in nitrogen budgeting, reducing the allowable biosolids application rate in fields with existing nitrogen reserves. This conservative approach prioritises environmental safety but may limit the potential for biosolids use in nutrient-depleted agricultural soils. The nitrogen-limited biosolids application rate (NLBAR) was calculated in accordance with the NSW EPA Biosolids Guidelines using the following equation (worked calculations are provided in Appendix B):

$$NLBAR t/ha = \frac{Crop Nitrogen Requirement (kg ha)}{Available Nitrogen (kg t biosolids)}$$
[1]

Available nitrogen in biosolids was estimated using the standard guideline formula:

For agricultural sites (WSU Richmond, Sydney Science Park, and Picton Farm), NLBAR is constrained by crop nutrient demand and residual soil nitrogen, and guidelines recommend calculating NLBAR based on crop nitrogen requirements. Nitrogen uptake rates for perennial rye-grass have been used as a reference species for this assessment. Using values from Table 3 from this report and the N-uptake values for perennial rye-grass from the NSW Guidelines, the calculated THB NLBAR is 128 dry t/ha using the five-year annualised approach (Appendix B). The value is higher than the anticipated annual ranges due to the low available nitrogen content in the THB material. The calculated NLBAR is also not beyond real-world examples, including those in Reichman et al. (2025), where annual application rates averaged 45 t/ha annually, with >200 t/ha used over four years. The calculated THB NLBAR of 128 dry t/ha value would provide some 200 kg N/ha to the soil and perennial rye-grass and is below the potential 1,200 kg N/ha limit. The NLBAR accounting for nitrogen requirements includes ammonium, oxidised nitrogen, and a fraction of organic nitrogen that mineralises in the first year. For example, if the maximum limit of 1,200 kg N/ha (as opposed to the 300 kg N/ha for perennial rye-grass) was applied as defined in the NSW Guidelines the NLBAR would be 138 dry t/ha. This assumes no existing soil N is present (raw NLBAR), and where soil nitrogen data is available, the 1,200 kg N/ha limit should be lowered by the corresponding residual amount (adjusted NLBAR).

When residual soil nitrogen is present, the maximum allowable biosolids application rate is lowered, which is a particular focus when repeated applications are being considered. Making these adjustments, the calculated NLBAR values indicate that Picton Farm would be reduced to 19 t/ha, WSU Richmond 29 t/ha and Sydney Science Park 24 t/ha due to elevated residual nitrogen (Table 9). The potential risk being accommodated is the potential for excessive nitrogen buildup, which could lead to nitrate leaching and environmental contamination. For the sites with low residual nitrogen, the adjusted NLBAR is near the raw NLBAR with Penrith Lakes at 121 t/ha and Western Sydney Parklands at 109 t/ha.

The non-agricultural lands of Penrith Lakes and Western Sydney Parklands may be exempt from the 1,200 kg N/ha restriction under specific conditions, provided regulatory approval is obtained. Exemptions are considered for land rehabilitation, forestry applications, and soil improvement in urban environments, where nitrogen is less likely to contribute to groundwater pollution due to higher sequestration potential. The distinction between agricultural and nonagricultural applications reflects broader differences in nitrogen cycling dynamics, where nonagricultural systems may benefit from higher biosolids loading without the same risks of overfertilisation. For the non-agricultural sites, biosolid applications may be a one-time event rather than repeated. The NSW EPA (2000) guidelines allow a maximum nitrogen load of 1,200 kg N/ha in single-use non-agricultural applications.

	Baseline Soil N (kg/ha)	Baseline Soil P (kg/ha)	Baseline Soil PBI	NLBAR adjusted (dry t/ha)	Total Nitrogen (kg/ha)	Total Phosphorus (kg/ha)
Penrith Lakes	202	107	44	121	1,200	4,518
Western Sydney Parklands	301	41	37	109	1,200	4,011
WSU Richmond	960	140	10	29	1,200	1,201
Sydney Science Park	997	238	160	24	1,200	1,135
Picton Farm	1,040	712	210	19	1,200	1,419

Table 9 – The adjusted Nitrogen-Limited Biosolids Application Rate (NLBAR) using hypothetical 1,200 kg/ha max levels based on the Environment Protection Authority (NSW EPA) (2000). Corrected for baseline soil N levels. Note: raw unadjusted NLBAR is 130 dry t/ha (see above). NLBAR calculations are provided in Appendix B

Applying biosolid materials will also increase the phosphorus load to the soil. The THB material contained >38,000 mg/kg of total phosphorus and loading to the soil relative to the NLBAR application rates, including the residual soil phosphorus levels, results in <1 t/ha to >4 t/ha in total phosphorus increase to the soil. These total phosphorus levels may be of concern where phosphorus mobility is deemed a potential risk. The Bray 1 and Colwell P extractions are used to measure the potential bioavailability and mobility of phosphorus (Table 3), and that part of the total phosphorus pool is mobile. Little data exist on equivalent measures in corresponding studies, which limits understanding of the broader implications. Furthermore, consideration of the Phosphorus Buffering Index (PBI) can provide insight into the ability of the soil to hold and retain soil phosphorus. The baseline soil PBI for Picton and Sydney Science Park indicates moderate phosphorus retention, while WSU Richmond, Penrith Lakes, and Western Sydney Parklands exhibit very low phosphorus retention. Higher PBI soils require more phosphorus for plant uptake, whereas low PBI soils are more prone to phosphorus loss through leaching or runoff. For sites receiving repeated biosolid applications, total phosphorus and phosphorus adsorption capacity and a measure of bioavailability should be determined to prevent overloading on an annual basis.

### Guideline CLBAR application rates

The Contaminant Limited Biosolids Application Rate (CLBAR) is designed to prevent excessive accumulation of heavy metals, organochlorine pesticides (OCP), polychlorinated biphenyls (PCB), dioxins (e.g., 2,3,7,8-TCDD), and hydrocarbons (e.g., benzopyrene) in soils following biosolids application. The CLBAR establishes site-specific application limits by calculating the maximum allowable soil contaminant concentration based on the difference between acceptable threshold limits and baseline and residual soil conditions. The range of

contaminants that must be considered in such an assessment is listed in the NSW EPA (2000) guidelines. The lowest calculated CLBAR for any individual contaminant determines the maximum biosolids application rate (worked calculations for each site are provided in Appendix C).

$$CLBAR t/ha = \frac{Max Allowable-Soil Baseline x Incorporated Mass}{Biosolid Concentration}$$
[3]

The CLBAR approach complements the NLBAR, ensuring that both nutrient and contaminant thresholds are managed effectively. While NLBAR prioritises agronomic nutrient requirements, CLBAR functions as an environmental safeguard, overriding NLBAR where contaminant levels pose a potential risk. The CLBAR varied substantially across the five study sites due to differences in soil contaminant concentrations, regulatory land-use classifications (agricultural or non-agricultural), and bulk densities (Table 10 and Appendix C). Copper was the limiting contaminant across all sites, with minimum CLBAR values ranging from 105 t/ha (Picton Farm) to a non-realistic high of 711 t/ha (Western Sydney Parklands). Sites designated as agricultural land (Picton Farm, Sydney Science Park, and WSU Richmond) were subject to more stringent contaminant thresholds compared to non-agricultural sites (Penrith Lakes and Western Sydney Parklands), resulting in consistently lower allowable biosolids application rates for Picton Farm (105 t/ha), Sydney Science Park (108 t/ha), and WSU Richmond (150 t/ha). The CLBAR application rate is lower than raw NLBAR and would therefore be the upper limit of application rate. However, when accounting for residual nitrogen, the adjusted NLBAR is lower than the CLBAR. Non-agricultural classifications allowed for higher application limits for several contaminants at Penrith Lakes (692 t/ha) and Western Sydney Parklands (711 t/ha). These high CLBAR rates are superseded by the lower NLBAR caps to application rates. These findings highlight the necessity of site-specific classification under regulatory frameworks and underscore the role of copper as the primary constraint in THB application strategies.

	NLBAR adjusted (t/ha)	CLBAR (t/ha)	THB Incubation (t/ha dry eq)
Penrith Lakes	121	692	126
Western Sydney Parklands	109	711	127
WSU Richmond	29	150	119
Sydney Science Park	24	108	123
Picton Farm	19	105	115

Table 10 – Contaminant Limited Biosolids Application Rate (CLBAR) based on the EnvironmentProtection Authority (NSW EPA) (2000). Adjusted NLBAR data provided for comparison from Table8. The lower of the NLBAR and CLBAR is the appropriate maximum application rate. Note: THBrates are based on 130 dry t/ha application which is corrected for density

The calculated NLBAR and CLBAR values are approximate and should not be applied rigidly. When considering actual application rates in real-world settings, the nitrogen requirements and potential export/loss depend on multiple factors, including:

*Soil Characteristics*: Variability in soil organic matter, texture, pH, and existing nitrogen levels affect nutrient availability.

*Climatic Conditions*: Rainfall, temperature, and seasonal variations impact nitrogen uptake efficiency and potential leaching losses.

*Management Practices*: Irrigation, fertilisation, and planting strategies influence nutrient demand.

While NLBAR provides indicative nitrogen requirements, site-specific soil assessments are essential. The calculated NLBAR value is 130 t/ha annualised over 5-year intervals or 26 dry t/ha if viewed as a single year. This aligns with application rates used in the Reichman *et al.*, (2025) field study. Calculating an adjusted NLBAR corrects for existing soil nitrogen values and setting a maximum limit of 1,200 kg N/ha results in an application for the study sites between 19 and 121 t/ha on a 5-year interval. The calculated CLBAR for the five sites was greater than the value of NLBAR and ranged between 105 and 711 dry t/ha, as potential contaminant levels were low risk. As the NLBAR is lower than the CLBAR, the default is to use the lower NLBAR as the field application rate. In the incubation study provided in this report, the NLBAR with a 1,200 kg N/ha limit is not used as an upper limit for application rate.

### Additions following National Environmental Management Plan (NEMP) 3.0

Per and polyfluoroalkyl substances (PFAS) are contaminants of concern and include a range of compounds persisting through the wastewater treatment processes. The PFAS National Environmental Management Plan (NEMP) 3.0 (HEPA, 2025) identifies PFOS, PFHxS, and PFOA as the key indicator compounds due to their prevalence, persistence, and associated human and ecological risks. The THB materials were tested for 34 different PFAS compound. Of the tested compounds only perfluorooctanesulfonic acid (PFOS) was detected. All others, including fluorotelomer alcohols (FTOHs), perfluorinated alkyl sulfonamides or polyfluoroalkyl phosphates (PAPs), were not detected (Table 11).

These compounds can accumulate in biosolids during wastewater treatment and persist through conventional treatment processes, resulting in measurable concentrations in land-applied biosolids. Because PFAS are resistant to degradation and can bioaccumulate, the land application of biosolids represents a diffuse but long-term source of environmental PFAS loading, necessitating regulatory controls and threshold limits to mitigate risk.

Compound	THB (initial) (n = 3)	THB (pre-mixing) (n = 2)	THB (post-exp) (n=2)
∑FTSA	< 0.01	< 0.01	< 0.01
∑FOSE	< 0.01	< 0.01	< 0.01
∑FOSA	< 0.01	< 0.01	< 0.01
∑FTOH	< 0.01	< 0.01	< 0.01
PFOS (1763-23-1)	0.031	0.030	0.032
PFOA (335-67-1)	< 0.01	< 0.01	< 0.01
PFHxS (355-46-4)	< 0.01	<0.01	< 0.01

**Table 11** – THB materials polyfluoroalkyl (PFA) compounds (ug/kg).  $\Sigma$  = sum total

The Margin of Safety (MOS) is a risk management factor applied in the PFAS NEMP 3.0 to account for uncertainties in exposure modelling, variability in environmental conditions, and the presence of multiple PFAS compounds beyond those specifically assessed. MOS ensures that biosolids application does not result in unacceptable human or environmental health risks and allows regulators to adjust thresholds based on local context and confidence level in input data. The MOS provides a conservative buffer by reducing the allowable PFAS concentrations in biosolids and soil through division by the selected MOS value (typically 1, 2, or 5), which are provided in Table 12.

 Table 12 – Criteria for PFOS+PFHxS and PFOA in Biosolids and Maximum Allowable Soil

 Contaminant Concentrations (MASCCs) from PFAS NEMP 3.0. Values in μg/kg

Criteria Type	Margin of Safety (MOS)	PFOS + PFHxS	PFOA
Biosolids – Restricted Use	5	6.2	16
	2	15	40
	1	31	81
Biosolids – Unrestricted Use	5	0.22	0.6
	2	0.55	1.5
	1	1.1	3
MASCC (Soil Thresholds)	5	0.22	0.6
	2	0.55	1.5
	1	1.1	3

A higher MOS reflects greater precaution and is particularly relevant when site-specific conditions, sensitive ecological receptors, or cumulative exposure risks are present. For the THB material, only PFOS was present at 0.03  $\mu$ g/kg, which results in a classification of

*Unrestricted Use* relative to PFAS NEMP 3.0 guidelines (HEPA, 2025). However, it is important to note that the Maximum Allowable Soil Contaminant Concentrations are at 0.22  $\mu$ g/kg for an MOS of 5 and consideration should be given to these thresholds when multiple application rates are considered.

### Western Sydney Soils following THB incubation

For this investigation, THB (see Section 4) were applied to soils collected from each of the five sites (see Section 2). Soil cores of 18cm diameter in PVC casing were collected by hand from the soil surface to the 'point of refusal' depth. The soil cores were extracted from the soil profile in complete form, with the surrounding material removed, and the soil core was extracted with PVC casing, holding the soil cores intact. This approach allows for the sampling of soils whilst maintaining their overall structure and existing condition. A total of 3 soil cores were sampled in each experimental run (triplicate cores) and allowed to air dry for 4 weeks before dissection and THB addition. The air-drying approach ensured accurate soil mass whilst limiting impact on existing microbial diversity. Drying was limited to a duration known to occur within the Sydney basin. Soil cores were then dissected, and subsoil and topsoil samples were separated from the parent core, with each layer homogenised.



Figure 2 – Typical soil core displaying topsoil and subsoil conditions

### **THB** incubation protocol

The raw NLBAR is 130 dry t/ha for a 5-year application period. This raw NLBAR not factoring in residual N loading in soil was a deliberate methodological choice to induce measurable responses within the 12-week incubation period. The Thermal hydrolysis biosolids (THB), produced under high temperature and pressure conditions, exhibit reduced short-term nitrogen mineralisation due to the breakdown and stabilisation of organic material during treatment and their chemical stability and recalcitrant structure closely resemble that of composted biosolids.

The incubation program aimed to accelerate soil and microbial responses, allowing clear differentiation between treatment effects and control conditions under controlled laboratory settings where environmental risks were eliminated. While the experimental THB application rates exceeded NLBAR limits, the findings provide critical insights into soil-THB interactions. These results will help guide long-term, sustainable application strategies at lower, field-appropriate rates while ensuring that THB use is optimised for soil improvement without exceeding environmental thresholds.

