Piggery pond sludge as a nitrogen source for crops. 1. Mineral N supply estimated from laboratory incubations and field application of stockpiled and wet sludge

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Abstract. Piggery pond sludge (PPS) was applied, as-collected (Wet PPS) and following stockpiling for 12 months (Stockpiled PPS), to a sandy Sodosol and clay Vertosol at sites on the Darling Downs of Queensland. Laboratory measures of N availability were carried out on unamended and PPS-amended soils to investigate their value in estimating supplementary N needs of crops in Australia's northern grains region. Cumulative net N mineralised from the long-term (30 weeks) leached aerobic incubation was described by a first-order single exponential model. The mineralisation rate constant (0.057/week) was not significantly different between Control and PPS treatments or across soil types, when the amounts of initial mineral N applied in PPS treatments were excluded. Potentially mineralisable N (N_0) was significantly increased by the application of Wet PPS, and increased with increasing rate of application. Application of Wet PPS significantly increased the total amount of inorganic N leached compared with the Control treatments. Mineral N applied in Wet PPS contributed as much to the total mineral N status of the soil as did that which mineralised over time from organic N. Rates of CO₂ evolution during 30 weeks of aerobic leached incubation indicated that the Stockpiled PPS was more stabilised (19-28% of applied organic C mineralised) than the Wet PPS (35–58% of applied organic C mineralised), due to higher lignin content in the former. Net nitrate-N produced following 12 weeks of aerobic non-leached incubation was highly correlated with net nitrate-N leached during 12 weeks of aerobic incubation ($R^2 = 0.96$), although it was <60% of the latter in both sandy and clayey soils. Anaerobically mineralisable N determined by waterlogged incubation of laboratory PPS-amended soil samples increased with increasing application rate of Wet PPS. Anaerobically mineralisable N from field-moist soil was well correlated with net N mineralised during 30 weeks of aerobic leached incubation ($R^2 = 0.90$ sandy soil; $R^2 = 0.93$ clay soil). In the clay soil, the amount of mineral N produced from all the laboratory incubations was significantly correlated with field-measured nitrate-N in the soil profile (0-1.5 m depth) after 9 months of weed-free fallow following PPS application. In contrast, only anaerobic mineralisable N was significantly correlated with field nitrate-N in the sandy soil. Anaerobic incubation would, therefore, be suitable as a rapid practical test to estimate potentially mineralisable N following applications of different PPS materials in the field.

Additional keywords: piggery manure, nitrogen mineralisation potentials, aerobic mineralisable N, anaerobic mineralisable N, ammonium-N, nitrate-N.

Introduction

Appropriate disposal of waste is a significant issue for the pig industry in Australia and worldwide due to increasing concerns of regulatory authorities to avoid environmental pollution (Kliese 2002). Commonly, piggery waste is collected in large earthen ponds, which eventually become overloaded with solids as liquid effluent is usually recycled through irrigation. Periodic removal of pond sediment or sludge is required to maintain pond performance. Piggery pond sludge (PPS) may be removed as a suspension by a

vacuum tanker and applied to land directly (wet), or allowed to dry in stockpiles and then applied to land using a solids spreader (Kliese 2002).

Piggery pond sludge applied to cropping land is a potential source of supplementary nutrients for crop production. In northern Australia, nitrogen (N) contained in PPS may provide some of the supplementary N that is now required for most broadacre field crops. Thus, knowledge of the kinetics of release of plant-available N is essential if PPS is to be used as a valued resource for crop production.

The kinetics of release of mineral N from animal wastes is also of importance for its land application to avoid nitrate (NO₃) pollution of surface and groundwater when applied in excess (Rate and Cameron 1992; Bernal and Roig 1993; Cameron *et al.* 1995; Carey *et al.* 1997; Estavillo *et al.* 1997). The kinetics of release of mineral N from PPS is controlled by soil microorganisms and environmental parameters that affect soil biological activity. Soil moisture, temperature, texture, pH, and PPS composition will influence the rate and extent of mineralisation (Zebarth et al. 1996; Zaman et al. 1998). Studies on N mineralisation from livestock waste applications, both in the laboratory and in the field, have revealed large variations in mineralisable N, making it difficult to develop guidelines for its application as a supplementary source of N (Castellanos and Pratt 1981; Serna and Pomares 1991; Paul and Beauchamp 1994; Gilmour et al. 2003; Yang et al. 2004).

Kinetic studies on N transformations in soils amended with pond sludge from intensive livestock facilities are few (Cameron *et al.* 1996; Zaman *et al.* 1998), and have been almost exclusively confined to temperate environments (Westerman *et al.* 1987; Ndayegamiye and Côté 1989; Rees *et al.* 1993; Paul and Beauchamp 1995; Cameron *et al.* 1996; Carey *et al.* 1997; Rochette *et al.* 2000). The objectives of our study were to: (1) compare the patterns of N release from wet or stockpiled PPS; (2) improve the prediction of N availability in the field for crop production following application of PPS to 2 contrasting soils in Australia's subtropical northern cereal region; and (3) evaluate PPS application in terms of equivalent rates of a synthetic N fertiliser (urea).

In this paper we report results that address objectives (1) and (2) above, while objective (3) is addressed in the following paper. We evaluated patterns of N release from PPS applied to a sandy soil and a clay soil under controlled moisture and temperature conditions in the laboratory. Mineralisation of N was assessed using long-term (30 weeks) aerobic leached incubation (Stanford and Smith 1972), short-term (12 weeks) aerobic non-leached incubation (Hart et al. 1994), and anaerobic (waterlogged) incubation for 7 days at 40°C (Keeney 1982). These laboratory assessments were then compared with field measurements of NO₃-N accumulated in the soil profile (0–1.5 m) during a 12-month weed-free fallow period after PPS applications.

