# Evaluation on the application of lime, cow manure with Rhizobium inoculum on arsenic accumulation and yield of white beans

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### Abstract

Arsenic (As) pollution of Soil and irrigation water is a serious problem when it comes to ameliorating crop yield and As accumulation. This resaerch was therefore carried at the field experiment in Phuoc Hung, An Phu suburban to find out impacts of lime, cow manure and Rhizobium on crop yield and As uptake of the white bean. The research was disposed in the local farm with four repeats. Four treatments were applied, which included doses of NPK-control (40kgN-60kgP-60kgK/ha), NPK + lime (CaCO3 at the ratio of 2t/ha)+ Rhizobium inoculant (108CFU/g), NPK + cow manure ( the ratio of 10 t/ha) + Rhizobium inoculant (108CFU/g) and co-application of a NPK, lime, cow manure and Rhizobium inoculant to soil was adequate influences on the white bean crop. The increase in fresh seed productivity over control treatment valued from 17.4 to 33.8%. The As uptake of stems significantly decreased from 20.3 to 63.6% compared to control. Especially, the As accumulation of white bean in seeds was not at all treatments. The alone or combined supplementation of lime, manure with Rhizobium inoculant should be practiced to increase crop yield and improve crop quality in As contaminated soil and irrigation water in Vietnam.

Keywords— arsenic, cow manure, lime, white bean

# Introduction

The white bean, which is a important export cereal in Asian country has universally used around the world. it plants in Asian countries such as China, Indonesia, Vietnam,.. and is an important nutrient source for humans [1]. White beans (*Phaseolus vulgaris* L.) are high nutrients of antioxidants, protein, lipide, minerals [2]. However, the production of white beans has significantly reduced in recent years. The Climate change such as salinity, drought, heat and compacted along with As pollution from agricultural soil and irrigation water has lessened the production and production qualities of white beans [3], [4]. 10.4 billion of global population by the next years, 81% of this development counted for Asia and Africa the world food need country [5] and predictability for this future orientation. effective methods to raise the crop yield in order to be performed immediately [6]. The effective use of local manure resources and firmly agronomical application for field manure, irrigation water and protection, an adequate decrease in the need for inorganic fertilizers, fresh and unpolluted water and pesticides in agricultural cultivation may be reached without ruinous yield and quality [7].

The white bean is a seed legume and raises the soil nitrogen thank to fix a biological nitrogen resource, through the symbiotic with Rhizobia [8] therefore decreasing the demand to fertilize a lot of inorganic nitrogen. The biological nitrogen fixation ability of this legume field is subsidiary to genotypes, Rhizobia, soil nutrient conditions, and the weight of amended nitrogen fertilizers [9,10]. Legumes are also decreased the As uptake of stems, seeds and increased the high yield by the lime amendment [11]. Application of lime combined organic manures (e.g., cow, earthworm, bat, chicken,...) could also improve the soil nutrient and raise crop yield. In generally, the use of lime, organic manures and Rhizobium inocolum which is significantly raised the legumes yield and reduced the As uptake on agricultural soils and irrigated waters which are highly polluted with the As toxicology [12]. One of the reasons for the decrease of soil nutritive is due to continuous cultivation and no use of organic manures, especially in the small field areas [13].

Local farmers also have not applied Rhizobium inoculants to their legumes seed before planting [14]. According to prior studies showed that Agricultural soil and irrigation water were polluted by the high As content. The As content of legume bodies and grains exceed the WHO standard. Crops may reduce As uptake when increasing the soil pH [15]. Therefore, the supplementation of lime, animal manure with inorganic fertilizers that lessened the As absorption of legume crops compared to no liming amendment [16]. The study goal attained to improve the quality and yield of white bean by lime, cow manure and Rhizobia on As pollution soil and irrigation water.

#### 2. I. Rhizobium strains

The white bean nodule of Rhizobia. whichwere isolated from five strains at the biology laboratory of An Giang university were used in this research.