The air-dried topsoil and subsoil were homogenised, and 150g was added along with 30g of THB material (as received with 37% moisture) to sterilised 250 ml Schott bottles (Figure 3). This equates to a hypothetical application rate of 115 to 127 dry t/ha due to soil density at each site. Each bottle was sealed, and the dry mix was homogenised with a mechanical tray roller for 1 hour. The irrigation water used for this study was simulated rainwater sourced from WSU labs. After mixing, each bottle received 30 mL of simulated rainwater for topsoils (to achieve field moist conditions) and 50 mL for subsoils (to reach saturated conditions), with the overall mass recorded. Additional water was supplied to subsoil samples to guarantee saturated moisture conditions that more accurately reflect deeper soil.



Figure 3 – Soil and TBH incubation protocol

Sealed bottles were incubated at 25°C with topsoil samples provided with limited natural light and subsoil samples kept away from light. Each sealed bottle was opened and vented twice a week to ensure adequate air mixing, and the mass of each bottle was monitored once a week. Where needed, additional water was added to return to the commencing original mass with an average of 10 ml added throughout the experiment. The incubation period ran for 12 weeks on this cycle. The homogenisation of the soil with added THB material was not straightforward, which has implications for land application (Figure 4). Figure 4(a) provides an example of the initial material unmixed, and (b) after mixing, showing incomplete mixing and aggregate formation due to the interaction of soil and THB.



Figure 4 – THB and soil homogenisation (a) Pre-homogenisation with dry soil mass added to THB material. (b) Post-homogenisation step with materials mixed

After the incubation period, bottles were immediately transferred to PC2 conditions and subsampled for microbiological and physicochemical properties. Soil chemical properties were determined using NATA-certified analytical laboratories. Soil DNA extractions were conducted on subsampled soil and THB mixes (250 mg) using the Qiagen DNeasy PowerSoil Pro Kit. DNA yield was assessed using a DeNovix DS-11+ spectrophotometer. The16S rDNA amplicons were processed using the QIIME2 platform using standard protocols. Quality control was conducted by truncating sequences at 245bp to remove low quality regions (<Q25) and the first 10bp of reads to ensure primer and adaptor removal. Amplicon Sequence Variants (ASVs) were determined using DADA2 denoising with QIIME2, which also merged paired reads and removed chimeric sequences. Taxonomy was assigned to representatives of each ASV using the QIIME2 Feature Classifier and the SILVA Database. Data was rarefied to 37,500 before statistical analysis to ensure even sampling depth. Multivariate statistics and diversity analyses were conducted within QIIME2 as follows. Bray-Curtis similarity was calculated between each sample and visualised as a Principle Coordinates Analysis (PCOA) using Emperer. The significance of clustering between each grouping was determined using ANOSIM with 999 permutations, and differences in alpha-diversity (ASV richness) were calculated using a Kruskal-Wallis test. The relationship between environmental variables and community structure was determined using BIOENV and presented as a redundancy analysis.

### Penrith Lakes response to THB additions

The landscape of the Penrith Lakes site has experienced soil replacement and landscape reshaping as part of remediation works occurring over the previous decade. The product of these works is the exposure of the sodic B-horizon to the near surface, along with the construction of artificial soil layers. The engineered nature of the Penrith Lakes site and the intentional compaction of soil materials to gain structural integrity of the bund areas have resulted in significant compaction of the soil materials.

The Penrith Lakes soils were circum-neutral upon collection (pH 6.20 and 6.70). Following 12 weeks of THB incubation, the pH increased in topsoil and subsoils (pH 6.93 and 7.21). A slight increase in soil salinity (EC) following incubation with THB is associated with increased water-soluble sodium, calcium, magnesium, and chloride levels (Table 13). The cation exchange capacity of the Penrith Lakes soil is typical of loam sands, which are low and have a limited capacity to adsorb and retain ions. Of the limited adsorption potential, the ESP at Penrith Lakes site was <5% and did not alter following THB addition. It is therefore considered "*non-sodic*" post THB incubation. Although the Penrith Lakes site does have soil sodicity issues across the site (as identified in WSU research work – Reynolds *et al.*, 2024), the samples used in this study do not possess these properties. The soil Dispersion Index of 5 and 11 for topsoil and subsoil, respectively, improved following THB addition to 1 and 3. The improvement in dispersion and categorisation as "*non-sodic*" is a marked improvement to the soil condition at Penrith Lakes.

Bioavailable phosphorus levels (measured using both Bray 1 and Colwell approaches) were below recommended guidelines for the original soil. Both increased following incubation, with greater increases in Colwell P matching the original THB material (Table 3). A similar result occurred with soil nitrate levels being low and remaining unchanged, whilst soil ammonium nitrogen increased following THB incubation. The bioavailability of (DTPA method) manganese, iron, and copper were within recommended guidelines for the original soil materials, with zinc below guideline values. Incubation with THB increased DTPA extractable zinc to within guideline limits, and manganese and iron increased tenfold to well above guideline values. The bioavailable (hot CaCl<sub>2</sub> method) boron levels remained unchanged at lower guidance values following THB addition to the upper limit of guidance values.

	Original		ТНВ Т	reated
	Topsoil	Subsoil	Topsoil	Subsoil
pH	6.20	6.70	6.93	7.21
Electrical Conductivity (dS/m)	0.6	0.4	0.3	0.5
Dispersion Index	5	11	1	3
Chloride Estimate (equiv. mg/kg)	39	28	175	98
Soluble Calcium (1:5 water)	489	407	1,096	1,083
Soluble Magnesium (1:5 water)	121	105	280	204
Soluble Potassium (1:5 water)	33	26	74	49
Soluble Phosphorus (1:5 water)	2	1	9.4	8.2
Phosphorus (Bray 1 mg/kg P)	13	2	58	11
Phosphorus (Colwell mg/kg P)	31	9	617	640
Nitrate Nitrogen (mg/kg N)	0	1	0.48	1
Ammonium Nitrogen (mg/kg N)	1	3	92	80
Sulfur (mg/kg S)	14	11	126	117
Zinc (DTPA method)	1	1	9.8	10
Manganese (DTPA method)	8	11	130	127
Iron (DTPA method)	25	29	345	352
Copper (DTPA method)	2	2	5.4	5.6
Silicon (Hot CaCl <sub>2</sub> method)	31	33	78	67
Boron (Hot CaCl <sub>2</sub> method)	1	1	1.2	1
Exchangeable Calcium	1,078	1,294	1,598	1,439
Exchangeable Magnesium	145	164	346	358
Exchangeable Potassium	60	64	139	130
Exchangeable Sodium	39	43	41	44
Exchangeable Aluminium	1	1	97	89
Exchangeable Hydrogen	2	1	2.6	1.9
Effective Cation Exchange Capacity	4	8	4	9

 Table 13 – Mean Penrith Lakes original soil and THB treated soils. Values in mg/kg unless otherwise stated

Soil carbon levels increased in response to incubation with THB (Table 14). The initial total soil carbon (LECO combustion method) at the Penrith Lakes was low and below the values expected for the original soil type before historic extraction activities. The topsoil *total* carbon value was 0.3% and the subsoil 0.1%. In the surface soil, the *organic* carbon value equated for all carbon pools at 0.3%, with the value below detection limits at <0.1% in the subsoil. Incubation with THB increased the total carbon pool ten-fold from 0.3 to 3.0% in the topsoil and from 0.1 to 2.1% in the subsoil. Total nitrogen also increased following incubation from below detection limits of <0.02 to 0.3%, still within recommended guideline values.

	Original		ТНВ Т	reated
	Topsoil	Subsoil	Topsoil	Subsoil
Total Carbon (%)	0.3	0.1	3.0	2.1
Total Nitrogen (%)	< 0.02	< 0.02	0.36	0.32
Carbon/Nitrogen Ratio	15	5	8.3	6.5
Organic Carbon (%)	0.3	< 0.1	2.5	1.9
Estimated Organic Matter (%)	<1	<1	5.3	3.6
Calcium/Magnesium Ratio	3	5	4.6	2.5

Table 14 – Mean carbon and nitrogen results for Penrith Lakes

The total elemental load, as determined by acid digestion for the Penrith Lakes soil, increased total calcium, sulfur, copper, zinc, and phosphorus following THB incubation (Table 15). Whilst the total elemental load is increased, it does not strictly result in a concern. The bioavailable and water-soluble fractions provide more detail on the potential release and mobility of elements. For the Penrith Lakes site, the sulfur, ammonium, phosphorus, copper and zinc had increased total concentrations and bioavailable fractions.

	Orig	ginal	THB T	reated
	(n =	= 3)	(n =	- 4)
	Topsoil	Subsoil	Topsoil	Subsoil
Total Calcium	2,104	1,381	2,953	1,762
Total Magnesium	1,715	1,429	1,471	1,519
Total Potassium	1,082	705	907	710
Total Sodium	81	169	154	201
Total Sulfur	110	<50	902	694
Total Phosphorus	92	22	3,159	4,395
Total Zinc	47	33	88	57
Total Manganese	532	354	433	501
Total Iron	19,949	18,315	20,880	19,954
Total Copper	14	10	53	40
Total Boron	<2	<2	<2	3
Total Silicon	767	870	389	692
Total Aluminium	8,601	7,024	7,491	7,011
Total Molybdenum	0.58	0.48	0.84	0.92
Total Cobalt	9	8	9.3	7
Total Selenium	0.74	0.68	0.56	0.51
Total Cadmium	< 0.5	< 0.5	< 0.5	< 0.5
Total Lead	14	12	13	13
Total Arsenic	5	5	4	3
Total Chromium	15	13	16	12
Total Nickel	12	10	12	10
Total Mercury	< 0.1	< 0.1	< 0.1	< 0.1
Total Silver	<1	<1	<1	<1

Table 15 - Mean total elemental concentrations for Penrith Lakes. All values in mg/kg

The Penrith Lakes soil microbial diversity was substantially changed in response to the incubation with THB materials (Figure 5). The major phyla were Proteobacteria (>30%), Bacteroidota (>10%), Chloroflexi (>10%), Firmicutes (>5%), Caldatribacterota (>5%), Desulfobacteria (>5%), and Euryarchaeota (>5%). These phyla were all maintained from the THB material into the incubated topsoils and subsoils. Firmicutes and Bacteroidota normally constitute the majority of gut microbiota, and Proteobacteria form a minor portion. These phyla persisted in the incubated topsoils and subsoils, suggesting limited competition from the soil in response to the THB addition. Firmicutes are one of the most common phyla and are reported to be abundant in rhizospheric soils. Bacteroidota interact with phosphorus in the rhizosphere, and Proteobacteria play a role in nitrogen cycling. In comparison, Synergistota, which was present in the THB and is known to be common in wastewater treatment, declined following incubation. Synergistota in the THB was >5%, decreasing to <2% in the incubated soils. This phylum is impacted by aerobic conditions found in the topsoil and, to a lesser extent, in the subsoil.



Figure 5 – Microbial community composition of Penrith Lakes. The biosolid is the original THB material and depths are the finished incubated soils grouped as the 'topsoil' (0-10 cm) and 'subsoil' (30-40 cm)

The Penrith Lakes soil phylum composition was unique compared to the other soils used in this study, with physicochemical data indicating the original soil to be nutrient-depleted. Actinobacteriota and Acidobacteriota, which are both common in the topsoils of the other sites in this study, were <2% in the subsoil and <2% in the topsoil, indicating low persistence in the Penrith Lakes soils. Acidobacteriota represent an underrepresented soil bacterial phylum distributed across nearly all ecosystems and may be involved in reducing nitrogen. Actinobacteriota are essential contributors to the process of plant biomass decomposition and are also ubiquitous in most environmental systems.

### Picton Farm response to THB additions

The landscape of the Picton Farm site has been modified and irrigated on a decadal timescale. The soil at Picton Farm has received irrigation and liquid biosolid addition. In turn, the soil has higher levels of nutrients and elements relative to the other study sites. The Picton Farm soils were circum-neutral (pH 6.44 and 6.52), and following 12 weeks of THB incubation, the pH interactions remained largely unaltered for both the topsoil and subsoils (pH 6.70 and 6.61). A slight increase in soil salinity (EC) following incubation with THB and increased water-soluble sodium, calcium, magnesium and chloride levels (Table 16).

The water-soluble concentrations of sodium, calcium, magnesium, and potassium were the highest for all the investigated sites and increased further following incubation with the THB. These concentrations were at the upper guideline values for the initial soil materials and doubled following incubation with THB. The potential risk of these higher salt levels is balanced against the leaching potential and loss of soluble salts from the soil material with drainage.

	Orig	Original		reated
	Topsoil	Subsoil	Topsoil	Subsoil
рН	6.44	6.52	6.70	6.61
Electrical Conductivity (dS/m)	0.25	0.15	0.35	0.45
Dispersion Index	1	2	0	0
Chloride Estimate (equiv. mg/kg)	127	99	223	176
Soluble Calcium (1:5 water)	1,874	1157	2,221	1,083
Soluble Magnesium (1:5 water)	146	149	400	204
Soluble Potassium (1:5 water)	41	<25	110	49
Soluble Phosphorus (1:5 water)	5.3	<1	7.0	8.2
Phosphorus (Bray 1 mg/kg P)	20	16	49	31
Phosphorus (Colwell mg/kg P)	227	89	574	440
Nitrate Nitrogen (mg/kg N)	5	4	33	31
Ammonium Nitrogen (mg/kg N)	19	16	33	27
Sulfur (mg/kg S)	43	30	522	487
Zinc (DTPA method)	6	3	22	15
Manganese (DTPA method)	45	33	50	41
Iron (DTPA method)	150	94	286	219
Copper (DTPA method)	2	1	13	8
Silicon (Hot CaCl <sub>2</sub> method)	33	33	83	67
Boron (Hot CaCl <sub>2</sub> method)	1	1	1.8	1
Exchangeable Calcium	3,154	2,213	3,910	1,739
Exchangeable Magnesium	203	191	515	358
Exchangeable Potassium	101	53	252	140
Exchangeable Sodium	257	268	242	254
Exchangeable Aluminium	3	2	9	8
Exchangeable Hydrogen	<1	<1	2.8	1.9
Effective Cation Exchange Capacity	22	14	26	18

Table 16 – Mean Picton Farm original soil and THB treated soils. Values in mg/kg unless otherwisestated

The Dispersion Index was 1-2, indicating a stable soil with low potential for soil loss under wetting. This was reduced to 0 for topsoils and subsoils in response to incubation with THB (Table 16). The Picton Farm surface soil effective cation exchange capacity (ECEC) was moderate (22 cmol<sup>+</sup>/kg), the highest ECEC measured in this study, and above expected levels in the Sydney basin. The increase in organic material due to the historic liquid biosolid addition is responsible for the higher ECEC. The ECEC decreased with depth below the organic amendment addition to 14 cmol<sup>+</sup>/kg, which aligns with Sydney-basin values. Incubation with THB increased the ECEC to 26 cmol<sup>+</sup>/kg for topsoil and 18 cmol<sup>+</sup>/kg for subsoil. The exchangeable sodium percentage (ESP) was >200 mg/kg for topsoils and subsoils, equating to an Exchangeable Sodium Percentage (ESP) of 4-8%. The values decreased following THB incubation. The moderate ESP and low Dispersion Index following THB incubation resulted in a soil with low sodicity risk.