Materials and methods

Soils

Two soils of contrasting texture were used for the field trials and the laboratory incubation experiments. These were Millmerran sandy soil (Sodosol; Isbell 1996; Typic Natrustalf; USDA 1975), and Formartin clay soil (Vertosol; Isbell 1996; Udic Haplustert; USDA 1975). The Millmerran site was coarse-textured (8% clay) and had a neutral surface pH (6.7 at 0–0.1 m depth), whereas the Formartin site was fine-textured (56% clay) and had an alkaline surface pH (8.1 at 0–0.1 m depth) (Table 1). Organic C and total N in the 0–0.1 m layers were

Table 1. Characteristics of the two soils of the Darling Downs of Queensland to which stockpiled and wet piggery pond sludge was applied

| Soil type | pН | Sand (%) | Silt (%) | Clay (%) | CEC (cmol/kg) |
|---------------------------------|-----|-------------|-------------|-------------|---------------|
| Millmerran sandy loam (Sodosol) | 6.7 | 86 | 6 | 8 | 7.0 |
| Formartin clay (Vertosol) | 8.1 | 32 | 12 | 56 | 51.3 |

0.54% and 0.04%, respectively, in the sandy soil and 1% and 0.08%, respectively, in the clay soil.

The Millmerran field trial site was located on a soil originally under bull oak (*Casuarina leuhmannii*) and iron bark (*Eucalyptus sideroxylon*) vegetation at Millmerran (27°88′S, 151°21′E), Qld, Australia. The climate of the region is subtropical, with mean annual rainfall of 666 mm and mean annual temperature of 19.9°C. Approximately 34% of the annual rainfall is received in the winter months (April–September inclusive) (Clewett *et al.* 1999). The site was cleared approximately 50 years ago and was cropped annually for 35 years, mainly to barley (*Hordeum vulgare*). Since 1985 the area was sown infrequently with oats (*Avena sativa*). The site received an application of poultry manure in 1993 at approximately 2.4 t/ha. Sheep have grazed the site regularly. The soil is a marginal agricultural soil with the main limitations to agricultural production being low N and P, low water-holding capacity, and a hardsetting surface.

The Formartin field trial site was located on a soil originally under grassland vegetation at Formartin (27°20′S, 157°23′E), Qld, Australia. The climate of the region is subtropical, with mean annual rainfall of 644 mm and mean annual temperature of 19.1°C. Approximately 33% of the annual rainfall is received in the winter months (April–September inclusive) (Clewett *et al.* 1999). The site was cleared between 1940 and 1942 and was cropped annually for approximately 60 years, mainly to cereal crops. The soil has few limitations with respect to agricultural capability, but is responsive to N and P applications. The site had never previously received manure applications nor had it been grazed by livestock during the last 50 years.

For site characterisation, soil samples were collected from each field trial site using a hydraulically operated core sampler with a 50-mm-diameter cutting edge. Six cores were sectioned into 7 depth intervals, 0-0.1, 0.1-0.2, 0.2-0.3, 0.3-0.6, 0.6-0.9, 0.9-1.2, and 1.2-1.5 m, and the respective depth intervals were combined from 2 cores to obtain 3 samples for each depth interval. The soil samples were sealed in polyethylene bags in the field and stored at 4°C until further processing. Samples were weighed, then placed in a forceddraught dehydrator and dried at 40°C for approximately 7 days. Following air-drying, the soil was ground to pass a <2-mm sieve. After mixing thoroughly and collecting a subsample, the samples were stored in sealed plastic containers at room temperature until further analysis. A soil subsample was further ground to pass a 0.25-mm sieve for organic C and total N analysis. Six additional samples were obtained for incubation studies from the 0-10 cm layer, sieved (<4 mm), and stored in field-moist conditions at 4°C until further analysis.

Piggery pond sludge (PPS)

Wet PPS was extracted from a storage pond located on a piggery farm at Millmerran, using a vacuum tanker, and 3 samples (3×20 -L drums) were collected from an outlet on the tanker and stored in air-tight containers at 4° C until further processing. Samples were homogenised and subsampled (1 L) for analysis. Stockpiled PPS was collected from a piggery located at Westbrook, Qld, Australia, after it had been

stockpiled for 12 months. It was dried further after collection on a concrete pad, turned frequently, and then sieved to <10 mm. Three samples of Stockpiled PPS (3 × 5 kg) were collected, homogenized, and subsampled (500 g). Samples of Wet PPS and Stockpiled PPS were each halved. One half was stored at 4°C in its moist state for analysis of PPS composition in an as-collected state; the other half was weighed into a forced-draught dehydrator and dried at 40°C for approximately 7 days. After air-drying, samples were reweighed in order to determine the air-dry moisture content. Stockpiled PPS was ground finely to <0.2 mm using a Glen Creston micro-hammermill to enable uniform representative small volumes to be analysed. A subsample was dried at 105°C for 48 h to determine the oven-dry moisture content of the PPS air-dried at 40°C. Properties of the Wet PPS and the Stockpiled PPS are expressed on an oven-dry weight basis and presented in Table 2. NH₄-N and NO₃-N contents of both PPS were determined from samples 'as collected', although air-drying of Stockpiled PPS at 40°C had little effect on mineral N content.

Design of laboratory studies

Laboratory incubations simulated the applications of PPS at the 2 field sites. Quantities of each sludge were mixed with each soil to establish a mixture of soil and sludge equivalent to those for each field application rate. Four replications of each sludge rate were used for each laboratory incubation experiment (aerobic leached, aerobic nonleached, and waterlogged mineralisation).

Aerobic leached incubation

The potentially mineralisable N (mineralisation potentials) in soil amended with PPS was measured by the aerobic leached incubation procedure proposed by Stanford and Smith (1972), with improvements developed by Bundy and Meisinger (1994). The method was modified in order to utilise a 75-mL syringe (2.5-cm diam. \times 13.5-cm length) as a combined filtration-incubation container. A glass-fibre filter (extra thick, 1 μm , 25 mm diam., no. 66075, Gelman Sciences Inc.), 20- μm frit for 75-mL syringe (no. AGS51FR75, Activon), untreated glass wool (Alltech), and acid-washed sand (AJAX) were used for filtration in the clay soil treatments. The glass-fibre filter and 20- μm frit were omitted for the sandy soil treatments due to clogging by fine clay particles during preliminary tests. Glass wool and acid washed sand were placed on top of the frit and glass-fibre filter to prevent soil particles clogging the filters and to decrease channelling of the leachate.

