

Full Length Research Paper

## Uptake kinetics of arsenic by lettuce cultivars under hydroponics

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Accepted 10 May, 2013

**Arsenic (As) uptake ability based on kinetic parameters by two lettuce cultivars; Sijibaiye (SJB Y) and Texuanyanlingsun (TX YLS) was investigated in nutrient solution containing eight levels of arsenic (As). Depletion of As from solution was monitored over a period of 24 h at regular time to estimate As uptake kinetics of the plants.  $K_m$  and  $V_{max}$  values were evaluated with the aid of the Lineweaver - Burk plot (that is the double reciprocal plot of initial velocity ( $1/v$ ) versus substrate concentration ( $1/[s]$ )). Both cultivars were capable to assimilate As(V), although with different kinetic parameters. The values obtained for  $K_m$  and  $V_{max}$  varied with plant cultivars. Using Lineweaver- Burk, values of  $K_m$  and  $V_{max}$  were found to be much lower in SJB Y ( $26.721 \text{ mg L}^{-1}$  and  $0.7737 \text{ mgAsg}^{-1} \text{ shoot d.wt.hr}^{-1}$ ) than that of TX YLS ( $81.922 \text{ mg L}^{-1}$  and  $1.8488 \text{ mgAsg}^{-1} \text{ shoot d.wt.hr}^{-1}$ ), indicating SJB Y had the lowest uptake ability than TX YLS plants. Furthermore, TX YLS had the highest  $K_m$  value than SJB Y, confirming that uptake in the plant cell plasma membrane of TX YLS had a higher affinity for As than SJB Y plants. The time (24 h) affects the depletion of As in both cultivars. It can be concluded that among the cultivars, SJB Y could be finally identified as the most suitable cultivar to be grown in polluted farmland. Therefore, it has been suggested that there is possibility to lower the As concentration in lettuce by selecting and breeding cultivars with less As concentration that can be safely grown in contaminated soils with slight levels of As for safe consumption.**

**Key words:** Arsenic (V), cultivars variation, kinetic parameters, Lineweaver Burk, uptake rate.

### INTRODUCTION

Arsenic (As) is one of the most dangerous toxic farmland that can be found. Besides various adverse effects of As on humans and animals, arsenic could also hamper normal growth of plants (Cozzolino et al., 2010).

Lettuce plant (*Lactuca sativa L.*) is an important leafy vegetable in the world, especially for Chinese people, due to the large consumption. It is considered as an excellent nutritive source of minerals and vitamins as is consumed as fresh green salad (Mulabagal et al., 2010). Crop with such promising potentialities for local markets, would necessitate much research in order to improve agricultural products' safety and to reduce arsenic threat

on human health.

In recent years, many researchers have paid much attention to the As uptake kinetic excised plant roots by different plant species, such as hyperaccumulation, rice (Mohamed and Andrew, 2008), *Brassica carinata* (Irtelli et al., 2008), *Pteris vittata* (Weifeng et al., 2011), maize (Abbas et al., 2008), adopting hydroponic and field experiments with intact roots, while there is lack of data on the kinetics of arsenic uptake by leafy vegetable. Plant species and even genotypes differ greatly in their ability to take up, transport and accumulate As within the plants. The genotypic variation in the different cultivars for

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**Table 1.** Name, abbreviation, origin, and seed provider of 32 leafy vegetable cultivars used for hydroponic experiments.

Cultivar name	Abbreviation	Origin	Seed provider
Texuanyanling sun	TXYLS	China	BGSRDC
Sijibaiye	SJBY	China	BGSRDC
Huayesun jing yong woju	HYSJYWJ	China	BGSRDC
Yeyong woju	YYWJ	China	BGSRDC
Guayuanyiyesun	GYYS	China	BGSRDC

IVF, CAAS: The Institute of vegetable and flowers, Chinese Academy of Agricultural Sciences; BGSRDC: Beijing Gardening Seeds of Research and Development Center; BGLATI: Beijing Golden Land Agricultural Technology Institute; BSVSPC: Beijing Special Vegetable Seed and Plant Co.; BSBC : Beijing seed and breed center.

