Original Research

Tank-mix application of p-hydroxyphenylpyruvate dioxygenase (HPPD) inhibiting herbicide (mesotrione, tembotrione or topramezone) with atrazine improves weed control in maize (Zea mays L.)

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FIELD experiments were conducted to identify the effective tank-mix combinations of HPPD (p-hydroxyphenylpyruvate dioxygenase) herbicides (Mesotrione, tembotrione and topramezone) with atrazine for post-emergence grass and broadleaf weeds control in maize crop during the three kharif seasons of 2013 to 2015. The dominant weeds infested the experimental plots were crow footgrass (Dactyloctenium aegyptium (L.) Willd.); large crabgrass, Digitaria sanguinalis (L.) Scop; barnyard grass, Echinochloa crus-galli (L.) Beauv.; Digera arvensis Forsk. and Horse purslane (Trianthema portulacastrum L.). All the three HPPD herbicides showed good crop safety and among them, for grass weed control, mesotrione 120 g ha⁻¹ applied alone was inferior to topramezone 50 g ha⁻¹ and tembotrione 120 g ha⁻¹ applications. However, reduced doses of the HPPD herbicide (mesotrione 90 g or topramezone 37.5 g or tembotrione 90 g ha⁻¹) with atrazine 900 g ha⁻¹ as post-emergent tank-mixture gave better weed control and maize yield than their solo applications. The uncontrolled weed competition reduced the maize yields by 31.5 to 68.5%. Overall, topramezone + atrazine provided comparable or superior control of annual grass and broad-leaf weeds than mesotrione + atrazine or tembotrione + atrazine. Post applied (15-18 days after sowing) atrazine 1000 g ha⁻¹ was better to pre-emergence atrazine in weed control and producing maize yield. In another field study, replacement of maize with puddle rice completely reduced the Johnsongrass {Sorghum halepense (L.) Pers.} density and therefore, in areas having its infestation, rice can be an alternative to contain its problem. The synergistic interactions between HPPD-inhibiting herbicides, and atrazine, were also observed against three grass weeds in pot experimentation as higher weed control achieved than what was expected from Colby’s equation. The results show that tank-mixture of topramezone or tembotrione with atrazine can be effectively used for diverse weed flora control in maize.

KEYWORDS

Atrazine
Grass weeds
HPPD herbicides
Johnsongrass
Tank-mix combinations

Introduction

Maize (Zea mays L.), the “Queen of Cereals” is grown under varied agro-climatic conditions in different countries, due to its wider adaptability. Globally, it occupies nearly 193.7 m ha area and

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contributes about 38.7% (1147.6 m t) in the total cereal grain production (FAO 2020). In view of the emerging water shortage in South Asia, maize seems to be a better alternative for the widely adopted water exhaustive rice crop. Although, maize has a high yield potential ranging 10-20 t ha\(^{-1}\) (Otegui et al. 1995; Dobermann et al. 2003; Liu et al. 2017), but its average productivity is low (5.75 t ha\(^{-1}\) globally and 3.11 t ha\(^{-1}\) in India). These yield differences are governed by genetics, environmental factors and management practices interactions. Among agronomic management practices, efficient weed management plays an important role in achieving potential yields under different environments. This has been shown by Oreke et al. (2006) that in spite of weed control measures, still globally, weeds caused an average 10.5% yield losses in maize and in absence of weed control measures, the worldwide potential average yield losses were 40.3%. Similarly, other research workers (Massing et al. 2003; Idziak and Woznica, 2014; Chhokar et al. 2019) have also reported weeds in maize cause huge grain yield losses ranging from 30 to 90% depending on the type of weed flora and their intensities. Some of the weeds like barnyard grass \(\textit{Echinochloa crus-galli}\) (L.) Beauv., horse-purslane \(\textit{Trianthema portulacastrum}\) L. and crow footgrass \(\textit{Dactyloctenium aegyptium}\) (L.) Willd., are highly competitive in rainy season maize in India (Chhokar et al. 2019). The wide spacing and slow initial growth of maize make it more vulnerable to weed competition. Hence, for realizing optimum maize yield, efficient weed control is a must.