Table 2. Composition of piggery pond sludge (PPS) applied to two grain-producing soils of subtropical northern Australia expressed on an oven-dry weight basis

| Property (air-dried at 40°C) | Wet PPS | Stockpiled PPS | |
|------------------------------|-----------|----------------|--|
| Total solids (%) | 8.6 | 85.9 | |
| рН | 7.5 | 6.6 | |
| EC (dS/m) | 5.4 | 1.9 | |
| Lignin (%) | 18.8 | 12.5 | |
| Acid digestible fibre (%) | 28.2 | 22.8 | |
| Cellulose (%) | 9.3 | 10.3 | |
| Total C (%) | 25.1 | 6.0 | |
| Total N (%) | 4.6^{A} | 0.77 | |
| C/N ratio | 5.5 | 7.8 | |
| C/organic N ratio | 8.8 | 9.2 | |
| Lignin/organic N ratio | 5.9 | 19.2 | |
| NH ₄ -N (mg/kg) | 189 | 58 | |
| NO ₃ -N (mg/kg) | 0 | 892 | |
| Mineral N (mg/kg) | 189 | 950 | |

^AWet PPS acidified with HCl and dried at 105°C overnight.

Field-moist soil was used in order to avoid artefacts from air-drying and to minimise reduction of the nitrifier population. Wet PPS and Stockpiled PPS were applied to soil in their as-collected state (i.e. they were not dried and ground prior to application). The PPS was applied at the intended field trial application rates of 6 t/ha (D1) and 18 t/ha (D2) of Stockpiled PPS and 17.5 t/ha (W1) and 52.5 t/ha (W2) of Wet PPS, therefore the treatments were comparable across soil types. The PPS rates were calculated on a mass of soil basis taking into account the bulk density of the field soil. Untreated soils were also included as control (C). The required amount of PPS was added to the field-moist soil and thoroughly mixed prior to the addition of acid-washed sand. Acid-washed sand was added in order to maintain favourable water transmission and aeration for the period of the incubation. The sandy soil was mixed at a 1:1 ratio (30 g soil: 30 g quartz sand), whereas twice the amount of quartz sand (20 g soil: 40 g quartz sand) was incorporated for the clay soil to compensate for its high clay content (Campbell et al. 1981; Dalal and Mayer 1987). Following thorough mixing of the soil-PPS-sand mixture, deionised water was added to bring the soil to field capacity (40% and 12.5% gravimetric water contents for the clay soil and the sandy soil). The soil-PPS-sand mixture was then transferred into the syringe, and glass wool and a glass-fibre filter were placed on top of the soil in order to minimise soil dispersion during leaching. Four blanks consisting of filters, glass wool, and acid-washed sand were included to account for trace amounts of N and C derived from the filtration materials.

Net N mineralisation of amended and unamended soil was monitored over 30 weeks of incubation. Soil-PPS-sand mixtures were incubated in a constant temperature room at $21^{\circ}\text{C} \pm 1^{\circ}\text{C}$. This temperature was similar to the mean annual temperature at the 2 field trial sites. The soil columns were leached immediately prior to the initial incubation, Time 0, and again after 1, 2, 4, 8, 12, 16, 22, and 30 weeks of incubation. An initial leaching was necessary to exclude inorganic N applied in PPS amendments and accumulated by soil prior to the commencement of the incubation study.

Syringes containing filters and soil-PPS-sand mixtures were weighed and placed on a vacuum manifold capable of accommodating 24 syringes per leaching. A vacuum regulator and manometer were used to control and monitor the vacuum level. Leachates were collected under a vacuum of $10\,kPa$ for the clay soil treatments, which leached slowly, and $3\,kPa$ for the sandy soil treatments, which leached rapidly, so that essentially similar leaching rates were achieved in both soils. Mineralised N in the soil columns was leached with $100\,mL$ of $0.01\,m$ calcium chloride (CaCl2) solution followed by $25\,mL$ of minus-N nutrient solution containing $0.002\,m$ CaSO4, $0.002\,m$ MgSO4, $0.005\,m$ Ca (H2PO4)2, and $0.0025\,m$ K2SO4 at the rate of $150\,mL/h$. A constant head was maintained on the soil during leaching.

Following leaching, a vacuum of 25 kPa was applied to the clay soil and 10 kPa to the sandy soil for approximately 15 min to remove excess moisture and to provide a uniform moisture content in the soil-PPS-sand mixture for the subsequent incubation period. The syringe was disconnected from the vacuum manifold, weighed, and the bottom sealed with Parafilm and the top of the syringe left unsealed to allow air exchange during incubation. Syringes were weighed prior to and after leaching to enable water loss between leaching events and leachate volume to be determined in order to calculate N leached on a mg/kg oven-dry soil basis. Water loss during incubation intervals was minimal. Leachates from the sandy soil treatments were cloudy as a result of the frit and glass-fibre filter being omitted. Therefore, the leachates were filtered through pre-rinsed Whatman no. 40 filter papers. All solutions were stored at 4°C prior to analysis. Percentage of organic N [mineralised from the PPS treatments was calculated as follows: 100 mineralised N at 30 weeks in the PPS treatment – (mineralised N at 30 weeks in the control treatment + initial mineral N in PPS + NO₃-N produced in the first 2 weeks due to soil disturbance)]/initial organic N in PPS.

Carbon mineralisation was determined simultaneously with N mineralisation by enclosing the incubation syringe inside a 750-mL glass jar and capturing the CO_2 evolved in an alkali trap (Anderson 1982). The alkali trap was a 10-mL volumetric vial containing 1 mL of 1 m NaOH. Approximately 40 mL of water was placed at the base of the glass jar to maintain humidity and minimise soil water loss during incubation. Blanks, consisting of glass jars containing water and alkali traps, were included to account for background CO_2 . Blank values were consistently low.

Aerobic non-leached incubation

Short-term laboratory incubations, such as the aerobic non-leached (12 weeks) and anaerobic incubation (7 days), are considered to be more practical than aerobic leached incubations that take 30 weeks as routine laboratory incubations to determine N availability following amendment of soil with PPS. The soil and sludge mixtures used in the aerobic leached incubation experiment mentioned above were similar to those used for aerobic non-leached incubations.