The Picton Farm site had a stable and productive soil structure with accrued soil biomass and nutrients over and above the original soil condition. Bioavailable phosphorus levels (measured using both Bray 1 and Colwell approaches) were within the recommended guidelines for Bray 1 and above the recommended guidelines for Colwell in the original topsoil (227 mg/kg). They increased following THB incubation to 574 mg/kg. Nitrate and ammonium nitrogen values increased following THB incubation but remained within guideline values. The initial soil bioavailable manganese and iron (DTPA method) were at the upper limit of recommended levels, whilst copper and zinc exceeded recommended levels. These values all increased in response to THB incubation. The boron and silicon (hot CaCl<sub>2</sub> method) were within recommended levels and typical of western Sydney soils, and these increased in response to THB addition but remained within guideline values. The bioavailable sulfur (KCl method) was higher than recommended for use in pasture systems, and these values increased in response to THB incubatiod. The incubated KCl sulfur values of 522 mg/kg and 487 mg/kg were above the recommended guidelines.

Total soil carbon (LECO combustion method) at Picton Farm was the highest measured in this study, with 5.8% in topsoil and 2.8% in subsoil (Table 17). This is attributed to the historic biosolid injection that has resulted in organic-rich topsoils. The soil *organic* carbon levels are the majority of the total soil carbon pool in the topsoil, at 5.6%, and are over half the total carbon pool in the subsoil, at 1.9%. These values are technically above the desired levels to sustain a healthy soil ecosystem, but are acceptable in a managed farm setting. The THB incubation increased the total soil carbon pool and organic carbon pool. Total nitrogen was within recommended values for the original Picton Farm soil at 0.4% in the topsoil and doubled following THB incubation to 0.8%.

	Orig	vinal	THB Treated	
	Topsoil Subsoil		Topsoil	Subsoil
Total Carbon (%)	5.8	2.8	6.9	3.9
Total Nitrogen (%)	0.4	0.2	0.8	0.5
Carbon/Nitrogen Ratio	14	14	8.8	7.8
Organic Carbon (%)	5.6	1.9	5.8	2.0
Estimated Organic Matter (%)	9	6	12	7
Calcium/Magnesium Ratio	9	8	4	5

Table 17 – Mean carbon and nitrogen results for Picton Farm

The total elemental load determined by acid digestion for Picton Farm soil had increased total calcium, magnesium, potassium, sulfur, copper, zinc, and phosphorus following THB incubation (Table 18). The original Picton Farm soil had the highest total elemental load measured in this study, and this is associated with historic biosolid liquid injection. Total sulfur, phosphorus, zinc, manganese, copper, and iron increased following incubation with THB. Of these increases in total load, the bioavailability data in Table 18 indicated that sulfur, phosphorus, zinc, copper, and iron had greater mobility in the THB incubated soils.

	Ori	rinal	THR Treated		
				reated	
	Topsoil	Subsoil	Topsoil	Subsoil	
Total Calcium	4,413	1,783	6,255	2.189	
Total Magnesium	614	517	938	782	
Total Potassium	848	639	947	891	
Total Sodium	295	111	367	207	
Total Sulfur	272	85	1,197	694	
Total Phosphorus	256	172	3,516	4,395	
Total Zinc	48	41	117	105	
Total Manganese	626	217	678	619	
Total Iron	17,051	12,196	24,549	20,064	
Total Copper	2	1	51	43	
Total Boron	2	<2	2.5	2.8	
Total Silicon	341	274	377	293	
Total Aluminium	13,553	9,183	15,096	10,039	
Total Molybdenum	1	0.2	1.2	0.8	
Total Cobalt	9	8	9.9	7	
Total Selenium	< 0.1	0.2	0.61	0.88	
Total Cadmium	<0.5	< 0.5	< 0.5	< 0.5	
Total Lead	20	9	22	9	
Total Arsenic	6	< 0.5	6.3	< 0.5	
Total Chromium	13	2	18	4	
Total Nickel	10	1	12	5	
Total Mercury	< 0.1	< 0.1	< 0.1	< 0.1	
Total Silver	<1	<1	<1	<1	

Table 18 - Mean total elemental concentrations for Picton Farm. All values in mg/kg
The Picton Farm soil microbial diversity did not substantially change in response to the incubation with THB materials (Figure 6). The THB material contained on average >5% Desulfobacteria, which utilise the available sulfur pool, but these did not persist following the 12-week incubation with the Picton Farm soil. The THB had elevated total sulfur and bioavailable sulfur (DTPA), supporting sulfur utilisation, whereas the Picton Farm soil had lower sulfur concentrations, in line with guideline values.



Figure 6 – Microbial community composition of Picton Farm. The biosolid is the original THB material and depths are the finished incubated soils grouped as the 'topsoil' (0-10 cm) and 'subsoil' (30-40 cm)

The THB also supported Caldatribacterota, which is common in wastewater treatment (>10%), and Euryarchaeota (>5%), which are methanogens associated with carbon cycling. These bacterial and archaeal taxa were diminished following incubation with the Picton Farm soil. While Actinobacteriota were >10% in the subsoil and >5% in the topsoil, they were only <2% in the THB, indicating persistence in the incubated soils. Acidobacteriota were also >10% in the incubated subsoil and >5% in the incubated topsoil, but <1% in the THB material. This provides evidence that the addition of THB to the Picton Farm soil did not alter the existing soil microbial diversity.

## Sydney Science Park response to THB additions

The landscape of the Sydney Science Park site has historically been utilised for agricultural activities, which has resulted in an eroded and depleted soil profile with a low retention capacity. The Sydney Science Park soils were mildly acidic (pH 5.73 and 5.50), and following 12 weeks of THB incubation, the pH remained largely unaltered for both the topsoil and subsoils (pH 5.86 and 5.61). There was an increase in soil salinity (EC) following incubation with THB and increases in water soluble sodium, calcium, magnesium and chloride levels (Table 19). The mildly acidic pH may induce further elemental release to the water-soluble fraction over and above the sodium, calcium, magnesium, and potassium.

	Original		ТНВ Т	reated
	Topsoil	Subsoil	Topsoil	Subsoil
pH	5.73	5.50	5.86	5.61
Electrical Conductivity (dS/m)	0.09	0.06	1.4	1.5
Dispersion Index	2	4	2	2
Chloride Estimate (equiv. mg/kg)	99	83	920	870
Soluble Calcium (1:5 water)	772	481	1,020	981
Soluble Magnesium (1:5 water)	432	421	508	499
Soluble Potassium (1:5 water)	82	81	75	81
Soluble Phosphorus (1:5 water)	2	<1	4.3	4.2
Phosphorus (Bray 1 mg/kg P)	12	4	46	23
Phosphorus (Colwell mg/kg P)	33	17	607	620
Nitrate Nitrogen (mg/kg N)	210	86	443	409
Ammonium Nitrogen (mg/kg N)	540	540 54		905
Sulfur (mg/kg S)	30	22	485	502
Zinc (DTPA method)	3	1	5.4	5.1
Manganese (DTPA method)	18	17	51	62
Iron (DTPA method)	158	92	465	298
Copper (DTPA method)	2	3	4	4
Silicon (Hot CaCl <sub>2</sub> method)	53	51	77	59
Boron (Hot CaCl <sub>2</sub> method)	1	<1	1.4	1
Exchangeable Calcium	1483	1154	1,947	1,382
Exchangeable Magnesium	701	759	667	758
Exchangeable Potassium	147	161	158	164
Exchangeable Sodium	104	81	205	196
Exchangeable Aluminium	2	39	3.2	37
Exchangeable Hydrogen	3	2	3.7	1
Effective Cation Exchange Capacity	15	7	16	8

 Table 19 – Mean Sydney Science Park original soil and THB treated soils. Values in mg/kg unless otherwise stated

The Dispersion Index was 2-4, indicating a stable soil with low potential for soil loss under wetting (Table 19). This lowered in response to THB incubation. The effective cation exchange capacity (ECEC) of the Sydney Science Park surface soil was moderate (15 cmol<sup>+</sup>/kg) and decreased with depth below the organic amendment addition to (8 cmol<sup>+</sup>/kg). The ECEC increased following THB incubation to 16 cmol<sup>+</sup>/kg for topsoil and 8 cmol<sup>+</sup>/kg for subsoil. The exchangeable sodium percentage (ESP) was >200 mg/kg for topsoils and subsoils, equating to

an Exchangeable Sodium Percentage (ESP) of 5-8% following THB incubation. The Sydney Science Park soils had a low potential for dispersion and moderate sodicity potential, and this was improved by THB addition.

The Sydney Science Park site had poor soil structure and limited fertility before THB incubation. Bioavailable phosphorus levels (measured using both Bray 1 and Colwell approaches) were below the recommended guidelines for Bray 1 and Colwell in the original soil materials and increased following THB incubation. Nitrate and ammonium nitrogen values were also below guideline values and increased to above guideline values following THB incubation. The bioavailable trace elements manganese, iron, copper and zinc (DTPA method) exceeded guideline values in the original soil materials. These values all increased in response to THB incubation. The boron and silicon (hot CaCl<sub>2</sub> method) were within recommended levels and typical of western Sydney soils and remained within guideline values in the original soils and increased in response to THB incubation. The bioavailable sulfur (KCl method) was at recommended values in the original soils and increased in response to THB incubation to exceed guidelines. The THB incubated sulfur values of 485 mg/kg in topsoil and 502 mg/kg in subsoil were above the recommended values.

Total soil carbon (LECO combustion method) at Sydney Science Park was 2.1% in topsoil and 0.8% in subsoil (Table 20). The soil *organic* carbon levels comprised 2% in the topsoil and <0.1% in the subsoil. Total carbon values increased in response to THB incubation to 5.3% in the topsoil and 4.9% in the subsoil. The increased carbon value is driven by *organic* carbon at 5.3% in the topsoil and 4.8% in the subsoil. Total nitrogen was within recommended values for the original soil at 0.2% in the topsoil and increased following THB incubation to 0.7%, which is elevated but still within acceptable limits.

			r		
	Orig	ginal	THB Treated		
	Topsoil	Subsoil	Topsoil	Subsoil	
Total Carbon (%)	2.1	0.8	5.3	4.9	
Total Nitrogen (%)	0.2	0.1	0.7	0.5	
Carbon/Nitrogen Ratio	12	9	10	11	
Organic Carbon (%)	2	< 0.1	5.3	4.8	
Estimated Organic Matter (%)	4	1.4	13	10	
Calcium/Magnesium Ratio	3	1	1.7	2	

Table 20 - Mean carbon and nitrogen results for Sydney Science Park

The total elemental load, as determined by acid digestion for the Sydney Science Park soil, increased following THB incubation (Table 21). Only aluminium, silicon, boron, mercury, silver and arsenic did not increase following THB incubation. The Sydney Science Park site has the greatest increased total elemental load following THB incubation. The bioavailable fractions provide more resolution on the potential mobility at the Sydney Science Park site, with sulfur, phosphorus, ammonium, iron, copper, and zinc increased in total and bioavailable concentrations.

	Orig	ginal	ТНВ Т	reated
	Topsoil	Subsoil	Topsoil	Subsoil
Total Calcium	637	584	3,703	3.123
Total Magnesium	575	462	1,245	1,182
Total Potassium	281	228	451	439
Total Sodium	132	103	291	304
Total Sulfur	100	50	1,359	1,562
Total Phosphorus	180	17	3,662	3,491
Total Zinc	5	6	91	88
Total Manganese	423	215	515	501
Total Iron	26,153	17,089	55,041	38,599
Total Copper	1	1	58	62
Total Boron	<2	<2	<2	<2
Total Silicon	288	305	367	354
Total Aluminium	9,994	9,892	12,933	12,843
Total Molybdenum	0.3	0.1	1.3	1.1
Total Cobalt	4	3	7	7
Total Selenium	0.1	0.1	0.7	0.7
Total Cadmium	< 0.5	< 0.5	< 0.5	< 0.5
Total Lead	12	8	22	15
Total Arsenic	7	< 0.5	10	5
Total Chromium	13	<1	26	8
Total Nickel	5	1	9.4	2
Total Mercury	< 0.1	< 0.1	< 0.1	< 0.1
Total Silver	<1	<1	<1	<1

Table 21 - Mean total elemental concentrations for Sydney Science Park. All values in mg/kg

The Sydney Science Park topsoil microbial diversity did not substantially change in response to the incubation with THB (Figure 7). However, the subsoil diversity did alter to align more closely with the THB material. The THB material contained on average >5% Desulfobacteria, which utilise the available sulfur pool. These did not persist following the 12-week incubation with the Sydney Science Park topsoil but did persist in the subsoil incubations. Although the THB material had elevated total sulfur and bioavailable sulfur (DTPA), it did not appear to support sulfur utilisation in the soil. Chloroflexi were present at >10% on average in the THB and this abundance was maintained in the subsoils but not in the topsoils. Members of the Chloroflexi, such as *Dehalococcoides*, which are common in THB, persisted under saturated subsoil conditions.



**Figure 7** – Microbial community composition of Sydney Science Park. The biosolid is the original THB material and depths are the finished incubated soils grouped as the 'topsoil' (0-10 cm) and 'subsoil' (30-40 cm)

The THB material also supported Caldatribacterota, which are common in wastewater treatment (>10%), and Euryarchaeota (>5%), which are methanogens associated with carbon cycling. These bacterial and archaeal groups were diminished following incubation with the Sydney Science Park topsoil but persisted in the subsoil. Firmicutes were dominant (>30%) in the Sydney Science Park topsoil, with lower levels observed in the subsoil and THB material; this phylum is known to be abundant in the rhizosphere. Actinobacteriota were present at >2% in the subsoil and >5% in the topsoil, but at <2% in the THB, indicating persistence in the incubated soils. Acidobacteriota were also present at >5% in the incubated topsoil and subsoil but were <1% in the THB material. This provides evidence that the addition of THB to the Sydney Science Park soil had a limited impact on topsoil diversity but did influence the subsoil microbial community.

## Western Sydney Parklands response to THB additions

The landscape of the Western Sydney Parklands site is that of a low-lying area within the South Creek catchment alluvial deposits. The soil is heavily compacted, and the soil structure has been lost over time. The soils were mildly acidic (pH 6.74 and 6.02) and following 12 weeks of THB incubation, the pH interactions remained largely unaltered for both the topsoil and subsoils (pH 6.21 and 5.59). A tenfold increase in soil salinity (EC) following incubation with THB and associated increases in water soluble sodium, calcium, magnesium and chloride levels (Table 22). The water-soluble sodium, calcium, magnesium, and potassium concentrations were elevated following incubation with the THB. These higher salt levels are balanced against the leaching potential and loss of soluble salts from the soil material with drainage.

