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Research Article

Application Timing of Premixed Formulation of Atrazine and Mesotrione in No-Tillage Corn Grown in Sandy Land

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Abstract

Background and Objective: Soil tillage and weed management are two factors that can be manipulated to increase the yield of corn. The objectives of this experiment were to study the application timing of atrazine and mesotrione mixture for weed control in no-tillage corn. **Materials and Methods:** The field experiment was organized in a split-plot design with 2 factors, the main plot was land preparation including no-tillage using glyphosate for land clearing, no-tillage using paraquat and conventional tillage, the sub-plot was the application timing of premixed formulation of atrazine and mesotrione which was 2, 3, 4 and 5 weeks after planting (WAP). **Results:** Using glyphosate is an appropriate measure for land clearing in the no-tillage system. The premixed formulation of atrazine and mesotrione applied at 2 and 3 WAP performed the highest efficacy of 95-100%. However, weeds were regrowth faster when the application was carried out at very early postemergence (2 WAP) indicated with the heaviest weeds biomass. The best growth and the highest yield of corn were found when the application time of the atrazine and mesotrione mixture was at 2 and 3 WAP. **Conclusion:** Glyphosate is an appropriate measure in no-tillage corn and the application time of 2 or 3 WAP of the atrazine and mesotrione showed the highest efficacy on weeds, the better growth and the highest yield of corn.

Key words: Weed control, no-tillage, postemergence, premixed herbicide formulation, sandy land

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Corn is an important commodity in agriculture because it functions like food, feed, fuel and fiber¹. Based on production per country, Indonesia is the 8th rank of corn producer in the world. However, maize production in Indonesia fluctuates due to various things such as global climate change, genetic resources and cultivation techniques². Various efforts can be made to push corn production to fulfill domestic needs, such as intensification by improving inputs of corn cultivation and extensification by an expansion of planting areas³.

One alternative that can be taken in the corn extensification program is to use marginal sandy coastal land for farming^{4,5}. Considering that Indonesia is an archipelagic country with 60% of its area in the form of ocean, there is a very wide availability of sandy land located around the coast that has the potential to be managed as agricultural land⁴. Various limiting factors that may occur are low soil quality such as the physical properties of land with a sandy texture that has low water and soil moisture-holding capacity, high evaporation, high salt content, low organic matter content and poor nitrogen and phosphorus content⁶. The problem in terms of cultivation techniques is that weeds grow fast and become the main competitors in cultivation areas. According to Oerke⁷, maize yield losses due to competition with weeds can reach 56%, while Gharde *et al.*⁸ reported that yield losses of corn caused by weeds in India reached 65%.

Soil tillage aims to condition the land to suit what plants need for root and growth development⁹. Land preparation techniques are classified as perfect tillage or conventional tillage, minimum tillage and no-tillage¹⁰. The soil tillage aims to kill weeds and loosen the soil so that aeration and soil infiltration capacity increase^{9,11}. However, for sandy land, intensive tillage can lead to a decrease in the water holding capacity and the potential for erosion of the land surface⁴. To prevent the adverse effects of conventional tillage, minimum or no-tillage is carried out, while weeds can be killed with herbicides^{10,12}. The most commonly used herbicides for no-tillage are glyphosate N-(phosphonomethyl) glycine and paraquat (paraquat dichloride)¹³. Glyphosate is a broad-spectrum systemic herbicide. The mechanism of action is inhibiting the Enzyme 5-enolpyruvylshikimate-3-phosphate Synthase (EPSPS) which plays a role in the biosynthesis of 3 aromatic amino acids namely tryptophan, tyrosine and phenylalanine. The symptoms and weed mortality caused were rather slow, usually more than 2 weeks after application (WAP) but the persistence was longer. Paraquat is a contact herbicide that damages plant tissue by killing the weeds that are exposed to the application. Symptoms appear faster in 3-4 days after application but the disadvantage is that weeds recover faster because

vegetative parts such as rhizomes or stolon that were not exposed to the herbicide will grow back¹⁴.

