Tolerance of young Loblolly pine (*Pinus taeda*) seedlings to post-emergence applications of MSMA

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Abstract

Nursery trials were conducted in 2005 and 2006 to determine the tolerance of bare-root loblolly pine (*Pinus taeda* L.) seedlings to the herbicide MSMA. Seedlings in five nurseries were treated 4 to 14 weeks after sowing at rates up to 2.24 kg/ha. Seedlings sampled in the fall showed no signs of shoot or root injury. MSMA treated seedlings were not statistically smaller than those lifted from control plots. MSMA is used in cotton (*Gossypium hirsutum* L.) fields and could prove useful for selectively controlling sedges in pine nurseries. However, since MSMA contains arsenic, the use of this herbicide will not be allowed in nurseries that supply seedlings to plantations certified by the Forest Stewardship Council.

Key Words: *Cyperus,* forest tree nurseries, weed control, herbicide, nutsedge

Introduction

Methylarsinic acid is an organic arsenical herbicide (CH_4AsNaO_3) that is used to control *Paspalum dilatatum* Poir. in turf. In Australia it is called monosodium methylarsonate and in the United States (U.S.) it is known as monosodium methanearsonate (MSMA). It some countries it is used to control sedges and grasses in cotton (*Gossypium hirsutum* L.), sugarcane (*Saccharum* spp.) and on non-cropland. In 1997, more than 2 million kg of MSMA were applied in the U.S. (Gianessi and Marcelli 2002), which ranks MSMA as 21st in terms of total amount of active ingredient applied. MSMA has medium to low mobility in sandy soils and might leach 50 cm in a sandy loam. The herbicide is strongly adsorbed to soil particles, and in some soils, there is apparently little or no leaching below the plow zone (Hiltbold *et al.* 1974). The reported half-life averages about 240 days in non-irrigated soils but with high irrigation levels, the half-life was reported to be about 55 days. Uptake by roots is limited and the primary pathway into plants is through the foliage. MSMA causes cell membrane destruction and rapid desiccation of treated weeds. It is classified as an acutely toxic substance.

MSMA is used to control weeds in Australia (Charles 1997), Africa (Hillocks 1995), India, the U.S. (Monks *et al.* 1999) and in New Zealand. This herbicide has activity on various weeds (Table 1) including the world's worst weed; purple nutsedge (*Cyperus rotundus*) (Holm *et al.* 1977). McElroy *et al.* (2003) found that 60 days after applying 2.2 kg/ha of MSMA, the numbers of nutsedge shoots were reduced by 48 percent for purple nutsedge and 68 percent for yellow nutsedge (*Cyperus esculentus*). While repeated applications of MSMA are often needed to provide effective control of yellow and purple nutsedge, a single application is sufficient to control flathead sedge (*Cyperus compressus* L.) (Belcher *et al.* 2002).

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Table 1. A partial list of plants susceptible to injury when MSMA is applied to foliage (broadleaves, grasses, sedges) or injected under the bark (conifers).

Common Name	Scientific Name
BROADLEAVES	
Redroot pigweed	*Amaranthus retroflexux* L.
Pitted morningglory	*Ipomea lacunose* L.
Ivyleaf morninglory	*Ipomoea hederacea* (L.) Jacq.
Prostrate spurge	*Euphorbia humistrata* Engel. ex Gray
GRASSES	
Large crabgrass	*Digitaria sanguinalis* (L.) Scop.
Goosegrass	*Eleusine indica* (L.) Gaertn.
Barnyardgrass	*Echinochloa crus-galli* (L.) Beauv.
Paspalum	*Paspalum dilatatum* Poir.
SEDGES	
Flathead sedge	*Cyperus compressus* L.
Purple nutsedge	*Cyperus rotundus* L.
Yellow nutsedge	*Cyperus esculentus* L.
CONIFERS	
Grand fir	*Abies Grands* (Dougl.) Lindl.
Silver fir	*Abies amabilis* (Dougl.) Forbes.
Jack Pine	*Pinus banksiana* Lamb.
Lodgepole pine	*Pinus contorta* Dougl. var *contorta*
Pondersoa pine	*Pinus ponderosa* Laws. var. *ponderosa*
Red pine	*Pinus resinosa* Ait.
Douglas fir	*Pseudotsuga menziesii* Franco.
Western hemlock	*Tsuga heterophylla* (Raf.) Sarg.