In rainy season, chemical weed control is a boon, due to non-feasibility of mechanical interculture, as well as, scarce and costly manual labour. For weed control in maize, photosynthesis (PS-II) inhibitor \textit{i.e.} atrazine is widely used because of its low cost, broad-spectrum weed control, application time flexibility (pre or post-emergence) and tank mix compatibility with different herbicides (Walsh et al. 2012; Chhokar et al. 2019). Nevertheless, atrazine is widely used as pre-emergence and its performance is variable depending on soil type, moisture and weed flora. The continuous use of atrazine is also resulting in weed flora shift and increased cases of herbicide resistance, as triazine herbicides with continuous use are more vulnerable to resistance evolution. Globally, 45 weed species across many corn growing areas have exhibited resistance against photosystem II (PSII) inhibiting herbicides, like atrazine (Heap, 2020). Therefore, alternative mechanisms of action herbicides in maize are needed to decrease the probability of herbicide resistance evolution and weed flora shift. The HPPD (p-hydroxyphenylpyruvate dioxygenase) enzyme inhibitor herbicides (mesotrione, topramezone and tembotrione) as post-emergence options have been recently made available to maize growers and they provide an opportunity to control weeds having resistance to glyphosate, triazine and ALS inhibiting herbicides (Sutton et al. 2002; Bollman et al. 2006; Vyn et al. 2006; Woodyard et al. 2009b; Walsh et al. 2012; Kumar and Jha, 2015; Kohrt and Sprague, 2017a). Herbicides of HPPD group are being preferred because of their broad-spectrum weed control, flexible application timing, tank-mix compatibilities, and better crop safety (Bollman et al. 2008; Walsh et al. 2012).
As, the dependence on the single herbicide may result in the escape of many weed flora, therefore, to tackle this issue, two or more herbicides either in mixture or in sequence having different mechanisms of action are desirable. Ready mixture or tank-mixing of two or more herbicides, a common practice is ideal due to wide spectrum weed control, reduction in application costs, and/or prevention and delay of evolution of herbicide-resistant weeds (Zhang et al. 1995; Damalas, 2004), as the mixture has less chances of resistance evolution compared to sequential usage (Diggle et al. 2003). Moreover, it will be better, if herbicide mixture show synergism, which further provides an opportunity to reduce the herbicide doses. Some of the workers have reported synergism between the mixtures of HPPD (mesotrione) and photosystem-II inhibitors herbicides (Sutton et al. 2002; Abendroth et al. 2006; Bollman et al. 2008; Chhokar et al. 2019). So, the combinations of different HPPD herbicides and atrazine should be explored for effective management of prevalent weed flora in maize. To determine the herbicide interactions, Colby’s method (Colby, 1967) can be used, as it calculates an expected weed control value for a mixture on the basis of the control achieved with the individual herbicide applied alone. Since, under South Asian conditions, there is paucity of information on comparative performance of these HPPD herbicides alone or in mixture with atrazine against prevalent weed flora in maize. Therefore, the present studies were undertaken with the aim to determine the comparative effectiveness of atrazine and HPPD herbicides (mesotrione, topramezone and tembotrione) applied alone or in tank-mix combinations at reduced doses against weeds in maize under field and pot trials.

Materials and Methods

Field and pot experiments were conducted at ICAR Indian Institute of Wheat and Barley Research (29° 43’ North, 76° 58’ East and an altitude of 245 above mean sea level), Karnal, India. Field experiments were conducted in a randomized complete block design (RCBD) having three replicates during three kharif seasons of 2013, 2014 and 2015. The experimental soil was sandy clay loam in texture and having pH 7.9-8.2, organic carbon content 0.39-0.43%, low available nitrogen, medium phosphorus and high potassium. The experimental site is having a hot and humid climate during Kharif season with an average annual rainfall of 750 mm and 77% of which is received during June to September.