	Orig	ginal	THB T	reated
	Topsoil	Subsoil	Topsoil	Subsoil
pH	6.74	6.02	6.21	5.59
Electrical Conductivity (dS/m)	0.06	0.04	0.9	0.5
Dispersion Index	4	6	2	2
Chloride Estimate (equiv. mg/kg)	89	103	579	629
Soluble Calcium (1:5 water)	101	193	1,963	1,981
Soluble Magnesium (1:5 water)	88	97	366	367
Soluble Potassium (1:5 water)	32	<25	79	56
Soluble Phosphorus (1:5 water)	1	<1	8.1	3.9
Phosphorus (Bray 1 mg/kg P)	2	13	46	31
Phosphorus (Colwell mg/kg P)	14	12	499	504
Nitrate Nitrogen (mg/kg N)	1	1	33	29
Ammonium Nitrogen (mg/kg N)	1	3	33	28
Sulfur (mg/kg S)	3	9	589	554
Zinc (DTPA method)	1	1	24	12
Manganese (DTPA method)	2	2	212	214
Iron (DTPA method)	18	67	240	229
Copper (DTPA method)	0	0	12	11
Silicon (Hot CaCl <sub>2</sub> method)	51	30	82	62
Boron (Hot CaCl <sub>2</sub> method)	1	1	2	1
Exchangeable Calcium	478	413	3,716	3,110
Exchangeable Magnesium	84	97	484	454
Exchangeable Potassium	<50	<50	207	201
Exchangeable Sodium	101	80	342	297
Exchangeable Aluminium	6	1	7	5
Exchangeable Hydrogen	3.0	<1	<1	<1
Effective Cation Exchange Capacity	6	3	25	14

 Table 22 – Mean Western Sydney Parklands original soil and THB treated soils. Values in mg/kg unless otherwise stated

The Dispersion Index was 4-6, indicating '*dispersion when wet*' with moderate potential for soil loss under wetting (Table 22). This improved in response to incubation with THB to values of 2, indicating '*stable soil*'. The effective cation exchange capacity (ECEC) of the Sydney Science Park surface soil was low (6 cmol<sup>+</sup>/kg) and decreased with depth to 3 cmol<sup>+</sup>/kg. The

ECEC increased following THB incubation to 25 cmol<sup>+</sup>/kg for topsoil and 4 cmol<sup>+</sup>/kg for subsoil. The exchangeable sodium percentage (ESP) was >300 mg/kg for topsoils and subsoils, equating to an Exchangeable Sodium Percentage (ESP) of 5-8% following THB incubation. The Western Sydney Parklands site had a low potential for dispersion and moderate sodicity potential.

The poor soil physical properties and degraded nature of the Western Sydney Parklands site soils result in unproductive soil with limited soil biomass and nutrients. Bioavailable phosphorus levels (measured using both Bray 1 and Colwell approaches) were below the recommended guidelines for Bray 1 and Colwell in the original soil materials. These increased to acceptable levels for Bray 1 and above the recommended values for Colwell following THB incubation. Nitrate and ammonium nitrogen values were low in the original soil and increased to the upper limits of guideline values following THB incubation.

The bioavailable trace elements manganese, iron, copper and zinc (DTPA method) were below recommended levels in the original soil materials. These values all increased in response to THB incubation to above guideline values. The boron and silicon (hot CaCl<sub>2</sub> method) were within recommended levels and typical of western Sydney soils. These increased in response to THB addition but remained within guideline values. The bioavailable sulfur (KCl method) was at recommended values in the original soils and increased in response to THB incubation to 589 mg/kg and 554 mg/kg, exceeding guideline values.

Total soil carbon (LECO combustion method) at Western Sydney Parklands was 1.0% in topsoil and 0.3% in subsoil (Table 23). The soil *organic* carbon levels were 0.4% in the topsoil and <0.1% in the subsoil. Total soil carbon increased in response to THB incubation to 5.4% in the topsoil and 5.0% in the subsoil. The increased carbon value was driven by *organic* carbon at 5.3% in the topsoil and 5.0% in the subsoil. Total nitrogen was within recommended values for the original soil at 0.3% in the topsoil and increased following THB incubation to 0.7%, which is within guideline values.

	Orig	ginal	THB T	reated
	Topsoil	Subsoil	Topsoil	Subsoil
Total Carbon (%)	0.3	0.05	5.4	5.0
Total Nitrogen (%)	0.03	< 0.02	0.7	0.6
Carbon/Nitrogen Ratio	5	15	10	11
Organic Carbon (%)	0.4	< 0.1	5.3	5.0
Estimated Organic Matter (%)	2	0.5	11	10
Calcium/Magnesium Ratio	1	3	4	4

Table 23 - Mean carbon and nitrogen results for Western Sydney Parklands

The total elemental load determined by acid digestion for Western Sydney Parklands soil showed increased total calcium, magnesium, potassium, sulfur, copper, iron, zinc, and phosphorus following THB incubation (Table 24). The bioavailable fractions provide more

detail on the potential mobility at the Western Sydney Parklands site, with sulfur, ammonium, phosphorus, copper, and zinc increased in total and bioavailable concentrations.

	Ori	ginal	ТНВ Т	reated
	Topsoil	Subsoil	Topsoil	Subsoil
Total Calcium	967	1,045	1,919	1,612
Total Magnesium	223	401	598	683
Total Potassium	255	199	429	329
Total Sodium	45	119	119	304
Total Sulfur	99	121	962	1,562
Total Phosphorus	50	49	3,387	3,491
Total Zinc	5	2	75	88
Total Manganese	1,179	567	1,078	501
Total Iron	8,293	7,921	16,854	18,599
Total Copper	1	1	52	48
Total Boron	<2	<2	<2	<2
Total Silicon	330	348	443	454
Total Aluminium	4,538	3,697	6,265	5,473
Total Molybdenum	<1	<1	<1	<1
Total Cobalt	12	1	14	5
Total Selenium	< 0.1	< 0.1	0.6	0.7
Total Cadmium	< 0.5	<0.5	<0.5	< 0.5
Total Lead	26	4	26	9
Total Arsenic	4	<0.5	3	2
Total Chromium	10	<1	16	4
Total Nickel	3	1	5	2
Total Mercury	< 0.1	< 0.1	< 0.1	< 0.1
Total Silver	<1	<1	<1	<1

Table 24 - Mean total elemental concentrations for Western Sydney Parklands. All values in mg/kg

The Western Sydney Parklands soil microbial diversity was substantially altered in response to the incubation with THB materials. Firmicutes (>5%), Bacteroidota (>10%), Chloroflexi (>10%), Caldatribacterota (>5%), and Desulfobacteria (>5%) were all maintained from the THB into the incubated topsoils and subsoils (Figure 8). These taxa persisted in both incubated horizons and dominated the microbial community. Bacteroidota were dominant members of the soil microbiome and are known to interact with phosphorus in the rhizosphere.



Figure 8 – Microbial community composition of Western Sydney Parklands. The biosolid is the original THB material and the depths are the finished incubated soils grouped as the 'topsoil' (0-10 cm) and 'subsoil' (30-40 cm)

There was a decline in Synergistota, from >5% in the THB to <2% in the incubated soils. Synergistota are common in wastewater treatment environments but are sensitive to aerobic conditions, which are more prevalent in the topsoil and, to a lesser extent, in the subsoil. Actinobacteriota and Acidobacteriota, which are commonly found in the topsoils of the other sites, were each <2% in both the topsoil and subsoil, and also <2% in the THB, indicating low persistence in the Western Sydney Parklands soils. This provides evidence that adding THB to the Western Sydney Parklands soil impacted microbial diversity.

## Western Sydney University Richmond response to THB additions

The landscape of the Western Sydney University (WSU) Richmond site is an alluvial plain under long-term management as a farming enterprise. These historic agricultural practices have resulted in a well-developed, curated topsoil including use of recycled water for irrigation. The WSU Richmond soils were mildly acidic (pH 6.16 and 6.58), and following 12 weeks of THB incubation, the pH interactions remained largely unaltered for both the topsoil and subsoils (pH 6.21 and 6.75). There was an increase in soil salinity (EC) following incubation with THB and increases in water soluble sodium, calcium, magnesium and chloride levels (Table 25). The soil EC increased in line with the increase in soluble cation values. The potential risk of higher EC is balanced against the leaching potential and is not considered a risk in these sandy soils.

	Orig	ginal	ТНВ Т	reated
	Topsoil	Subsoil	Topsoil	Subsoil
pH	6.16	6.58	6.21	6.75
Electrical Conductivity (dS/m)	0.11	0.07	0.9	0.6
Dispersion Index	2	3	2	1
Chloride Estimate (equiv. mg/kg)	78	41	427	413
Soluble Calcium (1:5 water)	301	285	886	917
Soluble Magnesium (1:5 water)	55	52	213	224
Soluble Potassium (1:5 water)	25	25	65	59
Soluble Phosphorus (1:5 water)	5	<1	34	42
Phosphorus (Bray 1 mg/kg P)	83	61	165	168
Phosphorus (Colwell mg/kg P)	102	65	574	504
Nitrate Nitrogen (mg/kg N)	7	7	9	10
Ammonium Nitrogen (mg/kg N)	410	13	527	509
Sulfur (mg/kg S)	20	11	334	313
Zinc (DTPA method)	3	1	16	13
Manganese (DTPA method)	10	4	19	20
Iron (DTPA method)	72	58	317	340
Copper (DTPA method)	0	0	3	4
Silicon (Hot CaCl <sub>2</sub> method)	19	16	44	51
Boron (Hot CaCl <sub>2</sub> method)	1	0	1	1
Exchangeable Calcium	512	365	1,260	887
Exchangeable Magnesium	88	55	250	115
Exchangeable Potassium	<50	<50	102	63
Exchangeable Sodium	118	44	138	101
Exchangeable Aluminium	3	2	5	2
Exchangeable Hydrogen	<1	<1	<1	<1
Effective Cation Exchange Capacity	7	6	10	9

 Table 25 – Mean WSU Richmond original soil and THB treated soils. Values in mg/kg unless otherwise stated

The Dispersion Index was 2-3, indicating a stable soil with low potential for soil loss under wetting (Table 25). This value was not reduced in the topsoil in response to THB incubation, whilst subsoil values improved to an index rating of 2. The effective cation exchange capacity (ECEC) of the WSU Richmond original surface soil was low (7 cmol<sup>+</sup>/kg). The ECEC increased following THB incubation to 10 cmol<sup>+</sup>/kg for topsoil and 9 cmol<sup>+</sup>/kg for subsoil.

Following THB incubation, the exchangeable sodium was >100 mg/kg for topsoils and subsoils, equating to an Exchangeable Sodium Percentage (ESP) of 6-8%. The incubation results for the WSU Richmond site indicated a low potential for dispersion and a moderate to high sodicity potential.

The sandy soils of the WSU Richmond site result in free-draining soils with limited retention capacity. Bioavailable phosphorus levels (measured using both Bray 1 and Colwell approaches) were at the recommended guidelines for Bray 1 and Colwell in the original soil and increased to above-guideline values following THB incubation. Nitrate and ammonium nitrogen values were at recommended values for the original soil, and both increased to above-guideline values following THB incubation. Nitrate and ammonium nitrogen values following THB incubation. The bioavailable trace elements manganese, iron, copper and zinc (DTPA method) were at the upper limit of guideline values in the original soil materials. These values all increased in response to THB incubation to above guideline values. The boron and silicon (hot CaCl<sub>2</sub> method) were within recommended levels and typical of the soils of western Sydney. Both increased following THB incubation but remained within guideline values. The bioavailable sulfur (KCl method) was within guideline values in the original soil and increased to above guideline values to 334 mg/kg in topsoil and 313 mg/kg in subsoil following THB incubation.

Total soil carbon (LECO combustion method) at WSU Richmond was 2.0% in topsoil and 0.1% in subsoil (Table 26). The soil *organic* carbon levels were 1.0% in the topsoil and <0.1% in subsoil. Total carbon values increase in response to THB incubation to 3.1% in the topsoil and 1.4% in the subsoil. The increased carbon value was driven by organic carbon at 2.9% in the topsoil and 1.4% in the subsoil. Total nitrogen was within recommended values for the original soil at 0.1% in the topsoil and increased following THB incubation to 0.4%, which is still within guideline values.

	Orig	ginal	ТНВ Т	reated
	Topsoil	Subsoil	Topsoil	Subsoil
Total Carbon (%)	2	0.1	3.1	1.4
Total Nitrogen (%)	0.1	< 0.02	0.4	0.4
Carbon/Nitrogen Ratio	20	5	8	4
Organic Carbon (%)	1.0	< 0.1	2.9	1.4
Estimated Organic Matter (%)	3.5	0.2	5.4	2.45
Calcium/Magnesium Ratio	1	3	3	4

Table 26 – Mean carbon and nitrogen results for WSU Richmond

The total elemental load, as determined by acid digestion, for WSU Richmond soil had increased total calcium, magnesium, potassium, sulfur, copper, iron, zinc, and phosphorus following THB incubation (Table 27). The bioavailable fractions provide more detail on the potential mobility at the WSU Richmond site, with calcium, magnesium, sulfur, ammonium, phosphorus, iron, copper, and zinc increased in total and bioavailable concentrations.

	Orig	Original		reated
	Topsoil	Subsoil	Topsoil	Subsoil
Total Calcium	276	291	2,041	2,152
Total Magnesium	49	87	380	441
Total Potassium	87	56	162	158
Total Sodium	31	30	131	149
Total Sulfur	100	40	914	876
Total Phosphorus	150	51	2,557	2,395
Total Zinc	5	3	49	37
Total Manganese	2	5	54	61
Total Iron	400	395	5,650	5,593
Total Copper	1	1	31	29
Total Boron	<2	<2	<2	<2
Total Silicon	378	410	441	449
Total Aluminium	818	918	2,300	2,415
Total Molybdenum	<1	<1	<1	<1
Total Cobalt	0.5	< 0.5	0.8	0.6
Total Selenium	< 0.5	< 0.5	<0.5	< 0.5
Total Cadmium	< 0.5	< 0.5	<0.5	< 0.5
Total Lead	1.6	< 0.5	3.7	1.2
Total Arsenic	<2	<2	<2	<2
Total Chromium	<1	<1	4.6	3.9
Total Nickel	< 0.5	< 0.5	2	2
Total Mercury	< 0.1	< 0.1	< 0.1	< 0.1
Total Silver	<1	<1	<1	<1

Table 27 - Mean total elemental concentrations for WSU Richmond. All values in mg/kg

The Western Sydney University Richmond microbial diversity was substantially changed in response to the incubation with THB materials (Figure 9). Firmicutes (>5%), Bacteroidota (>10%), Chloroflexi (>10%), Caldatribacterota (>5%), Desulfobacteria (>5%), and Euryarchaeota (>5%) were all maintained from the THB material into the incubated topsoils and subsoils. These taxa persisted in both incubated horizons and dominated the microbial community.



Figure 9 – Microbial community composition of WSU Richmond. The biosolid is the original THB material and the depths are the finished incubated soils grouped as the 'topsoil' (0-10 cm) and 'subsoil' (30-40 cm)

There was a decline in Synergistota, from >5% in the THB to <5% in the incubated soils. Actinobacteriota were present at >2% in the subsoil and >5% in the topsoil, but were <2% in the THB, indicating persistence in the incubated soils. Acidobacteriota were also present at >2% in the incubated topsoil and subsoil, but <1% in the THB material. This indicates that the addition of THB to the Western Sydney University Richmond soil impacted subsoil microbial diversity, while the original topsoil diversity was largely maintained.

