

# Transport of DMAA and MMAA into rice (*Oryza sativa* L.) roots

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1 **Transport of DMAA and MMAA into Rice (*Oryza sativa* L.) Roots**

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17

**18 Abstract:**

19 Arsenate (As(V)) transport into plant cells has been well studied. A study on rice (*Oryza sativa*  
20 L.) showed that arsenite is transported across the plasma membrane via glycerol transporting  
21 channels. Previous studies reported that the dimethylarsinic acid (DMAA) and  
22 monomethylarsonic acid (MMAA) uptake in duckweed (*Spirodela polyrhiza* L.) differed from  
23 that of As(V), and was unaffected by phosphate ( $\text{H}_2\text{PO}_4$ ). This article reports the transport  
24 mechanisms of DMAA and MMAA in rice roots. Linear regression analysis showed that the  
25 DMAA and MMAA uptake in rice roots increased significantly ( $p \leq 0.0002$  and  $\leq 0.0001$  for  
26 DMAA and MMAA, respectively) with the increase of exposure time. Concentration-dependent  
27 influx of DMAA and MMAA showed that the uptake data were well described by Michaelis-  
28 Menten kinetics. The MMAA influx was higher than that of DMAA. The DMAA and MMAA  
29 uptake in rice roots were decreased significantly ( $p \leq 0.0001$  and  $\leq 0.0077$  for DMAA and  
30 MMAA, respectively) with the increase of glycerol concentration indicating that DMAA and  
31 MMAA were transported into rice roots using the same mechanisms of glycerol. Glycerol is  
32 transported into plant cells by aquaporins, and DMAA and MMAA are transported in a dose-  
33 dependent manner of glycerol which reveals that DMAA and MMAA are transported into rice  
34 roots through glycerol transporting channels. The DMAA and MMAA concentration in the  
35 solution did not affect the inhibition of their uptake rate by glycerol.

36

37 **Keywords:** Arsenic, DMAA, MMAA, Rice (*Oryza sativa* L.), Aquaporins, Influx.

38

### 39 **Introduction:**

40           Although arsenic contamination in groundwater has been reported in many countries,  
41 Bangladesh (Acharyya et al., 1999; Alam and Sattar, 2000; Alam et al., 2002), West Bengal,  
42 India (Mandal et al., 1996; Chowdhury et al., 1999), China (Guo et al., 2001; Sun, 2004), and  
43 Taiwan (Schoof et al., 1998; Guo et al., 2001) are the mostly affected areas. In Bangladesh and  
44 West Bengal (India), the arsenic contaminated groundwater has been used not only for drinking  
45 purpose but also for crop irrigation, especially for rice cultivation. Presently, 75% of the total  
46 cropped area and 83% of the total irrigated area are used for rice cultivation in Bangladesh (Dey  
47 et al., 1996), which are mostly dependent on groundwater irrigation. Survey from Bangladesh  
48 show that irrigation with arsenic contaminated groundwater is leading to the elevation of arsenic  
49 in paddy soils (Alam and Sattar, 2000). Although the background levels of arsenic in soils of  
50 Bangladesh ranged between 4 and 8 mg kg<sup>-1</sup>, up to 83 mg kg<sup>-1</sup> soil arsenic has been reported in  
51 areas irrigated with contaminated water (Abedin et al., 2002). Irrigation of arsenic contaminated  
52 groundwater during dry season rice production has been adding > 1000 metric tons of arsenic to  
53 the soil per year in Bangladesh alone (Alam and Sattar, 2000; Meharg and Rahman, 2002). A  
54 substantial amount of arsenic is accumulated from soil and irrigation water and is deposited in  
55 rice grain, which has the potential to create health disaster for the population in Southeast Asia  
56 (Meharg, 2004). Worldwide market surveys show that rice grain contains considerably higher  
57 amount of arsenic than that in other food items (Schoof et al., 1999; Roychowdhury et al., 2002;  
58 Williams et al., 2007). Therefore, rice could be substantial for the population of arsenic epidemic  
59 areas.