Table 2. Soil texture, organic matter (OM) and acidity of soils at the five nurseries.

FACTOR	Elberta	Shubuta	White City	Taylor	Flint River
Texture	loamy sand	sandy loam	sand	sand	loamy sand
Sand (%)	80	77	95	86	84
Silt (%)	13	17	2	12	8
Clay (%)	7	6	3	2	8
Organic matter (%)	1.1	1.7	1.1	0.9	1.8
pH	5.2	5.0	4.9	4.7	6.1
Year	2005	2005	2005	2005	2005
Sowing date	May13	April 27	May 3	April 12	April 24
Treatment date	August 17	June 8	June 2	June 20	June 19
Sampling date	November 16	November 8	October 11	October 3	December 12

In Australia, Hingston (1990) applied MSMA to control epicormic shoots of *P. radiata*. In the U.S. and Canada, foresters may inject MSMA under the bark of undesirable trees. In Canada, MSMA is registered to assist in controlling beetle infestations in conifers (Machlauchlan *et al.* 1988) and for tree thinning. In the U.S., MSMA may also be applied to control annual weeds in walnut (*Juglans spp.*) and cherry (*Prunus spp.*) orchards. To reduce the potential for injury to young nut trees, some labels indicate that if necessary, the applicator should use a shield to protect the seedlings from injury.

MSMA is not registered for use in nurseries and therefore nursery managers use other methods to control sedges. One method is to fumigate the soil prior to sowing (Carey 2000; South 2007). Another method is to control nutsedge in fallow land with multiple applications of non-selective herbicides (Fraedrich *et al.* 2003). Selective herbicides like EPTC and fomesafen may be used in the U.S., but these treatments typically do not provide lasting nutsedge control. In some nurseries, glyphosate is applied between seedling drills using shields to protect the seedlings (South and Carey 2005; Vachowski 2006). Care must be taken during application since seedling mortality can result if glyphosate contacts young pines. For these reasons, managers of pine nurseries need a selective herbicide that will kill emerged nutsedge without injuring seedlings.

Newly transplanted pines have demonstrated tolerance to MSMA (Holt *et al.* 1973; Kane 1992) but use in nurseries has been limited to controlling weeds adjacent to irrigation pipes (Stanley 1970). MSMA apparently was not screened for use in seedbeds and therefore was not used in pine seedbeds during the 20th century (South 1995). The objective of this study was to determine tolerance of loblolly pine seedlings to MSMA. In addition, rate responses to MSMA were evaluated under a variety of nursery conditions.

Materials and Methods

Experiments were established at three nurseries in 2005 and at two nurseries in 2006. Nurseries were located in the southern U.S. on sands, loamy sands or sandy loams (Table 2). The Elberta Nursery (30° 27' N, 87° 31' W) and White City Nursery (32° 39' N, 86° 33' W) are located in Alabama while the Shubuta Nursery (31° 59' N, 88° 48' W), Flint River Nursery (32° 10' N, 83° 58' W) and Taylor Nursery (33° 44' N, 81° 49' W) are located in Mississippi, Georgia, and South Carolina, respectively. Seedbeds were formed and seeds were sown in either April or May. After sowing, soil was either covered with pine bark mulch (Shubuta, White City and Flint River) or treated with a polymeric bed stabilizer (Agrilock®). After sowing, seedbeds were treated with an operational treatment of oxyfluorfen. Operational postemergence herbicides were applied to suppress the competition from annual weeds.

For each study, the experimental design was a randomized complete block with five or six replications. Plot size was one bed wide (1.4 m) and 2.9 m long. The studies included at least two herbicide treatments plus an untreated control. The formulation of MSMA tested contained 720 g of active ingredient/liter with a surfactant. The herbicide solution was applied using a CO_2-backpack sprayer calibrated to apply 206 L/ha at 192 kPa through three 8003 nozzles. Treatments varied with nursery and rates ranged from 0.42 to 2.24 kg of MSMA per ha.

