

# Investigating the comparative effects of abattoir waste (Thallo™) organomineral fertilizer and inorganic NPK fertilizer on wheat grain and ryegrass yields and their nutrient uptakes

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## Original Research

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## Abstract:

**Purpose:** The study examines the efficacy of using recycled abattoir waste fertilizer as a sustainable nutrient input for crop Production.

**Method:** Two pot experiments were set up in a controlled environment room to examine biomass yields of ryegrass (*Lolium perenne* AberMagic), grain yields of spring wheat (*Triticum aestivum* KWS Cochise), and their micro and macronutrient uptakes, respectively, in an abattoir waste organomineral fertilizer amended soil compared to those treated with inorganic NPK fertilizer. Phosphorus was added at rates of 0, 25, 50, 100, 200, 300, 400, 500, 600, 800, 1000, and 1200 mg P kg<sup>-1</sup> in a low P south-west England soil.

**Results:** Total biomass yields of ryegrass in the NPK treatments were higher at application rates greater than 800 mg P kg<sup>-1</sup> soil compared with their corresponding abattoir waste organomineral fertilizer (Thallo™) treatments. Total wheat dry grain weights increased with P addition rates in both the Thallo™ and NPK amended soils until the application rates exceeded 800 mg P kg<sup>-1</sup> when the dry grain weights started to decline. Micronutrient concentrations in grass produced from Thallo™ fertilizer-amended soils were similar to those from their corresponding NPK fertilizer treatments.

**Conclusion:** This study demonstrated that if the Thallo™ is added at rates not exceeding 200 mg P kg<sup>-1</sup> soil to 300 mg P kg<sup>-1</sup> soil, it can serve as a sustainable and cost-effective alternative P source for ryegrass and wheat grain production. However, its application did not increase the micronutrient density of ryegrass or wheat grain any more than inorganic NPK additions.

**Keywords:** Biomass and grain yield; Inorganic NPK fertilizer; Micronutrient; Ryegrass; Spring wheat; Thallo™

## 1. Introduction

Agri-food systems contribute significantly to improving the health and nutritional well-being of individuals and populations (Devaux et al., 2021; Gashu et al., 2019) through the supply of nutrients contained in foods (McAuliffe et

al., 2020). Malnutrition occurs if agri-food systems do not supply the amounts of essential nutrients required to maintain good health. Crops grown in fertile soils are often well-nourished and have higher micronutrient density in edible portions than those grown in nutrient-deficient soils (Lockyer et al., 2018). A major focus of the UN sustainable

development goals (Eggersdorfer et al., 2018; Fróna et al., 2019) is to deal with hidden hunger, where deficiencies of minerals and vitamins in the diet affect human health. Imathiu (2021) reported that micronutrient malnutrition can impair mental and physical development in children, increase mortality rates, especially in pregnant women and children, increase morbidity, and reduce work productivity in adults. Agricultural strategies such as fertilizer application, plant breeding for enhanced nutritional quality, and improved cropping systems can be used to improve the micronutrient output of farming systems.

Inorganic fertilizer application has increased across the globe but excessive fertilizer application can cause pollution, algae proliferation, and the eutrophication of aquatic ecosystems (Chandini et al., 2019). Inorganic NPK fertilizers only contain very low concentrations of micronutrient elements as impurities, if any (Darch et al., 2019; Songkhum et al., 2018). Application of inorganic NPK fertilizers can boost crop yields but do not provide adequate amounts of micronutrients to address the micronutrient deficiency. Agronomic fortification or the application of mineral-rich fertilizers to soil can increase mineral concentration in crops (Lal et al., 2020; Singh et al., 2019). Mikula et al. (2020) suggested that the nutritional value of crops grown with basic NPK fertilizers can be improved if trace elements are applied together with the applied inorganic fertilizers. Well-informed and adequately resourced farmers may supplement inorganic NPK fertilizers with micronutrient fertilizers to correct micronutrient deficiencies to improve the yields and nutritional composition of crops. However, most farmers are unlikely to apply micronutrients, such as selenium that are essential for humans but not for the crop if no regulations or incentives exist that would make doing so mandatory or profitable (Wall and Plunkett, 2021).

Soil amendments such as abattoir waste fertilizer, farm-yard manure, and compost can be applied to soil to improve soil health and crop productivity. Their applications can alter soil structure, aggregation, and water-holding capacity, resulting in enhanced soil microbial activity and more extensive root development, and increase plant available micronutrient content in the soil (Atkinson, 2018; Zhang et al., 2020). Soil micronutrient concentration can influence micronutrient uptakes of crops growing on them (Kihara et al., 2020). A correct balance between macronutrients and micronutrients in pasture species is critical for sustainable livestock production. Deficiency in even a single nutrient will limit the optimization of grass biomass yield, so a balanced crop nutrition strategy is essential for achieving high crop yields. However, much more research is needed to understand how soil amendments affect the nutritional quality of food crops (Bonanomi et al., 2020; Celestina C, 2019; Mujtar et al., 2019).