# 2.2. Field Experiments

Field experiments were realized at An Phu suburban from the growing crop of 2022. Soil cproperties from the experimental location was analysed according to [17]. The analyse for the soil characteristics is presented in Table 1. White bean grains that were planted in the ratios of 800 kg grains per ha and were planted in field experiment. Each treament (0.5 m  $\times$  20 m) included four plots. One plot had  $10 \text{ m}^2$ and included 04 replications, distance 0.5 m between plots. Fungal pathogens, weeds and insects were controlled by the application of pesticides with suitable rates according to manufacturer's guide. The As content of irrigation water sample (360 µg/L) was highly WHO exceeded standard (10  $\mu g/L$ ). furthermore, the As concentration of the soil sample inside the dike, which contained 84.1 mg/kg exceeded about eight times compared to Vietnamese standard of agricultural soils (12,0 mg/kg) (Table 1). All white bean seeds that were taken from CANIFOOD company, Vietnam were used during the crop with the good growth high production. The field technology was studied from local farmers and harvests the september of 2021. Soil samples that were collected in the depth (0-20 cm) for determing soil characteristics was taken fifteen days before taking the experiment (Table 1). Plant samples were collected to determine the As concentration at the harvest.

	Table I	1. Soil propertie	s before the exp	erimental design	
Experimenta	al Soil	Value	Soil analysis		Value
	Sand (%)	60.5		Total N (%)	0.112
	Clay (%)	4.50		Available P (mg/kg)	2.53
Mechanics	Silt (%)	35.0	Soil nutrients	Exchangeble K	80.0
				(mg/kg)	
	Texture	Sandy loam		$CaCO_3(\%)$	2.60
Chemical	$pH_{soil}$	5.3	As contents	irrigation water (µg/L)	0.36

# 2. Materials and methods

**T** 11

pH deep well 4.9	experimental	Soil 84.1
water	(mg/kg)	

Тибле 2. Ар	pucation of a		nure with Knizobium ino	cuum
Treatment	Rhizobium	CaCO <sub>3</sub>	Cow manure (t/ha)	NPK
Treatment	(10 <sup>8</sup> CFU/g)	(ton/ha)		(kg/ha)
NPK (control)	No	0.00	0.00	
NPK + Lime	inoculant	2.00	0.00	
NPK + cow manure	inoculant	0.00	10.0	40-60-60
NPK + Lime+ cow	inoculant	2.00	10.0	
manure				

Table 2. Application of lime, cow manure with Rhizobium inoculum



Fig.1 The fieldFig. 2 The purebredexperimentRhizobium

NH<sub>4</sub>NO<sub>3</sub>, DAP, KCl, CaCO<sub>3</sub> and *Rhizobia* were used in Table 2. The whole area of experiment was 160 m<sup>2</sup> (0.5 m x 20 m x 04 repeats x all treaments). The soil properties such as pH, total N, total and available P, exchangeable K and texture were analysed by Carterand and Gregorich [18] and were analysed by Atomic Absorption Spectrophotometric for Total As level of all soil, water and plant samples. The growth and Yield of white bean was determined during the growth time of plants such as biomass, height and branch number, No. of pods per plant, weight of fresh nodule, fresh weight of fill and empty pods each plant (g). The fresh production was determined by t/ha for fresh seeds. The soil samples before the

experimental design were the sandy loam, low pH of soil and irrigation water (5.3 and 4.9, respectively), total nitrogen (0.112%), the Available phosphorus was quite rich (2.53 mg/kg).The exchangeble K and CaCO<sub>3</sub> in soil had the low contents (80.0 mg/kg and 2.60%, respectively). Rhizobium and white bean significantly needed Phosphorus, exchangeble K and CaCO<sub>3</sub> for the growth of white bean and Rhizobium life.

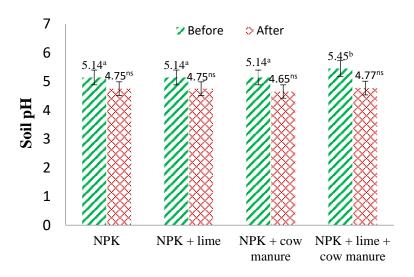
# 2.3 Statistical determination

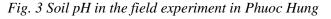
The experimental data was determined to analysis of variance by Statgraphics-Centurionxv to appreciate the sufficient differences among treatments at 5% level.

# Results and discussion. Soil pH

The results of Figure 3 presented that soil pH ranged from 5.14 to 5.45 at first time of the experiment and adequate diferrences among at 5%. However, the soil pH values, which ranged from 4.65 to 5.77 and inadequate diferrences among treatments at harvest reduced significantly soil pH compared to soil pH of experimentally first treatments. This result can explain that when As toxicology entered the agricultural soil

which may release large amounts of  $H^+$  ions and reduce soil pH [19].