arsenic concentration, as a strategy to reduce the movement of As from contaminated soils to edible parts of crop plants has been receiving close attention (Norton et al., 2012). Moreover, the fundamental requirement for breeding low As-accumulation cultivars is to know the genotypic differences and the mechanisms governing As accumulation in crops. It is therefore imperative to understand the variation in As accumulation of different lettuce cultivars and to evaluate the mechanisms involved in arsenic uptake kinetic and translocation in order to screen and find out excluder-cultivars which transport less As concentration to the shoot suitable for growing in the As-contaminated farmlands. Once identified, cultivars with high and less As accumulation will be tested for As uptake kinetic to confirm the result. The objective of this study was: (1) to screen and identify the cultivars with less As concentrations that can be grown in As contaminated farmland for food safety. (2) to compare the rates of As uptake ability between cultivars and to determine the kinetic parameters. (3) Because of the general lack of data on the uptake kinetics of arsenic in leafy vegetable cultivars, the main objective of this study was also to provide theoretical support and quantitative data that could offer an understanding of the differences in  $V_{max}$  and  $K_m$  among leafy vegetable.

## MATERIALS AND METHODS

### Plant material and preparation of seedlings

Seeds of five lettuce cultivars (Table 1) were purchased from Chinese Academy of Agricultural Sciences and cultivated at Institute of Environment and Sustainable Development in Agriculture CAAS, Beijing/China. In the experiment (I), seeds were surface sterilized with 30%  $H_2O_2$  (w/w) solution for 15 min, rinsed five times with distilled water. The seeds were germinated for 10 days in the plate containing autoclaved vermiculite. During the seed germination period, half-strength modified Hoagland's nutrient solution was supplied once a week.

### Hydroponic screening

On the 7<sup>th</sup> day, at third leaf stage, the healthy and uniform sized

seedlings were carefully removed from vermiculite, and washed with distilled water and then transferred to hydroponic bottles each containing 1500 ml of full-strength of modified Hoagland and Arnon (Hoagland and Arnon, 1950) with pH adjusted to 6.3 by using NaOH or HCl for 10 days. Nutrient solution was renewed twice a week. On the 21<sup>st</sup> day, the As treatment consisted of the basic nutrient solution with addition of 6 mg As L<sup>-1</sup>, As(V) (applied as  $Na_3AsO_4 \cdot 12H_2O$ ) for 14 days. There were four replicates for each cultivar and each replicate comprises four plants. The As treated nutrient solution was renewed once a week. Plants were grown for 35 days, and then harvested. The shoots and roots of all harvested plants were separated carefully, washed with water, and then rinsed severally using deionized water to remove adhering particles. These samples of roots and shoots were gently packed in envelopes. Plant parts were dried at 90°C for 1 h, then at 70°C in the oven until constant weight was obtained. Both dry and fresh weights were recorded. Dried plant materials were ground and digested and arsenic concentrations were determined using the atomic fluorescence spectrometry following the procedure described by Smith et al. (2009).

### Kinetics of As(V) uptake

The experiment (II) was carried out under the same conditions as described above. Two lettuce cultivars which have both been shown to absorb the highest and lowest arsenic in their shoots (Exp I) were used as materials.

After 4 weeks of growth before the uptake kinetic experiment (II), the seedlings were transferred to the deionized water for 5 days, and then uniform seedlings from TXYLS and SJBY were selected, washed from root system and transferred to plastic bottles containing 1200 ml of As uptake solution. Each bottle was wrapped with opaque plastic membrane. Eight different concentrations of arsenic (0, 0.5, 1, 2, 4, 6, 8, and 10 mg As L<sup>-1</sup>) were used to study the uptake kinetics of arsenic. Each treatment was replicated three times. Each 1200-mL container had 4 plants. The pH of the nutrient solution was maintained at 6.3. At each time interval (0, 0.5, 1, 2, 4, 6, 8, 10, 12, 22, and 24 h), sample of 1 ml aliquot was withdrawn from each uptake solution to measure depletion in As, as a result of its uptake by plants, the depletion period was for 24 h, and 1.0 ml of deionized water was then added to each container at set time intervals (Wang et al., 2002). Water losses through transpiration were compensated by additions of deionized water at hourly intervals. After 24h of uptake experiment, the plants were harvested and quickly rinsed with distilled water. The plants were separated into roots and shoots, blotted dry with paper tissue and dried at 65°C for 72 h, and the dry weights were recorded. Arsenic concentrations were determined as previously described.