Thallo™ is a novel fertilizer product manufactured by Elemental, incorporating abattoir waste (including Ca- and P-rich bone material) and fortified with trace elements from industrial by-products with fertilizer value. Despite varied inputs, Thallo™ meets international standards as the product is subjected to real-time spectroscopic analysis during the production process. Thallo™ can be viewed as a sus-

tainable P fertilizer because it is not obtained by mining, but instead recovered from abattoir waste, including bones, hooves, tails, and skin. Thus, replacing inorganic P fertilizers with Thallo™, can reduce the amount of abattoir waste sent to landfill and contribute to greener farming. Darch et al. (2019) found that Thallo™ fertilized crops contained significantly more selenium and zinc and lower cadmium than inorganic fertilized ones. They concluded that Thallo™ has the potential to maintain yields and improve essential element concentrations in wheat and grass. However, its efficacy and optimal application rates are yet to be demonstrated with the appropriate crops. This study examined the effect of different application rates of Thallo™ fertilizer on grass and wheat yield and nutrient uptakes compared to conventional NPK fertilizer. The study was underpinned by the hypotheses that; i) grass/wheat yield is unaffected by the fertilizer type, ii) Yields of crops will increase to a point and decline afterwards, and iii) Micronutrient-enriched Thallo™ fertilizer will produce crops with greater concentrations of micronutrients compared to NPK fertilizers.

## 2. Materials and methods

### Pot experiment set up.

Two pot experiments were set up in a controlled environment room with grass (*Lolium perenne* AberMagic) and spring wheat (*Triticum aestivum* KWS Cochise) grown as test crops. The grass and the wheat were both grown in soil collected from a permanent pasture located in the South West of England that had not received any P fertilizer in the recent past. The soils were taken from de-turfed soil at the 0 – 10 cm depth and sieved to remove all stones and coarse roots (> 4 mm diameter). The soil was classified by the Soil Survey of England and Wales as Halstow and Hallsworth series (clay loam or clay topsoil over slowly permeable clayey, fine silty or fine loamy, mottled subsoil). Using standard protocols (DEFRA Reference Book 427) for analysis, the soil pH was 6.4, with available P, K, and Mg contents being 5.6, 274.3, and 60.3 mgL<sup>-1</sup>, respectively. According to the DEFRA fertilizer Recommendation RB 209, 9<sup>th</sup> edition, the P, K, and Mg contents represented indexes of 0, 3, and 2, respectively. This indicates that the soil had low P content, so it was likely to respond to P fertilizer application.

### Wheat and grass planting

All of the plants (wheat and grass) were grown in plastic rose pots with saucers, the pots measuring 17.4 cm in height and 14.8 cm in diameter, with the soil filled to a depth of approximately 15 cm. A layer of finely woven plastic mesh was placed at the base of the pot to ensure the soil was contained. The pots were filled with either 1.5 kg soil (air-dried) plus 0.5 g *L. perenne* seed and a further 50 g air-dried soil used to lightly cover the grass seeds, or 1.5 kg air-dried soil with eight *T. aestivum* seeds pushed into 1.5 cm depth. Once wheat plants emerged, the number of seedlings was reduced to five, with any ungerminated seeds removed. The pots were placed under LED grow lights,

with a 16 h/8 h day/night cycle and a 30-minute brightening and dimming period between the day and night cycles. A total of 1460 W LED fixtures, each with 119 individual LEDs operating at 42 V, were positioned approximately 90 cm above the soil. Temperatures were set to 20° C during the day and 16° C during the night periods. Pots were watered with an artificial rainwater solution (Darch et al., 2019) either every other day or daily depending on rates of transpiration, with soil maintained at 60% of water holding capacity (assessed by mass).

In both the wheat and grass trials two fertilizer types: Thallo™ and a conventional NPK fertilizer were applied at 12 different rates; 0, 25, 50, 100, 200, 300, 400, 500, 600, 800, 1000, and 1200 mg P kg<sup>-1</sup> soil. The rates of NPK applied were formulated from individual fertilizers (N as Extra N 33.5% N, P as triple super phosphate (46% P<sub>2</sub>O<sub>5</sub> as Ca(H<sub>2</sub>PO<sub>4</sub>)<sub>2</sub>), and K as muriate of potash (60% K<sub>2</sub>O as KCl) to match the NPK concentrations of the Thallo™ fertilizer applied. All four fertilizer types were analyzed for their N, P, and K content via ICP-OES at Rothamsted Research, UK. Therefore, the concentrations of N, P, and K applied as Thallo™ or Extra N, triple super phosphate, and muriate of potash were equal. All fertilizers were evenly incorporated into the soils of each pot prior to sowing grass or wheat seeds. Concentrations of different elements in the four types of fertilizers, and their rates applied are shown in Table 1 while the quantities of each of the fertilizer sources added at each rate in the study are presented in Table 2.

### Grass and wheat data collection

Data were collected on wheat grain yield, grass dry matter yield, and the elemental composition of the grass biomass and wheat grain. The grass was cut at 4 cm above soil level every four weeks for a total of six cuts (Data not included). At the 6th cut, the remaining stubble was also cut to soil level. In the wheat trial, wheat ears per pot were counted and their height above the soil level was measured. Wheat grain was separated from the chaff before being assessed for the total grain yield. All harvested plant materials (i.e. grass, wheat grain, wheat chaff, and wheat straw) were dried on the day of cutting at 85° C for 48 hours for ICP-OES and ICP-MS analyses. Elemental analysis was conducted on the grass biomass and wheat grains to determine aluminium (Al), arsenic (As), Ca, cadmium (Cd), chromium (Cr), cobalt (Co), copper (Cu), Fe, K, magnesium (Mg) manganese (Mn), molybdenum (Mo), sodium (Na), nickel (Ni), P, lead (Pb), S, selenium (Se), titanium (Ti), and zinc (Zn) contents after being finely milled and digested with concentrated nitric and perchloric acids before analysis using inductively coupled plasma atomic emission spectroscopy (ICP-AES) (Zhao et al., 1994). Micro- and macronutrient uptakes in each crop were calculated as a product of bio-mass and nutrient concentration.