On June 22, a visual estimate of *Cyperus compressus* cover (i.e. percent ground cover) was recorded for plots at the White City Nursery. In the fall, plots at all nurseries were evaluated for seedling production. Seedling densities (i.e. number of plantable seedlings per square meter) were recorded using a 30.4 x 122 cm counting frame and seedlings with root-collar-diameters greater than 3.1 mm were classified as plantable. Seedling samples were hand-lifted from the center of each plot and transported to Auburn for analysis. Heights and root-collar diameters were measured on 25 seedlings per plot. Oven-dry weights of shoots and roots were recorded for each 25-seedling sample. ANOVA was conducted on all data using SAS Proc Anova (SAS 1998). Data were analyzed separately for each trial and linear and quadratic contrasts were used to evaluate the

Figure 1. Untreated plot in foreground was covered by Cyprus compressus L. on July 13, 2005 (20 days after treatment) at the White City Nursery. Treated plot (top) was sprayed with 1.68 kg/ha MSMA on June 31, 2005.

significance of herbicide rate on seedling morphology. The null-hypothesis was rejected when *p*-values for contrast tests were significant ($\alpha = 0.05$). When the objective is to examine seedling response to herbicide rates, contrast statements are preferred over the use of multiple range tests (Mize and Schultz 1985; Saville 2003).

Results and Discussion

MSMA treatments did not stunt loblolly pine seedlings at any nursery and seedling production was not significantly reduced at application rates of 1.68 to 2.24 kg/ha. None of the MSMA rate studies produced a negative linear effect on seedling attributes (Table 3). At time of treatment, seedlings ranged from 4 weeks (White City Nursery) to 14 weeks after sowing (Elberta Nursery). Therefore, seedling age did not appear to be a factor in tolerance to MSMA. Positive effects of MSMA rates on seedling density were indicated by significant quadratic effects at two nurseries in Alabama (Table 3). At the Elberta Nursery, the middle rate (0.56 kg/ha) produced the most seedlings and at the White City Nurseries the higher rate (1.68 kg/ha) produced the most seedlings (Table 4).

Weed populations were low at four nurseries due to the application of operational herbicides and the low amount of weed cover did not justify conducting visual ratings of weed cover. At nurseries with minimal weed competition, the average shoot weight of untreated seedlings was more

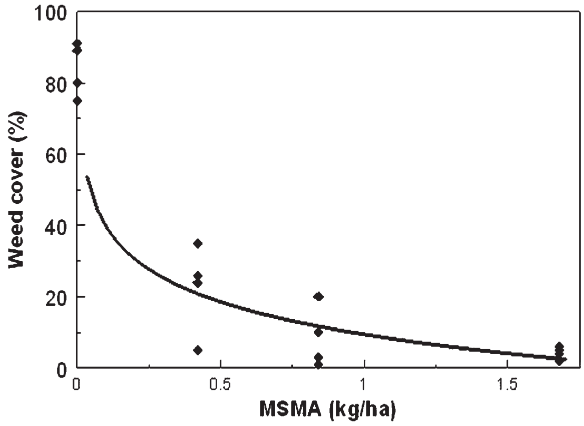

Figure 2. The effect of MSMA rate on the amount of seedbed covered by Cyprus compressus L.
20 days after treatment at the White City Nursery. Contrast statements indicate significant linear (P=0.0001), quadratic (P=0.0001) and lack-of-fit (P=0.0278) effects.

than 2.4 g (Table 4). However, at the White City Nursery, the post-sowing application of oxyfluorfen did not prevent the emergence of flathead sedge. Therefore, the low shoot weight of untreated seedlings (2.04 g) was due at least in part to competition from flathead sedge (Figure 1). The MSMA treatments at the White City Nursery produced positive responses with rates greater than 0.8 kg/ha producing the tallest seedlings and greater shoot and root mass (Table 4). Seedlings treated with 1.68 kg/ha of MSMA were as large in diameter as any treatment from the nurseries with low weed pressure and total dry mass (3.17 g per seedling) was equal to or greater than untreated seedlings from three other nurseries. However, the positive quadratic response to MSMA and the comparison of dry mass among nurseries does not prove that MSMA did not injure seedlings at the White City Nursery. The possibility that MSMA stunted seedlings remains since one cannot prove a null-hypothesis.

At the White City Nursery, plots treated with 0.84 kg/ha of MSMA had 9 percent cover while untreated plots averaged 84 percent cover (Figure 2). These findings indicate that recently emerged flathead sedge is susceptible to MSMA at rates as low as 0.5 kg/ha.

