

# Recycling of Phosphate from Animal Bones

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**Abstract:** Phosphorus is essential for food production and modern agriculture currently sources phosphorus fertilizers from finite phosphate rock. The 2008 food and phosphate fertilizer price spikes triggered increased concerns regarding the depletion timeline of phosphate rock reserves. While estimates range from 30 to 300 years and are shrouded by lack of publicly available data and substantial uncertainty, there is a general consensus that the quality and accessibility of remaining reserves are decreasing and costs will increase. Increasing environmental, economic, geopolitical and social concerns about the short and long-term use of phosphate rock in agriculture means there is a need to reassess the way crops obtain their phosphorus and humanity is fed. The animal bones are generally disposed of as waste by abattoirs in large cities and towns and take many years to decompose and yet they are a rich source of phosphate that can be harvested and used in fertilizer production. This study set out to prepare bone phosphate enriched phosphoric acid from the otherwise discarded animal bones. The extracted bone-phosphate enriched phosphoric acid was reacted with ammonia to generate the diammonium phosphate ( $(\text{NH}_4)_2\text{HPO}_4$ ) fertilizer.

**Keywords:** Animal bones, Enriched phosphoric acid, Diammonium Phosphate Fertilizer

## 1. Introduction

Bones are structurally complex materials that comprise of both organic (Collagen and other proteins) and inorganic chemicals (calcium, phosphate, carbonate, fluorine) calcium and phosphorus forms the greater part of the inorganic matter of bones. They have the chemical formula  $\{\text{Ca}_{10}[(\text{PO}_4)_6(\text{CO}_3)_2](\text{OH})_2\}$ . Recycling of phosphate from animal bones is a very attractive and promising method for mitigation of the phosphate problem. Each year, large amounts of phosphate fertilizers are applied to the fields to provide the suitable amounts of this macronutrient to ensure the proper development and growth of crop plants (Jastrzebska, *et al.*, 2018 & Rolewicz, *et al.*, 2017). The primary method of phosphate fertilizer production is digestion of high grade phosphate rock by sulphuric acid in a wet process producing phosphoric acid and super phosphates (Jastrzebska, *et al.*, 2018). Fertilizer soils are the key to sustainable commercial scale production of crops for feed fiber (Dawser and Hilton, 2011). However, the reserves of high quality phosphorite could be depleted in the next 3 – 4 decades and after only a low grade deposits, contaminated by cadmium or uranium and costly to extract and purify will remain (DeRidder, *et al.*, 2012 & Rittmann, *et al.*, 2011). Additionally, environmental problems related to creation of a huge amount of waste phosphogypsum contaminated by radionuclides (Rolewicz, *et al.*, 2017 & Smith *et al.*, 2006). Common responses to resource scarcity problems include higher prices, more efficient resource use, the introduction of alternatives, and the recovery of the resource after use. Phosphorus is essential and unsubstitutable nutrient for plants and animals, (Dawson & Hilton, 2011) and Schroder *et al.*, 2000). Animals can gain P with food, while plant from soil. Because soil has been intensively exploited during the last 50 years it is necessary to supplement the deficiency caused by intense agriculture. Fertile soils are the key to sustainable commercial-scale production of crops for food, feed and fiber (Dawson & Hilton, 2011). The most concentrated form of organic phosphorus are bones (Schroder, *et al.*, 2000 and SuX, *et al.*, 2003). Utilization of bones as a source of phosphorus has already a long history. After 1770, when phosphorus was discovered in bones and

many other parts of various animals, many attempts were made to obtain phosphorus from them. Until the middle of nineteenth century, bone and guano were used as the raw material to produce phosphorus and phosphoric acids (Demirbus and Abali, 1999). In 1842 John B. Lawes had the first British patent in this field on the manufacture of superphosphate by reacting bone with sulphuric acids (Demirbus and Abali, 1999).

Bone meal or gelatinized bone meal are among the fertilizers allowed in organic farming (European Commission, 1991). Cayuela reported the soil amendment with meat bone meal resulted with an immediate and remarkable increase of C mineralization and N availability (carbon and nitrogen mineralization are the main process regulating the availability of nutrients for plant) and microbial biomass size and activity in the soil, that fatherly control decomposition dynamics. They are the main source of enzymes in the soil (Enowashu, *et al.*, 2009). Biogenic apatite (bone) has the lowest total concentrations of all elements. Low concentrations of impurity in biogenic apatite are the result of a short accumulation time (few weeks are needed for bone formation) (Tutken, *et al.*, 2008). However, after this material becomes deposited in sediments, it incorporates trace elements over geological timeframes at concentration levels that are enriched by many orders of magnitude over initial levels (Sneddon, *et al.*, 2006). Another advantage of application of bones is the lack of fluorine in bones that plays a major role in the formation of sedimentary apatite and contributes to its preservation in sediments (Soundry and Nathan, 2000).