Phosphorus Use Efficiency at each of the rates of P

application was calculated as follows:

$$PUE = \frac{\left( \text{P uptakes in} \right)_t - \left( \text{P uptakes in} \right)_c}{\text{P applied (kg)}}$$

where;

PUE = Phosphorus Use Efficiency

t = P uptake from grass biomass or wheat grain from the Thallo or NPK-treated soils

c = P uptake from grass biomass or wheat grain from the unamended control soil

P applied (kg) = Kilograms of phosphorus applied to the soil

P uptake = P content (mg P kg<sup>-1</sup>) in grass biomass or wheat grain x grass biomass or wheat grain dry weight (kg)

### Statistical analysis

Wheat grain yield, grass dry matter yield, and the elemental composition of the grass and wheat grain samples in the different treatments were statistically compared using regression by the GENSTAT software package. All except the number of wheat ears analysis were done via Multiple Linear Regression with Groups. Most of the variables were analyzed after square root transformations. The final model used has separate lines with an estimation of differences from the reference level (in this case the reference being NPK values). The number of ears of wheat being counted data did not follow a normal distribution, but rather followed a Poisson distribution, and as such a Generalized Linear Model with Log fit function was used to analyze them.

## 3. Results and discussion

### Grass biomass and wheat grain yield responses to Thallo™ and inorganic NPK fertilizer additions

Total grass biomass yields in the Thallo™ and NPK amended soils (Fig. 1) were similar at application rate ≤ 800 mg P kg<sup>-1</sup> soil, increasing with P addition. At application rates greater than 800 mg P kg<sup>-1</sup> soil, total biomass yields in the NPK treatments were higher than their counterpart Thallo™ treatments, with total biomass yields in the 1000 mg P kg<sup>-1</sup> soil and 1200 mg P kg<sup>-1</sup> Thallo™ amended soils being lower than the 800 mg P kg<sup>-1</sup> Thallo™ treatment.

Total grass biomass yields in the Thallo and NPK amended soils were similar at P addition rates less or equal to 600 mg P kg<sup>-1</sup> soil, but at higher application rates of 800 mg kg<sup>-1</sup> soil to 1200 mg P kg<sup>-1</sup> soil, total grass biomass dry matter in the NPK treatments were higher than Thallo amended soils at similar rates. Basically, both of the fertilizer types showed a significant regression slope (i.e., they differ from a no-response flat line), and the initial part as assessed by the linear function showed that the two fertilizers did not differ from each other, but the quadratic function showed that the curves differ from each other at the higher rates of application. The Thallo treatment curves downwards illustrating a reduction in the amount of biomass produced,

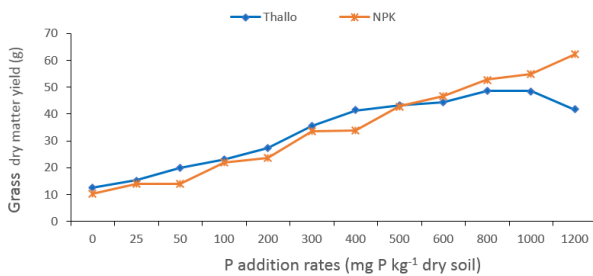
**Table 1.** Concentration of individual elements within the four types of fertilizer used.

	Mean values (mg/kg)																				
	Al	As	Ca	Cd	Co	Cr	Cu	Fe	K	Mg	Mn	Mo	Na	Ni	P	Pb	S	Se	Ti	Zn	N
Thalco™	860	1.90	23908	0.83	7.98	57.25	86.96	1893.60	38178	38564	143.99	2.84	109798	58.08	43595	25.83	137668	12.60	98.70	641.56	35010
Extra N	31	1.76	5151	0.03	0.03	0.50	BD	98.00	BD	6621	9.12	0.21	62	0.36	36	0.20	93	BD	7.80	2.40	335000
TSP	1811	3.14	269904	23.57	0.41	132.07	35.41	1041.00	2491	2782	17.32	17.57	4162	46.34	184121	BD	21772	23.30	39.30	512.30	1020
MoP	95	1.94	3484	0.05	BD	0.37	1.54	1181.00	500284	1143	7.85	-0.22	8394	0.21	246	1.80	3264	BD	14.60	3.20	470

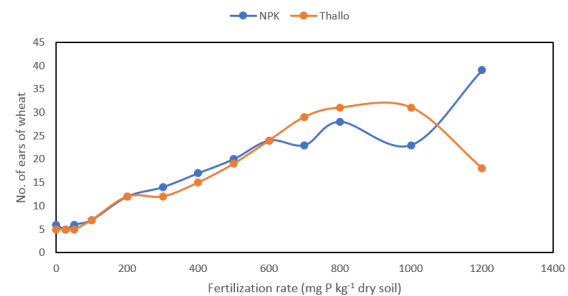
N values from Carlo Erba NA1500 Elemental Analyser, other elements via ICP-MS. (BD-below detection, TSP-triple super phosphate, MoP-muriate of potash).