MSMA is an effective herbicide for controlling emerged annual and perennial sedges. It might prove useful in pine nurseries as part of an integrated weed management program. Prior to sowing pines, sedge populations could be reduced by applying glyphosate to nutsedge patches on fallow land (Fraedrich *et al.* 2003). After pine seedlings and weeds have emerged, spot applications of MSMA could be applied to selectively suppress sedge growth. Desiccating the shoots of nutsedge plants will reduce their ability to produce new tubers. Preventing the reproduction of nutsedge tubers is an integral part of a successful nutsedge control program.

Table 3. Analysis of Variance for loblolly pine seedlings as affected by MSMA.

Source	df	Density	RCD	Height	Shoot	Root
		---------------------- Probability of a greater F-value ------------------------				
---------- Elberta ----------						
Replication	4	0.7003	0.0106	0.0074	0.2341	0.4797
MSMA Treatment	2	0.0933	0.8522	0.6715	0.3117	0.4249
Linear	(1)	0.6030	0.5991	0.3876	0.1989	0.2898
Quadratic	(1)	0.0377	0.8741	0.9584	0.4133	0.4524
Error	8					
---------- Shubuta ----------						
Replication	4	0.7133	0.5774	0.6997	0.6828	0.3766
MSMA Treatment	2	0.2593	0.6609	0.3212	0.4273	0.9072
Linear	(1)	0.3675	0.9300	0.1564	0.2898	0.7579
Quadratic	(1)	0.1679	0.3796	0.6828	0.4574	0.7654
Error	8					
---------- White City ----------						
Replication	5	0.0001	0.1871	033473	0.1869	0.5536
MSMA Treatment	3	0.0211	0.0044	0.6025	0.0146	0.0564
Linear	(1)	0.4011	0.8543	0.8841	0.6259	0.9625
Quadratic	(1)	0.0034	0.0021	0.2623	0.0056	0.0177
Lack of fit	(1)	0.5819	0.0237	0.4764	0.0661	0.1473
Error	15					
---------- Taylor ----------						
Replication	4	0.6990	0.0944	0.0928	0.4071	0.3921
MSMA Treatment	2	0.1485	0.1015	0.5393	0.6512	0.3245
Linear	(1)	0.0828	0.0801	0.3166	0.3890	0.8966
Quadratic	(1)	0.3555	0.1799	0.6712	0.7899	0.1468
Error	8					
---------- Flint River ----------						
Replication	4	0.0425	0.1348	0.0006	0.0421	0.0577
MSMA Treatment	2	0.8632	0.9093	0.3642	0.7166	0.4728
Linear	(1)	0.7632	0.6771	0.7838	0.6111	0.3307
Quadratic	(1)	0.6647	0.9421	0.1748	0.5375	0.4700
Error	8					

Table 4. Morphological characteristics for loblolly pine seedlings at five nurseries. Fisher's LSD values are provided in italics to provided information on the relative power of each statistical test.

Treatment	rate kg/ha	Density (#/m2)	RCD (mm)	Height (cm)	Shoot (g)	Root (g)
	---------- Elberta ----------					
Control	0	201	4.4	24.5	2.49	0.51
MSMA	0.56	244	4.4	24.4	2.51	0.47
MSMA	1.12	188	4.4	24.2	2.01	0.91
	(LSD)	*53*	*0.23*	*0.99*	*0.80*	*0.81*
	---------- Shubuta ----------					
Control	0	278	4.0	29.2	2.83	0.35
MSMA	0.56	270	4.1	30.0	2.88	0.37
MSMA	1.12	274	4.0	30.3	2.29	0.36
	(LSD)	*10*	*0.28*	*0.58*	*1.08*	*0.07*
	---------- White City ----------					
Control	0	95	3.7	24.6	2.04	0.28
MSMA	0.42	105	4.3	25.3	2.64	0.36
MSMA	0.84	120	4.3	25.5	2.72	0.38
MSMA	1.68	125	4.4	25.4	2.79	0.38
	(LSD)	*20*	*0.36*	*1.65*	*0.47*	*0.08*
	---------- Taylor ----------					
Control	0	228	4.4	29.5	4.10	0.55
MSMA	1.12	214	4.4	29.3	4.47	0.64
MSMA	2.24	213	4.3	29.5	4.59	0.55
	(LSD)	*17*	*0.14*	*0.48*	*1.23*	*0.15*
	---------- Flint River ----------					
Control	0	186	3.8	30.8	2.41	0.23
MSMA	1.12	182	3.8	31.7	2.52	0.23
MSMA	2.24	184	3.8	31.0	2.48	0.20
	(LSD)	*16*	*0.21*	*1.43*	*0.32*	*0.05*

Table 5. Estimated arsenic inputs from various amendments to a pine nursery.