It is found that biogenic apatite (bone) has the largest solubility with a log (Ksp) of 45.2, compared with 57.0 for the mined rock phosphate and 48.0 for beneficiated phosphate rock (Coutand, *et al.*, 2008). Also, biogenic apatite had the highest P concentrations in the water soluble fraction (505 mg/kg compared with 307 mg/kg for processed phosphate and 22 mg/kg for mined phosphate). Biogenic apatite has lower concentrations of impurities than mined and beneficiated phosphate, especially mined and processed phosphates from older deposits (Knox, *et al.*, 2006). As

described by Jeng *et al.*, (2007) and Warren *et al.*, (2009), bone meal is a source that is rich in P, as much as phosphates of mineral origin are, and it has a high concentration of Ca ( $83.7-111.0 \text{ g kg}^{-1}$ ), an interesting alternative for nutritional supplementation of plants such as sugarcane in soils with first-year crops. A study by Wesley *et al.*, (2019) demonstrated the efficiency of phosphate fertilizers with bone meal in forage sugarcane, which showed a response similar to that of triple superphosphate fertilization in attributes such as plant height, number of tillers, fresh mass and dry mass. The authors reported that this effect possibly occurred due to some factors such as the climatic condition of the region, planned soil fertility and crop management practices (minimum planting and proximity between rows), whose main objective is to control undesirable plants and incorporate nutrients via topdressing fertilization, thereby improving the effect of phosphates.

In this work, bone phosphate enriched phosphoric acid is prepared from the otherwise discarded animal bones.

## 2. Materials and Methods

### 2.1 Animal bone preparation

Bones were collected from Kachok Municipal dump site, Kisumu West District, Kenya. They were washed with running tap water, rinsed with de-ionized water and dried in a temperature-controlled oven (WTC binder, 150533, Germany) at  $110^\circ\text{C}$  for twelve hours. They were reduced in size using a hammer at Kenyatta University Physical Chemistry Research Laboratory and ground using a grinding machine at the Geology and Mines laboratories, Kenya, to enhance the rate of digestion in phosphoric acid

### 2.2 Bone phosphate extraction

This was done following the procedure highlighted in the article "Manufacture of phosphoric acid from hydroxyl apatite contained in the ashes of the incinerated meat – bone ashes" by Kinga Krupa – Zuczek *et al.*, 2008. The process of evaluating the lowest concentration of phosphoric acid for total dissolution of bones was carried out by varying the concentrations from 0.1 M, 0.2 M, 0.25 M, 0.275 M and 0.3 M. These concentrations were each exposed to 10 g of bones each at  $75^\circ\text{C}$  and agitated for two hours. The concentrates were filtered, the unreacted bones were rinsed with deionized water and dried by squeezing between two filter papers. The filtrate was reacted with 6 M  $\text{H}_2\text{SO}_4$  acid to precipitate the  $\text{Ca}^{2+}$ . It was filtered and the acid concentration in the filtrate determined by back titration. The % P in the filtrate was determined and results recorded in Table 4.3. From the results it was determined that the lowest concentration of phosphoric acid to completely dissolve bones was 0.275 M. Therefore, volumes of 10000 ml of 0.275 M phosphoric acid were used to dissolve 10 kg of bones in plastic containers for a period of seven days. A stirring bar was used for stirring the solutions after every twenty four hours to ensure that all the bones were dissolved. The laboratory temperatures varied between  $21 - 25^\circ\text{C}$  during this period of experiment. Ten twenty-litre plastic containers were used in dissolution of bones and replicate measurements were done for a period of two

months to dissolve about 130 kg of bones and to confirm the solubility data. The resulting mixture was filtered and stored. 5.0 ml of the sample was measured and dissolved in 50 ml of deionized water each. The solution was then transferred to three 100 ml of volumetric flasks separately and diluted up to the mark with deionized water. A solution of each sample (1.0 ml) was transferred to a 25 ml volumetric flask followed by 2 ml of 2.5 % ammonium molybdate and 0.5 ml of 1 M sulphuric acid solutions. The mixture was shaken before adding 1.0 ml of 0.5 M hydrazine hydrate solution and the volume made up to mark with deionized water. The solution was allowed to stand for about 45 minutes for maximum colour development. Absorbance was taken at a wavelength of 830 nm. A calibration curve (Figure 4.1) for the standard phosphate solutions was used to calculate the concentration of phosphate in the enhanced phosphoric acid. The values obtained for  $\text{PO}_4^{3-}$  were converted to Total phosphorus by multiplying with 0.3261 and the values were converted to  $\text{P}_2\text{O}_5$  by multiplying by 2.2915.