Substrate concentrations, with which the uptake rates were

**Table 2.** Mean As concentrations and As uptake of five cultivars (Hydroponic experiment, 2 weeks of As exposure).

Cultivar name	As concentration (mg kg <sup>-1</sup> dwt)		Dry weight (g pot <sup>-1</sup> )		As uptake (µg pot <sup>-1</sup> dwt)
	shoot	Root	Shoot	Root	Shoot
Texuanyanling sun	20.56a	255.91	2.70	0.21	55.51a
Sijibaiye	14.07c	442.52	2.14	0.10	30.25c
Huayesun jing yong woju	16.27b	478.31	2.77	0.11	45.07ab
Yeyong woju	17.67b	534.06	2.75	0.11	47.97ab
Guayuanysesun	16.22b	482.65	2.51	0.13	40.62bc

Values (mean n=4) with different letters in the same column are significantly different according to the duncan's test (P<0.05)

compared, was calculated as the arithmetic mean of the initial (t = 0) and final concentration for a time interval.

### Calculations of As uptake rates and kinetic parameters

The amount of uptake rate (V [mg As g<sup>-1</sup> shoot d.wt.hr<sup>-1</sup>]) was calculated from the depletion of As in the uptake solution. As uptake by the plant per unit time per gram dry weight was calculated according to the following formula: (Shimono and Bunce, 2009; Ajaelu et al., 2011)

$$V = \frac{(V_i \times C_i) - (V_f \times C_f)}{t \times Bdw} \quad (1)$$

Where, V<sub>i</sub>, C<sub>i</sub>, V<sub>f</sub>, C<sub>f</sub> and Bdw are the volume (V) and quantities in mg L<sup>-1</sup> of substrate in solution at the start and the end of uptake experiment, and t is the time in hours and Bdw is the biomass dry weight of the plants, respectively. The reduction in the volume was very small, so this was not taken into account in the calculation.

The Michaelis-Menten Equation is:

$$V_0 = \frac{V_{\max} [S]}{K_m + [S]} \quad (2)$$

Where, V<sub>0</sub> is the initial velocity of the reaction at substrate concentrations (S); [S]: is the substrate concentration; V<sub>max</sub>: is the maximum uptake rate (mgAsg<sup>-1</sup> shoot d.wt.hr<sup>-1</sup>) achieved at which the enzyme is saturated with substrate, and K<sub>m</sub>: is the Michaelis-Menten constant (Briggs and Haldane, 1925).

The parameters K<sub>m</sub> and V<sub>max</sub> characterize the ability of plant to absorb nutrients from their soil environment (Glass, 1989). The equation describes the relationship between the uptake rate of the nutrient and the nutrient concentration. Uptake rate reaches a constant or saturated rate (V<sub>max</sub>) at high ambient concentration. Lineweaver and Burk (1934) pointed out that equation (2) becomes linear in form upon taking the reciprocal of both sides of the equation to calculate the key parameters in the Michaelis-Menten equation:

$$V_0 = \frac{V_{\max} [S]}{K_m + [S]} \quad \longrightarrow \quad \left(\frac{1}{V}\right) = \left(\frac{K_m}{V_{\max}}\right) \cdot \left(\frac{1}{[S]}\right) + \left(\frac{1}{V_{\max}}\right)$$

That is (y= m x+ c), where (y=1/V and x= 1/[s])

In order to obtain the kinetic uptake parameters representing the maximum uptake rate (V<sub>max</sub>) and the half saturation constant (K<sub>m</sub>), by plotting 1/V against 1/[S] will give a straight line graph having a slope of K<sub>m</sub>/V<sub>max</sub> and a y- intercept on the ordinate at 1/V<sub>max</sub>. From

this plot, the K<sub>m</sub> and V<sub>max</sub> values of arsenic uptake were determined by linear regression analysis, thus giving a convenient method for obtaining both V<sub>max</sub> and K<sub>m</sub>.

### Statistical analysis

All data were subjected to analysis of variance (ANOVA) using SPSS 16.0. Differences at p<0.05 were considered significant. All curve fitting was completed using a computerized, linear regression on Lineweaver-Burk transformed data to estimate the V<sub>max</sub> and K<sub>m</sub>.