60           Studies on the kinetics of arsenic uptake in plant roots have focused almost entirely on  
61 arsenate as this is the dominant form of plant available arsenic in aerobic soils (Meharg and

62 [Jardine, 2003](#)). In flooded condition, arsenite becomes the predominant species of arsenic  
63 ([Takahashi et al., 2004](#)). There is evidence of arsenic methylation in paddy soil systems by  
64 microorganisms ([Takamatsu et al., 1982](#)). A number of studies have been investigated the  
65 mechanisms of arsenic uptake by different plant species ([Meharg and Macnair, 1992](#); [Meharg  
66 and Macnair, 1994](#); [Rahman et al., 2008a](#); [Rahman et al., 2008c](#)). Plants take up arsenate through  
67 the phosphate transporters ([Meharg and Hartley-Whitaker, 2002](#); [Wang et al., 2002](#)). Although  
68 the exact mechanisms of arsenite uptake in higher plants has not been identified, physiological  
69 studies suggests that arsenite is transported in rice by aquaporins ([Abedin et al., 2002](#); [Meharg  
70 and Jardine, 2003](#)). A recent molecular study explained more clearly that arsenite is transported  
71 into rice roots by nodulin 26-like intrinsic membrane proteins (NIPs), one of the major  
72 subfamilies of aquaporins transporter that facilitates the transport of neutral molecules such as  
73 water, glycerol, and urea ([Ma et al., 2008](#)).

74 Uptake of organoarsenic species by plants is lower than that of inorganic species  
75 ([Odanaka et al., 1987](#); [Rahman et al., 2007](#)). [Marin et al. \(1992; 1993\)](#) observed high uptake of  
76 inorganic arsenic species, dimethylarsinic acid (DMAA) and monomethylarsonic acid (MMAA),  
77 in rice plant in hydroponic culture. Whatever the amount was, previous studies confirmed the  
78 uptake of organoarsenic species (DMAA and MMAA) in rice and other plant species. Although  
79 the uptake mechanisms of inorganic arsenic species such as arsenate and arsenite in rice have  
80 been studied ([Abedin et al., 2002](#); [Meharg and Jardine, 2003](#); [Ma et al., 2008](#)), the uptake  
81 mechanisms of organoarsenic species are overlooked. [Rahman, et al. \(2008a\)](#) observed that the  
82 DMAA and MMAA uptake in duckweed (*Spirodela polyrhiza* L.) was much lower than that of  
83 As(V) and As(III), and the uptake was not influenced by phosphate. This might be because the  
84 mechanisms of organoarsenic species uptake in plants differed from that of inorganic arsenic

85 species, and the physicochemical adsorption would be one of the possible mechanisms of  
86 DMAA and MMAA uptake in aquatic plants. [Robinson, et al. \(2003\)](#) also proposed  
87 physicochemical adsorption as an alternative mechanism for DMAA and MMAA uptake in New  
88 Zealand watercress (*Lepidium sativum*).

89 Uptake mechanisms of DMAA and MMAA in rice have not studied extensively. [Abedin](#)  
90 [et al. \(2002\)](#) studied the uptake kinetics of arsenic species in rice, and mostly focused on  
91 inorganic arsenic species, arsenate and arsenite. This study investigates the uptake kinetics of  
92 DMAA and MMAA into rice roots to observe how these species are taken up into the plant cells.  
93 Since plant aquaporins transport neutral molecules such as water, glycerol, and urea ([Dean et al.,](#)  
94 [1999; Ma et al., 2008](#)), and organoarsenic species are not taken up into plants by phosphate  
95 uptake pathway; there is a possibility of DMAA and MMAA uptake through the aquaporins  
96 water channels. Studies showed that the rice aquaporin Lsi1 mediates uptake of methylated  
97 arsenic species ([Li et al., 2009](#)). In the present study, we investigated the competition between  
98 glycerol and DMAA and MMAA for uptake into rice roots.

99

## 100 **Materials and Methods:**

### 101 **Seed sterilization**

102 Rice seeds of BRRI (Bangladesh Rice Research Institute) dhan 29 were collected from  
103 Bangladesh Rice Research Institute, Gazipur. The seeds were surface-sterilized before using  
104 them in the experiment. For sterilization, about 100 g seeds were soaked in 200 mL of 1%  
105 methyl-1-butylcarbamoyl-2-benzimidazole carbonate solution for 10 min. After that, the seeds  
106 were washed by deionized water (using an E-pure system (Barnstead)) and kept in deionized

107 water at 20 °C for 24 h. The seeds were then washed and transferred to deionized water of 45 °C  
108 for 2 min, and of 52 °C for 10 min.