# **Outcomes and Benefits of THB additions**

The use of THB in this experimental program altered the physicochemical and biological properties of the soils following the 12-week incubation period. All study sites had demonstrable physicochemical changes to topsoil and subsoil with increases in carbon, nitrogen, phosphorus, *bioavailable* metals, sulfur, and *total* elemental load. The sites regarded as heavily degraded and/or modified were the Penrith Lakes, Sydney Science Park, and Western Sydney Parklands. These five sites had improvements to baseline physicochemical properties and altered soil microbial biodiversity following THB incubation. The THB incubation resulted in increased nutrient pools, with the transplanted THB microbial biodiversity persisting at Penrith Lakes, Western Sydney Parklands, and WSU Richmond sites for the 12-week incubation. The lack of microbial biodiversity in the original soil at the Penrith Lakes, Sydney Science Park, and Western Sydney Parklands resulted in less competition with the biodiversity in the introduced THB.

The microbiological diversity for some sites did not change in response to THB incubation. These sites were where the pre-existing physicochemical properties of the soil supported microbial biodiversity. The Picton Farm site had the highest total elemental load and nutrient pools in the study. The higher loading was attributed to previous biosolid injection. The pre-existing nutrient pools supported a soil biodiversity that did not alter in response to THB incubation. The Sydney Science Park topsoil also had limited change to microbial biodiversity following THB incubation. The underlying Sydney Science Park subsoil did alter in response to THB incubation, which may be due to the lack of microbial diversity in the heavy clay subsoils.

# Aggregate stability

Soils with dispersive and sodic characteristics are typical in the western Sydney region. The key measurements to understand these behaviours are the Dispersion Index (Loveday and Pyle, 1973) and the 'exchangeable sodium percentage' (ESP). The methodology used for the THB incubation incorporated additional mixing steps to produce a homogenised material. The complete homogenisation of the THB-soil mix under the experimental conditions was not easily achieved, and this may have implications for larger projects (Figure 3). Overall, the incubation of THB with soil materials improved soil aggregate stability.

The Dispersion Index of materials were categorised with values 0-4 being 'stable soil', 4-6 'soil will disperse when wet', 6-9 'strongly dispersive when wet' and 9-14 'may need gypsum'. The pure THB materials had a value of 0, which indicated no dispersion potential. The original Picton Farm, Sydney Science Park, and WSU Richmond sites were within the desired 'stable soil' values (rating between 0-4) for surface and subsoil materials. The Western Sydney Parklands soil materials were within the 'will disperse when wet' category (ratings between 4 and 6) and were lowered in the topsoil and subsoil to a rating of 2 'stable soil' following THB incubation. The Penrith Lakes site had the highest overall ratings with surface soils of 5 'will

*disperse when wet*', ranging to subsoils at 11 '*may need gypsum*'. The application of THB reduced the dispersion values at Penrith Lakes to <3 '*stable soil*'.

The ESP of materials were categorised with values <6% being 'non-sodic', 7-10 'transition to sodic', 10-15% 'moderate to high sodicity' and >15% 'highly sodic'. The pure THB materials had an ESP value of 4.6, which indicated no sodicity concerns. The original topsoils from both Penrith Lakes and Western Sydney Parklands were 'non-sodic' with ESP <6%, and subsoils were 'transition to sodic' with ESP 7-10%. The Picton Farm, Sydney Science Park, and WSU Richmond soils had topsoil ESP values of 'transition to sodic' (values 7-10%). This remained unchanged for the subsoils at Picton Farm, but at WSU Richmond, the subsoils were 'moderate to high sodicity (values 10-15%). The assigned ESP values used to define soil sodicity are broad due to several factors, including electrolyte concentration, pH, organic matter content, and clay mineralogy. The THB incubation did increase the amount of soil exchangeable sodium. However, although the exchangeable sodium amount increased, this was relative to the overall increase in the effective cation exchange capacity and in exchangeable magnesium and calcium. Therefore, overall ESP values did not increase following THB incubation even though sodium levels increased.

# **Acidity and Salinity**

Acidity and salinity levels were determined by the electrical conductivity (EC) and pH probe and elemental analysis of 1:5 (soil:water) extracts. The pH for the THB materials was 6.47. The pH for all soil materials across all sites ranged from 5 to 6.5. The THB incubation resulted in <0.5 pH increase for all soils across the study. Soil salinity in a 1:5 extract (as opposed to bulk paste measurements) is categorised as <0.25 being '*non-saline*', 2.6-0.5 *slightly saline*', 0.5-1.0 as '*moderately saline*', 1.0-2.0 as '*highly saline*', 2.0-4.0 as '*severely saline*', and >4 as '*extremely saline*'. The pure TB material had an EC at 1.3 dS/m, which was '*highly saline*'. The single application of THB introduced a soluble salt load. The EC across all sites and original soils was <2 ds/m and '*non-saline*'. Following THB incubation, the EC levels for Penrith Lakes topsoils and subsoils increased to '*slightly saline*' and Picton Farm, Sydney Science Park, Western Sydney Parklands, and WSU Richmond these increased to '*moderately saline*' levels.

Understanding where the soluble salt load is partitioned in the soil can provide some resolution on the potential for salt mobility over the longer term. The correlation between the Cation Exchange Capacity (CEC) and the sum of soluble salts provides some information on the association between salt and soil mineralogy (Figure 10). As the soluble salt procedure is a 1:5 soil-to-water extraction, only the readily solubilised components are determined, and this can be used to identify potential leaching effects as soil moisture is increased.



Figure 10 – Relationship between mean cation exchange capacity (CEC) and sum soluble salts (Ca, Mg and K) for incubated soils with THB

The soluble salt levels in the soils following THB incubation were at the upper limit for soils of western Sydney. These values would likely reduce in real-world circumstances where leaching occurs, and the salt loading is not an immediate concern where THB application is annualised across 5-year applications.

#### Soil nutrients

The THB application rate was at a maximum of 5-year NLBAR, between 115 and 127 t/ha (Table 10). The pure THB material is predominantly organic matter, and biosolids are recognised to contribute to increased soil organic matter and nutrient cycling efficiency. Yet, multiple factors influence the extent to which these benefits persist. In soil, the carbon fractions, including soil organic matter, organic, and inorganic carbon fractions, play an essential role in soil health and fertility. The organic matter fraction is known to modify different physicochemical reactions and has direct and indirect impacts on nutrient transformations. During soil organic matter and plant residue decomposition, nitrogen can be removed (immobilised) from plant-available forms or released (solubilised or mineralised) into a plantavailable form. The topsoil organic carbon at Picton Farm was the highest in the study due to historic amendment additions and greater than Sydney Science Park, noting both are on the Blacktown soil landscape (9029bt and 9030bt, respectively). The lower bounds of soil carbon at Sydney Science Park, WSU Richmond, and Penrith Lakes were all in the typical range for soils in the Sydney basin, between 0.9 and 2.5%. The Western Sydney Parklands site had SOC levels well below typical ranges and can be considered low from a landscape productivity standpoint. Adding THB increased total carbon and soil organic carbon levels for Penrith Lakes, Sydney Science Park, and Western Sydney Parklands sites, which had low soil carbon levels. These increases were significant and improved the soil carbon pool. The Picton Farm and WSU Richmond sites had modest changes to the soil carbon pool in response to THB incubation.

The significant increases in response to THB incubation in total nitrogen and carbon support its potential as an effective soil conditioner. However, consideration to an excess nutrient loading is a potential risk. For each of the five sites, the overall and mean values for total nitrogen, phosphorus, carbon, and sulfur are provided in Table 28. The mean topsoil total nitrogen increased by 0.38%, total carbon increased by 2.6%, total phosphorus increased by 1,497 mg/kg, and total sulfur by 803 mg/kg across the five sites following the THB incubation.

	Total N	Total C	Total P	Total S	C:N	N:P
THB	3.9	28.0	38,152	9,512	7:1	1:1
Penrith Lakes	0.4	3.0	3,159	902	8:1	1:1
Picton Farm	0.8	6.9	3,516	1,197	9:1	2:1
Sydney Science Park	0.7	5.3	3,662	1,359	8:1	1:1
Western Sydney Parklands	0.7	5.4	3,387	962	8:1	2:1
WSU Richmond	0.4	3.1	2,557	334	8:1	2:1
Mean	0.6	4.7	3,256	1,067	8:1	2:1
Indicative threshold	>0.3	>2.0	400 - 1,500	100 - 1,000	10:1	8:1

Table 28 -Summary total nitrogen, carbon, and phosphorus in soil following THB-incubation

The carbon-to-nitrogen (C:N) ratio is a key factor influencing the decomposition and nutrient cycling of biosolids in soils. Biosolids typically have a lower C:N ratio than plant residues, often ranging between 5:1 and 15:1, accelerating decomposition and nitrogen mineralisation. The original soil results were within the expected ratio for typical Australian topsoils having C/N ratios between 11 and 16. When applied at high rates, biosolids can rapidly increase soil nitrogen availability, but low C:N ratios may also lead to nitrogen losses through leaching and volatilisation, particularly in sandy soils with low organic matter. In contrast, biosolids with a higher C:N ratio, such as those stabilised through composting, decompose more slowly and contribute to longer-term soil carbon sequestration while reducing nitrogen losses.

Optimal C:N ratios facilitate efficient organic matter decomposition and nutrient mineralisation, enhancing soil fertility. Deviations from these optimal ratios can lead to issues such as nitrogen immobilisation or leaching, adversely impacting crop productivity. Soils with C:N ratios higher than this threshold may experience nitrogen immobilisation, wherein microbes sequester available nitrogen, rendering it inaccessible to plants. Conversely, soils with lower C:N ratios may undergo rapid nitrogen mineralisation, increasing the risk of nutrient leaching. Studies indicate that standard biosolids application rates ranging from 16 to 119 t/ha may result in carbon losses through microbial respiration ranging from 53% to 71% within the first 30 days. Net sequestration effects vary, with some systems storing equivalent total carbon to the biosolids application rate, while others exhibit negligible or negative sequestration rates. The original topsoil from Sydney Science Park (11:1) and Picton Farm (15:1) were near ideal

values, with WSU Richmond (20:1), Penrith Lakes (8:1), and Western Sydney Parklands (8:1) at levels just below recommended for a healthy soil. The THB material with a C:N of (7:1) following incubation did not alter Penrith Lakes C:N ratio (8:1) or Western Sydney Parklands (8:1). The THB incubation lowered the C:N ratio for Sydney Science Park (9:1), Picton Farm (8:1) and WSU Richmond (8:1). All variations to C:N were towards recommended ratios.

The optimal N:P ratios range between 10:1 and 20:1 for balanced nutrient availability. Ratios below this range may indicate nitrogen limitation, leading to stunted growth and reduced yields. In contrast, ratios above it can suggest phosphorus limitation, potentially causing poor root development and delayed maturation. The original soil from Picton Farm (16:1) and Sydney Science Park (11:1) was within optimal ranges. Whilst WSU Richmond (7:1) and Western Sydney Parklands (6:1) were below optimal, and Penrith Lakes (2:1) was well below optimal, indicating a relative excess of phosphorus. The THB incubation increased the nitrogen and phosphorus loading to the soils. The THB N:P ratio (1:1) may lead to a disproportionate increase in phosphorus relative to nitrogen if used as a soil amendment, potentially necessitating additional nitrogen inputs to balance the nutrient profile. Following THBincubation with the soil, the N:P for all sites was lowered to a consistent ratio of 2:1 for Picton Farm, Sydney Science Park, WSU Richmond, and Western Sydney Parklands, with Penrith Lakes at 1:1. This was well below optimal, indicating a relative excess of phosphorus. Nitrogen availability is often limited in soils with disproportionate phosphorus levels, potentially reducing vegetative growth due to insufficient nitrogen to support plant growth. An imbalance may also impede microbial activity and disrupt nutrient cycling processes, as the surplus phosphorus can alter microbial community structures and reduce organic matter decomposition rates.

Unlike carbon and nitrogen, phosphorus from biosolids is more resistant to losses but can still contribute to environmental contamination if applied in excess. Research shows that biosolids contain 70-90% inorganic phosphorus, which is primarily retained in the soil due to strong adsorption onto iron and aluminium oxides. However, excessive biosolids application can lead to phosphorus saturation, increasing the risk of runoff losses, particularly in landscapes prone to erosion. The bioavailability of phosphorus also varies depending on the biosolids stabilisation process; for example, heat-dried biosolids tend to have lower P solubility, whereas biologically treated biosolids release phosphorus more readily. To explore mobility within the THB and incubated soils, 'plant available' phosphorus was measured using two different methodologies (Colwell and Bray 1), with both having values for the original soil being near the recommended values for productive soil (Table 29). The Bray 1 and Colwell approaches aim to determine the bioavailable phosphorus in soil, with Bray considered more suitable for mildly acidic soils and Colwell more suitable for circumneutral soils.

The THB incubation results show that a reserve of 'plant available' phosphorus is available where sufficient topsoil is present at Picton Farm, Sydney Science Park, and WSU Richmond topsoils, where larger pools of potentially bioavailable phosphorus are present, whilst the Penrith Lakes and Western Sydney Parklands pools of phosphorus are limited. Following incubation with THB, both Bray and Colwell P values increased tenfold. The Bray 1 test is

more effective at determining bioavailable phosphorus in the mildly acidic soils across the sites. Following THB incubation, the pH of the soil increased, and Colwell extraction may provide more accurate estimations of phosphorus mobility. Both analytical techniques were applied, and the original soil materials had lower Bray 1 values, with a mean across all five sites of 26 mg/kg and Colwell at 81 mg/kg. Post THB-incubation, the Bray 1 values increased with a mean across all five sites of 73 mg/kg and Colwell at 574 mg/kg. The Colwell P was greatest in the pure THB material and is the best estimation of bioavailable phosphorus for soils at neutral to alkaline pH, which is appropriate for the soil incubated with THB, as the pH reached circumneutral. The overall phosphorus loading to the soil is high, however Bray 1 and Colwell results indicate that phosphorus may be primarily in a recalcitrant phase and not immediately bioavailable and the water-soluble phosphorus (1:5 water extraction) provide some support for this argument with values not higher than the recommended. This indicates that immediate phosphorus loss due to leaching may not be an immediate risk with phosphorus being retained in the soil/THB matrix.

	Soluble P	Bray 1	Colwell	Ammonium	Nitrate	Sulfate
THB	51	434	1447	2765	3.7	4909
Penrith Lakes	7	58	617	92	0.48	126
Picton Farm	6	49	574	33	33	522
Sydney Science Park	7	46	607	882	443	485
Western Sydney Parklands	4	46	499	30	31	589
WSU Richmond	5	165	574	527	9.0	334
Mean	6	73	574	313	103	411
Indicative Threshold	12	45	80	20	15	10

 Table 29 –Summary bioavailable phosphate, nitrogen, and sulfate following THB-incubation Values are in mg/kg

Soil mineral nitrogen has the dominant forms of nitrate and ammonium. The bioavailable nitrogen pools were extracted using a KCl approach along with sulfur. The original topsoil ammonium and nitrate levels were low for all sites except for Sydney Science Park and WSU Richmond, where residual fertiliser likely contributes to higher nitrogen loading to the soil. The nitrate anion is considered the more reactive and mobile pool for nitrogen in soil systems, with rainfall being the dominant input to soil systems. Soil nitrate levels remained below recommended values following THB incubation. The measured ammonium levels for the original soil material were within expected ranges for open grassland areas in Australia, except for WSU Richmond and Sydney Science Park. Ammonium is typically the larger pool for soil nitrogen, is considered less mobile, and is more likely to be retained by the soil. The ammonium and nitrate levels for all sites were above guideline values, but land management practices confounded the data.