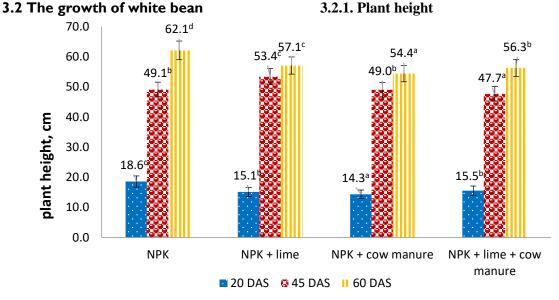


Fig. 4 The height of white beans during the growth

Effects of lime, cow manure, *Rhizobia* and water irrigation on the height of white beans (Fig. 4) showed that there was significant difference at LSD <0.05 among studied treatments. The highest height (62.1 cm) of control treatment (NPK) and lowest highest (54.4 cm) of NPK + cow manure treatment at harvest. The height of white bean was affected

by the soil nutrient and environment. According to Aipa and Michael, [20] was shown that useful effects of cow manure, lime, *Rhizobium* increased sufficiently on yield components of white beans such as height and effective branches.

### 3.2.2. Plant branches of white beans

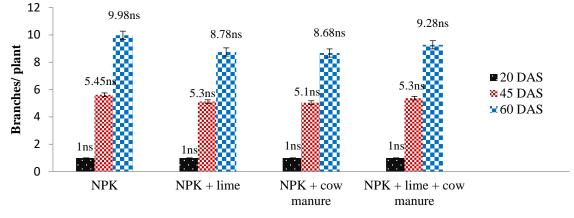


Fig. 5 The branches of white beans during the growth

Results in Figure 5 presented that white bean branches of all treatments were insufficiently differences druring the growth at  $LSD \le 0.05$ . However, branches of white beans regularly increased from 1 to 9.98 branches during the experiment at the whole treatments (Fig. 5). This study results were in agree with prior study results of the same plant and organic manure [21]. Other studies found that cow manures raised the development, seed yield but not change branches in white bean followed by NPK fertilizer compared to the control [22].

*Table 3. Effect of cow manure, lime, rhizobium on the production components and yield production of white beans* 

Parameters (each plant)			
Treatment	No. of pods	seed of pods (g)	Wt. of 1,000 seeds (g)
NPK (control)	27.8ª	10.5ª	188ª
NPK + Lime	36.3 <sup>bc</sup>	9.50ª	200 <sup>b</sup>
NPK + cow manure	33.5 <sup>b</sup>	11.0 <sup>a</sup>	194 <sup>ab</sup>
NPK + Lime+ cow manure	39.5°	14.3 <sup>b</sup>	199 <sup>b</sup>
Ftest	**	**	*
CV(%)	15.2	20.6	3.50
*,**: significant differenc	the at $P$ value $\leq 0$	0.05 and 0.01%	

Influences of cow manure, lime, Rhizobium on the pod number and pod weight of wihte beans were similar to that of the of 1,000 seeds weight of white bean. A mixture of lime, inorganic, organic fertilizer and rhizobium inculant influenced the sufficient result on the plant height, No.of shoots, No. of pods, seed of pods and weight of 1,000 seeds (Fig. 4, 5 and Table 3). Number of pods, seed of pods and weight of 1,000 seeds had a significant difference among fertilization treatments. The highest values of number of pods (39.5), seed of pods (14.3g) and weight of 1,000 seeds (199g) obtained at the treatmen of NPK + Lime+ cow manure compared to other treatments (Table 3). Treatment (NPK + Lime+ cow manure) using 2.0 tons  $CaCO_3/ha + 10$  tons cow manure /ha with Rhizobium inoculum showed the highest yield (5.59 t/ha). Meanwhile, the lowest yield (3.70 t/ha) of white bean was found in control treatment (NPK) using 40 kg N+60 kg P+ 60 kg K /ha. The following high values of white bean yield were obtained by using 40 kg N +60 kg P + 60 kg K /ha +10.0 tons cow manure/ha (4.48 t/ha) and 40 kg N +60 kg P+ 60 kg K /ha +2.0 tons CaCO<sub>3</sub>/ha (5.13 t/ha), respectively (Table 4). The production of white bean that was calculated by the growth of the crop from the sapling to the end of experiment where the soil nutrient was an important role. Co-application of lime, cow manure and inorganic fertilizers could affect on production components and yield production of white beans such as No. of pods and grains per plant, weight of 1,000 seeds (**Table 3 & 4**).