Amendment	Amount per ha	Arsenic concentration	Area treated	Arsenic applied
8 cm of topsoil	1,000,000kg	5 mg/kg*	1 ha	5,000 g
MSMA - broadcast	2 kg	463 g/kg	1 ha	926 g
Chicken manure	10,000 kg	25 mg/kg*	1 ha	250 g
7-3-7 fertiliser	300 kg	80 mg/kg*	1 ha	24 g
20 cm of irrigation	2,000,000 kg	5 μg/kg*	1 ha	10 g
MSMA - spot	2 kg	463 g/kg	0.01 ha	9.3 g
Diammonium phosphate	100 kg	4 mg/kg*	1 ha	0.4 g

* Concentration will vary depending upon source.

Arsenic

The phrase "the dose makes the poison" certainly applies to arsenic. Arsenic could be considered an essential element for adult humans (Nielsen 2001). Major food sources of arsenic include fish, grain and cereal products and therefore the weekly NZ adult consumption in food is approximately 10 μg/kg of body weight. Of course a high dose (1 to 3 g/kg) of pure arsenic can result in death (Pais and Jones 1997). Some view natural arsenic compounds as "contaminants" when found in volcanic thermal pools, oceans, plants and in soil even though arsenic is the 20th most abundant element in Earth's crust and the 12th most abundant element in the biosphere. Arsenic usually exists as an inorganic compound that contains oxygen, chlorine or sulfur. Inorganic arsenic compounds are typically more toxic than organic forms. For example, when using rats, the lethal dose (LD_{50}) for MSMA is 2,449 mg/kg while that for As_2O_5 is 763 mg/kg.

There are environmental concerns over the application of pesticides that contain arsenic. For example, thousands of arsenic-dip sites exist in New Zealand-Australia and soil adjacent to these sites might contain an average of 290 mg/kg of arsenic (Smith et al. 2003). One sheep dip site in Canterbury, contained 4,390 mg/kg of arsenic (EC 2003). MSMA contains 46 percent arsenic and some are concerned about its use in forestry (Morrissey *et al.* 2007). Regarding natural and organic forms of arsenic, the U.S. Environmental Protection Agency (USEPA 2000) reports that:

> *Arsenic is released to the environment from a variety of natural and anthropogenic sources. In the environment, arsenic occurs in rocks, soil, water, air, and in biota. Average concentrations in the earth's crust reportedly range from 1.5 to 5 mg/kg (Cullen and Reimer 1989). Higher concentrations are found in some igneous and sedimentary rocks, particularly in iron and manganese ores (Welch et al. 1988). In addition, a variety of common minerals contain arsenic, of which the most important are arsenopyrite (FeAsS), realgar (AsS), and orpiment (As_2S_3). Natural concentrations of arsenic in soil typically range from 0.1 to 40 mg/kg, with an average concentration of 5 to 6 mg/kg (NAS 1977). Through erosion, dissolution, and weathering, arsenic can be released to ground water or surface water.*
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> *Geothermal waters can be sources of arsenic in ground water, particularly in the Western United States (Nimick et al. 1998, Welch et al. 1988). Other natural sources include volcanism and forest fires. Anthropogenic sources of arsenic relate to its use in the lumber, agriculture, livestock, and general industries. Most agricultural uses of arsenic are banned in the United States. However, organic arsenic is a constituent of the organic herbicides monosodium methanearsonate (MSMA) and disodium methanearsonate (DSMA), which are currently applied to cotton fields as herbicides (Jordan et al. 1997). Organic arsenic is also a constituent of feed additives for poultry and swine, and appears to concentrate in the resultant animal wastes (NAS 1977). The potential impact of arsenic in animal wastes used to fertilize crops is uncertain.*
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> *Most of the arsenic used in the United States is for the production of chromated copper arsenate (CCA), the wood preservative (Reese 1998). CCA is used to pressure treat lumber and is classified as a restricted use pesticide by the USEPA. A significant industrial use of arsenic is the production of lead-acid batteries, while small amounts of very pure arsenic metal are used to produce the semiconductor crystalline gallium arsenide, which is used in computers and other electronic applications.*