### 2.3 Synthesis of Diammonium Phosphate

An average amount (156.09 g) of lithium nitride was ground in a mortar and placed in a flat bottomed flask and a separatory funnel attached to the flask. 150 ml of deionized water was added to the flat-bottomed flask through the separatory funnel to initiate hydrolysis. The ammonia gas produced passed through a delivery tube to a beaker containing 200 ml of 4.58 M  $\text{H}_3\text{PO}_4$  extracted from bones. This experiment was repeated eight times to enable use of all the lithium nitride solid that had been prepared. The beaker containing 4.58 M phosphoric acid (200 ml) was immersed in basin containing ice because the reaction was highly exothermic. The highest temperature recorded during the reaction was  $95^\circ\text{C}$  but the crystals formed at  $51^\circ\text{C}$ . The crystals formed were filtered through Whatman No 1 filter paper, air dried at room temperature for seven days and weighed. A total of 3170.60g of the crystals formed were analysed for percentage composition of phosphorus spectrophotometrically.

### 2.4 Characterization of diammonium phosphate fertilizer

#### Determination of phosphorus in the fertilizer

The sample of synthesized fertilizer (5.0 g) was weighed out and dissolved in 50 ml of deionized water. The solution was then filtered through a Whatmann-41 filter paper and the filtrate transferred to a 100 ml of volumetric flask and diluted up to the mark with deionized water. A solution of sample (1.0 ml) was transferred to a 25 ml volumetric flask followed by 2 ml of 2.5 % ammonium molybdate and 0.5 ml of 1 M sulphuric acid solution. The mixture was shaken before adding 1.0 ml of 0.5 M hydrazine hydrate solution and the volume made up to mark with deionized water. The solution was allowed to stand for about 45 minutes for maximum colour development. Absorbance was taken at a wavelength of 830 nm. Similarly, the absorbance values for the fertilizer samples were measured and the corresponding concentrations of phosphorus were obtained using the equation from the calibration curve. The procedure was replicated for the commercial fertilizer. From the experimentally determined phosphorus concentration, the

percentage  $P_2O_5$  in the fertilizer was calculated from the equation:

$$\%P_2O_5 = \text{Conc of P (g/l)} \times \frac{100l}{g \text{ of fertilizer}} \times 10^{-6} g/mg \times$$

$$M_r \text{ of } \frac{P_2O_5}{2g} \text{ at. wt. P}$$

Where g. of fert = weight of fertilizer measured

$M_r$  of  $P_2O_5$  = Molecular weight of phosphorus pentoxide

g. at. wt. of P = gram atomic weight of phosphorus

Three replicate determinations were obtained for each different fertilizer. The value for the percentage  $P_2O_5$  were then compared to the commercial fertilizer.

#### Determination of Nitrogen in the fertilizer

The nitrogen in the fertilizer samples was determined by the Kjeldahl method. A mass of 1.0 g of the diammonium phosphate fertilizer prepared was weighed and transferred to a two-necked digestion flask and set up for distillation. A receiver flask containing 25.0 ml of 4% boric acid solution was attached to the distillation unit. About 50.0 cm<sup>3</sup> of NaOH (40%) solution added to the fertilizer in the flask to initiate evolution of ammonia which was absorbed into the boric acid in the receiver flask. Distillation took 20 minutes after which the receiver flask was removed; 5 drops of screened methyl red indicator solution were added to the distillate receiver flask and titrated against 0.05 M sulphuric acid to a grey end point. The procedure was repeated three times and results averaged. Two blank titrations were run and the average blank value used for subsequent calculations. The content of nitrogen, ( $W_N$ ), in milligrams per gram, was calculated using the formula:

$$\frac{(V_1 - V_0) \times [H^+] \times M_N \times 100}{(m \times m_t)} = W_N \text{ Where,}$$

$V_1$  = the volume, in ml of the sulphuric acid used in the titration of the sample

$V_0$  = the volume, in ml of the sulphuric acid used in the titration of the blank test

$[H^+]$  = the concentration of  $H^+$  in the sulphuric acid in moles per litre (e.g. if 0.01 mol/l sulphuric acid is used,  $[H^+] = 0.01$  mol/l)

$M_N$  = the molar mass of nitrogen, in grams per mole (=14)

$m$  = the mass of test sample

$m_t$  = the dry residue, expressed as g / 100g on the basis of oven dried material according to the standard of the special material.