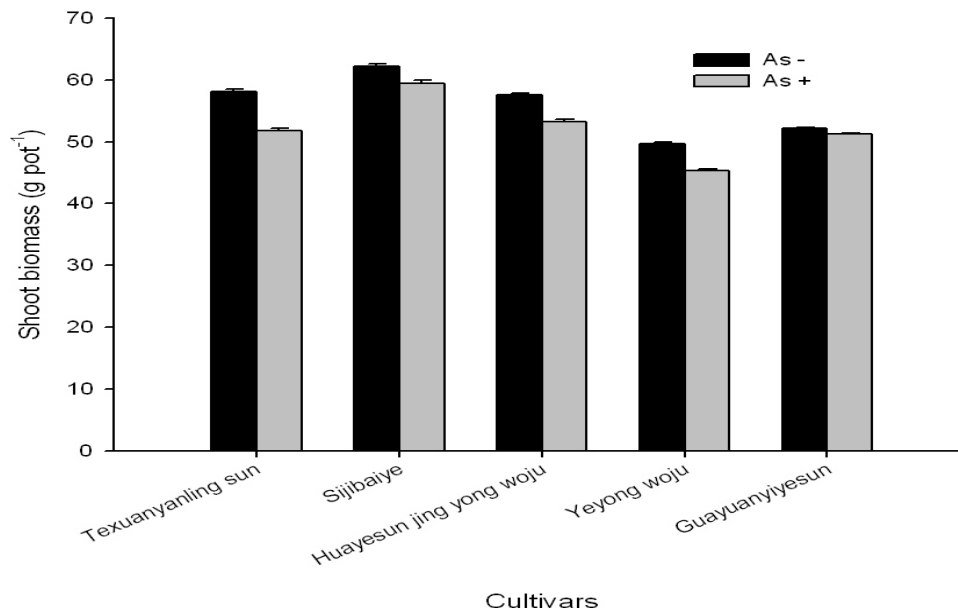
## RESULTS

### Genotypic variation in shoot As concentrations and accumulation

In this preliminary experiment (I) we designed to determine cultivars with less and high shoot As concentration. Significant genotypic variations were observed in the shoots and roots As concentration of 5 cultivars studied under As treatment (6 mg L<sup>-1</sup>). Of the 5 cultivars, As concentration ranged from 14.07 to 20.56 mg kg<sup>-1</sup> dry weight (1.5 variation). The main results were shown as follows: SJBV had the lowest shoot As concentration (14.065 mg kg<sup>-1</sup> dry weight). While the highest shoot As concentration were detected in TXYLS (20.56 mg kg<sup>-1</sup> dry weight) (Table 2). After taken up by roots, arsenic was accumulated and sequestered in the roots in much higher amounts than in the shoots (root>shoot), only about 4-6% of As was translocated to shoots of cultivars. The average As concentration in root was nineteen to twenty-six times higher than that in shoot (root>shoot) as an indicator for translocation ability from root to shoot. Therefore absolute majority of As accumulated by cultivar plants was restrained in root and only a very small portion was transferred into shoot.

### Arsenic accumulation

The data presented in Table 2 shows the amounts of arsenic (As) accumulated in the shoot of 32 cultivars. The values are expressed in µg<sup>-1</sup>dry weight each calculated from the formula: (dry weight × As concentration) and they show significant difference (p<0.05) ranging from 30.25 to 55.11 µg<sup>-1</sup> pot dry weight. Arsenic accumulated



**Figure 1.** Shoot biomass (FW) of 5cultivars exposed to 6mg L<sup>-1</sup> As for 14 days. Data are expressed as means±S.E. of four replicates.  $p < 0.05$ .

**Table 3.** Tolerance index, % (TI) based on shoot biomass of 5 lettuce cultivars exposed to 6mg/L As for 14 days.

Cultivar	Shoot biomass
Texuanyanling sun	89.03c
Sijibaiye	95.58a
Huayesun jing yong woju	92.45b
Yeyong woju	91.26bc
Guayuanliyeesun	98.17a

Values (mean n=4 with different letters in the same column are significantly different according to the Duncan's test ( $P < 0.05$ )).

to the greatest extent in TXYLS had the highest As accumulation while SJBV had less arsenic accumulation.

### Variation in shoot biomass trait under As stress

By growing 5 lettuce cultivars in the greenhouse, the responses of the selected cultivars differs. The shoot biomass (FW) of tested cultivars varied between 46.340-60.505, respectively under 6 mg L<sup>-1</sup> (Figure 1). On the other hand GYYYS and SJBV showed greatly higher shoot biomass (Figure1). The maximal reduction in shoot biomass was observed in TXYLS (11%), followed by YYWJ (9%), HYSJYWJ (8%), SJBV (4.5%), and GYYYS (1.8%) (Table 3). The tolerance index, in terms of the biomass ratio of As treatment to control, was high in all cultivars except TXYLS. By contrast, 80% of tested culti-

vars showed a tolerance index higher than 90%. Therefore, the effects of As on leafy vegetable growth and development appear to be cultivar dependent.