The total sulfur levels increased with a mean value above recommended values. The bioavailable sulfur was determined using the KCl approach, with the original topsoil having a mean across the five sites of 27 mg/kg. The sulfate levels were within range for Penrith Lakes, Western Sydney Parklands and WSU Richmond soils before THB incubation. Higher sulfate was found in the original Picton Farm and Sydney Science Park soils due to their relatively higher organic matter content. Following THB incubation, this increased to a mean of 411 mg/kg and was above recommended levels and may impact some plant species. The bulk of sulfur in soil is present within organic matter and consequently, biological activity regulates sulfate availability. Whilst the total sulfur pool increased and was still within guideline values. Bioavailable sulfur levels increased and exceeded guideline values. Bioavailable sulfur content in the THB and may become mobile with organic matter decomposition over time in real-world applications.

# Soil contamination

The THB materials used in this study have a low potential for contamination. The key concerns for contamination were nitrogen (primarily ammonium), phosphorus, sulfur, and DTPA (bioavailable) arsenic, iron, zinc, manganese and copper. While the CLBAR approach focuses on total concentration, bioavailable fraction extraction techniques such as DTPA highlight the potential bioavailability within the total pool. The DTPA results highlight that even if *total* concentration is low, the potential *mobile* fractions are higher than recommended. In a broadscale application sense, these elements would be mobile and available for biologic uptake. The total elemental load determined by acid digest did not identify any heavy metal contamination issues, although chromium, lead, nickel, and cobalt were detected in the THB. Soil screening for organic contaminants was undertaken with no result above detection limits. The PFAS compounds were investigated, with only perfluorooctanesulfonic acid (PFOS) detected at low levels in the pure THB material and undetectable in the THB-soil incubation due to the dilution effect. The PFOS concentrations in the THB material, with reference to the NEMP 3.0, are considered limited in risk. Still, attention should be paid to monitoring when repeated applications are to be implemented.

The DTPA approach is often utilised to determine the bioavailability of iron, manganese, copper and zinc. The THB incubation increased bioavailable iron, copper, manganese, and zinc for all five sites. These bioavailable fractions are associated with organic matter and may be released during the decomposition of the THB in the soil. Subsequent mobility in the soil is dictated by cation exchange, pH, and oxidation potential. Following THB incubation, the DTPA zinc, manganese, iron, and copper were all above recommended levels with iron being ten times greater than recommended (Table 30).

	Zinc	Manganese	Iron	Copper	Silicon	Boron
THB	121	1,250	2,434	55	443	13
Penrith Lakes	9.9	129	349	5.5	73	1.1
Picton Farm	19	46	253	11	75	1.5
Sydney Science Park	5.3	57	382	4	68	1.2
Western Sydney Parklands	18	213	235	12	72	1.5
WSU Richmond	15	20	329	4	48	1.0
Mean	13	93	309	7	67	1.2
Threshold	6	25	25	2	50	2

Table 30 - Summary bioavailable zinc, manganese, iron, copper, silicon and boron. Values in mg/kg

To a lesser extent, boron and silicon mobility can also be determined using the hot  $CaCl_2$  approach. Boron deficiency is typical in western Sydney soils, and the original soil materials had low boron levels at <1 mg/kg (hot  $CaCl_2$  approach). The THB boron content was 13 mg/kg, and the THB incubation increased soil boron levels to 1 mg/kg, which is ideal for plant/vegetation health. Potential silicon mobility, including soluble and colloidal materials, increased in response to THB incubation to threshold limits.

#### Soil microbial biodiversity drivers

The key physicochemical parameters that drive microbial biodiversity in the incubated soils were modelled, and redundancy analysis was undertaken to identify key influences on soil microbial diversity (Figure 11). While biosolids provide a slow-release source of organic and inorganic N, significant losses occur through volatilisation, leaching, and denitrification. High biosolids application rates (exceeding 70 t/ha per year) have been shown to shift soil nitrogen cycling dynamics, leading to increased organic nitrogen dominance and reducing nitrate availability for plant uptake. Denitrification also contributes to atmospheric N<sub>2</sub>O emissions, and gaseous nitrogen losses are strongly linked to microbial community structure and oxygen availability in the soil matrix. The decomposition and microbial transformation of biosolids-derived organic matter create dynamic nutrient fluxes, which can result in substantial C and N losses. Carbon loss primarily occurs through microbial respiration, which converts a portion of added organic matter into CO<sub>2</sub>. While biosolids increase soil organic carbon, particularly in agroecosystems with low initial carbon stocks, microbial mineralisation often accelerates the release of CO<sub>2</sub>, reducing long-term carbon stabilisation.



Figure 11 – Redundancy analysis for microbial community and physicochemical properties. Pure THB is identified as 'control' in yellow highlight

The redundancy analysis identifies the key drivers as ammonium (left side) to nitrate (right side), acidity (downwards), and sodicity (upwards). The oxidation potential of the soil regulates the nitrogen cycling in the soil, and this provides some separation between the microbial diversity in the Penrith Lakes, Western Sydney Parklands and WSU Richmond incubations (left side) when compared to the Picton Farm and Sydney Science Park (right side). The acidity (downwards) separates the Penrith Lakes and Picton Farm from the sodicity of Sydney Science Park, Western Sydney Parklands, and WSU Richmond incubations. The exchangeable sodium percentage measures sodicity, and as sodium is a base cation, it is typically associated with neutral to alkaline conditions and is related to soil alkalinity.

The outcome of the redundancy analysis indicates that the Picton Farm and Sydney Science Park sites had a microbial biodiversity that is robust enough to introduce microbial populations. However, the Penrith Lakes, Western Sydney Parklands, and WSU Richmond sites were heavily influenced by the microbial biodiversity introduced via the THB incubation. This provides insight into the potential to add THB to degraded landscapes and introduce microbial biodiversity, which may expedite soil health improvements. The possibility of taking THB and influencing the microbial diversity to suit potential applications to soil systems is a possible avenue of further research, noting the potential low risk of pathogen contamination from the THB material.

## **Broadscale THB application considerations**

A summary of broadscale management considerations is provided in Table 31 with interpretation provided here. The addition of THB can be beneficial when applied to soils with existing poor soil fertility. The primary benefits of THB were the nutrient loading (nitrogen, phosphorus, and potassium) and the aggregate stability. The nutrient-loading benefits of THB need to be balanced against the bioavailability and potential mobility of these nutrients, which may result in rapid export/loss following THB decomposition. The bioavailable sulfur and trace metals in the THB are also a potential risk and should limit the amount of THB applied to land. The nutrient load in THB supported the initial THB microbial biodiversity and allowed for its persistence in soil. When applied to soils with low nutrient status, this diversity persisted for the duration of the 12-week experiment. When applied to soils with moderate to high nutrient status the THB microbial biodiversity was not maintained.

The THB also provide the positive benefit of aggregate stability, which aids in providing physical support to dispersive and erodible soils. The Dispersion Index was improved in the THB incubated soils, and this is an important finding when considering western Sydney soils are recognised for their levels of degradation and sodicity. The THB provides structural aggregate support and improves soil cation exchange capacity, which is also typically low in western Sydney soils. The increased cation exchange capacity is a result of the organic matter in the THB, and sodium is present on exchange sites. This places some constraint on the feasibility of utilising THB on extremely sodic soils.

Management considerations need to balance the existing soil constraints and limitations, the elemental loading and bioavailability of the THB product, THB mixing and homogenisation with soil, the soil microbial ecosystem upon application, and the persistence and retention of THB organic matter over time. These considerations are all part of the broader guidelines and regulatory frameworks for the use and application of THB on land surfaces. These considerations are outlined in Table 31. THB application must therefore be carefully managed to optimise nutrient retention while mitigating losses. The interaction between biosolids and soil microbial communities is central to nutrient transformation processes, influencing the carbon mineralisation rate, nitrogen cycling, and phosphorus retention rate. The balance between nutrient availability and loss is affected by soil texture, climate, and the chemical properties of the biosolids applied. Long-term studies highlight the need for adaptive management strategies, where biosolids application rates are adjusted based on site-specific conditions to minimise adverse environmental impacts. Developing improved biosolid products, such as the THB processing technologies, may further refine nutrient retention and reduce environmental risks. The results in this report demonstrate that even at high raw NLBAR application rates, nutrient loading is indeed high, but not extraordinary. Ongoing research should explore the long-term interactions between biosolids amendments, soil microbial populations, and biogeochemical cycles to refine best management practices and optimise biosolids land application, particularly in degraded landscapes.

### **Contaminant and Metal Mobility**

The CLBAR results showed that copper is the limiting metal, but the limitations were at application rates >100 t/ha for the five sites and were greater than the raw NLBAR. No other CLBAR limitation existed for common contaminants, and the revised PFAS guidelines, when applied to the THB materials, did not present a significant limitation to application rate. PFOS was the only PFAS compound detected in the THB and was below all guideline thresholds after soil mixing. According to national guidance (HEPA, 2025), biosolids reuse must apply appropriate margins of safety and consider site-specific environmental values. While THB presents a low immediate risk from PFAS, monitoring for cumulative loading is recommended, particularly if reuse is repeated over time. Post-incubation analysis indicated elevated soil DTPA bioavailable iron, manganese, copper, and zinc. These bioavailable metals in the THB and soil matrix suggest that trace element mobility may increase under field conditions, particularly where decomposition and organic matter turnover are high. While total element concentrations did not exceed relevant limits, the potential for trace element movement in surface or drainage waters requires further consideration, especially in sandy or low pH soils.

#### **NLBAR** limitations

In this report, the THB was applied at rates aligned with the *raw* 5-year nitrogen-limited biosolids application rate (NLBAR), which ranged from 115 to 127 t/ha. This is compared to the adjusted NLBAR, where residual nitrogen at each site lowers the overall NLBAR to values from 19 to 121 t/ha. These NLBAR rates reflect the maximum allowable application under current nutrient management guidelines, and the findings from this study support their agronomic relevance. Across all five sites, THB improved soil condition, particularly in soils with low fertility and poor structure. Increases in total nitrogen, total and bioavailable phosphorus, sulfur, and soil organic carbon were consistently observed, with the most substantial gains occurring in degraded soils. Improvements in aggregate stability and microbial richness were also noted, reinforcing the role of THB as a soil conditioner.

The application of THB altered elemental ratios in the soil, with important implications for nutrient cycling. The THB material had a low carbon-to-nitrogen (C:N) ratio of approximately 7:1, which shifted all soils towards narrower C:N values (8:1 to 9:1) after incubation. These lower ratios can support enhanced microbial turnover and nitrogen mineralisation but may also increase microbial respiration and carbon loss as CO<sub>2</sub>, potentially limiting long-term carbon retention. Similarly, the high phosphorus content of THB relative to nitrogen resulted in a product nitrogen-to-phosphorus (N:P) ratio of 1:1, well below the recommended agronomic range of 10:1 to 20:1. Following incubation, all soils exhibited similarly low N:P values, suggesting that, if used alone, THB may induce nitrogen limitation and phosphorus oversupply. Two standard extraction methods were used to better understand the potential bioavailability of phosphorus following THB application: the Colwell method, suited to circumneutral soils, and the Bray 1 method, better suited to mildly acidic soils. Both extractions confirmed increased plant-available phosphorus content of the THB material.

Field-scale implementation of THB requires careful consideration of soil type, nutrient demand, water balance, and baseline condition. To ensure long-term sustainability, annual monitoring should extend beyond traditional nutrient and metal parameters to include microbial indicators (including pathogens) and trace element bioavailability. Given the emerging evidence of microbial risks, particularly in sensitive environments or food production zones, a precautionary monitoring framework is recommended. THB presents a valuable resource for improving soil health, but its broad-scale application must be grounded in sound planning and regulatory compliance.

### Soil Structural and Fertility Benefits

The soils of western Sydney are broadly recognised as being limited by sodicity issues. TheTHB improved soil structure at all sites by reducing dispersion indices and enhancing aggregate stability. These effects are most beneficial in erosion-prone and sodic soils, such as those found at Penrith Lakes and Western Sydney Parklands. The inherent stability of THB material contributed to these gains and suggests that structural benefits can be achieved when THB is applied in solid form and when consideration is given to incorporation at appropriate depths. The addition of THB increased exchangeable sodium, which may pose a structural risk in soils with shallow A-horizons or those already classed as sodic or natric. However, THB also contributed calcium and magnesium and increased the effective cation exchange capacity, offsetting sodicity. Where sodicity risk is identified, co-application of gypsum or the use of subsoil mixing strategies can reduce the likelihood of structural breakdown and erosion. The improvement in slaking and dispersion results is a promising improvement to the overall soil condition. However, caution should be applied to avoid application to exposed subsoils or compacted clay horizons due to the potential existing sodicity issues being exacerbated by additional salt load.

In parallel, THB added significant amounts of organic carbon and nutrients, supporting microbial activity and improving the physical quality of nutrient-poor soils. These effects were more pronounced in sites with low baseline fertility, with evidence of persistent microbial communities introduced with the THB that persisted through incubation. This study cannot confirm whether these communities persist in longer-term and real-world applications. The THB incubation altered the microbial composition of all soils, particularly in degraded sites with low initial microbial richness. In these soils, the microbial communities introduced during the THB incubation persisted and contributed to increased microbial diversity. In contrast, in higher fertility soils (e.g., Picton and Sydney Science Park), the existing soil microbiota appeared more resistant to change. Recent metagenomic research has highlighted that biosolids may increase the abundance of antibiotic resistance genes and virulence factors in soil. Although these organisms were not detected in this study, the persistence of biosolids-derived microbiota, particularly in low-fertility soils, suggests a need for cautious application in sensitive locations. Application strategies should aim to support beneficial microbial development while avoiding repeated inputs that could drive unwanted microbial shifts. Annual microbial monitoring is recommended to be included along with the traditional soil chemical monitoring suite. Table 31 provides an overview of these management considerations.