#### Determination of Moisture in the fertilizer

The moisture content of the fertilizer prepared was determined gravimetrically (Masayoshi Koshino, 1998). About 50 g of the sample was transferred to a weighing dish and first dried in a desiccator for twenty four hours to a constant mass before subjecting it to the oven temperature. It is believed that the sample would "crawl" if dried rapidly hence cause inaccurate results (Masayoshi Koshino, 1998). The sample was then placed in an oven set at 110 °C for five hours and dried to a constant mass. It was then transferred to a desiccator for cooling. Five replications were done using the same procedure. The % moisture content was calculated using the equation

$$\frac{(\text{Mass of initial sample} - \text{Mass of dried sample})}{\text{Mass of initial sample}} \times 100$$

### 3. Results

#### 3.1 Extraction of bone phosphate

The evaluation of the effect of change of concentration of phosphoric acid on bone dissolution was as shown in Table 3.1

**Table 3.1:** Change of acid concentration on bone dissolution

Molarity of phosphoric acid (g/l)	Bones: Volume of acid ratio (g: ml)	Acid concentration after dissolution (g/l)	Mass of unreacted bones (g)	% P in the filtrate	Concentration of $H_2SO_4$ used (g/l)
0.1	10:35	0.135	4.3	19.41	6
0.2	10:35	0.241	4.0	19.39	6
0.25	10:35	0.290	4.0	19.78	6
0.27	10:35	0.355	0.0	19.84	6
0.3	10:35	0.356	0.0	19.83	6

Dissolution of bones was not complete when concentrations of 0.1, 0.2, 0.25 M  $H_3PO_4$  was used. Complete dissolution was observed when 0.275 M and 0.3 M were used. This indicated that the extent of dissolution was directly proportional to the concentration of phosphoric acid used. The mass of dissolved bones in the solvent was expressed as the difference between the initial mass of bones weighed before the experiment and the mass of unreacted bones. The calculation done for the dissolved bones gave the following results % P (19.45), %  $P_2O_5$  (44.58) and concentration of phosphoric acid was raised from 0.275 M to 4.58 M signifying significant phosphate enrichment.

#### 3.2 Synthesis and characterization of diammonium phosphate fertilizer

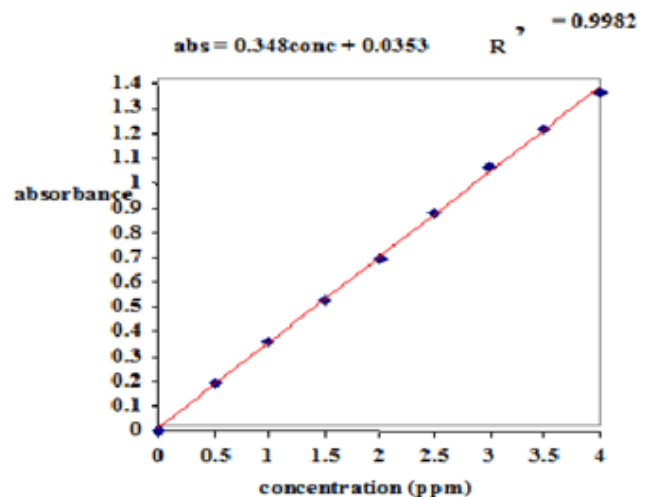
The yield of the dissolved phosphorus in g/l was determined as a mass percentage of the phosphorus content of the

crushed bone which was equivalent to 28.11 %  $P_2O_5$ . Reaction of phosphoric acid with ammonia in the ratio of 1.4:1 was a very exothermic reaction with temperatures recorded reaching 95 °C. The reaction was exothermic with the highest temperature recorded during the reaction as 95 °C but the crystals formed at 51°C. After the reaction, the temperatures of the reaction vessel were allowed to cool to 26 °C. The crystals were filtered from the mother liquor and dried in air giving a mass of 3170.60 g of diammonium phosphate, equivalent to 48.06 % yield. The calibration curve for the standard solutions was calibrated and the equation used to calculate the concentrations of  $PO_4^{3-}$  in the fertilizer prepared and commercially obtained fertilizer. The values for  $PO_4^{3-}$  obtained were converted to Total Phosphorus by multiplying with 0.3261 whereas, Total Phosphorus was converted to  $P_2O_5$  by multiplying the values obtained by 2.2915. Table 3.2 shows the average absorbance

and concentrations of the standard solutions while Figure 3.1 shows the calibration curve.