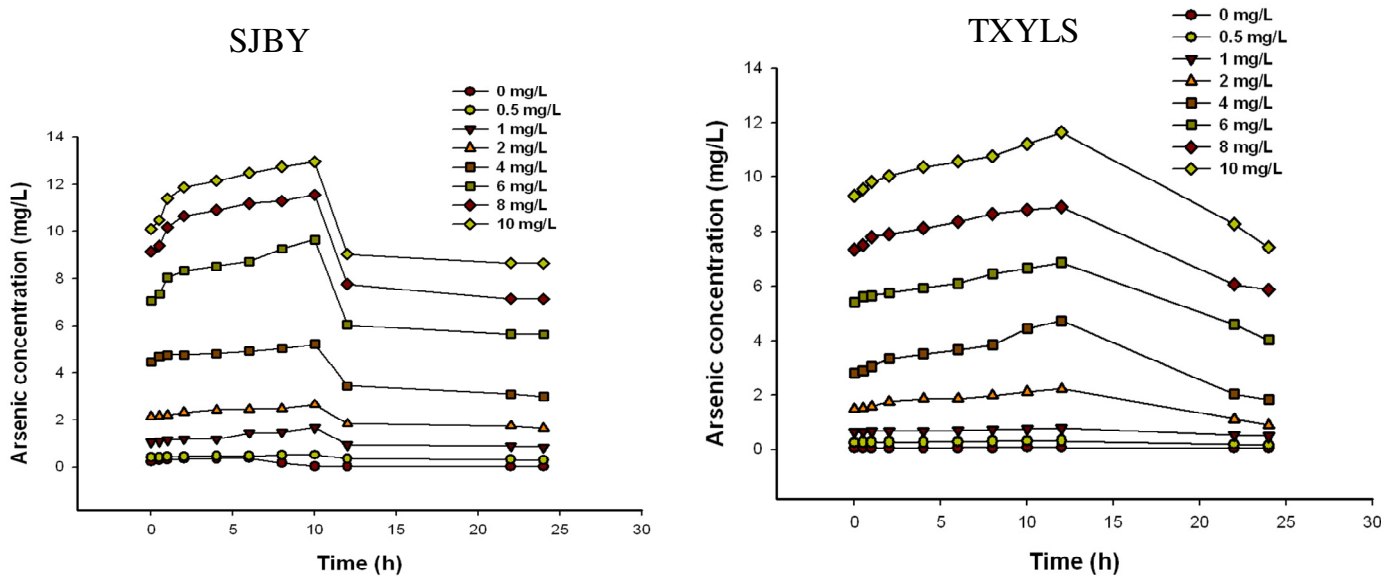
### Variation in root-to-shoot transport and nutrient solution-to-plant

#### Translocation factor (TF)

The TF can be used to evaluate the ability of the plant to translocate As from the root to shoot. The average TF of various cultivars differed among cultivars (Table 4). The root-shoot translocation factor ranged from 0.03 to 0.08. There were no significant ( $P > 0.05$ ) differences found in As TF of SJBV, HYSJYWJ, YYWJ and GYYYS. While TXYLS showed the highest TF similar to As accumulation and BF (Tables 2 and 4). The translocation factor (TF) was calculated by dividing the As concentration in the shoot by its concentration in the root.

#### Bioaccumulation factor (BCF)

The quotient of the As concentration in shoots to that in nutrient solution, can be used to evaluate the ability of plants to accumulate heavy metals. Table 4 shows the average arsenic bioaccumulation factor of 5 cultivars under As treatment (6mg L<sup>-1</sup>), ranging from 3.43 to 2.34 in the cultivars. The lowest BCF was found in SJBV, similar to the TF, and the highest BCF was detected in TXYLS. There were no significant differences between HYSJYWJ, YYWJ and GYYYS.



**Figure 2.** Changes of As concentration with time (h) in the cultivars with high and low-As Accumulation (TXYLS and SJB Y) grown in nutrient solutions with different As(V) concentrations for 24 h.