In maize cultivation with minimum or no tillage, the method of weed control is preferred with selective postemergence herbicides¹¹. A premixture packaged formulation of atrazine and mesotrione with a ratio of (500:50) g/L is recommended for postemergence herbicides in corn^{15,16}. Atrazine is one of the herbicides in the triazine group which is effective in controlling grass and broadleaf weeds in corn^{14,17}. The mechanism of action of atrazine is to inhibit electron transport in photosystem II. Mesotrione is a new organic herbicide in the triketone group and is effective in mixing with atrazine because it can control weed species that are resistant to triazine herbicides¹⁸. The mechanism of action of mesotrione is to inhibit the enzyme of p-hydroxyphenylpyruvate dehydrogenase (HPPD) which functions in the biosynthesis of carotenoid pigments so that photosynthetic metabolism will be inhibited and cause symptoms of bleaching in the leaves¹⁹. This herbicide is selective in maize and effective for grass and broadleaf weeds¹⁹⁻²¹. The effectiveness of the postemergence herbicide is highly dependent on the time of application²²⁻²⁴. The application timing of postemergence herbicide in related to crop growth stages is a crucial factor to maximize the efficacy of herbicide on weeds and minimize crop injuries²⁴⁻²⁷.

The experiment was aimed to find the best combination of weed management in corn cultivation with no-tillage in coastal sandy land with the application time of the premixed formulation herbicide of atrazine and mesotrione for postemergence weed control.

MATERIALS AND METHODS

Study area: The research was conducted on a sandy coastal land around Bengkulu University, Indonesia, with a geographical position of 101°01'-103°46' east longitude and 2°16'-2°16'-5°31' south latitude and an altitude of 5-10 m above sea level, starting from June until December, 2020. The land is coarse soil and has been used for crop cultivation.

Vegetation analysis was carried out before starting the research following the method of Simarmata *et al.*²⁸. A total of 10 squared plots measuring 0.5 × 0.5 m of each were randomly assigned to the research area. Then, the density of weeds for each species and the frequency of weed appearances were counted based on the number of weed occurrences in the 10 squared plots. The summed dominance ratio (SDR) for each weed species was determined by the sum of the relative density (RD) and relative frequency (RF) divided by two Eq. 1:

$$SDR = \frac{RD + RF}{2} \times 100$$

Table 1: Application time of atrazine and mesotrione mixture and corn life stages

Herbicide application times	Number of leaves	Plant height (cm)
2 WAP/very early postemergence	3.80	27.69
3 WAP/early postemergence	6.34	57.85
4 WAP/mid postemergence	8.00	92.69
5 WAP/late postemergence	9.15	124.64

WAP: Week after planting

Experimental design: The experiment was arranged in a split-plot design with two treatment factors. The main plot consists of 3 ways of land preparation, namely no-tillage with the application of the herbicide glyphosate for land clearing (NTG), no-tillage with the application of paraquat herbicide for land clearing (NTP) and 2 times tillages or conventional tillage (CT). Four treatments of the sub-plot were four application timing of post-emergence herbicide of a premixed herbicide formulation of atrazine and mesotrione at a very early postemergence, early postemergence, mid postemergence and late postemergence of 2, 3, 4 and 5 weeks after planting (WAP), respectively (Table 1). The study consisted of 3 blocks as replications. Each block is further divided into 3 main plots and each main plot is divided into four sub-plots each measuring $(3 \times 2) \text{ m}^2$. The treatment of no-tillage using glyphosate and paraquat was applied on the main plot at a dose of 3 and 2 L ha^{-1} , respectively. The spray volume of 200 L ha^{-1} was calibrated before herbicide application based on the required spray volume for each unit plot using a blue flat nozzle at a pressure of 15 psi.

Two weeks after land clearing using glyphosate and paraquat, the dried weeds were laid down by rolling the banana stems repeatedly. Then the sub-plots were bounded with plastic ropes. Two seeds of corn *var.* Pioneer-23 were planted in a planting hole with 3 cm depth and spacing of $(75 \times 20) \text{ cm}^2$. Carbofuran insecticide was given to prevent the insects before covering the hole with soil. Irrigation was carried out at the beginning of planting until 2 WAP. The intensity of watering was once a day using a water pump machine and a hose to spray water to the planting area. Thinning was done at 2 WAP by selecting one vigorous plant in one planting hole. Fertilization was carried out at the time of planting with urea, KCl and TSP fertilizers of 100, 100 and 150 kg ha^{-1} , respectively. Fertilization of the urea fertilizer was repeated with the same dose at 5 WAP. Fertilizers were given in holes at a distance of 10 cm from the planting hole. Pest and disease control were carried out if a significant attack or symptoms of disease occurred. Weed control was carried out according to the treatment applied using the premixed formulation of atrazine and

mesotrione at 2 kg a.i./ha . The spray volume of 200 L ha^{-1} was mixed with 1% (v/v) of a non-ionic surfactant, calibrated before being applied as previously described. The application was carried out during the day so that it was easily absorbed and sticks to the weed leaves. Corn harvesting was done when the plants reach the age of 110 days after planting, which was characterized by 95% of the leaves turning yellow or dry, the cob hairs turning black and the corn kernels hardened.