Table 3	1 –	Consid	lerations	when	considerin	g THB	application	when	working	with	regulatory	guidelines
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Identified issue	Potential Risk	Revised Risk	THB Management Options
Homogenisation of THB and soil	Incomplete mixing between added THB and soil materials. May result in soil instability and high nutrient zones.	The homogenisation between added THB materials (as a moist solid) and soil materials in this study required additional mixing steps under laboratory conditions. Increasing moisture content aided mixing and the visual appearance of homogenisation. However, when re-dried the THB:soil mix was shown to not be homogenised. Additional tumbling was required which may be difficult in non-laboratory settings.	Use of THB as a dry product (moisture levels ~40% as exist in finished product supplied) may require additional mixing for complete incorporation into soil. Consideration to soil mixability to be encouraged in designing deployment programs. Slurry/injection approaches may improve mixability to soil but may reduce aggregate stability benefits provided by THB. Consideration to overall soil depth for THB addition to soil as per standards. Consideration to planting (root depth) and potential for roots to contribute to mixing and/or be impacted by the THB clumping.
Water balance	The narrow range between known rainfall and pan evaporation rates within the Sydney-basin.	<ul> <li>The pre-existing physical soil restrictions at Penrith Lakes, Sydney Science Park and Western Sydney Parklands may result in water logging.</li> <li>Application of THB materials may increase hydraulic conductivity if used appropriately. This may improve free drainage.</li> <li>Application of THB may retain/improve soil moisture where burial depth is below root zone. This may assist in water retention in some soils. This may also promote anaerobic zones where excess THB is applied or where homogenisation is incomplete.</li> </ul>	<ul> <li>Water balance calculations used for irrigation programs at Picton Farm and WSU Richmond should be applied to potential THB sites.</li> <li>Modifications to improve shallow topsoil and A-horizons to be considered prior to THB addition. THB application rates to meet recommended guideline values. Higher dosage rates may be possible where homogenisation with soil is possible.</li> <li>Post-application investigations in THB homogenisation with soil and assessment of water retention, sodicity, salinity, redox status, and soil microbiome should be undertaken on a yearly basis. Repeat application of THB materials to a site should consider these parameters prior to re-dosing.</li> </ul>

Soil type T an ra ex pr T	The existing data and analysis in this report show a ange of soil types. Pre- existing soil constraints may provide variable response to THB addition.	Soil types are originally mapped as Kurosols ranging to <i>natric</i> Kurosols and the <i>natric</i> component was present in the studied soils for all sites, but the acidic characteristic was not always present. The definition for Kurosol which is present in at the Sydney Science Park, Western Sydney Parklands, and Western Sydney University Richmond site is: <i>Soils with <u>strong texture contrast</u> between A</i> <i>horizons and <u>strongly acid B</u> horizons. Many of these soils have unusual subsoil chemical features (high Magnesium, Sodium and Aluminium).</i> As reported in this study, no site other than the Sydney Science Park had a soil pH <5.5, meaning all other sites did not meet the 'strongly acid' criteria. For a 'natric' Kurosol the strong texture contrast soil also presents with having an exchangeable Sodium percentage (ESP) greater than 6% and is present in some soils. The definition for Dermosol which dominates the Penrith Lakes and Picton Farm sites is: <i>Soils with <u>structured B2 horizons</u> and <u>lacking a strong texture-contrast</u> between the A and B <i>horizons. Although there is some diversity</i> within the order, it brings together a range of soils with some important properties in <i>common.</i> The elevated ESP at Penrith Lakes classifies as <i>natric</i> and the shallow subsoil is a potential sodicity risk.</i>	Acidity was not a significant risk in the studied soils contrasting existing soil data. Where soil acidity is present the use THB may act as a mild buffer. The potential for THB pH to lower in response to material decomposition/oxidation was not evident in this study but caution to potential organic acid production in response to prolonged drying periods should be tested. Where <i>natric</i> soils are present the potential risk of sodium addition from THB is a risk. Soils with elevated ESP should be avoided. This is especially important where shallow A-horizons into sodic B-horizons exist such as occur at Penrith Lakes. Sodium levels are a concern in the THB. This is balanced by a high cation exchange capacity in the THB and the sodicity ratio is balanced against corresponding levels of calcium and magnesium. In non-sodic soils where the THB is managed and soil moisture is maintained to avoid rapid decomposition, potential sodicity risk is low. Improvements to overall soil depth and development of deeper A-horizons may decrease risk associated with <i>natric</i> soils. The Dispersion Index is low for THB. The aggregate stability is a potential benefit to soils that have moderate soil dispersion. The THB aggregates provide a supporting architecture to the soil. These aggregates should be maintained when deployed to soils. Slurry/injection type deployment programs where the aggregate is eliminated may not provide this benefit. Monitoring of surface and subsoil ESP and dispersion should be undertaken at an annual interval just upon amendment anplication
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Soil sodicity (ESP %).	Risks include but not limited to: surface-sealing, loss of unbedded topsoils, impeded	Soil sodicity issues arise where sodium ion accumulates, and the exchangeable Sodium percentage (ESP) is elevated.	THB materials increase sodium load to soils. This is counter-balanced by increasing CEC and improving dispersion index.
	water holding capacity due to compaction.	An increase in soil sodicity (as ESP) may have negative effects including a decline in soil aggregate stability, swelling and dispersion of clay colloids, an increase in surface sealing, hard	The transplant of 'heathy' soil materials to areas where topsoil is shallow ( $<10$ cm) may expediate soil microbiome repair that may aid in soil stability.
		pan formation, and susceptibility to erosional activities. These structural instability problems are common in soils irrigated with saline-sodic	Three general improvements that would be needed to support implementation of an irrigation program are:
		water.	1) Ensure A-horizon depth is >80cm when identifying sites for THB application. Where >80cm increase the soil depth through transplant of 'heathy' soil materials.
			2) Application of low-dose calcium amendments (e.g. gypsum) to reduce soil sodicity with THB. THB is a moderate sodicity risk which is not a direct concern, however decomposition of organic matter may encourage release of sodium to the soil.
			3) Avoiding dehydration of the THB product. Application at depth and management of soil moisture will support THB organic matter. Caution to be applied to potential for over-watering which may have a negative consequence of encouraging anaerobic conditions where soil free drainage is poor.
			Monitoring of surface and subsoil pH along with ESP and dispersion should be undertaken at an annual interval just prior to amendment application and monitored over time.
			Any use of irrigation waters, any fertiliser, or amendment should be assessed for salinity and sodium levels prior to use with THB. Water balance assessments to be performed to understanding leaching and nutrient retention over time.

Potential for soil dispersion and erosion due to subsoil dispersion	Sheet, rill, gully, tunnel erosion events because of soil dispersion.	For the studied sites at Penrith Lakes, Sydney Science Park, and Western Sydney Parklands sites have high erosion potential as the subsoil is often very close to the surface. Where a shallow A-horizon is present above a sodic B-horizon there is a likelihood of erosion events to occur. The shallow A-horizons are a result of long-term land degradation and exposure of the underlying B-horizons induce instability. The use of THB with sodium/potassium on exchange sites may increase soil structural breakdown and may induce dispersion and erosion. This is counterbalanced with the low Dispersion Index and aggregate stability of the THB which may aid in soil coherence. The Dispersion Index for all soils was lowered in response to THB addition.	Erosional activities are evident at Penrith Lakes, Sydney Science Park, and Western Sydney Parklands. Exposure of sub-surface areas will be an on-going problem if ground cover is not maintained. This is currently achieved at Picton Farm and WSU Richmond. The THB material contains excess sodium and potassium on exchange sites. This is a potential risk for sodicity and salinity. Deployment to soil at low THB:soil ratios may reduce soil dispersion. This is achieved through increased soil aggregate stability. The addition of calcium amendments (e.g. gypsum) to the THB to aid in further lowering sodicity risk is recommended. Monitoring depth to sodic subsoils and measurement of sodicity is required to avoid erosional events that may impact future land use. The improvement of soil depth and improvement of soil drainage may decrease the risk for irrigation.
Soil acidity and salinity	High salinity levels may restrict biomass production. Mobility of salts may result in areas of spatial concentration. Acidity issues may arise in depleted heavily leached soils where buffering capacity is exploited.	Acidity and salinity levels for all the sites was lower than expected and these issues are not a risk for the sites currently. Salinity levels were also low for the sites not considered a risk. The use of THB may introduce salinity to the soil system. Soil acidification presents as a lower soil pH (<5.5), and this is not the case in the studied soils. The potential for stored acidity was evaluated by measuring cation exchangeable H <sup>+</sup> and Al <sup>3+</sup> equating to <1% of total exchangeable cations. The THB materials have elevated EC salinity and water-soluble ions. These increased the EC levels of the THB incubated soil.	Investigation has shown that salinity and acidity risks are minimal across the sites. There is limited risk associated with stored acidity in the soil materials. Acidity is not considered a significant risk in the studied soils. The potential for THB pH to lower/acidify in response to material decomposition/oxidation was not evident in this study but caution to potential organic acid production in response to prolonged drying periods and organic matter decomposition should be tested. The THB increase soil salinity levels. Application rates should minimise the salinity loading to soil. The soil moisture and salt should be monitored. Particular attention to water movement through deeper soil layers where sodicity is identified. Salt accretion in the soil profile may occur if/when soil structural collapse (e.g. through sodicity) limits water infiltration. Water balance calculation and induced leaching to remove excess salts is a common practice utilised at Picton Farm

			and WSU Richmond and used as a best practice approach for other sites.
Soil drainage limitations	Poor subsoil infiltration rates. Increase sub-soil bulk density. Heavy clay textures in sub-soil.	The Penrith Lakes, Sydney Science Park, and Western Sydney Science Park had subsoil drainage constraints. This may result in water retention and lack of effective drainage. This occurs where a restrictive (typically heavy clay) layer exists impeding downward flow	Soil drainage to depth may be impeded where soil structural breakdown occurs, and this is common in sodic soils. This results in water pooling/ponding at shallow depth and the potential for lateral water movement.
		Use of excessive volumes of irrigation water may result in water logging of the soil and associated impacts on redox and biogeochemical processes in operation within the soil profile	Applications to address sodicity and dispersion issues should be monitored to investigate potential improvements to soil deep drainage and soil moisture. Any improvement to soil sodicity will result in an improvement in soil drainage at the site.
		the son prome.	Where soil A-horizon is shallow water storage limits are likely to be low and water logging is likely to occur. This can be improved with soil re-engineering and use of THB to improve infiltration and water storage. Caution needs to be applied to this approach but may result in benefit to soil function if utilised at low THB application rates.
			Soil moisture should be monitored. Particular attention to water movement through deeper soil layers where salinity and sodicity are identified.
Soil nutrient levels	Risk of plant growth and fertility restrictions based on soil nutrients	The soils from Penrith Lakes, Sydney Science Park, and Western Sydney Parklands had limited A-horizons and topsoil depth was shallow. This limits the capacity of the soil to accrue and retain nutrients.	The THB incubation did result in higher soil C, N, P, S, and K. Caution should be prescribed for application rates to soil due to these increases. The total and bioavailable S in the THB is the restricting constraint when deciding land application rates.
		The THB had appreciable bioavailable forms of C, N, P, S, and K. Bioavailable forms are available for uptake and are generally considered to be more mobile in the environment. Boron was present in THB and may provide benefit to vegetation.	Increases to bioavailable soil N and P may result in nutrient export where land application rates are above nutrient retention capacity of the soil. N and P export should be monitored. At sites other than Picton Farm, P is limited (which is not uncommon for Australian soils) and N is not efficiently utilised. Consideration to

		The C:N ratio ranged from 5 to 20 in the soils across the western Sydney sites. Ideal soil values are between 10-12. The C/N ratio for THB is 7.2. THB incubation moved soils towards C/N ratios towards ideal ranges.	N:P:K:S ratio for biomass productivity needs to be assessed when N utilisation is to be maximised. The total boron levels are recognised to be low in Sydney soils. The addition of boron via THB may serve a beneficial purpose for biomass production.
Soil contaminants	The retention of contaminants from THB may result in toxicity issues within the soil profile.	<ul> <li>Bioavailable nitrogen, phosphorus, sulfur, zinc, copper, manganese, iron are present in the THB at levels that limit potential application rates to land.</li> <li>The presence of PFAS were detected in THB as PFOS. These levels were low and decreased to below detection levels once mixed with soil (dilution effect). THB application may result in cumulative loading of PFAS in soil, especially with repeated use.</li> <li>Subsoil leaching of trace metals such as copper and zinc has been documented in long-term field sites. Anaerobic conditions may promote sulfur mobilisation from biosolids.</li> </ul>	The bioavailability of elements in the THB is associated with organic matter breakdown. The bioavailability of nutrients N and P may restrict the application rates to land where soil nutrient status is moderate to high. The bioavailability of S should be monitored in land application of THB. THB microbial biodiversity include sulfur utilising bacteria, and these may persist under soil anaerobic conditions. Caution should be applied to soil redox condition and S mobility in land applications. The potential for organic matter decomposition in THB may result in elevated metals (including zinc, copper, manganese, and iron) release to the soil. THB materials may have bioavailable metal loads that may become mobile in response to organic matter decomposition or involved in biologic uptake. Consideration to vegetative plantings that may support biological accumulation and removal of excess metals may serve a beneficial purpose. The mobility of trace elements should be monitored. The presence of PFAS was detected in THB. The PFAS concentrations were below detection limits once mixed with soil. PFAS may adhere to THB and subsequently soil organic matter. Caution should be applied to existing PFAS levels in sites where THB is to be applied and/or where re-application of THB is considered. Follow PFAS NEMP 3.0 guidance including application of Margin of Safety (MOS) and pre-screening for legacy PFAS. Monitor PFAS accumulation over time in high- risk or sensitive areas. Include subsurface sampling

			(e.g., 30–60 cm) to detect vertical movement of trace metals. Track sulfate concentrations and redox conditions where waterlogging may occur.
Soil microbial biodiversity	The use of THB may adversely impact soil microbial biodiversity. Persistence of introduced microbial communities and potential presence of low- level pathogens.	The THB material has a distinct microbial composition. The major phyla were Proteobacteria, Bacteroidota, Chloroflexi, Firmicutes, Caldatribacterota, Desulfobacteria, and Euryarchaeota. All common in gut microbiome and THB materials. The THB-incubation altered the microbial biodiversity in sites where the soil condition was poor and nutrient limited. In sites with moderate nutrient levels the application of THB had limited impact on soil microbial biodiversity. Sites with low nutrient status at Penrith Lakes, and Western Sydney Parklands the microbial diversity from the THB materials was maintained over the 12-week incubation. The soils of Picton Farm did not support the introduced THB microbial diversity. The Sydney Science Park and WSU Richmond sites topsoils also did not alter in response to THB, but subsoils did respond to the THB incubation. Low levels of human-associated pathogens and virulence genes may persist after application in degraded soils.	The THB have a unique microbial composition. This does not directly match the desired composition of soil. The THB microbial diversity was primarily driven by N cycling anaerobes which are less common in aerobic soil systems. Where the soil system has a moderate to high nutrient status the application of THB microbial biodiversity has limited impact. The existing soil biodiversity is established and introduced microbial diversity is unlikely to benefit. Where the soil system has a low nutrient status the application of THB microbial biodiversity may proliferate. The introduced nutrient load allows the existing THB microbial biodiversity to be maintained. The duration to return to pre-existing soil microbial biodiversity may take several months to years. Avoid application to soils used for food production where pathogen risk is a concern. Monitor soil microbial community changes, particularly after repeated THB use. Consider pathogen assays in risk-sensitive applications.
Monitoring and implementation	Lack of consistent sampling may miss nutrient or contaminant migration.	Repeated application requires structured monitoring for defensible nutrient and contaminant tracking.	Establish fixed soil sampling grids with depth intervals (e.g., 0–10, 10–30, and 30–60 cm). Include untreated control areas where possible. Monitor for nutrient loading, metal leaching, PFAS, and microbial change over annual intervals.