**Table 4.** Translocation and bioaccumulation factor of As in 5 lettuce cultivars exposed to  $6\text{mgL}^{-1}$  As.

Genotype	BCF	TF
Texuanyanling sun	3.43a	0.08a
Sijibaie	2.34c	0.03b
Huayesun jing yong woju	2.71b	0.04b
Yeyong woju	2.95b	0.04b
Guayuanyiyesun	2.71b	0.03b

Different letters in a column indicate significant differences between the cultivars at the 0.05 level according to the Duncan's test.

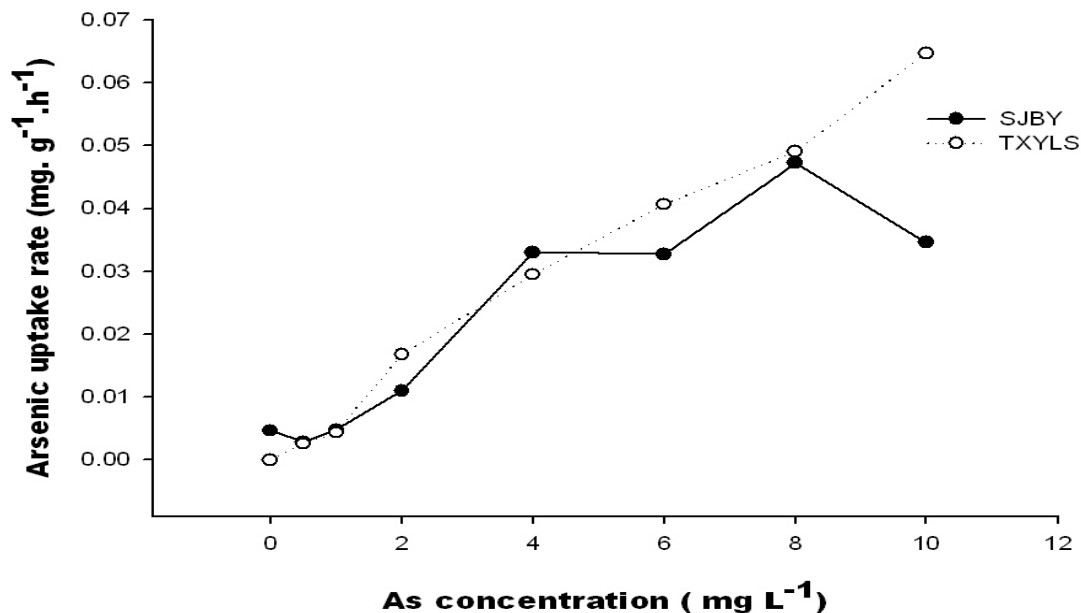
### Depletion of As concentration in nutrient solution

The uptake time is one of the most important factors affecting the uptake of heavy metals by plants. Generally, in the initial period of exposure metal is absorbed and accumulated at a high rate, and then the level stabilizes when equilibrium of metal uptake rates is attained. Short-term, time-dependent arsenic depletion from hydroponic solution was measured at a series of different time intervals (after 0 h, 0.5, 1, 2, 4, 6, 8, 10, 12, 22 and 24 h respectively) with various concentration of arsenic. Figure 2 show that the depletion of As in TXYLS gradually increased with increasing uptake time for at least 12 h and then starts decreasing rapidly with the passage of time until 24 h, while SJB Y showed the same trend for the first 10 h then starts decreasing until 24 h. After 24 h of uptake, the depletion of As became slower for all cultivars and showed a significant difference in the arsenic accumulation. It can be explained that arsenic was ab-

sorbed by cultivars. Both the cultivars and treatments influenced concentrations of As in nutrient solution. At the end of the depletion (24 h), TXYLS and SJB Y have up taken 23.09% and 28.40% of arsenic from nutrient solution (figure 2). Interaction between treatments and cultivars influenced depletion of As concentration.