**Table 3.2:** Average absorbance and concentrations of standard solution

Volume (ml) of standards	Average absorbance	Concentration of PO <sub>4</sub> <sup>3-</sup> (ppm)
1	0.2295	0.5580
2	0.3904	1.0204
3	0.5605	1.5092
4	0.7313	2.0000
5	0.9185	2.5379
6	1.0990	3.0566
7	1.2550	3.5049
8	1.3970	3.9129



**Figure 3.1:** Calibration curve for phosphorus standards

Concentration of phosphate levels in the fertilizer was determined using the equation of the calibration curve and results recorded in Table 3.3.

**Table 3.3:** Levels of % P<sub>2</sub>O<sub>5</sub> in Synthesized DAP and commercial DAP

Synthesized DAP	Absorbance	Concentration (ppm)	% P <sub>2</sub> O <sub>5</sub>	Commercial DAP	Absorbance	Concentration (ppm)	% P <sub>2</sub> O <sub>5</sub>
Sample 1	0.2428	0.5964	44.56	Sample 1	0.2467	0.6082	45.45
Sample 2	0.2433	0.5976	44.66	Sample 2	0.2471	0.6086	45.48
Sample 3	0.2426	0.5957	44.52	Sample 3	0.2470	0.6083	45.46
Average	-	-	44.58	Average	-	-	45.46
Rsd (%)	-	-	0.23	Rsd (%)	-	-	0.23

Synthesized DAP contained 44.58 % P<sub>2</sub>O<sub>5</sub> as compared to 45.46 % P<sub>2</sub>O<sub>5</sub> for commercial DAP. The difference could be accounted for by the fact that bones contain 15 – 19 % P and rock phosphate contains 15 ± 1 % P (Coutand *et al.*, 2008).

The % N levels in the prepared DAP and commercial DAP were analysed using Kjeldahl process and results recorded in Table 3.4

**Table 3.4:** Levels of % nitrogen in Synthesized and commercial DAP

Laboratory fertilizer	M <sub>sample</sub> (g)	V <sub>sample</sub> (ml)	%N	Commercial fertilizer	M <sub>sample</sub> (g)	V <sub>sample</sub> (ml)	%N
Sample 1	0.6034	14.73	17.07	Sample 1	0.6680	16.55	17.35
Sample 2	0.5787	14.22	17.21	Sample 2	0.6979	17.14	17.20
Sample 3	0.6045	14.94	17.31	Sample 3	0.6758	16.78	17.39
Average	-	-	17.19	Average	-	-	17.31
Rsd (%)	-	-	0.77	Rsd (%)	-	-	0.77

Synthesized DAP contained 17.19 % N as compared to commercial DAP (17.39 % N). This could probably be as a result of the effect of poor storage facilities for the synthesized DAP where some levels of ammonia were lost. The values for the moisture content for the fertilizers determined was recorded in Table 3.5.

**Table 3.5:** % Moisture content in synthesized and commercial DAP

	Mean Weight of Wet Fertilizer (g)	Mean Weight of Dry Fertilizer (g)	% Moisture content
Synthesized DAP	50	23.5	0.53 ± 0.14
Commercial DAP	50	30.5	0.39± 0.06

The synthesized fertilizer contained a higher percentage of moisture (0.53 ± 0.14) as compared to the commercial fertilizer (0.39 ± 0.06). This could be attributed to the solubility levels of the fertilizer. It is assumed that

commercial fertilizer contained low moisture content due to the treatment to which it was subjected to in the process of manufacture.

The yield of the dissolved phosphorus in g/l was determined as a mass percentage of the phosphorus content of the crushed bone which was equivalent to 28.11 % P<sub>2</sub>O<sub>5</sub>. An acid concentration of 0.27M at a liquid/solid ratio of 10:1 was found to be the most suitable in preparation of bone phosphate enriched phosphoric acid. The mass of phosphorus was found to be 19.45 g which on conversion gave 44.58 % P<sub>2</sub>O<sub>5</sub>. This calculations show that 99.84 % of phosphorus in the bones was recovered during dissolution.

#### 4. Conclusion

This study reveals that animal bones which are generally disposed of as waste by abattoirs in large cities and towns and take many years to decompose are a rich source of



phosphate that can be harvested and used in fertilizer production.

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