Mineralisable N in soil amended with PPS was measured by the aerobic non-leached procedure described by Hart $\it et al.$ (1994). Experimental treatments were obtained by applying Wet PPS and Stockpiled PPS at the required rate to field-moist soil (equivalent to 200 g of oven-dry soil). Soil-PPS mixtures were thoroughly mixed and placed into containers (8-cm diam. \times 16-cm height) and the moisture content was adjusted to field capacity by the addition of deionised water. Treatments were incubated aerobically for 12 weeks in the dark in a controlled-temperature growth cabinet maintained at $20\pm0.6^{\circ}{\rm C}$ and at $94\pm2\%$ humidity. Containers were covered with perforated plastic wrap, which allowed gas exchange and minimised water loss. Water loss was monitored every 2–3 days by weighing containers, and the moisture content was adjusted by returning the container to its original weight with deionised water.

Concentrations of mineral N were determined following 0, 0.5, 1, 2, 4, 8, and 12 weeks of incubation. At each sampling time, 4 replications of each Control and soil-PPS treatment were randomly selected for analysis and analysed using 2 $\,$ M KCl as an extractant (1:4 soil: extractant), and filtered through pre-rinsed Whatman no. 40 filter papers. All extracts were stored at 4 $\,$ C prior to analysis for mineral N.

Anaerobic (waterlogged) incubation

Mineralisable N in soil amended with PPS was measured by incubation under waterlogged (anaerobic) conditions for 7 days at 40° C (Keeney 1982). Control and PPS-amended field-moist soil equivalent to 5 g of oven-dry soil was extracted immediately (Time 0) and an equivalent to 5 g of oven-dry soil was anaerobically incubated for 7 days at 40° C in 12.5 mL of water. To assess the effect of air-drying soil prior to waterlogged incubation, the remaining Control and PPS-amended field-moist soil, 10 g of oven-dry soil, was air-dried at 40° C for 7 days.

Methodology presented by Bundy and Meisinger (1994) was modified and ammonium-N (NH₄-N) was extracted by the addition of 37.5 mL of $2.67 \, \text{m}$ KCl to the test tube containing the waterlogged soil mixture and shaking for 1 h. Extracts were filtered through prerinsed Whatman no. 40 filter papers and stored at 4°C prior to analysis for NH₄.

Soil nitrate accumulation in field experiments

Field experiments consisted of 5 PPS treatments: 2 rates of Wet PPS (17.5 and 52.5 t/ha, W1 and W2, respectively), 2 rates of Stockpiled PPS (6 and 18 t/ha, D1 and D2, respectively), and an unamended control (C). Four replicates of each treatment were arranged in a randomised block design. The plot size at both sites was 9 by 25 m, and the plots were split

longitudinally (4.5 by 25 m each) for summer and winter crops. Plot width was determined by both the wet sludge spread width of 8.5 m and the planter/harvester width of 4 rows of 2.25 m each. Plot locations within the randomised block design remained the same throughout the experiment.

Stockpiled PPS was applied by hand on the 17 and 18 December 1997 at Formartin and Millmerran, respectively. Wet PPS was collected from a non-agitated lagoon, using a vacuum tanker, and was applied on 19 December 1997. The vacuum tanker used a splash plate for spreading the Wet PPS. The Wet PPS was incorporated to approximately 0.15-m soil depth within 30 min of application at each site, with offset discs in order to minimise NH₃ volatilisation. Stockpiled PPS was incorporated at the same time as the Wet PPS. Subsamples of both PPS were taken for laboratory studies and chemical analysis. Weeds were controlled on the trial sites by manual chipping along with the selective use of herbicides. Roundup (glyphosate) was used for total control of weeds in fallow areas

After application of PPS, plots were sampled for NO₃-N in January, February, March, and September 1998 during the fallow period. Two cores were collected per plot by a hydraulically operated core sampler with a 50-mm-diameter cutting edge. Cores were sectioned into 7 depth intervals, 0–0.1, 0.1–0.2, 0.2–0.3, 0.3–0.6, 0.6–0.9, 0.9–1.2, and 1.2–1.5 m, and the respective depth intervals from the 2 cores were combined. The soil samples were sealed in polyethylene bags in the field and stored at 4°C until further processing. Samples were weighed, then placed in a forced-draught dehydrator and dried at 40°C for approximately seven days. Following air-drying, samples were ground (<2 mm) for analysis. Nitrate was extracted with 2 m KCl at a soil : extractant ratio of 1 : 10 (10 g soil : 100 mL KCl) (Bremner 1965). Extracts were filtered through pre-rinsed Whatman no. 40 filter papers, and stored at 4°C until analysed for NO₃.

Analytical procedures

Particle size distribution was measured by dispersing soil (<2 mm) with a Cullex cation exchange resin (Edwards and Bremner 1965). Clay (<0.002 mm) and silt (0.002-0.02 mm) were determined by a pipette and sedimentation procedure (Day 1965). Sand (0.02-2.0 mm) was calculated by difference. Soil pH was measured in the supernatant liquid of a 1:5 (soil: deionised water) suspension (Rayment and Higginson 1992). Cation exchange capacity was measured using alcoholic 1 M ammonium chloride at pH 8.5 (Rayment and Higginson 1992). Finely ground (<0.25 mm) soil samples were analysed for organic C by the dichromate oxidation method (Walkley and Black 1934), followed by measurement of chromium (Cr III) produced relative to sucrose standards (Sims and Haby 1971). Soil total N was determined using a semimicro Kjeldahl digestion with colourimetric N quantification using the indophenol blue method (Rayment and Higginson 1992). Ammonium (NH₄) concentration in extracts was determined by an automated procedure using a modified Berthelot indophenol reaction with alkaline Na salicylate/nitroprusside/isocyanurate (Crooke and Simpson 1971). Nitrate-N in KCl extracts was determined by an automated hydrazine reaction procedure for the Griess-Ilosvay reaction for NO2 (Best 1976). Soil profile NO₃-N content was expressed on an oven-dry weight basis (kg/ha) from soil moisture content and BD for each layer. Sorbed CO2 in the soil respiration experiment was determined by adding 60 mL of deionised water to the NaOH, precipitating the carbonate with 1 mL of 2 M BaCl2, and the remaining NaOH (excess alkali) was back-titrated with 0.05 M HCl using an automatic titration unit (Anderson 1982).