### Short-term Arsenic uptake kinetics

Short-term (24h) uptake rate of the cultivars over a range of arsenic concentrations ( $0.5, 1, 2, 4, 6, 8$  and  $10\text{mg L}^{-1}$ ) was calculated as the ratio of the amount of absorption arsenic by cultivars in a unit time to the dry weight of cultivars and its unit is  $\text{mg} \cdot \text{g}^{-1} \cdot \text{h}^{-1}$ . The results showed that there was variation in uptake rate between two cultivars. Lower uptake rate was found at the rate of 0.5, 1 and 4  $\text{mg As L}^{-1}$  in SJB Y and at 2, 6, 8 and  $10\text{mg L}^{-1}$  in TXYLS (Figure 3). The data for kinetic parameters were derived from a linear regression on Lineweaver-Burk plot transformations (Table 5), indicated that uptake characteristics were different between cultivars. TXYLS exhibited about 2-fold higher  $V_{\text{max}}$  value ( $1.8488\text{mg As g}^{-1}\text{shoot d.wt.hr}^{-1}$ ) than that ( $0.7737\text{mg As g}^{-1}\text{shoot d.wt.hr}^{-1}$ ) in SJB Y, similar TXYLS possesses higher value of  $K_m$  ( $81.92\text{mg L}^{-1}$ ) as compared to that in SJB Y ( $26.721\text{mg L}^{-1}$ ). The uptake ability is jointly decided by the  $V_{\text{max}}$  and  $K_m$ ; hence the  $V_{\text{max}}/K_m$  represents the absorption ability of cultivars. The  $V_{\text{max}}/K_m$  values indicated that the arsenic uptake ability of cultivars followed TXYLS > SJB Y. The slope of the linear component of TXYLS was higher (44.312) than SJB Y (34.537) (Table 5). Therefore, we could conclude TXYLS's cell wall had a relatively strong arsenic binding capacity, while SJB Y had relatively lower.



**Figure 3.** As uptake rate ( $\text{mg.g}^{-1}.\text{h}^{-1}$ ) by plants of TXYLS and SJBY grown with 0, 0.5, 1, 2, 4, 6, 8 and 10  $\text{mg L}^{-1}$  As for 24 h. (values are means of 3 replicates).

**Table 5.** Comparison in the low-As-accumulating cultivar 'SJBY' and the high-As-accumulating cultivar 'TXYLS' in kinetic parameters ( $K_m$ ,  $V_{\max}$  and  $K_m/V_{\max}$ ) for concentration-dependent uptake of As into intact plants immediately after 24 h of As.

Cultivar	$V_{\max}$ ( $\text{mgAsg}^{-1}$ shoot d.wt.hr <sup>-1</sup> )	$K_m$ ( $\text{mg/L-1}$ )	$K_m/V_{\max}$	$R^2$
TXYLS	1.8488	81.9227	44.312	1
SJBY	0.7737	26.7211	34.537	0.9999

Data are presented as mean (n=3).

## DISCUSSION

In this study, the transport of As from nutrient solution to root and shoot of the cultivars were the important factors which lead to the cultivar differences (Tables 2 and 4). The plant is able to move the As from the root to the shoot, but the mechanism through which these leafy vegetable plants do this is unknown. The average As concentration in the roots was nineteen to twenty six times higher than that of shoot (root>shoot), this might be due to the inability of the plants to translocate the As beyond the roots (Smith et al., 2009). The result reveals that all the cultivars had TFs less than 1, (3 fold variation) in TF ability and (1.2 fold variation) in BCF, indicating that As was limitedly transported into the shoot. According to the study of Sinha et al. (2004), the plants act both as "accumulators" and "excluders". Therefore, these five cultivars can be considered as a As excluder, comprising avoidance of metal uptake and restriction of metal transport to the shoots (Sinha et al., 2004; Devos et al., 1991). This is consistent with the results reported by Huang et al. (2005) using lettuce and when compared with other

plants such as *Pteris vittata* (Francesconi et al., 2002), the As concentrations were high in the shoot (aboveground) than in root, also the TFs were greater than 1.0. Wei et al. (2005) found that the exclusion of metals from shoot has been regarded as a metal tolerant strategy. Metals, once taken up by roots, can either be stored in the root or exported to the shoot. Resistance of plants to heavy metals can be achieved by an avoidance mechanism resulting in low metal concentration in the shoots as shown in this study.

The mean TFs for five cultivars were 0.043. Ming et al. (2012) found that TFs of rice were 0.20, 0.25, and 0.24 and Williams et al. (2007) reported that TF of As to the shoots of rice was 0.05–3.8, with mean 0.76. Su et al. (2010) demonstrated that rice had a higher efficiency of As translocation from root to shoot than wheat and barley. Our result revealed that lettuce cultivars had a less efficiency of TF than rice. These data suggest that As was transported at a much slower rate to the shoot of cultivars. Liu et al. (2004) reported that the large cultivars variation in rice was caused by As uptake and shoot As transport, which could also lead to genetic differences in

As uptake. In the present study cultivars may affect remarkably TF of As. The results indicated that As concentration in shoot was somewhat related to As uptake and the transport of As to shoot.