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Appendix A – Contaminant and stabilisation thresholds

Contaminant	Grade A	Grade B	Grade C	Grade D
Arsenic (As)	20	20	20	30
Cadmium (Cd)	3	5	20	32
Chromium (Cr, total)	100	250	500	600
Copper (Cu)	100	375	2000	2000
Lead (Pb)	150	150	420	500
Mercury (Hg)	1	4	15	19
Nickel (Ni)	60	125	270	300
Selenium (Se)	5	8	50	90
Zinc (Zn)	200	700	2500	3500
DDT/DDD/DDE	0.5	0.5	1.0	1.0
Aldrin	0.02	0.2	0.5	1.0
Dieldrin	0.02	0.2	0.5	1.0
Chlordane	0.02	0.2	0.5	1.0
Heptachlor	0.02	0.2	0.5	1.0
Hexachlorobenzene (HCB)	0.02	0.2	0.5	1.0
Lindane	0.02	0.2	0.5	1.0
Benzene hexachloride (BHC)	0.02	0.2	0.5	1.0
Polychlorinated Biphenyls (PCBs)	0.3	0.3	1.0	1.0

A.1 Biosolid contaminant thresholds as per Environment Protection Authority (NSW EPA) (2000) Environmental Guidelines: Use and Disposal of Biosolids Products

A.2 Biosolid stabilisation thresholds as per Environment Protection Authority (NSW EPA) (2000) *Environmental Guidelines: Use and Disposal of Biosolids Products* 

Stabilisation Grade	Pathogen Reduction	Vector Attraction Reduction
	Requirements	Requirements
A	One of several advanced processes such as thermal treatment, high pH-high temperature treatment, or 3+ year storage; must meet microbiological standards	Volatile solids reduction ≥38%, or alternatives like specific oxygen uptake rate <1.5 mg O₂/h/g, or high solids content
В	Processes like anaerobic or aerobic digestion, air drying, or lime stabilisation; no requirement to meet microbiological standards	Same options as Grade A, or additional options like extended aeration plus 6-month lagoon storage, or injection/incorporation into soil
С	Does not meet the criteria for Grade A or B; insufficient treatment to reduce pathogens or vector attraction	Does not meet any vector attraction reduction requirements

Land Use	Sum of PFOS and PFHxS (mg/kg)	PFOA (mg/kg)	Comments and Source
Residential with garden / accessible soil (HIL A)	0.003	0.06	Assumes home-grown produce is 10% of diet; excludes eggs, milk, meat from home livestock. Applies to residences, childcare centres, preschools, schools. If produce makes up 50% of diet, use 0.001 mg/kg for PFOS+PFHxS and 0.01 mg/kg for PFOA.
Residential with minimal soil access (HIL B)	2.0	20	Assumes no home- grown produce consumption. Includes high-rise or dense dwellings.
Public open space (HIL C)	1.0	10	Parks, ovals, playgrounds, paths; excludes bushland and unmaintained areas.
Industrial / Commercial (HIL D)	20.0	50	Based on adult worker exposure assumptions.

# A.3 PFAS contaminant thresholds as per Human Health Investigation Levels Heads of EPAs Australia and New Zealand (HEPA). (2025). *PFAS National Environmental Management Plan Version 3.0*.

Appendix B – NLBAR calculations

### **B.1 - Crop Assumption**

Crop: Perennial Rye-Grass

Nitrogen Requirement: 210 kg/ha (from NSW Biosolids Guidelines)

<b>Biosolids Nitrogen Inputs</b>	
Parameter	Value
Total Nitrogen (LECO)	3.9% = 39.0  kg/t
Oxidised Nitrogen (NO3 <sup>-</sup> )	3.8  mg/kg = 0.0038  kg/t
TKN (Total N – NO <sub>3</sub> -)	39.0 - 0.0038 = 39.0  kg/t
Ammonium N (NH4+)	2761  mg/kg = 2.761  kg/t
Organic N (TKN – NH4 <sup>+</sup> )	39.0 - 2.761 = 36.239  kg/t
Mineralisation Rate (MR)	15%
Mineralised Organic N (× MR)	$36.239 \times 0.15 = 5.436 \text{ kg/t}$

#### **Available Nitrogen Calculation**

Available N (Year 1) = 2.761 (NH<sub>4</sub><sup>+</sup>) + 0.0038 (NO<sub>3</sub><sup>-</sup>) + 5.436 (Organic N × MR) = 8.201 kg/t

Annualised Available N  $(\div 5) = 8.201 / 5 = 1.640 \text{ kg/t}$ 

#### **Raw NLBAR**

NLBAR = 210 / 1.640 = 128.0 t/ha, based on annualised available N of 1.640 kg/t and crop N requirement of 210 kg/ha.

#### **B.2 - Adjusted NLBAR**

On sites which receive frequent biosolids applications, the residual nitrogen in the soil should also be determined and considered in calculating the NLBAR. Although no explicit calculation is provided in the NSW Biosolids Guidelines (2000) an estimate is provided here.

The adjusted Nitrogen-Limited Biosolids Application Rate (NLBAR) was calculated by subtracting the baseline soil nitrogen (kg/ha) from a hypothetical maximum nitrogen loading limit of 1,200 kg/ha, as specified in the NSW EPA (2000) guidelines. The remaining nitrogen allowance was then divided by the estimated nitrogen contribution per tonne of biosolids, calculated as 8.201 kg N/t based on the total available nitrogen content of the thermal hydrolysis biosolids (THB). This yielded the following adjusted NLBAR values (in dry tonnes per hectare) for each site:

- Penrith Lakes: 1,200 202 = 998 kg N remaining  $\rightarrow 998 \div 8.201 = 121.7$  t/ha
- Western Sydney Parklands:  $1,200 301 = 899 \text{ kg N remaining} \rightarrow 899 \div 8.201 = 109.6 \text{ t/ha}$
- WSU Richmond:  $1,200 960 = 240 \text{ kg N remaining} \rightarrow 240 \div 8.201 = 29.3 \text{ t/ha}$
- Sydney Science Park: 1,200 997 = 203 kg N remaining  $\rightarrow 203 \div 8.201 = 24.8$  t/ha
- Picton Farm:  $1,200 1,040 = 160 \text{ kg N remaining} \rightarrow 160 \div 8.201 = 19.5 \text{ t/ha}$

These adjusted values ensure that the combined nitrogen load from both the biosolids and the pre-existing soil nitrogen does not exceed the regulatory cap of 1,200 kg N/ha.

Appendix C – CLBAR calculations

Contaminant	Maximum Allowable Soil Contaminant Concentration (mg/kg)	Measured In-Situ Soil Contaminant Concentration (mg/kg)	Available Capacity of Soil to Assimilate Contaminants (mg/kg)	Biosolids Contaminant Application Concentration (mg/kg)	CLBAR (dry t/ha)
Arsenic (As)	20	2.97	17.03	2.93	5797
Cadmium (Cd)	5	0.5	4.5	0.79	5682
Chromium (Cr)	250	9.94	240.06	52.8	4535
Copper (Cu)	375	32.03	342.97	494.12	692
Lead (Pb)	150	7.7	142.3	19.57	7253
Mercury (Hg)	4	0.1	3.9	0.48	8104
Nickel (Ni)	125	6.83	118.17	21.33	5526
Selenium (Se)	8	0.49	7.51	4.65	1611
Zinc (Zn)	700	55.7	644.3	697.61	921
DDT/DDD/DDE	0.5	0.02	0.45	0.05	9567
Aldrin	0.2	0.005	0.195	0.005	38902
Dieldrin	0.2	0.005	0.195	0.005	38902
Chlordane	0.2	0.005	0.195	0.005	38902
Heptachlor	0.2	0.005	0.195	0.005	38902
Hexachlorobenzene	0.2	0.005	0.195	0.005	38902
Lindane	0.2	0.005	0.195	0.005	38902
BHC	0.2	0.005	0.195	0.005	38902
PCBs	0.3	0.05	0.25	0.05	4987
Assumptions		Soil bulk density (dry tonnes/m <sup>3</sup>	3)		1.34
		Incorporation depth (m)			0.075
		Incorporated soil mass (SM), dr	y tonnes/ha		997
Maximum CLBAR fo	or Penrith Site				692 t/ha

## C.1 - Calculation of CLBAR for Non-Agricultural Land – Penrith Lakes Site

C.2 - Calculation of CLBAR for	<b>Agricultural Land - Picton Farm</b>
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Contaminant	Maximum Allowable Soil Contaminant Concentration (mg/kg)	Measured In-Situ Soil Contaminant Concentration (mg/kg)	Available Capacity of Soil to Assimilate Contaminants (mg/kg)	Biosolids Contaminant Application Concentration (mg/kg)	CLBAR (dry t/ha)
Arsenic (As)	20	4.38	15.62	2.93	4897
Cadmium (Cd)	1	0.5	0.5	0.79	579
Chromium (Cr)	100	12.59	87.41	52.8	1514
Copper (Cu)	100	39.56	60.44	494.12	111
Lead (Pb)	150	15.56	134.44	19.57	6285
Mercury (Hg)	1	0.1	0.9	0.48	1715
Nickel (Ni)	60	8.46	51.54	21.33	2210
Selenium (Se)	5	0.43	4.57	4.65	899
Zinc (Zn)	200	88.06	111.94	697.61	146
DDT/DDD/DDE	0.5	0.02	0.45	0.05	8784
Aldrin	0.02	0.005	0.015	0.005	2745
Dieldrin	0.02	0.005	0.015	0.005	2745
Chlordane	0.02	0.005	0.015	0.005	2745
Heptachlor	0.02	0.005	0.015	0.005	2745
Hexachlorobenzene	0.02	0.005	0.015	0.005	2745
Lindane	0.02	0.005	0.015	0.005	2745
BHC	0.02	0.005	0.015	0.005	2745
PCBs	0.3	0.05	0.25	0.05	4575
Assumptions		Soil bulk density (dry tonnes/m?	3)		1.22
		Incorporation depth (m)			0.075
		Incorporated soil mass (SM), dr	y tonnes/ha		915
Maximum CLBAR fo	or Picton Site				111 t/ha

Contaminant	Maximum Allowable Soil Contaminant Concentration (mg/kg)	Measured In-Situ Soil Contaminant Concentration (mg/kg)	Available Capacity of Soil to Assimilate Contaminants (mg/kg)	Biosolids Contaminant Application Concentration (mg/kg)	CLBAR (dry t/ha)
Arsenic (As)	20	5.52	14.48	2.93	4932
Cadmium (Cd)	1	0.5	0.5	0.79	632
Chromium (Cr)	100	15.81	84.19	52.8	1591
Copper (Cu)	100	46.38	53.62	494.12	108
Lead (Pb)	150	16.88	133.12	19.57	6789
Mercury (Hg)	1	0.1	0.9	0.48	1871
Nickel (Ni)	60	6.39	53.61	21.33	2508
Selenium (Se)	5	0.52	4.48	4.65	962
Zinc (Zn)	200	68.05	131.95	697.61	189
DDT/DDD/DDE	0.5	0.02	0.45	0.05	8982
Aldrin	0.02	0.005	0.015	0.005	2994
Dieldrin	0.02	0.005	0.015	0.005	2994
Chlordane	0.02	0.005	0.015	0.005	2994
Heptachlor	0.02	0.005	0.015	0.005	2994
Hexachlorobenzene	0.02	0.005	0.015	0.005	2994
Lindane	0.02	0.005	0.015	0.005	2994
BHC	0.02	0.005	0.015	0.005	2994
PCBs	0.3	0.05	0.25	0.05	4990
Assumptions		Soil bulk density (dry tonnes/m3	)		1.31
		Incorporation depth (m)			0.075
		Incorporated soil mass (SM), dry	/ tonnes/ha		998
Maximum CLBAR fo	r Sydney Science Park Site				108 t/ha

## C.3 - Calculation of CLBAR for Agricultural Land - Sydney Science Park

Contaminant	Maximum Allowable Soil Contaminant Concentration (mg/kg)	Measured In-Situ Soil Contaminant Concentration (mg/kg)	Available Capacity of Soil to Assimilate Contaminants (mg/kg)	Biosolids Contaminant Application Concentration (mg/kg)	CLBAR (dry t/ha)
Arsenic (As)	20	2	18	2.93	5801
Cadmium (Cd)	1	0.5	0.5	0.79	598
Chromium (Cr)	100	3.29	96.71	52.8	1729
Copper (Cu)	100	21.46	78.54	494.12	150
Lead (Pb)	150	2.51	147.49	19.57	7118
Mercury (Hg)	1	0.1	0.9	0.48	1773
Nickel (Ni)	60	1.66	58.34	21.33	2587
Selenium (Se)	5	0.5	4.5	4.65	915
Zinc (Zn)	200	35.77	164.23	697.61	222
DDT/DDD/DDE	0.5	0.02	0.45	0.05	8505
Aldrin	0.02	0.005	0.015	0.005	2835
Dieldrin	0.02	0.005	0.015	0.005	2835
Chlordane	0.02	0.005	0.015	0.005	2835
Heptachlor	0.02	0.005	0.015	0.005	2835
Hexachlorobenzene	0.02	0.005	0.015	0.005	2835
Lindane	0.02	0.005	0.015	0.005	2835
BHC	0.02	0.005	0.015	0.005	2835
PCBs	0.3	0.05	0.25	0.05	4725
Assumptions		Soil bulk density (dry tonnes/m3	3)		1.26
		Incorporation depth (m)			0.075
		Incorporated soil mass (SM), dr	y tonnes/ha		945
Maximum CLBAR fo	or Richmond Site				150 t/ha

## C.4 - Calculation of CLBAR for Agricultural Land – WSU Richmond

Contaminant	Maximum Allowable Soil Contaminant Concentration (mg/kg)	Measured In-Situ Soil Contaminant Concentration (mg/kg)	Available Capacity of Soil to Assimilate Contaminants (mg/kg)	Biosolids Contaminant Application Concentration (mg/kg)	CLBAR (dry t/ha)
Arsenic (As)	20	2.53	17.47	2.93	6213
Cadmium (Cd)	5	0.5	4.5	0.79	5935
Chromium (Cr)	250	14.11	235.89	52.8	4655
Copper (Cu)	375	37.64	337.36	494.12	711
Lead (Pb)	150	20.6	129.4	19.57	6890
Mercury (Hg)	4	0.1	3.9	0.48	8466
Nickel (Ni)	125	5.57	119.43	21.33	5834
Selenium (Se)	8	0.46	7.54	4.65	1690
Zinc (Zn)	700	60.03	639.97	697.61	956
DDT/DDD/DDE	0.5	0.02	0.45	0.05	9378
Aldrin	0.2	0.005	0.195	0.005	40638
Dieldrin	0.2	0.005	0.195	0.005	40638
Chlordane	0.2	0.005	0.195	0.005	40638
Heptachlor	0.2	0.005	0.195	0.005	40638
Hexachlorobenzene	0.2	0.005	0.195	0.005	40638
Lindane	0.2	0.005	0.195	0.005	40638
BHC	0.2	0.005	0.195	0.005	40638
PCBs	0.3	0.05	0.25	0.05	5210
Assumptions		Soil bulk density (dry tonnes/m3	)		1.35
		Incorporation depth (m)			0.075
		Incorporated soil mass (SM), dry	/ tonnes/ha		1,013
Maximum CLBAR f	for Western Sydney Parklands S	lite			711 t/ha

#### C.5 - Calculation of CLBAR for Non-Agricultural Land - Western Sydney Parklands