**Table 2.** Phosphorus application rates and the corresponding amounts of Thallo fertilizer and each of the three inorganic fertilizer sources used in the study.

Rate (mg P/kg soil)	Thallo™ applied to each pot (g)	Extra N applied to each pot (g)	TSP applied to each pot (g)	MoP applied to each pot (g)
0	0.00	0.00	0.00	0.00
25	0.86	0.09	0.20	0.07
50	1.72	0.18	0.41	0.13
100	3.44	0.36	0.82	0.26
200	6.88	0.71	1.63	0.52
300	10.32	1.07	2.44	0.78
400	13.76	1.43	3.26	1.04
500	17.20	1.78	4.07	1.29
600	20.65	2.14	4.89	1.55
700	24.09	2.49	5.71	1.81
800	27.53	2.85	6.52	2.07
1000	34.41	3.57	8.15	2.59
1200	41.29	4.28	9.78	3.10



**Figure 1.** Total grass biomass dry weight following application of Thallo™ and inorganic NPK fertilizers.



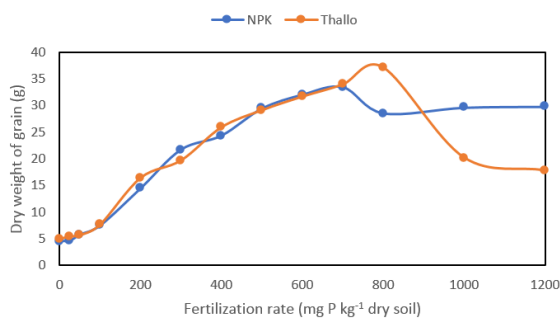
**Figure 3.** Number of ears of spring wheat following application of Thallo™ and Inorganic NPK fertilizers.

presumably, due to the negative effects of accumulating too much of a particular element (or elements) present in the Thallo that is not supplied to the same extent in the NPK fertilizer. This requires further investigation.

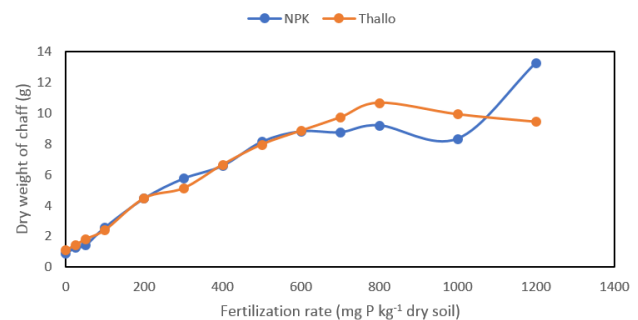
Total wheat grain dry weights increased with P addition rates in both the Thallo™ and NPK amended soils until the application rates exceeded 800 mg P kg<sup>-1</sup> as shown in Fig. 2. At application rates greater than 800 mg P kg<sup>-1</sup> soil, wheat grain dry weight decreased with P addition rates, especially in the Thallo™ treatments.

Wheat stubble dry matter only increased considerably

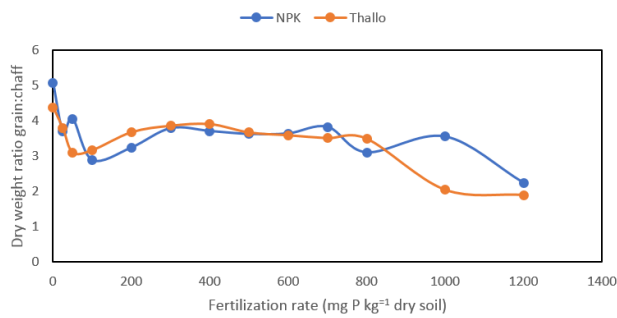
at application rates  $\geq 300$  kg P ha<sup>-1</sup> in both the NPK and Thallo amended soils. This trend was similar to that observed in the total biomass yields, suggesting that at application rates of  $\leq 300$  kg P ha<sup>-1</sup> nutrient release from both fertilizer types followed a similar pattern. At application rates  $\geq 300$  mg P kg<sup>-1</sup> soil, total grass biomass (cut at 4 cm above soil surface) was higher than stubble biomass, but stubble: aboveground biomass ratios at these higher application rates were lower than in the lower application rates of both fertilizer types.



**Figure 2.** Total wheat grain dry weight following the application of Thallo™ and Inorganic NPK fertilizers.



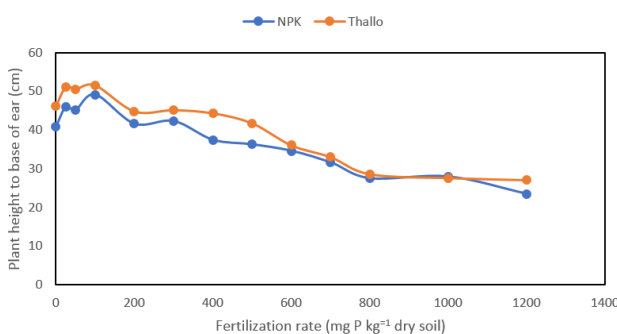
**Figure 4.** Spring wheat chaff dry weight following application of Thallo and inorganic NPK fertilizers.