Data collected included the efficacy of herbicide control on weeds and the injuries on corn plants at 2 and 3 weeks after treatment (WAT). Observations were made visually by looking at the colour and shape changes in the weeds and corn plants. Data of efficacy and injury were counted in percent²⁸. At the end of the study, weeds were harvested from 2 squared plots taken from each plot measuring $(0.5 \times 0.5) \text{ m}^2$. The weeds within the square plots were cut and oven-dried for $3 \times 24 \text{ hrs}$ at 70°C . Data of plant growth and yield parameters included plant height, the number of leaves, stalk diameter, leaf area, tasseling time, cob dry weight, cob length, cob diameter and kernels dry weight.

Statistical analysis: Data were statistically analyzed by ANOVA at a 95% level of significance. If there is a significant effect, then the means of data were separated by the least significant test (LSD) at 5%. Data on herbicide efficacy and crop injuries were only analyzed descriptively.

RESULTS

The assessment of weeds in the research area calculated as the summed dominance ratio (SDR) was presented in Table 2. *Imperata cylindrica* (cogon grass) is the dominant weed with an SDR of 68.29%. Other weeds in the research area were *Digitaria ciliaris*, *Crassocephalum crepioides*, *Asystasia intrusa*, *Eleusine indica* and *Phyllanthus niruri* with SDRs of 8.05, 7.46, 5.92, 5.23 and 5.04%, respectively. The six weed species observed were grass and broadleaf groups for 3 species of each group.

Efficacy of postemergence herbicide: Tables 3 and 4 showed the herbicide efficacy and crop injuries, respectively, at 2 and 3 WAT. The highest efficacy was found when the mixture of atrazine and mesotrione was applied at 2 WAP or a very early postemergence, then the efficacy decreased by delaying the application time and the lowest efficacy was found when the herbicide application was carried out at 5 WAP or late postemergence. The maximum efficacy was observed at 3 weeks after treatment (WAT). Overall, the efficacy was

Table 2: Weed assessment on the experiment field before land preparation

Weed species	Weed groups	Frequency	Relative frequency	Density	Relative density	SDR (%)
<i>Imperata cylindrica</i>	Grasses weeds	9	0.43	1,493	0.94	68.29
<i>Digitaria ciliaris</i>	Grasses weeds	3	0.14	29	0.02	8.05
<i>Crassocephalum crepidioides</i>	Broadleaf weeds	3	0.14	10	0.01	7.46
<i>Asystasia intrusa</i>	Broadleaf weeds	2	0.10	37	0.02	5.92
<i>Eleusine indica</i>	Grasses weeds	2	0.10	15	0.01	5.23
<i>Phyllanthus niruri</i>	Broadleaf weeds	2	0.10	9	0.01	5.04
Total	-	21	1.00	1,593	1.00	100.00

Table 3: Weed efficacy due to the different application times of atrazine and mesotrione mixture on weeds

Herbicide application time	Weed efficacy (%)		Dry weight of weed biomass (g m ⁻²)
	2 WAT	3 WAT	
2 WAP/very early postemergence	75.8	100	573.87 ^a
3 WAP/early postemergence	65.4	95	554.88 ^{ab}
4 WAP/mid postemergence	60.0	94	525.10 ^b
5 WAP/late postemergence	60.9	90	448.76 ^c
LSD (5%)	ns	ns	*

WAT: Weeks after treatment, WAP: Weeks after planting, ns: Non-significance and *Numbers followed by different letters are significantly different at LSD test 5%

Table 4: Corn injuries due to the different application times of atrazine and mesotrione mixture

Herbicide application times	Crop injury (%)	
	2 WAT	3 WAT
2 WAP/very early postemergence	20.0	0
3 WAP/early postemergence	9.0	0
4 WAP/mid postemergence	5.0	0
5 WAP/late postemergence	0	0
LSD (5%)	ns	ns

ns: Non-significance

60.0-75.8 after 2 weeks of herbicide application and the efficacy increased to 90-100% after 3 weeks of herbicide application. The total dry weight of weeds harvested at the same time as the harvesting crop yields was also shown in Table 3. The highest weed biomass was harvested when a herbicide mixture of atrazine and mesotrione was applied at 2 WAP or very early postemergence, the biomass of weeds decreased with delaying the application time of postemergence herbicide and the lowest weed biomass was harvested when herbicide application time was carried out as late postemergence or 5 WAP.