109

## 110 **Plant growth**

111 Sterilized rice seeds were soaked in deionized water for 48 h, and were germinated on  
112 moistened filter paper placed within petri dishes. When the germinated seeds produced enough  
113 roots and about 2 cm of shoot, the small seedlings were transferred to a 500-mL polystyrene  
114 beaker filled with 400 mL of distilled water. The seedlings were placed on the water with a  
115 support in such a way that only the roots of the seedlings emerged into the water. The rice  
116 seedlings were allowed to grow for 1 wk in the distilled water. Nutrient salts and other  
117 osmoregulators were not added to the water so that they could not alter the arsenic transporter  
118 regulation in an unknown manner (Meharg and Jardine, 2003). Rice seedlings were grown in a  
119 plant growth chamber, and the conditions in the chamber were set as 14:10 h light/dark schedule,  
120 100-125  $\mu\text{E m}^{-2} \text{s}^{-1}$  light intensity, 22( $\pm$ 2) °C temperatures.

121

## 122 **Uptake kinetics**

123 After 1 wk growth, sufficient numbers of roots were produced from the basal node.  
124 Replicated rice seedlings were then transferred to aerated water solution (having no nutrient  
125 salts) for 30 min at room temperature. They were then incubated in aerated test solutions  
126 (distilled water without nutrient salts) for 1 h with different concentrations (ranged between 0.1  
127 and 0.7 mM) of DMAA and MMAA for concentration-dependant uptake experiment. Replicated

128 rice seedlings were incubated in test solution (distilled water without nutrient salts) with 0.1 mM  
129 arsenic for time-dependant uptake experiment, and the samples were collected at a 10 min  
130 interval. In DMAA and MMAA transport assay, 0.3 mM or 10  $\mu$ M DMAA or MMAA was  
131 added to 10, 50, 100, 500, and 1000 mM glycerol solutions. Replicated rice seedlings were  
132 incubated into these solutions for 1 h. The test solution was adjusted to pH 7 using weak  
133 solutions of HCl or NaOH. Stock solutions of DMAA and MMAA were prepared from  
134 dimethylarsinic acid ( $((\text{CH}_3)_2\text{AsO}(\text{OH}))$ ) and  $(\text{CH}_3\text{AsO}(\text{OH})_2)$ , respectively. Glycerol was  
135 purchased from Kanto Chemical Co., Japan (purity 99.0%).

136

### 137 **Sample preparation and chemical analysis**

138 After the set time, the roots were quickly rinsed in ice cold distilled water, and then  
139 placed in aerated ice cold distilled water for 20 min (Meharg and Jardine, 2003). The roots were  
140 washed once again with distilled water and blotted dry with tissue papers. Now the roots of the  
141 rice seedlings were excised at the basal node, and the fresh weight of the roots were determined.  
142 The roots were then taken into 50-mL polyethylene digestion tubes, and 3 mL of 65%  $\text{HNO}_3$   
143 were added to the samples and allowed to stand for 12 h. The samples were heated on a heating  
144 block at 95  $^\circ\text{C}$  for 90 min. After cooling to room temperature, 2 mL of 30% hydrogen peroxide  
145 were added, and heated again at 105  $^\circ\text{C}$  for 30 min. On cooling, the residue was taken was  
146 diluted to 10 mL with deionized water, and analyzed for total As. At least one reagent blank and  
147 two certified standard reference materials (1573a, tomato leaf from National Institute of  
148 Standards and Technology (NIST), Department of Commerce, United States of America) were  
149 included in the digestion. Chemical analysis for arsenic was performed by graphite-furnace

150 atomic absorption spectrometer (Z-8100, Hitachi, Japan). Certified standard reference material  
151 1573a (tomato leaf from NIST, USA) was used to check the accuracy of analysis. Arsenic  
152 concentration in certified standard reference materials was  $0.112 \pm 0.004 \mu\text{g g}^{-1}$  dry weight while  
153 the measured concentration was  $0.114 \pm 0.002 \mu\text{g g}^{-1}$ . The concentrations detected in all samples  
154 were above the instrumental limits of detection ( $\geq 0.01 \mu\text{M}$  in water sample).