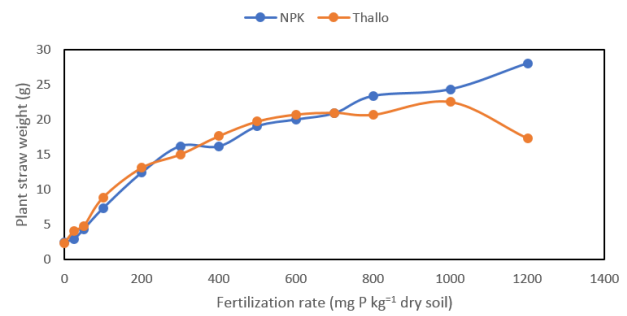
**Figure 5.** Spring wheat grain: chaff ratio following application of Thallo™ and inorganic NPK fertilizers.

The number of wheat ears (Fig. 3) in the Thallo™ and NPK amended soils were similar, ranging from 5 to 39 in the NPK treatments, and 5 to 31 in the Thallo™ amended soils, respectively. Heights to the base of the ear increased with application rates only up to 300 mg P kg<sup>-1</sup> in the NPK treatments but increased with application rates up to the P addition rate of 700 mg P kg<sup>-1</sup> in the Thallo treatments. Dry chaff weights (Fig. 4) in both the NPK and Thallo amended soils increased with increasing P application rates, except in the 1000 mg P kg<sup>-1</sup> and 1200 mg P kg<sup>-1</sup> Thallo fertilizer treatments, where dry chaff weights recorded were lower than the 800 g P kg<sup>-1</sup> treatment. Dry grain: chaff ratios (Fig. 5) decreased consistently from 4.38 in the control to 1.89 in the 1200 g P kg<sup>-1</sup> Thallo treatments, but no consistent trend was found in the NPK treatments.

In the wheat trial, all the yield and growth variables (grain dry weight, chaff dry weight, number of ears, height to base of ear, and ratio of chaff to grain) increased with increasing P application rate, indicating that higher P application led to increased vegetative growth and grain yield. The highest number of spring wheat ears was found at a P addition rate of 200 g P kg<sup>-1</sup> in the NPK treatment compared to 800 mg P kg<sup>-1</sup> in the Thallo treatment, which indicates that NPK supplied adequate amounts of nutrients to maximize ear formation at lower rates than Thallo. This is not surprising as the inorganic NPK was soluble, making the nutrients more readily available than the Thallo fertilizer, in which the nutrients needed to be mineralized before they could be taken up by the plant. The rate of increase in both spring wheat and grass growth exemplified by their biomass yields



**Figure 6.** Height of Spring wheat following application of Thallo™ and inorganic NPK fertilizers.

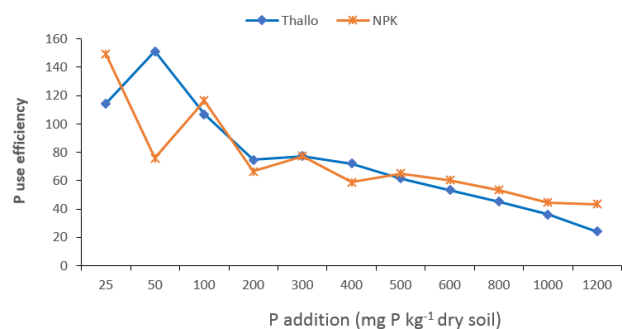


**Figure 7.** Spring wheat straw weight following application of Thallo™ and inorganic NPK fertilizers .

declined considerably at P application rates exceeding 800 mg kg<sup>-1</sup> soil in both the Thallo and NPK amended soils. The observed PUE values partly explain this observation because PUE in grass was greater than 100% only at application  $\leq 100$  mg P kg<sup>-1</sup> treatments, declining to less than 50% at rates  $> 600$  mg P kg<sup>-1</sup> in both Thallo and NPK fertilizer-amended soils. Wheat dry matter yields increased with increasing P addition rates in the NPK and Thallo treatments up to 800 kg P kg<sup>-1</sup>, respectively, indicating that higher P additions stimulated nutrient assimilation and photosynthate production for greater grain yields only up to rates not exceeding 800 mg P kg<sup>-1</sup> soil. Thus, at P addition rates of 1000 mg P kg<sup>-1</sup> and 1200 mg P kg<sup>-1</sup>, the wheat grew rather more vegetatively or suffered toxicity effects in the NPK and Thallo amended soils, respectively, resulting in lower grain yields.

Wheat heights (Fig. 6) were highest at P application rates of 100 mg P kg<sup>-1</sup> in both the Thallo and inorganic NPK treatments but declined with increasing P application rates afterward. Strawweight increased with increasing P application rates in both Thallo and inorganic NPK-amended soils (Fig. 7). However, at an application rate of 1200 mg P kg<sup>-1</sup> soil, the strawweight in the Thallo amended soil reduced to the level of the 800 mg P kg<sup>-1</sup> treated soil.