Statistical analysis

The experimental design for all incubation experiments was a randomised complete block design with 4 replicates. Statistical analysis

of inorganic N (NH₄ and NO₃), leached or extracted at each sample time, was conducted using a one-way ANOVA. Cumulative inorganic N leached during the 30 weeks of incubation was calculated by summing the inorganic N leached from each sample interval. Cumulative inorganic N leached from treatments over time was analysed using repeated measures ANOVA with a Greenhouse-Geisser adjustment of the degrees of freedom (Greenhouse and Geisser 1959).

Each field experiment was a randomised complete block (RCBD) with four replicates (Blocks). Statistical analysis was performed using the statistical package Genstat 5 (Release 4.1, 4th Edn) (Lawes Agricultural Trust 1996). In some cases, a log transformation of the data improved the distribution of residuals. Means were separated using least significant differences (l.s.d.). For uniformity, l.s.d. values were calculated at $P \le 0.05$, when main effects or interactions were significant at or less than 5%. The relationships between measured parameters were investigated using single regression analysis (Steel and Torrie 1980).

Results

Aerobic non-leached soil incubation

Organic N mineralised from the soil and PPS during the 30-week incubation varied from 76 mg/kg in the unamended clay soil (control) to 274 mg/kg in amended (W2) sandy soil (Fig. 1). Sandy soil mineralised larger amounts of N than clay soil. From the Stockpiled PPS, almost 50% of organic N was mineralised in the sandy soil, whereas only 10% was mineralised in the clay soil (Fig. 1). All of the organic N from the Wet PPS was mineralised in the sandy soil but much less organic N was mineralised (<50%) in the clay soil (Fig. 1). In the former, there appears to be an apparent priming effect on soil organic N from the application of the wet PPS.

Cumulative organic N mineralised during the 30 weeks' incubation from the 2 soils amended and unamended with PPS was successfully described by the following first-order

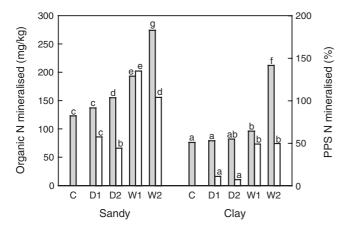


Fig. 1. Organic N mineralised during 30 weeks of leached incubation (shaded bars) and proportion of organic N from applied PPS mineralised after discounting for the first week of N mineralisation due to soil disturbance (open bars) from sandy and clay soils. Within bar shading and soil type, same letters at the top of the bar denote no significant difference at P = 0.05.

exponential equation ($R^2 \ge 0.98$ and low standard error of the estimate, Table 3), modified to exclude the initial flush (Beauchamp *et al.* 1986):

$$N_{\rm m} = N_{\rm o} - (N_{\rm o} - N_{\rm i}) \exp(-kt)$$
 (1)

where N_m is the organic N mineralised at time t (week), N_0 is the potentially mineralisable N or mineralisation potential (Stanford and Smith 1972) at infinite time (\propto), and N_i is the mineral N initially present or mineralised in the first week due to soil disturbance. The mineralisation rate constant of organic N is given by k (/week) in Eqn 1.

As for the 30-week cumulative N mineralised, potentially mineralisable N was higher for the sandy soil (156 mg/kg) than for the clay soil (96 mg/kg), although soil total N in the 0–0.1 m layer of the sandy soil was lower (0.043%) than for the clay soil (0.080%). The sandy soil also had higher amounts of net organic N mineralised from the applied PPS than the clay soil (Table 3). However, mineralisation rate constants, k values, showed little variation between the PPS-amended and unamended soils or across soil types (mean k value = 0.057/week) (Table 3). Estimated potentially mineralisable N values, using the mean k value, were significantly ($R^2 > 0.99$) increased by wet PPS application and remained higher in the sandy soil than in the clay soil (Table 3).

The total amount of CO₂ evolved was similar for comparable treatments on the two contrasting soils during the 30 weeks of incubation, except for the W2 treatment (Fig. 2). The W2 treatment evolved more CO₂ when applied

Table 3. Mineralisation rate constant (k, per week) and potentially mineralisable N (N_o , mg/kg) estimated from the 30-week leached incubation of unamended and PPS-amended sandy and clay soils $N_m = N_o - (N_o - N_i)$ exp(-kt), where N_m is the organic N mineralised at time t (week), N_o is the potentially mineralisable N or mineralisation potential at infinite time (\propto), and N_i is the mineral N initially present or mineralised in the first week due to soil disturbance. D1 and D2, Stockpiled PPS applied at 6 and 18 t/ha equivalent; W1 and W2, Wet PPS applied at 17.5 and 52.5 t/ha. Within column, values followed by the same letter are not significantly different at P = 0.05

| Treatment | k | N _o | R^2 | s.e.e. ^A | N _o ^B | PPS N _o ^C |
|-----------|-------|----------------|----------|---------------------|-----------------------------|---------------------------------|
| | | | Sandy se | oil | | |
| D1 | 0.044 | 193 | 0.997 | 2.87 | 169 bc | 13 a |
| D2 | 0.044 | 221 | 0.997 | 3.07 | 193 cd | 37 b |
| W1 | 0.069 | 237 | 0.986 | 8.05 | 256 d | 100 d |
| W2 | 0.076 | 299 | 0.985 | 10.7 | 335 e | 179 f |
| | | | Clay so | il | | |
| D1 | 0.032 | 137 | 0.998 | 1.15 | 98 a | 2 a |
| D2 | 0.032 | 141 | 0.997 | 1.48 | 102 a | 6 a |
| W1 | 0.044 | 174 | 0.997 | 1.95 | 158 b | 62 c |
| W2 | 0.054 | 250 | 0.996 | 3.17 | 245 d | 149 e |

^AStandard error of estimate.

 $^{{}^{\}rm B}{\rm N}_{\rm o}$ estimated from mean k value (0.0569/week).

^CNet mineralisation potential of PPS applied to soil (control soil value subtracted; l.s.d. (P = 0.05) = 14.7 mg/kg soil).