155 All chemical reagents used in this experiment were of analytical grade. Glassware and  
156 dishes were washed with detergent and 1 N HCl solution, and rinsed with deionized water for  
157 eight times before use.

158

## 159 **Statistical Analysis**

160 Data were analyzed for linear and nonlinear regression using GraphPad Prism (v5)  
161 (GraphPad Software, Inc., CA, USA). Kinetic parameters for DMAA and MMAA uptake were  
162 calculated from mean arsenic influx ( $n = 3$ ) by linear and nonlinear regression models.

163

## 164 **Results and discussions:**

### 165 **Uptake kinetics of DMAA and MMAA**

166 Time- and concentration-dependent uptake kinetics of DMAA and MMAA were  
167 determined to assess the pattern and efficiency of organoarsenic species influx in rice roots.  
168 Since the uptake kinetic is calculated from the influx (uptake into the plant cells) across the  
169 plasma membrane, it is important to measure the adsorption of arsenic on rice root surfaces. The

170 adsorption of As(V) and As(III) on roots of terrestrial and aquatic plants has been reported by  
171 several researchers (Otte et al., 1995; Hansel et al., 2002; Blute et al., 2004; Chen et al., 2005;  
172 Rahman et al., 2007; Rahman et al., 2008c). A significant amount of As(V) is adsorbed on Fe-  
173 oxides (Fe-plaques) on rice root surfaces (Chen et al., 2005; Rahman et al., 2008b) because of  
174 high adsorptive affinity of As(V) to Fe-oxides, while DMAA and MMAA adsorption, either on  
175 Fe-plaques or on rice roots, is negligible (Rahman et al., 2007).

176 Even though there was a little chance of chemical adsorption of arsenic on Fe-oxides in  
177 the Fe-free experimental solution (distilled water without nutrient salts), Meharg and Jardine  
178 (2003) reported significant physical adsorption of As(V) and As(III) on rice roots. Meharg and  
179 Jardine (2003) evaluated the desorption of arsenic from rice root surface by ice cold distilled  
180 water and NaCl (0.1 M) solution. Since there was no significant differences of these two washing  
181 methods in arsenic desorption from rice root surface, they proposed ice cold distilled water  
182 washing as the appropriate method. Therefore, rice roots were washed with ice cold distilled  
183 water in this study to remove arsenic physically adsorbed on rice roots.

184 Time-dependent uptake showed that the influxes of both DMAA and MMAA were linear  
185 upon 60 min. of exposure (Fig. 1). The DMAA and MMAA uptakes were well described by a  
186 linear function ( $r = 0.688$  and  $0.756$  for DMAA and MMAA, respectively), and their uptakes  
187 were increased significantly ( $p = 0.0002$  and  $< 0.0001$  for DMAA and MMAA, respectively)  
188 with the increase of exposure time (Table 1). Meharg and Jardine (2003) reported that As(III)  
189 influx was linear up to 30 min, and further influx did not occurred probably due to the  
190 toxicological inhibition as As(III) exhibits phytotoxicity through binding with protein -SH  
191 groups. Present result showed that DMAA and MMAA did not show phytotoxicity up to 60 min.  
192 Although organoarsenic species are generally considered to be less toxic than inorganic species

193 to a wide range of organisms including aquatic plants, animals and humans (Tamaki and  
194 Frankenberger, 1992), long-term arsenic uptake studies showed that the phytoavailability of four  
195 arsenic species to *Spartina patens* in hydroponic systems followed the trend: DMAA < MMAA  
196  $\cong$  As(V) < As(III), while the order of phytotoxicity was As(V)  $\cong$  As(III) < MMAA < DMAA.  
197 Studies also suggests that organoarsenicals would be more toxic than inorganic arsenic species  
198 (Carbonell-Barrachina et al., 1998). In another study with arsenate, arsenite, and DMAA influx  
199 in maize (*Zea mays* L.), Abbas and Meharg (2008) found low toxicity of DMAA compared with  
200 arsenate and arsenite, and the relative toxicity of arsenic species on maize was As(V) > As(III) >  
201 DMAA. The phytoavailability of arsenic by rice, however, in long-term hydroponic culture was  
202 DMAA < As(V) < MMAA < As(III), and the order of phytotoxicity was the same as the order of  
203 phytoavailability (Marin et al., 1992). Moreover, short-term uptake of MMAA and DMAA was  
204 considerably less than that of As(V) and As(III) in rice (Abedin et al., 2002), which is consistent  
205 to the time-dependent DMAA and MMAA uptake in rice roots of present study. Thus, from the  
206 above discussions it could be assumed that the uptake and toxicity of arsenic species are related  
207 to the plant species as well as to the exposure time depending on their resistance mechanisms.