Wheat grain dry weight and all the growth parameters observed for the wheat generally increased with fertilizer application rates but only the ear-to-base height was influenced by fertilizer type (Table 3). The interactive effect between fertilizer type and application rate was not found in the wheat dry weight nor the uptake of any of the



**Figure 8.** P Use efficiency of grass grown in Thallo™ and inorganic NPK amended soils.

**Table 3.** Coefficients of regressions for relationships between spring wheat grain dry weight and growth parameters versus P addition rate, Fertilizer type, and P addition rate x fertilizer type interactions.

Parameter Variable	P addition rate	Fertilizer type t. pr.	P addition rate x fertilizer type
Grain (dry weight)	0.038	0.991	0.901
Chaff (Dry weight)	< .001	0.718	0.708
Straw (Dry weight)	< .001	0.494	0.294
Number of ears	0.038	0.991	0.909
Height to base of ear	< .001	0.007	0.283
Ratio of grain to chaff	0.029	0.927	0.565

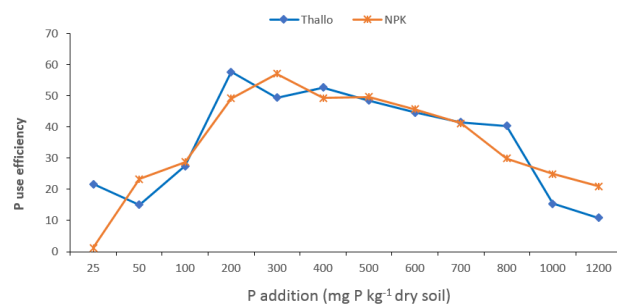
elements.

Regression analysis (Table 3) of the data confirmed that, in general, grass biomass yields and spring wheat grain yields increased with increasing P addition rates, but there was no variation between the biomass yields from the 2 different P sources until P was added at rates exceeding 800 mg P kg<sup>-1</sup> soil. However, a significant interactive effect of fertilizer type and amount of P added on biomass yield was observed, especially at the higher P addition rates.

Our findings are in good agreement with Darch et al. (2019) who reported that application of Thallo to soil was beneficial for grass and wheat production, and when applied to the soil, conventional NPK fertilizer did not outperform Thallo on grass biomass, wheat grain yields nor micronutrient contents. Additionally, the present study showed that dry straw and dry chaff weights increased with increasing P addition rates only up to 800 mg P kg<sup>-1</sup> soil in the Thallo amended soils, whereas they increased to 1000 mg P kg<sup>-1</sup> soil in the NPK treatments. This supports the assertion that higher NPK fertilizer application rates promote greater vegetative growth, increasing aboveground biomass yield (Chen et al., 2020). Lower vegetative growth at the Thallo addition rates exceeding 1000 mg P kg<sup>-1</sup>, however, points to a possible toxicity effect of other elements that were probably applied in excess in the Thallo that were not supplied by the inorganic NPK fertilizer.

### Macronutrients uptake and P use efficiency following Thallo and NPK fertilizer additions

N uptake rates were relatively higher in the NPK treatments compared with their corresponding Thallo

**Figure 9.** P Use Efficiency of spring wheat grown in Thallo<sup>TM</sup> and inorganic NPK amended soils.

treatments, confirming that N was more readily available in soluble inorganic NPK fertilizer-amended soils than in the recycled abattoir waste Thallo<sup>TM</sup> fertilizer in which organic N forms needed to be mineralized before they could be absorbed by plants (Leye and Omotayo, 2014; Sogn and Haugen, 2011). Similarly, at higher P addition rates, Ca uptake rates were higher in NPK treatments than Thallo treatments. At P addition rates exceeding 800 mg P kg<sup>-1</sup> soil, Ca uptakes were 8 to 10 times higher in NPK-amended soils than in Thallo treatments. The organic matter associated with Thallo may have chelated the Ca in the Thallo-amended soil application (Hammér and Kirchmann, 2015). In this regard, Ca was expected to be less available for uptake in the Thallo soils as was observed at higher rates of application. High organic matter from Thallo, especially at higher rates of application, possibly contributed to Ca chelation, reducing uptake. In contrast, S uptake rates were 2 to 3 times greater in Thallo treatments than NPK amended soils at all rates.

P Use Efficiency (PUE) (Fig. 8 and 9) determined as the differences between P uptakes in the total grass biomass or wheat grain from the inorganic NPK or Thallo<sup>TM</sup> amended soils and those from the un-amended control treatment was expressed as a percentage of the amount of P applied in each Thallo<sup>TM</sup> and inorganic NPK treatment, respectively. In both the NPK and Thallo amended soils, P Use Efficiency in grass was greater than 100% in the 25 mg P kg<sup>-1</sup> to 100 mg P kg<sup>-1</sup> treatments (Fig. 2). This reduced to between 60% and 80% in the 200 mg P kg<sup>-1</sup> to 500 mg P kg<sup>-1</sup> treatments, and declined further to < 50% at P addition rates ≥ 600 mg P kg<sup>-1</sup> soils of both amendments.