Evaluation of atrazine and HPPD herbicides alone or in combinations against weeds in maize

Field experiments were conducted during kharif seasons of 2013 and 2014 for evaluation of HPPD (mesotrione, topramezone and tembotrione) herbicides alone or in tank-mix combinations with atrazine for diverse weed flora control in maize. Ten weed control treatments (Table 1) replicated thrice and tested in randomized block design consisted of atrazine 1000 g ha⁻¹ and HPPD herbicides (mesotrione 120 g, topramezone 50 g and tembotrione 120 g ha⁻¹) alone and their reduced doses in combinations along with other treatments comprising of standard herbicide 2,4-D-Na at 1000 g ha⁻¹,
protected spray of non-selective herbicide paraquat 500 g ha\(^{-1}\) between maize rows and untreated weedy check. The reduced doses of herbicides in combinations consisted of 75% of HPPD herbicide’s sole doses and 90% of solo atrazine dose i.e. mesotrione 90 g, topramezone 37.5 g and tembotrione 90 g ha\(^{-1}\) each in combination with 900 g ha\(^{-1}\) atrazine. Among three HPPD herbicides, mesotrione in combination with atrazine was applied using ready-mix combination of atrazine plus mesotrione i.e. Calaris Xtra 275 SC (Mesotrione 2.27% + Atrazine 22.72% w/w) at 1000 (90+909) g a.i. ha\(^{-1}\) in mixture treatment. Cationic surfactant (Leader Mix) at 1000 ml ha\(^{-1}\) was used with HPPD herbicides applied either alone or in combinations with atrazine. Whereas, paraquat was sprayed as protected between maize rows using single flat fan nozzle fitted in a spray hood. Each weed control treatment had a plot size of 3 × 10 m. Maize hybrid ‘DKC 9108’ was sown during last week of June using a seed-cum fertilizer planter fitted with horizontal inclined plate seed metering system. Seed rate of about 20 kg ha\(^{-1}\) was drilled at 5-6 cm depth in 60 cm row to row and about 20 cm plant to plant spacing. Gap filling and thinning were accomplished immediately after germination to maintain uniform plant stand. The crop was fertilized with 150 kg N, 26.2 kg P and 33.3 kg K ha\(^{-1}\) through urea, diammonium phosphate and muriate of potash, respectively. Full doses of phosphorus and potash were applied as basal at the sowing time, whereas, N doses were applied in four equal splits, at sowing, around 4 leaf, knee high and tasseling stages. Post-emergence herbicides were applied 18-20 DAS (days after sowing). All the herbicides, except paraquat, were applied as blanket spray solution of 400 lit ha\(^{-1}\) using backpack sprayer fitted with flat fan nozzles. The dry weights of major individual weed species were recorded by placing a quadrat (50 cm × 50 cm) at two spots in each plot at 3 and 6 WAS (weeks after spray). The above ground portion of weeds collected were first sun dried and then dried to constant weight in an oven at 65 °C, for recording dry weight. Based on weed dry weight, weed control efficiency (WCE) was also calculated. Finally, the cobs harvested from the net plots and after sun drying were shelled using small plot maize sheller for recording the grain yield at 16% moisture after cleaning and winnowing.

**Comparative performance of pre and post-emergence atrazine and its combination with tembotrione and topramezone against weeds**

During 2015, in three fields having larger plots (100 m\(^{2}\) area for each treatment), atrazine at 1000 g ha\(^{-1}\) as pre and post-emergence along with its post-emergence combinations with tembotrione (90 g ha\(^{-1}\)) or topramezone (40 g ha\(^{-1}\)) were evaluated in comparison to untreated weedy control. Atrazine 1000 g ha\(^{-1}\) was applied 0-1 DAS in pre-emergence treatment and post- applied either alone or in combination with tembotrione or topramezone at 18-20 DAS. The method followed for growing of crop, herbicide application and recording observations on weed dry weight and crop yield were similar as mentioned before. The weed and crop observations were recorded at three spots in each plot. Based on the observations from three fields, mean and standard error of the mean were worked out. The mean of
weird dry weight at 3 WAS and maize grain yield are based on 9 observations and weed dry weight at 6 WAS data are based on 6 observations (Figure 1).