Soil, irrigation water, fertiliser, chicken manure and MSMA typically contain arsenic (Table 5). When soils are treated with 2 kg/ha of MSMA, the organic arsenic in the soil initially will be increased by 962 g/ha. If this amount were completely degraded into the inorganic form, then a hectare of soil (15 cm deep), would increase the arsenic content by approximately 0.5 mg/kg. However, over time, the level of arsenic in the topsoil can decline due to gaseous losses. Volatile organic arsenic compounds can be formed by soil microbes and plants and emitted into the atmosphere (Bentley and Chasteen 2002; He *et al.* 2005). As a result, in some cases the arsenic level in the soil three months after applying 2.24 kg/ha of MSMA is no different from that just prior to herbicide application (Zandstra and De Kryger 2007). Losses due to volatile compounds may explain why some researchers could not account for 40- to 60 percent of the applied arsenic (Hiltbold 1975).

At some nurseries, irrigation water is also a source of arsenic. It is estimated that about 3 percent of surface water sources in the U.S. contain more than 5 μg/L of inorganic arsenic (USEPA 2000; Ayotte *et al.* 2003). Groundwater sources typically have higher levels of arsenic than surface waters. When contamination from sheep dip chemicals occurs, groundwater might contain 150 to 2,420 μg/L of arsenic (EC 2003). The maximum acceptable amount of arsenic in drinking water in New Zealand is 10μg/L and most groundwater sources are below this level. However, some well water samples from Waikuku, Saltwater Creek, and Kaikoura contain more than 10 μg/L of arsenic. If well-water contains 5 μg/L, then 2 cm of irrigation would add 1 g of inorganic arsenic per ha. This management practice would add the same amount of arsenic as treating an 11 m^2 nutsedge patch with MSMA at a rate of 2 kg/ha. The main difference being the 1 g added by irrigation would be in the more toxic, inorganic form while the 1 g added by the spot application of MSMA would be in the less toxic, organic form.

Some nursery managers apply poultry manure to the soil in order to increase the organic matter content (Davis *et al.* 2006). Since organic arsenic (ie. roxarsone) is added to poultry feed to control intestinal parasites (thereby improving feeding efficiency and weight gain per unit of feed), arsenic ends up in the chicken manure. Studies have shown arsenic concentrations in poultry manure from some farms can range from 15 to 35 mg/kg (Morrison 1969). Therefore, if a nursery manager applied 10,000 kg/ha of chicken manure to the soil, the amount of arsenic could range from 150 to 350 g/ha. In contrast, some types of inorganic fertilisers may contain 100 mg of arsenic per kg. When 300 kg/ha of fertiliser are applied to seedbeds, the arsenic applied might equal 30 g/ha.

Weed management with MSMA in pine nurseries

At some nurseries, applying a spot application of MSMA to a few sedge patches might add less arsenic to a nursery than a year's worth of irrigation. The herbicide application might add less than the practice of adding chicken manure to increase soil organic matter (Table 5). Since more effective non-selective herbicides can be applied to effectively control perennial weeds on riserlines, fencerows and on ditch banks, there is no need to use MSMA to control weeds in fallow land. Likewise, there is no need to use MSMA to control grasses since several selective herbicides are effective on both annual and perennial grasses. However, sedges are troublesome nursery weeds and they are difficult to control using herbicides that are safe to spray over young pine seedlings. A few diphenylether herbicides have some activity on sedges (e.g. oxyfluorfen and fomesafen), but they typically do not kill the shoots. MSMA might prove useful to managers when patches of *Cyperus spp.* become established in pine seedbeds. However, some nursery managers may not be able to use MSMA due to registration restrictions or to certain forest certification programs.

The Forest Stewardship Council (FSC) is an international non-profit organization created to support environmentally appropriate, socially beneficial, and economically viable management of the world's forests and plantations. FSC has developed a list of herbicides that may not be used (FSC 2005). MSMA is on this list and therefore plantation owners seeking FSC certification might not be allowed to obtain seedlings from nurseries that use spot applications of MSMA. Therefore, plantation managers desiring FSC certification might choose to purchase seedlings that are produced using herbicides that are allowed such as EPTC and glyphosate.

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