P Use Efficiencies (PUE) estimated for the different P addition rates peaked at approximately 200 mg P kg<sup>-1</sup> and 300 mg P kg<sup>-1</sup> soil, in the Thallo and NPK fertilizer-amended soils, respectively. The results showed that PUE declined steadily after the peaks in both Thallo and NPK treatments, reaching values less than 50% at P addition rates ≥ 500 mg P kg<sup>-1</sup> soil. Plants can access variable forms of soil phosphorus including organic P forms and those tightly bound to the soil surface, so P use efficiency can be enhanced through breeding more P-efficient cultivars, by conventional P fertilizer application, or through new technologies such as Thallo fertilizer addition. Strategies that improve soil P use by plants are likely to help reduce excess P loss from soil into water, which causes choking algal blooms and eutrophication in

**Table 4.** Coefficients of regressions showing the relationship between grass biomass yield and nutrient uptakes versus P addition rate, fertilizer type, and fertilizer type x P application rates interactions.

Parameter Variable	P addition rate t. pr.	Fertilizer type	P addition rate x fertilizer type
Biomass yield	< 0.001	< 0.144	< 0.001
P	< 0.001	< 0.001	< 0.016
K	< 0.001	< 0.085	< 0.025
Ca	< 0.001	< 0.848	< 0.001
Mg	< 0.001	< 0.528	< 0.348
Al	< 0.002	< 0.733	< 0.468
Cd	< 0.001	< 0.114	< 0.001
Co	< 0.001	< 0.430	< 0.004
Cr	< 0.002	< 0.297	< 0.139
Cu	< 0.001	< 0.090	< 0.001
Fe	< 0.001	< 0.439	< 0.022
Mn	< 0.001	< 0.019	< 0.001
Mo	< 0.001	< 0.001	< 0.227
Na	< 0.042	< 0.001	< 0.021
Ni	< 0.001	< 0.373	< 0.002
Zn	< 0.001	< 0.210	< 0.585

many freshwater and marine environments across the world (Ngatia and Taylor, 2019). Contrary to our expectations, Se and Cd uptakes in the Thallo treatments were similar to uptake in the NPK treatments.

Here we contend that comparing results obtained and conclusions drawn from a pot experiment with pure stands to field situations with species mixtures requires caution. This controlled environment study can offer more reliable nutrient uptake data than a field trial as plant growth conditions (temperature, light, water) were controlled in the greenhouse. In this study, we assumed that regular water supply and relatively high soil temperature, coupled with the homogenization of the soil prior to sowing the seeds, potentially increased the mineralization rates of the inherent soil organic matter, thereby increasing the availability of essential plant nutrients. However, compared to the field conditions, the soil volume of the pots was limited and may have affected the root growth and the uptake of micronutrients. Thus, the low uptakes observed for many micronutrients were not unexpected. Our observation is in good agreement with Agathokleous et al. (2019) who contended that aboveground biomass: root biomass ratios widely reported in many independent trials involving a variety of annual and perennial plant species, often reflect significant stimulation at low doses and inhibition at high doses when plants are exposed to different chemicals and physical stressors.

#### Effect of Thallo and NPK fertilizers on plant micronutrient uptake

Pooling all the data together, regression analyses (Tables 4 and 5) showed that uptake of all the elements increased with increasing fertilizer application rate. Also, P, Mn, Mo, and Na uptakes differed between the two fertilizer types. Interactive effects of fertilizer type and their

application rates were observed for all the nutrients except Mg, Al, Cr, Mn, and Zn. In the wheat grains, P, K, Al, Cd, Cu, Mn, Ni, Zn, and N uptakes increased with increasing P addition rates but no significant effects were found in Ca, Mg, Cu, Cr, Fe, Mo, Na, S, and Se up-takes. Uptakes of Ca, Fe, Mo, Na, S, and Se rates varied between Thallo and NPK treatments but those of the remaining elements were not affected by fertilizer type. The interactive effect of fertilizer type and application rates was observed in P, Al, Cd, Cu, Mn, Ni, Zn, and N up-takes but not in the others.

There is a paucity of information regarding micronutrient uptakes in typical UK pastures; however, the micronutrient uptakes found in this study were compared to the ranges reported in other studies (Brennan et al., 2019; Oviasogie et al., 2007; Ward, 2000), and unpublished data from the North Wyke Farm Platform, Rothamsted Research, North Wyke, Devon, SW England. Regardless of the type of fertilizer applied, uptakes of all the micronutrients, with the exception of chromium, even at application rates > 600 mg P kg<sup>-1</sup>, were lower than the ranges reported in other studies.

Assessment of micronutrient concentrations in forage is critical for livestock nutrition purposes as micronutrients influence livestock productivity in ways such as livestock health, milk, and meat production. Suttle (2010) contended that, alongside macronutrients, essential micronutrients such as cobalt (Co), copper (Cu), iron (Fe), manganese (Mn) and zinc (Zn), and beneficial molybdenum (Mo) and nickel (Ni) are important components of nutrition. McBride (1994) argued that plant micronutrient concentrations are influenced by both the total amounts and the availability of the micronutrients in the soil, which are affected by factors such as soil pH, clay content, and organic matter. In this study, the grass was watered from the base of the soil, and water holding capacity was constantly maintained at 60% so leaching of elements did not likely occur. Therefore,



**Table 5.** Coefficients of regressions showing the relationship between spring wheat nutrient uptakes versus P addition rate, fertilizer type, and fertilizer type x P application rates interactions.