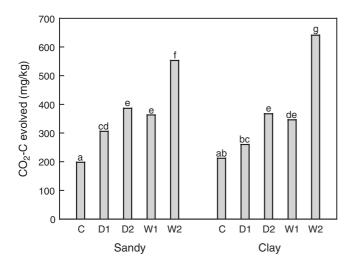


Fig. 2. The relationship between total CO_2 evolved during 30 weeks of incubation and the organic C content of unamended and PPS-amended sandy and clay soils. Within a soil type, same letters at the top of the bar denote no significant difference at P = 0.05.

to the clay soil than when applied to the sandy soil. Addition of PPS increased the amount of CO_2 evolved, except for the 6 t/ha Stockpiled PPS treatment (D1) on the clay soil. Organic N mineralised and CO_2 -C evolved during the 30-week incubation were significantly correlated ($R^2 = 0.60$).

Aerobic non-leached incubation

Application of Wet PPS resulted in a net increase in NO₃-N extracted following 12 weeks of incubation in both soils (Table 4). As the application rate of Wet PPS increased, the amount of NO₃-N produced also increased. The proportion of organic N mineralised from the Wet PPS was 17% for the W2 treatment on the sandy soil and 22% for the W2 treatment on the clay soil. No net mineralisation of organic N occurred from the Stockpiled PPS treatments.

The amount of NO_3 released during 12 weeks of incubation by non-leached and leached studies was strongly correlated ($R^2 > 0.96$) on both soils (Fig. 3). However, net NO_3 extracted following 12 weeks of aerobic non-leached incubation was <60% of that released during 12 weeks of aerobic leached incubation from both sandy and clay soils (Fig. 3).

Anaerobic (waterlogged) incubation

Similar to aerobic non-leached incubation, Wet PPS treatments significantly increased anaerobically mineralisable N whether field-moist or air-dried soil samples were used for incubation (Table 4). However, the amounts of mineralisable N in air-dried soil were higher than in the field-moist soil, and higher in the clay soil than in the sandy soil. The anaerobically mineralisable N was strongly correlated with N mineralised during aerobic leached incubation

Table 4. Net change in nitrate-N under aerobic non-leached incubation (Net NO₃-N, mg/kg) and ammonium-N produced under anaerobic incubation (NH₄-N, mg/kg) in two PPS-amended soils

D1 and D2, Stockpiled PPS applied at 6 and 18 t/ha equivalent; W1 and W2, Wet PPS applied at 17.5 and 52.5 t/ha. The sandy soil in the control treatment contained 7.0, 4.7, and 15.1 mg/kg net amount of NO₃-N, field-moist soil NH₄-N, and air-dried soil NH₄-N, respectively; the corresponding values in the clay soil were 4.7, 7.4, and 31.0 mg/kg. Within a column, values followed by the same letter are not significantly different at *P* = 0.05

| Treatment | Aerobic non-leached | Anaerobic incubation | | |
|-----------|------------------------|----------------------|--------------------|--|
| | incubation | Field-moist | Air-dried | |
| | Net NO ₃ -N | NH ₄ -N | NH ₄ -N | |
| | Sandy soi | l | | |
| D1 | 1.3 a | 1.5 a | 0.8 ab | |
| D2 | −1.5 a | 2.3 a | -0.7 a | |
| W1 | 35.8 b | 5.0 c | 9.1 c | |
| W2 | 101.0 c | 11.7 d | 22.7 e | |
| | Clay soil | | | |
| D1 | 0.9 a | 1.7 a | 3.4 b | |
| D2 | -0.3 a | 2.0 a | 1.9 b | |
| W1 | 33.6 b | 3.3 bc | 10.0 c | |
| W2 | 98.3 c | 12.4 d | 13.8 d | |

(Fig. 4), but the regression slope of the air-dried soil (0.105) was twice the slope of the field-moist soil (0.05). In contrast, the anaerobically mineralisable N from field-moist clay soil was better correlated ($R^2 = 0.93$) with net N mineralised during aerobic leached incubation than anaerobically mineralisable N from air-dried clay soil ($R^2 = 0.65$) (Fig. 4). However, the slopes of the 2 relationships were similar (0.045) but the intercepts were significantly different, indicating that air-drying released mineralisable N, possibly from the killed microbial biomass.

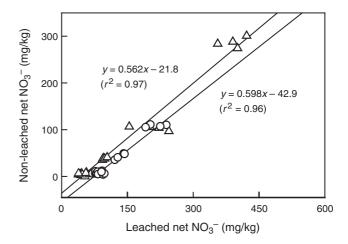


Fig. 3. The relationship between net NO_3 -N extracted during 12 weeks of aerobic non-leached incubation and 12 weeks of aerobic leached incubation from unamended and PPS-amended sandy (O) and clay (\triangle) soils.

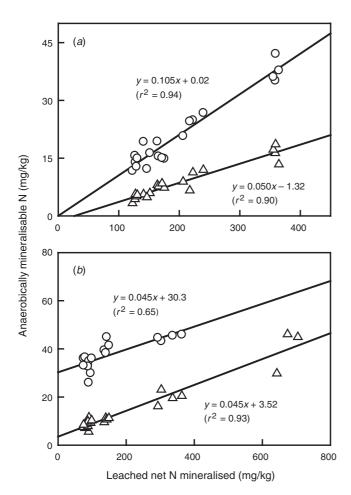


Fig. 4. The relationship between anaerobically mineralisable N and net N mineralised during 30 weeks of aerobic leached incubation from unamended and PPS-amended air-dried (\bigcirc) and field-moist (\triangle) (a) sandy soil and (b) clay soil.

Soil profile nitrate accumulation

Amounts of NO₃ in the sandy soil (0-1.5 m) after fallowing (December 1997–September 1998) were 52, 47, 58, 56, and 119 kg N/ha (l.s.d., P < 0.05, 31 kg N/ha) in the control, D1, D2, W1, and W2 treatments, respectively. The amounts of NO₃ in the clay soil profile after fallowing for a similar period in the corresponding treatments were 36, 68, 111, 83, and 127 kg N/ha (l.s.d., P < 0.05, 34 kg N/ha). In neither soil, however, was there a significant increase in NO₃ in the 1.2–1.5 m layer (Kliese 2002) after application of PPS. The net change in NO₃ content as a proportion of organic N in the Stockpiled PPS for the D1, D2, and W1 treatments on the sandy soil was negligible (-5.3, 4.5, and 4.6%, respectively). On the clay soil, however, 81% of the organic N applied in the D1 treatment and 59% of the organic N applied in the D2 treatment appeared mineralised. The 25% of total N applied in the W2 treatment available on the sandy soil (66 kg NO₃⁻-N/ha) was similar to the amount of mineral N contained in the applied Wet PPS (81 kg NH₄⁺-N/ha). Similarly, the proportion of NO₃-N present in the W1 treatment (29%) and the W2 treatment (19%) on the clay soil 9 months after application of PPS was similar to the quantity of mineral N contained in the applied Wet PPS, thus emphasising the importance of mineral N contained in Wet PPS applied directly from the pond.