208 Concentration-dependent influx showed that the DMAA uptake was poorly described by  
209 Michaelis-Menten kinetics ( $r^2 = 0.688$ ), but well explained by linear function ( $r^2 = 0.837$ ) (Fig.  
210 2; Table 1). On the other hand, MMAA uptake showed a hyperbolic increase with the increase of  
211 MMAA in the experimental solution (Fig. 3). The MMAA uptake fitted well to the Michaelis-  
212 Menten kinetics ( $r^2 = 0.914$ ) as well explained as linear function ( $r^2 = 0.904$ ) (Table 1). These  
213 results are also in consistent with those of Abedin et al. (2002). The MMAA influx was higher  
214 than that of DMAA (Figs. 2 and 3). At a substrate concentration of 0.7 mM, the uptake rates of  
215 DMAA and MMAA were 1.25 and 1.74  $\mu\text{mol g}^{-1}$  fresh weight  $\text{h}^{-1}$ , respectively. Kinetic

216 parameters also showed that  $V_{\max}$  for DMAA and MMAA were 0.757 and 3.619  $\mu\text{mol g}^{-1}$  fresh  
217 weight  $\text{h}^{-1}$  (Table 1). [Abedin et al. \(2002\)](#) also reported similar results for DMAA and MMAA in  
218 rice roots.

219

### 220 **Inhibition of DMAA and MMAA uptake by glycerol**

221 Water channels or water channel proteins (WCPs) are transmembrane proteins that have a  
222 specific three-dimensional structure with a pore that permeates water molecules ([Benga, 2009](#)).  
223 The WCPs belong to the superfamily of major intrinsic proteins (MIPs) (over 800 members) that  
224 are present in plants, animals, and microorganisms. The WCPs include three subfamilies: i) the  
225 water specific aquaporins (AQPs), ii) aquaglyceroporins (permeable to water, glycerol, and/or  
226 other small, neutral molecules), and iii) superaquaporins or subcellular AQPs ([Agre, 2004](#);  
227 [Benga, 2009](#)). In addition to water, some MIPs seem to be specific to other molecules such as  
228 urea, glycerol or even  $\text{CO}_2$  ([Maurel et al., 1994](#); [Baiges et al., 2002](#)).

229 The competition between glycerol and arsenite for uptake into rice (*Oryza sativa* L.)  
230 ([Meharg and Jardine, 2003](#)) and *Saccharomyces cerevisiae* ([Wysocki et al., 2001](#)) reveal that  
231 arsenite is transported across the plasma membrane through WCPs/aquaporins. Previous studies  
232 reported that the DMAA and MMAA uptake mechanisms into plant tissues differ from those of  
233 arsenate ([Mkandawire and Dudel, 2005](#); [Rahman et al., 2008a](#)). In the present study, we  
234 investigated the effect of glycerol on DMAA and MMAA uptake in rice roots to understand the  
235 uptake mechanisms of these organoarsenic species. Results showed that glycerol inhibited  
236 DMAA and MMAA uptake in rice roots significantly ( $p \leq 0.0001$  and 0.0077 for DMAA and

237 MMAA, respectively; [Table 2](#)) in a concentration dependant manner ([Fig. 4](#)), which is consistent  
238 to the arsenite uptake in rice roots ([Meharg and Jardine, 2003](#)).