The animal manures, which content a large number of microorganism could raise the positive impacts of soil fertility. Inoculation of *Rhizobia* a key important role on plant growth impulse through nitrogen fixation in legumes [21].Other characteristics were shown in the current research, such as No. of pods and grains per the crop also related to the soybean

yield. The prior research, higher results in weight of 1,000 seeds of soybeans presented that the larger seed size that could raise to higher yield [22]. This research also indicated that the number of pods, seeds and weight of 1,000 seeds per plant significantly affected on the bean yield and this was suitable to the prior research presenting that the production yield was significantly related to the yield component and increase the growth and yield of soybean [23].

Parameters per plant	Yield (t/ha)	As conte	As contentrations (mg/kg)			
Treatment		stems	seeds	experimen	nt Soils	
		After ex	After experiment		After	
NPK (control)	3.70 <sup>a</sup>	1.18 <sup>c</sup>	negative	86.0°	96.7 <sup>b</sup>	
NPK + Lime	5.13 <sup>c</sup>	0.940 <sup>b</sup>	negative	85.6ª	113°	
NPK + cow manure	4.48 <sup>b</sup>	0.430 <sup>a</sup>	negative	86.1°	96.6 <sup>b</sup>	
NPK + Lime+ cow	5.59 <sup>d</sup>	0.940 <sup>b</sup>	negative	85.8 <sup>b</sup>	89.1 <sup>a</sup>	
manure						
Ftest	**	**	-	**	**	
CV(%)	16.2	22.4	-	2.6	9.19	
**: adequately different	at 0.01%	•	1	•	•	

Table 4. Effect of cow manure, lime, rhizobium on the yield and As accumulation of white beans

The As uptake of white bean in stems valued from 0.43 to 1.18 mg/kg and significant differences at 1%. The maximum As ccontent of white bean stems (1.18 mg/kg) in the control treatment and lowest value (0.43 mg/kg) in using 40 kg N +60 kg P + 60 kg K /ha +10.0 tons cow manure/ha. On contrary, there had not absorbed the As toxicology of white bean from the agricultural soil to bean seeds at all treaments (Table 5). However, the As contents before and after experiment had of soils significant differences at 1% (LSD < 0.01). Arsenic contents of agricultural soils before and after experiment ranged from 85.6 to 86.1 mg/kg and 89.1 to 113 mg/kg, respectively. In generally, the minimum and maximum As accumulation of soils was 89.1 mg/kg in using NPK, cow manure, lime and Rhizobium and 113 mg/kg in NPK and lime treatment, respectively (Table 4). This above result in Table 4 could show that co-application of lime, cow manures could decrease the As accumulation and increase production yield of white bean. The negtive relationship between the pH and As uptake of plant was adequately raised pH of soil by lime and organic manures [15]. According to prior resarch of Tuan & Chuong, (2022) [12] showed that amendment of lime, earthworm manure combined with *Rhizobium* inoculant reduced the As store and raised the peanut.production.

# 4. Conclusion

Application of lime, cow manure combined with *Rhizobium* inoculant significantly obtained the fresh yield compared to the control treatment. This current research presented the positive impacts of liming, animal manure and chemical fertilizer on the yield increase of white bean yield. A amendment of lime, cow manure combination of NPK fertilizer was higher yield than the yield in comparison to the application of NPK only. Application of using NPK (40:60: 60 kg /ha) combined with 2.0 tons  $CaCO_3/ha + 10$  tons cow manure /ha and *Rhizobium* inoculum showed an increase in 33.8% fresh seed yield to control treatment. Futhermore, the single or mixable amendment of lime and Cow manure reduced As accumulation of white bean in stems from 20.3 to 63.6%. These research results should encourage that co-application of lime, cow manure, *Rhizobium* inoculum and chemical fertilizers has the positive to increase the high seed yield and decrease As uptake of white bean mitigating the environmental effects caused by the As pollution of agricultural soil and irrigation water.

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