*Johnsongrass (Sorghum halepense (L.) Pers.) density as affected by crop sequence*

The effect of two crop sequence (rice-wheat and maize-wheat) on emergence and establishment of Johnsongrass (*S. halepense*) was evaluated during 2014 (two fields) and 2015 (one field). Fields having the infestation of *S. halepense* were selected and divided into two parts. In one part, maize-wheat and in another half, rice-wheat sequence was followed. For sowing maize, cross harrowing and cross cultivator operations were performed at optimum soil moisture conditions. For rice, after cross harrowing and cross cultivator operations, puddling (wet tillage) was performed using rotary tiller in 7-10 cm standing water. At the time of cultivator and harrowing operations, the rhizomes of *Sorghum halepense* present on the soil surface were uniformly spread. During both the years, sowing of maize in maize-wheat sequence field and transplanting of 25 days old rice seeding in rice-wheat sequence were performed between 20 to 25 June, following the standard package of practices for these crops. The transplanting of rice cultivar ‘HKR-47’ seedlings was done at 20 cm row spacing and within row, two seedlings per hill were transplanted at 15 cm spacing using a plastic rope having a 15 cm markings. Both the crops were fertilized with 150 kg N, 26.2 kg P and 33.3 kg K ha$^{-1}$. *S. halepense* panicle densities were recorded from 100 m$^2$ area at 100 days after sowing/transplanting and converted to number ha$^{-1}$. The observations were recorded at three spots in each plot. Based on the observations from three fields, mean and SEM were worked out.

*Pot experiments: Tank-mix interactions of HPPD herbicides with atrazine against grass weed species*

Pot experiments were conducted to determine the comparative efficacy of tank-mix combinations of HPPD herbicides + atrazine. The test species namely crow footgrass (*Dactyloctenium aegyptium* (L.) Willd); barnyard grass, *Echinochloa crus-galli* (L.) Beauv and large crabgrass, *Digitaria sanguinalis* (L.) Scop. were sown in the pots of 4.5 kg soil capacity at a depth of 0.5-1.0 cm. After emergence, 30 plants pot$^{-1}$ of *E. crus-galli* and 40 plants pot$^{-1}$ of *D. aegyptium* and *D. sanguinalis* were maintained for herbicide spraying. In first set of pot studies, herbicides and their rates consisted of atrazine at 500 and 1000 g ha$^{-1}$, mesotrione at 60 and 120 g ha$^{-1}$, topramezone at 25 and 50 g ha$^{-1}$, tembotrione at 55 and 110 g ha$^{-1}$, and their combinations (mesotrione + atrazine at 60 + 500 and 120 + 1000 g ha$^{-1}$; topramezone + atrazine at 25 + 500 and 50 + 1000 g ha$^{-1}$; tembotrione + atrazine at 55 + 500 and 110 + 1000 g ha$^{-1}$) along with untreated control. In second set of pot studies, in addition to 0.5 X and 1.0 X (recommended) herbicide rates, 0.125 X and 0.25 X rates of herbicides alone and in combinations were also included. Both the sets of experiments were run twice with three replications. Cationic surfactant (Leader mix) at 1000 ml...
ha⁻¹ was used with mesotrione, topramezone and tembotrine and their combinations with atrazine. Herbicides were applied 15-18 days after sowing. Fresh biomass pot⁻¹ was recorded 3 WAS and from which, the % biomass reduction compared to control was worked out for determining the herbicide responses. Colby’s method (Colby, 1967) was used to assess herbicide interactions as per equation below.

\[ E = (X + Y) - \frac{XY}{100} \]

Here, \( E \) is the expected level of control of a given weed species when two herbicides are applied as mixture, and variables \( X \) and \( Y \) denotes the observed control level of a weed species provided by individual herbicides. The mixture’s expected and observed values were compared and when the expected value was significantly lesser than the observed value, the mixture was considered synergistic.