239         Among DMAA and MMAA, the DMAA influx was about two times greater than that of  
240 MMAA. DMAA and MMAA influx in rice roots were higher at low glycerol concentrations (10-  
241 50 mM) than those at higher concentrations (100-1000 mM) ([Fig. 4](#)). [Meharg and Jardine \(2003\)](#)  
242 elucidated the possible explanations for the inhibition of arsenite uptake in rice roots by glycerol  
243 which can be applicable for DMAA and MMAA too. The explanations are: i) glycerol closes  
244 aquaporin channels, ii) glycerol causes general physiological stress disrupting arsenite  
245 transporter, and iii) high levels of glycerol rapidly down-regulate aquaporin channels. Since  
246 glycerol has been widely used for aquaporins assay ([Biela et al., 1999](#); [Dean et al., 1999](#)) and  
247 since glycerol has low phytotoxicity ([Meharg and Jardine, 2003](#)), the first two explanations do  
248 not elucidate adequately. Moreover, the third explanation is also not agreeable because transport  
249 activity would be constant for the short exposure time (1 h) ([Meharg and Jardine, 2003](#)).  
250 Therefore, inhibition of DMAA and MMAA influx by glycerol indicates that they are  
251 transported across the plasma membrane via same transporter such as MIPs/aquaglyceroporins.

252         Inhibition effect of glycerol on arsenic uptake at low (10  $\mu$ M) and high (0.3 mM) DMAA  
253 and MMAA concentration was investigated. Results show that the rates of DMAA and MMAA  
254 influx were almost similar for both concentrations, and the uptake of both arsenic species was  
255 linear rather than hyperbolic ([Figs. 4 and 5](#)). Linear regression analysis of mean arsenic influx in  
256 rice roots reveals that the mean  $r^2$  values for DMAA and MMAA at 0.3 mM concentration were  
257 0.891 and 0.525, respectively. The values were 0.509 and 0.354 for DMAA and MMAA at 10  
258  $\mu$ M concentration, respectively ([Table 2](#)). Thus, it can be revealed that the aquaglyceroporin  
259 channels facilitate DMAA uptake more frequently compared to that of MMAA.

260

## 261 **Conclusions**

262 Uptake of organoarsenic species in plants is lower than those of inorganic species.  
263 Although several reports have been described the uptake mechanisms of arsenate and arsenite in  
264 plants, little is known about uptake mechanisms of organoarsenic species. The results of this  
265 study show that the DMAA and MMAA follow the uptake mechanisms of glycerol in rice roots.  
266 The glycerol transporter in plasma membrane (aquaglyceroporins) facilitates DMAA and  
267 MMAA uptake in rice roots indicating that these arsenic species are transported in rice via  
268 MIPs/aquaglyceroporins.

269

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273

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407 Table 1: Kinetic parameters for time-dependent DMAA and MMAA uptake in rice roots. Kinetic  
 408 parameters were calculated from mean As influx ( $n = 3$ ) by Michaelis-Menten function  
 409 (nonlinear regression) and linear regression model using the GraphPad Prism (v5)  
 410 (GraphPad Software, Inc., CA, USA).

As Species	Nonlinear Regression			Linear Regression			
	$V_{\max}$ ( $\mu\text{mol g}^{-1} \text{ f. wt.}$ )	$K_m$ (mM)	$r^2$	$a$	$b$	$r^2$	$p$
Time-dependent							
DMAA				0.007±0.001	0.119±0.045	0.688	0.0002
MMAA				0.002±0.000	0.014±0.007	0.756	< 0.0001
Con.-dependent							
DMAA	0.757	0.140	0.688	0.644±0.116	0.253±0.048	0.837	0.0014
MMAA	3.619	0.762	0.914	2.399±0.318	0.186±0.133	0.904	0.0003

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419 Table 2: Kinetic parameters for the uptake inhibition of DMAA and MMAA in rice roots by  
 420 glycerol. Kinetic parameters were calculated from mean As influx ( $n = 3$ ) by linear  
 421 regression model using the GraphPad Prism (v5) (GraphPad Software, Inc., CA, USA).