Our objective was to characterize the uptake kinetic aspects of arsenic in two cultivars which have both been shown to absorb the highest and lowest arsenic in the experiment I (Table 2) in order to verify the cultivar with less As accumulation through kinetic experiment II. Uptake kinetics characteristics can be considered as one of the important criteria for selecting a cultivar to use in As-contaminated areas because it gives the information on how fast arsenic is taken up by plant. There are a number of studies investigating uptake kinetics of arsenate, both in higher and lower plants (Weifeng et al., 2011; Mohamed and Andrew, 2008; Geng et al., 2006). In general, arsenic uptake kinetics has not been described for leafy vegetable. Researchers using the depletion method have used time periods ranging from 15 min to several days. Mcfarlane and Yanai (2006) selected two-hour interval because in earlier experiments they found that two hours were sufficient to cause a measurable change in solution concentrations. Wang et al. (2002) found between 20 and 40% of arsenic was depleted over the 8 h period. In this study, the result showed that at the end of the depletion experiment (24 h), between 23.09 and 28.40% of arsenic from nutrient were depleted. Interaction between treatments and cultivars influenced depletion of As concentration. The value of  $V_{max}$  represents the plant's ability to absorb a nutrient. The higher the  $V_{max}$  value, the more could be absorbed. The value of  $K_m$  corresponds to the plant's affinity for a certain element, and the lower the  $K_m$  value, the greater the affinity (Chao et al., 2008).

In this study, there was a clear difference between cultivars with regards to the uptake kinetic parameters over the short time period (24 h) investigated here. Interestingly, SJBV, which had low shoot As accumulation in experiment I (Table 2), had a lower As uptake kinetic than TXYLS which shows higher As accumulation. The value of  $V_{max}$  in SJBV was ( $0.7737 \text{ mg As g}^{-1} \text{ shoot d.wt.hr}^{-1}$ ) and ( $1.8488 \text{ mg As g}^{-1} \text{ shoot d.wt.hr}^{-1}$ ) in TXYLS, and the value of  $K_m$  was ( $81.92 \text{ mg/L}^{-1}$ ) in TXYLS and ( $26.721 \text{ mg/L}^{-1}$ ) in SJBV. These values varied with plant species (Xiaozhang et al., 2005). SJBV had the lowest  $V_{max}$ , explained the lowest uptake ability in SJBV and TXYLS plants had the highest  $V_{max}$ , which explains the higher uptake ability. Furthermore, TXYLS had the highest  $K_m$  value than SJBV, confirming that uptake in the plant cell plasma membrane of TXYLS have a higher affinity for As (Poynton et al., 2004). The  $V_{max}/K_m$  values (Table 5) indicated that the arsenic uptake ability of cultivars followed TXYLS > SJBV. The difference between these two  $K_m$  values indicates that different cultivars of lettuce have different As affinity (Table 5). Yu and Gu, (2006) found that different cultivars had different kinetic parameters in the same substrate. The results

show that TXYLS was more As efficient to accumulate arsenic than SJBV. The higher As efficiency of TXYLS can be explained by the higher As uptake rate of the shoot with the passage of time (Table 5).

## Conclusion

This study clearly established the differences in  $K_m$  and  $V_{max}$  between two cultivars. In addition, the present study shows that, during 24 h of exposure, arsenic was depleted from solution, suggesting uptake of arsenic by the plants. The present study provides insights into the underlying mechanisms of As uptake kinetic in two different cultivars of lettuce which have both been shown to absorb the highest and lowest arsenic in their shoots. It is concluded that SJBV is the suitable cultivar plant to be developed on arsenic contaminated areas, which has a lower ability to accumulate a lesser concentration of arsenic from the hydroponic solution. Therefore, results suggest the possibility to lower the As concentration in lettuce through local cultivar by selecting and breeding cultivars in order to produce cultivars for safe consumption from soils with slight and moderate levels of As.

## ACKNOWLEDGEMENTS

We thank Dr Yi bing Ma, (China), Dr Liaqat Ali, scientist research officer(Pakistan) and Dr Paul Held Laboratory manager for their guidance in kinetic experiment and designing and performing Michaelis-Menten equation. This study was financially supported by the National Scientific and Technological Program of the "12th Five-Year" Plan of China (Grant No. 2011BAD04B01).

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