Value of laboratory tests to estimate N release in PPS-amended soils

Relationships between laboratory estimates mineralisation potential, NO₃ released during the 30week incubation (leached), NO₃-N released during the 12-week incubation (non-leached), and NH₄-N released during the 7-day anaerobic incubation, and the quantities of NO₃ determined in situ after PPS field application, are shown in Fig. 5. For the sandy soil, all laboratory incubations were significantly correlated ($R^2 = 0.9-0.95$) with NO₃ measured in the field. In contrast, for the clay soil, laboratory incubations were poorly correlated $(R^2 = 0.46 - 0.50)$ with NO₃ measured in the field, except for anaerobic mineralisable N ($R^2 = 0.59$). Thus, it is possible to improve the predictability of N uptake by crops from PPS-amended soils by including the immediately available source such as NO₃ in the soil and potentially mineralisable N such as anaerobically mineralisable N, which may become available during the crop growth.

Discussion

Influence of sludge treatment on N release in soil

Two differences between stockpiled and wet piggery pond sludges applied in these studies might explain apparent difference in N release after their application to soil. Firstly, the higher lignin content of stockpiled sludge than of wet sludge, evident from ratios of lignin to C, N, or organic N, could be one reason for the decreased release of N after application of stockpiled than after wet application. Secondly, the high content of mineral N, mainly ammonium-N, in wet sludge would have exacerbated this difference in available N supply after applications of the 2 sludges.

The low proportions of organic N mineralised from the Stockpiled PPS, applied to the clay soil (11% for the D1 treatment and 7% for the D2 treatment), were similar to the 8% for composted and 15% for anaerobically digested sewage sludge applied to a silt loam soil, reported by Parker and Sommers (1983), and to the 13–18% reported by Zaman *et al.* (1998) from the incubation of dairy pond sludge with silt loam soil. Similar low proportions of organic N mineralised on fine textured soils were probably due to these organic materials all being stabilised during storage.

To effectively utilise PPS as an N source for crop production, the rate of N release by mineralisation must

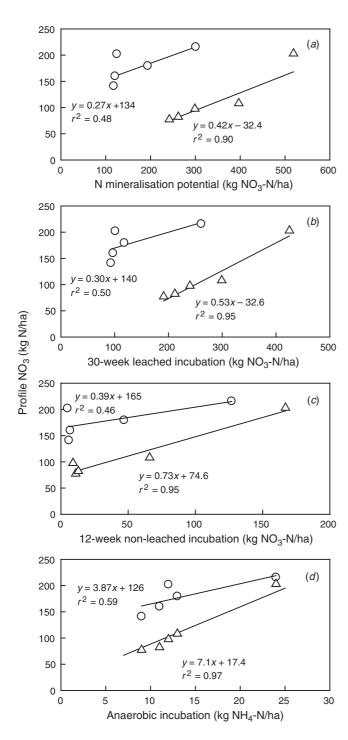


Fig. 5. Relationship between accumulated NO₃-N in the soil profile and NO₃-N estimated from (a) N mineralisation potential, (b) 30-week leached incubation, (c) 12-week non-leached incubation, and (d) NH₄-N produced during anaerobic incubation in unamended and PPS-amended sandy (\bigcirc) and clay (\triangle) soils.

be predicted. Thus, it would be beneficial to determine the reasons why the rates of C and N mineralisation differ among sludges. According to the total C/total N and the total C/organic N ratios, the 2 forms of PPS should

behave similarly when applied to soil (Table 2). Incubation studies showed that the proportion of organic N from the Wet PPS mineralised more than that from Stockpiled PPS (Tables 3 and 4; Fig. 1). Others have also shown that the C/N ratio is not well correlated with decomposition of some substrates and that materials with similar C/N ratios may decompose differently (Gilmour *et al.* 1985; Levi-Minzi et al. 1990; Fauci and Dick 1994). Therefore, it is considered that the C/N ratio as an indicator of organic matter (OM) decomposition is no longer relevant when stabilised materials such as PPS are being considered. However, if the lignin/total N, lignin/organic N (Table 2) and lignin/total C ratios are considered, then a difference exists between the Wet PPS and Stockpiled PPS. For example, the lignin/organic N ratio of the Wet PPS was lower (5.9) than of the Stockpiled PPS (19.2). The higher lignin content of the Stockpiled PPS would suggest that its OM would be more stable or recalcitrant than the OM contained in Wet PPS. Hence, land-applied Wet PPS would be expected to be a better source of mineralisable N for crop use. Moreover, the high content of inorganic N in Wet PPS, largely as NH₄ (1240 mg N/kg on as-collected basis) would further elevate Wet PPS over Stockpiled PPS as a nutrient source for land application.

Influence of soil texture on N release from applied sludge

As has been previously reported (Castellanos and Pratt 1981; Chae and Tabatabai 1986; Ladd et al. 1992), net N mineralisation was lower in the soil with the higher clay content, due possibly to the combined effect of soil aeration and immobilization, since ammonium fixation appears to be <5% of applied NH₄-N in the clay soils of the region (Black and Waring 1972). All 4 laboratory estimates of N release after application of the 2 sludges, organic N mineralised over 30 weeks (Fig. 1), potentially mineralisable N (Table 3), and aerobic (12 weeks) and anaerobic (7 days) mineralisable N (Table 4), indicated higher net mineral N values after application to sandy soil than after application to a clay soil. There may have been a priming effect on soil organic N in the sandy soil from the application of Wet PPS due to its relatively labile organic N (and C), whereas in the clay soil these constituents would have been rapidly sorbed, retained, or immobilised by the clay. For example, Sørensen and Jensen (1995) found that net N immobilisation from cattle slurry increased with increasing clay content. Thus, effect on immobilisation due to contrasting clay content could explain the considerable difference in apparent release of N from PPS applied to these two soils.