As Species	<i>a</i>	<i>b</i>	$r^2$	<i>p</i>
Glycerol + As (0.3 mM)				
DMAA	$-3.04 \times 10^{-4} \pm 3.36 \times 10^{-5}$	$3.34 \times 10^{-1} \pm 1.54 \times 10^{-2}$	0.891	< 0.0001
MMAA	$-4.11 \times 10^{-4} \pm 1.23 \times 10^{-4}$	$11.66 \times 10^{-1} \pm 5.68 \times 10^{-2}$	0.525	0.0077
Glycerol + As (10 $\mu$ M)				
DMAA	$-1.73 \times 10^{-5} \pm 5.38 \times 10^{-6}$	$3.15 \times 10^{-2} \pm 2.46 \times 10^{-3}$	0.509	0.0092
MMAA	$-6.96 \times 10^{-5} \pm 2.97 \times 10^{-5}$	$1.22 \times 10^{-1} \pm 1.36 \times 10^{-2}$	0.354	0.0412

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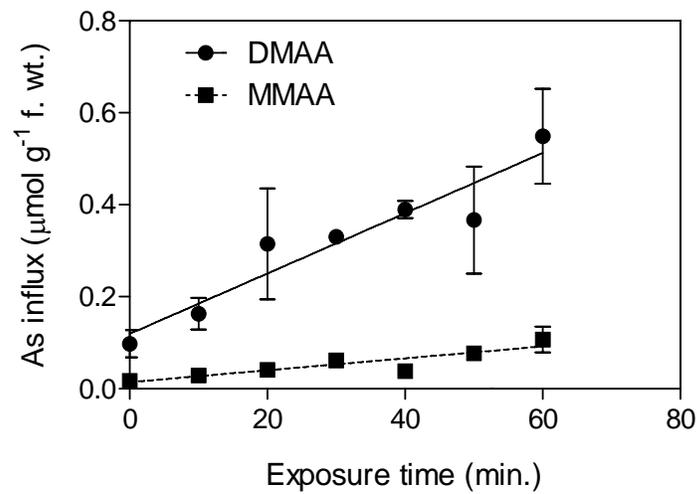
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433 Fig. 1: Time-dependent influx of DMAA and MMAA in rice roots. Arsenic concentration in the  
434 solution was 0.30 mM. The graph shows the linear regression lines, and the kinetic  
435 parameters are given in Table 1. Each point is the average value of three replicated  
436 treatments. Bares represent  $\pm$  standard error of the mean (SEM) of the replicates.

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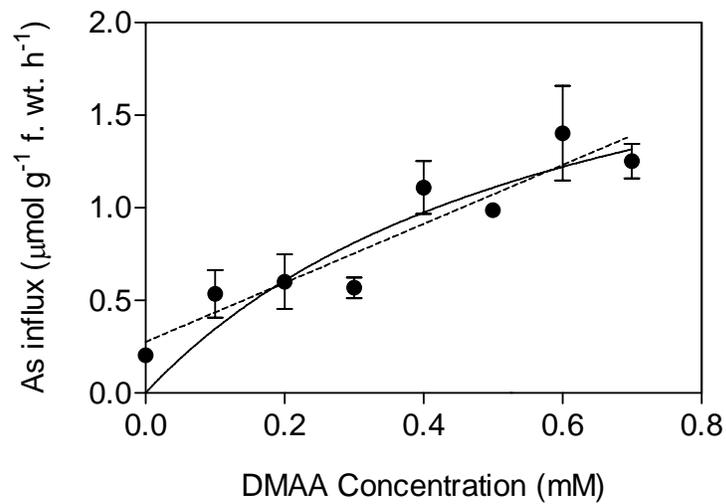
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446 Fig. 2: Concentration-dependent influx of DMAA in rice roots. The graph shows the Michaelis-  
447 Menten (nonlinear regression) curve and linear regression line, and the fits are given in  
448 Table 1. Each point is the average value of three replicated treatments. Bares represent  $\pm$   
449 standard error of the mean (SEM) of the replicates.