Postemergence application times of weed control using a herbicide mixture of atrazine and mesotrione caused an injury to corn plants which was observed at 2 WAT (Table 4). A mild toxicity level of 5-20% injury was found when atrazine and mesotrione mixture was applied at the very early postemergence (2 WAP), decreasing to a very low toxicity level of <5% injury in the application of early and mid-postemergence application or 3 and 4 WAP and there was no toxicity if the application timing was late postemergence or 5 WAP. The corn plants were recovering from herbicide injuries or growing normal after 3 weeks of herbicide application.

Effect of no-tillage systems on corn: Statistical analysis showed that there was no interaction between the combination of tillage systems and the application time of atrazine and mesotrione mixture on the growth and yield of maize. However, the tillage systems showed a significant effect on plant height, leaf area, cob dry weight, cob length and kernel dry weight (Table 5). The data indicated that a no-tillage system using glyphosate for land clearing was an appropriate technology in corn cultivation on sandy land compared to using paraquat. This was indicated by the positive impacts of plants on the growth and yield parameters. In general, the no-tillage using glyphosate was not significantly different from that of conventional tillage. Yields of kernels dry weight obtained with no-tillage with glyphosate (NTG), no-tillage with paraquat (NTP) and conventional tillage (CT) were 89.65, 74.74 and 94.29 g/plant, respectively, where the NTG was not significantly different from CT.

Effect of the application time of postemergence herbicide on corn: The application time of premixed atrazine and mesotrione for weed control showed a significant influence on some parameters of corn growth and yield including leaf area, cob dry weight and kernels dry weight (Table 6). The better growth of corn was observed when the application time of herbicide was carried out at the very early and early postemergence (2 and 3 WAP), then the growth parameters decreased when the weed control was delayed to mid and late postemergence (4 and 5 WAP). The heaviest yield of dry kernels was obtained when weed control was conducted at the early postemergence or 3 WAP, which was 89.13 g/plants and the yield decreased if weed control was delayed and the

Table 5: Effect of land preparation on the growth and yield of corn

Land preparation	Plant height (cm)	Number of leaves	Stalk-diameter (cm)	Leaf area (cm ² /plant)	Tasseling time (days)	Cob dry weight (g)	Cob length (cm)	Cob diameter (cm)	Kernels dry weight (g/plant)
NTG	187.94 ^a	10.42	2.71	570.72 ^a	46.5	165.39 ^a	16.80 ^a	5.03	89.65 ^a
NTP	154.27 ^b	9.57	2.54	522.32 ^b	47.8	126.78 ^b	14.55 ^b	4.88	74.74 ^b
CT	182.30 ^a	10.41	2.62	550.18 ^{ab}	45.8	174.32 ^a	17.25 ^a	5.44	94.29 ^a
LSD (5%)	*	ns	ns	*	ns	*	*	ns	*

NTG: No-tillage with glyphosate application, NTP: No-tillage with paraquat application, CT: Conventional tillage, ns: Non-significance and *Numbers followed by different letters are significantly different at LSD test 5%

Table 6: Effect of the application time of atrazine and mesotrione mixture on the growth and yield of corn

Herbicide application time	Plant height (cm)	Number of leaves	Stalk-diameter (cm)	Leaf area (cm ² /plant)	Tasseling time (days)	Cob dry weight (g)	Cob length (cm)	Cob diameter (cm)	Kernels dry weight (g/plant)
2 WAP	178.64	10.22	2.65	571.36 ^a	46.2	161.61 ^a	16.25	5.17	89.13 ^a
3 WAP	178.42	10.35	2.63	566.52 ^a	46.0	163.35 ^a	16.26	5.07	88.09 ^a
4 WAP	173.83	9.94	2.61	522.96 ^b	46.9	152.92 ^b	16.26	5.20	86.06 ^a
5 WAP	168.46	10.32	2.61	530.11 ^b	47.2	144.12 ^c	16.03	5.02	81.64 ^b
LSD (5%)	ns	ns	ns	*	ns	*	ns	ns	*

WAP: Week after planting, ns: Non-significance and *Numbers followed by different letters are significantly different at LSD test 5%

lowest kernel yield was found when the weed control was done at the late postemergence (5 WAP), which was 81.64 g/plant.