Protection of OM, in particular PPS, in many small pores of the clay soil, as suggested by Hassink *et al.* (1993), is an unlikely explanation for lower net N mineralisation than in the sandy soil. Unamended soils displayed similar difference in net N mineralisation. Furthermore, application of PPS to field-moist soil immediately prior to incubation would

have given little opportunity for PPS to occupy smaller soil pores.

Comparison of estimates of N mineralisation with those of other studies

Estimated potentially mineralisable N for unamended sandy soil was 160.5 mg N/kg and for unamended clay soil was 115.6 mg N/kg soil. These values were within the range of mean values (110-270 mg N/kg soil), reported for 7 soil orders by Jones et al. (1982). A Waco Black Earth similar to the clay soil used in this study was found by Dalal and Mayer (1987) to have higher potentially mineralisable N, 203 mg N/kg soil, than the clay used in these studies, 115.6 mg N/kg soil. This difference was due probably to higher total N content (0.14% v. 0.08%) and a higher incubation temperature (40°C for 30 weeks) used by Dalal and Mayer (1987). Therefore, the value for potentially mineralisable N determined at 20°C in this study at approximately half of the value reported by Dalal and Mayer (1987), would seem comparable.

Similarly, mineralisation rate constants were similar (0.032–0.076/week) (Table 3) to those of other workers who used a long incubation period (30 weeks) and a lower temperature (20°C) (Chae and Tabatabai 1986; Simard and Ndayegamiye 1993).

Parameter estimates from studies with longer incubation times invariably result in larger potentially mineralisable N and lower and less variable mineralisation rate constants (Paustian and Bonde 1987; Sierra 1990). The mineralisation rate constants for unamended sandy soil (0.053/week) and unamended clay soil (0.04/week) were similar to the mean mineralisation rate constants of Waco Black Vertosols (0.050/week) but lower than of Langlands-Logie and Cecilvale Grey Vertosols (0.075–0.078/week) incubated for 30 weeks at 40°C by Dalal and Mayer (1987).

Comparison of estimates of C mineralisation with those of other studies

In contrast to substantial effects on N mineralisation, soil type had little effect on the total CO₂ released during 30 weeks of incubation from unamended or PPS-amended soil. Hassink *et al.* (1993) also reported no significant effect of soil texture on the percentage of organic C mineralised, even though N mineralisation was affected by soil texture as found in our studies. Comparable results have been reported where sewage sludge was applied to a wide range of soils differing in pH, clay, and OM contents; soil properties were not found to be significant factors in determining the extent of C mineralisation after sewage sludge incorporation into soil (Gilmour *et al.* 2003). However, some researchers have reported greater C stabilisation in soils with higher clay contents (Sørensen and Jensen1995,). When estimating parameters to describe organic N mineralisation from

application of PPS, it is essential to ensure that the effects of initial inorganic N are excluded.

The low proportion of organic N from Wet PPS mineralised in aerobic non-leached incubation in comparison with that mineralised in the aerobic leached incubation (Fig. 1), is more than likely due to the closed system of the aerobic non-leached incubation. This resulted in higher N immobilisation in the non-leached system because C and N are not regularly displaced as they were in the aerobic leached incubation (Garau *et al.* 1986; Hart *et al.* 1994). However, organic N mineralised as NO₃ was closely correlated (Fig. 3) between the aerobic leached incubation and aerobic non-leached incubation.

Use of laboratory estimates to determine supplementary crop N needs

Anaerobic incubation was sensitive enough to detect significant differences between mineralisable N following amendment of sandy and clay soils with PPS. Short-term (7 day) anaerobic incubation resulted in N release that was well correlated with that derived from a 30-week aerobic incubation, leached regularly. Soil drying prior to incubation increased N release from both soils as has been previously observed (Westerman et al. 1988). Since soil drying had a somewhat different influence on N released anaerobically (Fig. 4a, b) when using this test across different soils fieldmoist soil would be preferable. Short-term (7 day) anaerobic incubation also has been shown to be a useful comparative measure of N release for soil amended with plant residues by Hossain et al. (1996), who investigated the effects of leguminous ley pastures on N release in soils of south-eastern Queensland (Warra).

Net N supply as NO₃-N/ha produced from PPS application can be estimated from the relationship between field-measured NO₃ and laboratory measures of mineral N released from PPS-amended and unamended soils (Table 5). For example, additional 20 kg NO₃-N/ha can be supplied by the application of 17.5 t/ha of the wet PPS in both soils. This can be rapidly assessed from the 7-day anaerobic incubation.

Table 5. Net N supply (kg/ha soil NO₃-N) from Wet PPS application estimated from the relationship between field NO₃ and the four laboratory measures of mineral N release from PPS-amended and unamended soils in Fig. 5
W1 and W2, Wet PPS applied at 17.5 and 52.5 t/ha

| Treatment | N_o | 30 weeks leached | 12 weeks aerobic | 7 days anaerobic |
|-----------|-------|---------------------|---------------------|---------------------|
| | | Sandy | | |
| W1 | 20.8 | 29.8 | 17 | 19.3 |
| W2 | 37.5 | 53.5 | 43 | 45.3 |
| | | Clay | | |
| W1 | 24.4 | 32.8 | 35.9 | 23.5 |
| W2 | 56.3 | 79 | 82.6 | 88.1 |

Conclusions

Differences in susceptibility to decomposition of Wet PPS and Stockpiled PPS are evident from both the aerobic non-leached and anaerobic incubation methods, with Wet PPS being more susceptible to decomposition. Results from both incubation methods were well correlated with the results from aerobic leached incubation, except for the net N mineralised during aerobic non-leached incubation. Since anaerobic incubation is also better correlated with field NO₃, anaerobic incubation would, therefore, be suitable as a rapid practical test to detect differences in potentially mineralisable N following additions of PPS on a field scale. Anaerobic mineralisable N was determined on field soil samples where Stockpiled and Wet PPS were applied, and along with NO₃, was correlated with crop N uptake and crop yield. The results are presented in the subsequent paper. In conclusion, wet PPS provides a readily available mineral N source for crops.

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