Parameter Variable	P addition rate F.pr	Fertilizer type	P addition rate x fertilizer type
P	0.004	0.145	0.001
K	0.001	0.803	0.001
Ca	0.989	0.001	0.146
Mg	0.294	0.117	0.168
Al	0.001	0.689	0.003
Cd	0.001	0.733	0.001
Co	0.229	0.179	0.163
Cr	0.190	0.332	0.192
Cu	0.028	0.062	0.001
Fe	0.472	0.003	0.094
Mn	0.005	0.361	0.004
Mo	0.953	0.001	0.070
Na	0.686	0.027	0.298
Ni	0.001	0.958	0.001
Zn	0.002	0.269	0.001
N	0.001	0.806	0.001
Pd	0.987	0.088	0.933
S	0.732	0.004	0.123
Se	0.669	0.038	0.331
Ti	0.282	0.137	0.176

as we used the same soil type for all the treatments in this study, we can conclude that the relative availability and hence uptakes of most micronutrients in this study most probably did not reflect the amounts added in each treatment. Further work is required to assess the long-term uptake following the application of the micronutrients to the soil.

#### Interactive effects of fertilizer type and P application rates on nutrient uptakes

Regression analysis (Tables 4 and 5) revealed that uptake of P and all the other elements significantly ( $P < 0.05$ ) varied with P addition rates, while the fertilizer type only significantly affected P, Mn, Mo, and Na uptakes in the grass. The trends in macronutrient ( $N > P > Ca$ ) uptakes clearly confirmed the long-established fact that plants require N followed by P in relatively higher amounts than all other macronutrients (Jacoby et al., 2017). Greater N uptake with increasing Thallo and NPK fertilizer application rates was also reflected in the increasing grass biomass that was associated with higher fertilizer application rates.

Some authors have reasoned that in addition to their soil concentrations and bioavailability, plant uptake of trace elements is influenced by other elements in the soil, resulting in positive (synergism) or negative (antagonism) interactions. For instance, previous trials have demonstrated antagonistic interactions between Zn uptake and uptakes of P, Ca, Cu, Mg, and K (Cai et al., 2017; Kashem and Singh, 2002; Tangahu et al., 2011). Here, interactive effects of P addition rates and fertilizer types were observed in uptakes

of P, Ca, Cu, Fe, Mn, Co, Na, K, Cd, and Ni in the grass, but not in the Mg, Al, Cr, nor Zn uptakes. Our results were not in agreement with those reported by Tangahu et al. (2011), Kashem and Singh (2002) and Cai et al. (2017). In the present study, P application rates in the NPK treatments were matched to those in the Thallo-amended soils, but Ca uptakes were relatively higher in the NPK treatments compared to their counterpart Thallo-amended soils. However, no obvious interactions were observed between Zn uptake and those of P, Ca, Cu, Mg, or K. Furthermore, in agreement with Darch et al. (2019) no synergism was observed between Cd and Zn, Fe, or Cu, although Cd is reported to be positively correlated with Zn, Fe, and Cu (Jackson et al., 2012). Similarly, we found no antagonism between Mn and Ca, Mg, or Zn contrary to the report by Smialowicz et al. (1987) that these elements are negatively correlated with Mn. In this regard, we agree with Darch et al. (2019) who concluded that there is a dearth of understanding regarding synergism or antagonism among uptakes of elements from controlled growth trials.

#### 4. Conclusion

In accordance with our hypothesis, the study has demonstrated that i) grass/wheat yield is un-affected by the fertilizer type unless they are applied at rates  $\geq 1000$  mg P kg<sup>-1</sup> soil ii) wheat grain and grass biomass yields increased with increasing fertilizer application up to a point and declined afterward, especially at very high rates of Thallo fertilizer addition possibly due to the toxicity effects of some compounds or minerals added with the Thallo fertilizer. Contrary to our expectation, the micronutrient-enriched Thallo fertilizer did not produce

grass with greater concentrations of micronutrients compared to NPK fertilizers nor were Cd up-takes in the NPK-amended plants greater than their respective Thallo-treated plants. Furthermore, the study showed that, added at rates not exceeding 200 mg P kg<sup>-1</sup> soil to 300 mg P kg<sup>-1</sup> soil, recycled abattoir waste fertilizer (Thallo) can be considered a sustainable and cost-effective P alternative source for ryegrass and wheat production based on optimal PUE found between those application rates. This research demonstrates that Thallo<sup>TM</sup> can be a sustainable and cost-effective alternative to conventional fertilizers for ryegrass and wheat production on a global scale. However, further field-scale investigations are important to enable a better formulation and a more informed recommendation of the fertilizer. In conclusion, our results indicated that even in the short term, the recycled abattoir fertilizer can serve as a sustainable alternative to conventional fertilizers, sustainably contributing to global phosphorus fertilizer needs.

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#### Ethical approval

This manuscript does not report on or involve the use of any animal or human data or tissue. So the ethical approval is not applicable.

#### Author contribution

The authors confirm the study conception and design: KAF, MB, RD; data collection: KAF, EAB, CAP, AN; analysis and interpretation of results: KAF, EAB, MB, RD, CAP, AN; draft manuscript preparation: KAF, CAP, MB, EAB. The results were evaluated by all authors, and the final version of the manuscript was approved.

#### Availability of data and materials

Data presented in the manuscript are available via request.

#### Conflict of interest statement

The authors declare that there are no conflicts of interest associated with this study.

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