Statistical analyses

Statistical Analysis System (SAS version 9.2) software was used for data analyses. The data on the field evaluation of herbicides were statistically analyzed in a simple block design. The pooling of data was not done, since; there were variations in the weed intensity and diversity. The treatments in pot experiments were arranged in a completely randomized design with three replications (pots) for each treatment. The experiments were run twice. Fresh weight data were expressed as a percent reduction compared to the un-treated control. The mixture’s expected (Colby’s method) and observed values were compared, using a two-sided t-test (\( \alpha = 0.05 \)). The mixture was considered synergistic, if the expected value was significantly lesser than the observed value. In addition to the herbicide interactions analysis using Colby’s method, the per cent fresh biomass data were subjected to analyses of variance (ANOVA), separately for each species to compare mixtures with their individual herbicides. Weed and crop data in various experiments were subjected to analyses of variance, and the Fisher’s Protected Least Significant Difference (LSD) was used to detect treatment differences at the \( P = 0.05 \) level. The data on the weed dry weights in field studies were square root \( \sqrt{x+1} \) transformed before subjected to the ANOVA in RCBD. The original weed data are presented in the results tables with a comparison of means for significant differences using superscript letter(s). Also, in larger plots field evaluation trials, SEM was worked out based on the number of observations and for significance comparison of two treatments, Fisher’s t test was used.

Results and Discussion

Evaluation of atrazine and HPPD herbicides applied alone or in combination against weeds in maize

Major weeds infested the experimental plots were crow footgrass, Dactyloctenium aegyptium (L.) Willd.; barnyard grass, Echinochloa crus-galli (L.) Beauv; large crabgrass, Digitaria sanguinalis (L.) Scop;
horse purslane, *Trianthema portulacastrum* L.; *Digera arvensis* Forsk. and *Phyllanthus niruri* L. Among these weeds, the dominant were *E. crus-galli*, *T. portulacastrum*, *D. arvensis* and *D. aegyptium*. Various herbicide treatments significantly (<0.0001) affected the dry weights of major weeds (Tables 1 to 4). The maximum weed dry weight accumulation was observed in untreated weedy check and the values were 454.0 and 284.9 g m⁻² at 3 and 6 WAS, respectively, during first year and 659.1 and 481.8 g m⁻² at 3 and 6 WAS, respectively during second year. Under untreated weedy control at 3 WAS, *D. aegyptium*, *E. crus-galli*, *D. arvensis* and *T. portulacastrum* constituted 19.3, 2.2, 11.0 and 67.0%, respectively, of the total weed dry weight accumulation, during first year and the respective values at 6 WAS were 38.3, 10.8, 19.6 and 29.6%. Whereas, during second year of study, of the total weed dry biomass accumulation, *D. aegyptium*, *E. crus-galli* and *T. portulacastrum* accounted for 13.9, 1.5 and 83.5% at 3 WAS and 31.6, 12.0 and 44.7% at 6 WAS stage, respectively. *T. portulacastrum* dry weight was reduced at 6 WAS compared to 3 WAS due to its maturity because of short life cycle.

Balyan and Bhan (1986) reported *T. portulacastrum* as short duration weed, which starts production of flowers and seeds 20 to 30 DAS. Short duration and fast early growth of *T. portulacastrum* make it more competitive during early crop growth stages. Atrazine at 1000 g ha⁻¹ applied as post-emergence (18-20 DAS) significantly reduced the weed dry weight at both (3 and 6 WAS) the stages (Tables 1 to 4).

**Table 1.** Performance of early post-emergence application of HPPD herbicides alone and in combination with atrazine against weeds in maize at 3 WAS during 2013.