DISCUSSION

Imperata cylindrica (cogon grass) is the dominant weed in the research area. Cogon grass is one of the noxious weeds in the Poaceae family that is widespread in tropical areas. It has high adaptability to a stressful and nutrient-poor environment²⁸. In some areas of Indonesia with conventional agriculture, intensive land management with perfect tillages is a common practice for the land preparation of crop cultivation. Soil tillage is aimed not only to optimize the physical properties of the soil for plant root growth but also to kill weeds⁹. In sandy soil conditions that are coarse, the soil is possible for no-tillage cultivation and weeds are cleaned by using herbicides^{12,25,29}. Dead weeds remain on the land as mulches to cover the soil surface that may serve to withstand rain and maintain soil friability, inhibit surface erosion and reduce soil evaporation. These mulches also have the benefits to improve soil nutrition as well as biological benefits that may increase soil microbial development^{5,6}. The degradable mulches of dry weeds will be favourable for crops, thus providing good growth and increased yields. Results showed that conventional tillage was still the best in terms of corn yield. Of two herbicides used for land clearing of no-tillage treatments, the use of glyphosate is better than paraquat. One of the possible reasons for better glyphosate performance is that glyphosate is a broad spectrum and systemic herbicide so weed death is more total than the contact paraquat treatment^{11,30,31}. The herbicide paraquat killed weeds quickly but the vegetative parts of perennial weeds, such as *Imperata cylindrica*, that were not exposed to herbicide will

grow rapidly^{13,14}. If this condition occurs, the no-tillage treatment with paraquat did not provide optimal benefits to crops.

The application time of weed control using a premixed formulation of atrazine and mesotrione showed a significantly different efficacy on weeds. The highest efficacy was found when the application time was conducted at a very early postemergence (2 WAP). In this treatment, the growth stage of corn has not covered yet the planting area so that the targeted spraying on the weed surface can be reached properly³². On the other hand, a light injury of corn was observed if the application time was conducted at a very early postemergence (2 and 3 WAP) because the corn height was still lower than the height of the nozzle spray. However, the crops were recovered from herbicide injuries after 3 weeks of herbicide application. The corn injuries were not found during the late postemergence because the height of the corn was reaching 124 cm, so the herbicide droplets could not reach the leaf surface. The symptoms of injury were indicated by slowing of growth and changes in leaf colour to white (bleaching). This was caused by the mechanism of action of mesotrione which inhibits the HPPD enzyme in the biosynthesis of carotenoid pigments^{19,33}. Similar symptoms were also reported in the sorghum plants^{23,24}. Corn plants were recovered lately from the symptoms of injuries. The ability of corn plants to recover from bleaching symptoms was also stated in previous studies that described the injury of maize plants due to the application of premixed herbicide atrazine and mesotrione immediately recovered a few weeks after application^{21,32}.

When the application time of the herbicide mixture was conducted at the very early postemergence (2 WAP), the highest weed biomass was harvested from the cornfield. The biomass of weeds decreased with delaying the application

time of postemergence herbicide and the lowest weed biomass was harvested at a late postemergence or 5 WAP. In the early weed control, weed seeds grew quickly and the grace period is sufficient for weed development until the end of the season^{20,22,23}. On the other hand, the application time of herbicide for weed control lately at 5 WAP will limit the time for the emergence and development of weeds.

Delaying weed control caused a longer time for weeds to grow and compete with corn plants³⁴. Weeds are a strong competitor for plants to obtain nutrients, growing space and sunlight¹³. A critical period for weed control of crops is a period in which crops are very sensitive to weeds. During this period, the presence of weeds can damage crop growth and reduce yields³⁵. Weed control at the very early and early postemergence effectively controlled weeds during the critical periods and prevent weeds-crops competition. These circumstances will bring benefits for corn plants because the nutrients and other growth requirements were available to the crops without any competition from weeds, thus affected to the maximal growth and yield of crops^{35,36}. The impacts were found in the significant increase of corn yields harvested of 89.13 and 88.09 g/plant when the herbicide application time was carried out at 2 and 3 WAP, respectively.

CONCLUSION

Glyphosate is an appropriate measure to be used in no-tillage corn grown on sandy land as indicated by the growth and yield of corn plants that were not significantly different from the conventional tillage. The premixed formulation of atrazine and mesotrione applied 2 or 3 WAP or very early or early postemergence is an appropriate measure for postemergence weed control in no-tillage corn grown in sandy land and performed a persistent highest efficacy of weed control. Even though it caused mild injuries, the corn plants recovered to normal growth in several weeks. Better growth and the highest dry kernels yield of corn were produced when the premixed formulation of atrazine and mesotrione was carried out 2 or 3 WAP.

SIGNIFICANCE STATEMENT

This research provides important information when the extensification of the corn cultivation program extends to the coarse sandy coastal lands. A no-tillage system can be adopted by using glyphosate herbicide for land clearing because it is not only a broad spectrum and systemic herbicide that persistent killed weeds but also is environmentally safe. Chemical weed control using the

premixure formulation of atrazine and mesotrione applied at an early postemergence is recommended to maintain the advantages for coarse soil.

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