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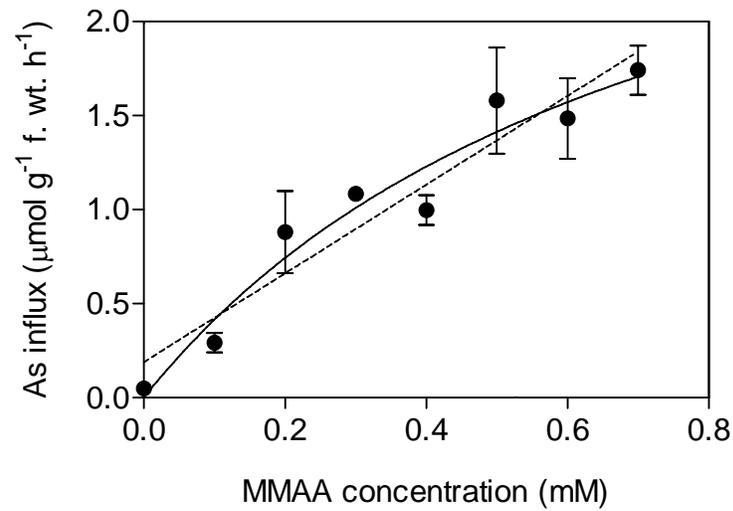
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459 Fig. 3: Concentration-dependent influx of MMAA in rice roots. The graph shows the Michaelis-  
460 Menten (nonlinear regression) curve and linear regression line, and the fits are given in  
461 Table 1. Each point is the average value of three replicated treatments. Bares represent  $\pm$   
462 standard error of the mean (SEM) of the replicates.

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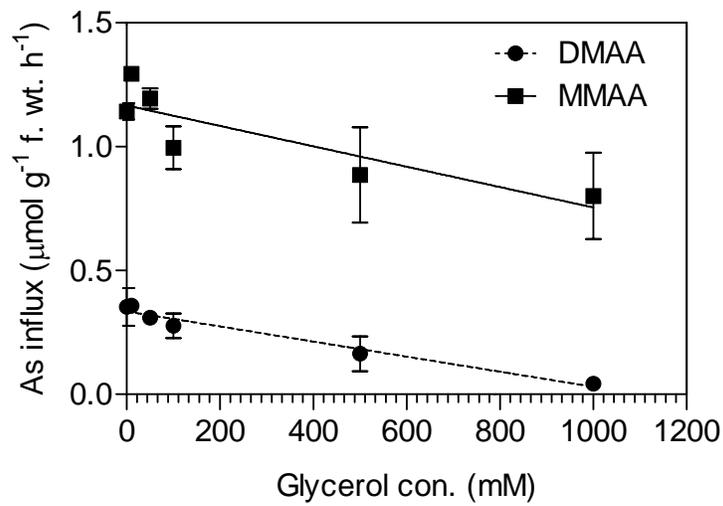
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472 Fig. 4: Inhibition of DMAA and MMAA influx (0.3 mM) in rice roots by different concentration  
473 of glycerol. The graph shows the nonlinear regression lines, and the fits are given in  
474 Table 2. Each point is the average value of three replicated treatments. Bares represent ±  
475 standard error of the mean (SEM) of the replicates.

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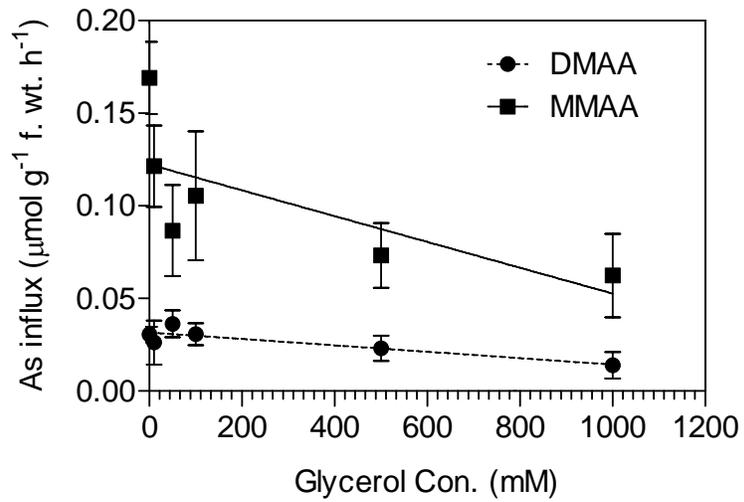
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485 Fig. 5: Inhibition of DMAA and MMAA influx (10 μM) in rice roots by different concentration  
486 of glycerol. The graph shows the nonlinear regression lines, and the fits are given in  
487 Table 2. Each point is the average value of three replicated treatments. Bares represent ±  
488 standard error of the mean (SEM) of the replicates.