| Herbicide                  | Dose ha⁻¹ (g. a.i.) | E. crusgalli | D. arvensis | D. aegyptium | T. portulacastrum | P. niruri | Other weeds | Total weeds |
|----------------------------|---------------------|--------------|-------------|--------------|-------------------|-----------|-------------|-------------|
| Atrazine                   | 1000                | 0.7 C        | 0.0 C       | 62.9 C       | 0.0 C             | 0.0       | 6.7         | 70.2 C      |
| Topramezone+S‡             | 50                  | 0.1 C        | 0.0 C       | 0.0 C        | 0.0 C             | 0.0       | 1.5         | 3.2 D       |
| Atrazine+Topramezone+S     | 900+37.5            | 0.2 C        | 0.0 C       | 0.0 C        | 0.0 C             | 0.0       | 2.1         | 2.3 D       |
| Mesotrione+S               | 120                 | 0.1 C        | 0.0 C       | 41.8 B       | 0.0 C             | 0.1       | 1.1         | 43.1 C      |
| Atrazine +mesotrione+S     | 900+90              | 0.0 C        | 0.0 C       | 0.2 C        | 0.0 C             | 0.0       | 2.2         | 2.5 D       |
| Tembotrione+S              | 120                 | 0.4 C        | 0.0 C       | 4.0 C        | 0.0 C             | 0.0       | 2.8         | 7.9 D       |
| Atrazine +Tembotrione+S    | 900+90              | 0.0 C        | 0.0 C       | 0.7 C        | 0.0 C             | 0.0       | 0.6         | 1.4 D       |
| Paraquat (Protected spray) | 500                 | 3.8 B        | 16.5 B      | 75.7 A       | 145.1 B           | 0.4       | 3.6         | 245.0 B     |
| Untreated weedy check      | 10.0 A              | 49.8 A       | 87.6 A      | 304.3 A      | 1.9               | 0.4       | 454.0 A     |

† Original values of weed dry weight data were square root transformed \(\sqrt{(x+1)}\) before statistical analysis and based on the analysis of the transformed data letter have been assigned to original values for interpretation. Means with at least one letter common within a column are not statistically significant using Fisher's Least Significant Difference at 5%. ‡ S= Cationic surfactant (Leader Mix) at 1000 ml ha⁻¹.
Table 2. Performance of early post-emergence application of HPPD herbicides alone and in combination with atrazine against weeds in maize 6 WAS during 2013.

| Herbicide                        | Dose ha⁻¹ (g. a.i.) | \(^{1}\text{Weed dry weight g m}^{-2}\) | \(E.\ crusgalli\) | \(D.\ arvensis\) | \(D.\ aegyptium\) | \(T.\ portulacastrum\) | \(P.\ niruri\) | Other weeds | Total weeds |
|----------------------------------|---------------------|----------------------------------------|--------------------|-------------------|-------------------|------------------------|----------------|-------------|------------|
| Atrazine                         | 1000                |                                        | 3.3 \(^{C}\)       | 0.0 \(^{C}\)      | 81.8 \(^{A}\)     | 0.0 \(^{C}\)           | 0.0            | 3.3         | 88.4 \(^{C}\) |
| Topramezone+S\(^{\ddagger}\)    | 50                  |                                        | 4.3 \(^{C}\)       | 0.0 \(^{C}\)      | 1.7 \(^{B}\)      | 0.0 \(^{C}\)           | 0.1            | 6.5         | 12.5 \(^{DE}\) |
| Atrazine+Topramezone+S           | 900+37.5            |                                        | 1.5 \(^{C}\)       | 0.2 \(^{C}\)      | 0.5 \(^{B}\)      | 1.0 \(^{C}\)           | 0.0            | 2.3         | 5.5 \(^{DE}\) |
| Mesotrione +S                    | 120                 |                                        | 8.3 \(^{BC}\)      | 0.0 \(^{C}\)      | 79.3 \(^{A}\)     | 0.0 \(^{C}\)           | 0.5            | 5.7         | 93.3 \(^{C}\) |
| Atrazine + Mesotrione +S         | 900+90              |                                        | 0.9 \(^{C}\)       | 0.4 \(^{C}\)      | 0.0 \(^{B}\)      | 0.0 \(^{C}\)           | 0.2            | 1.5         | 2.8 \(^{E}\) |
| Tembotrione +S                   | 120                 |                                        | 4.3 \(^{C}\)       | 0.2 \(^{C}\)      | 6.5 \(^{B}\)      | 0.0 \(^{C}\)           | 0.2            | 5.7         | 16.7 \(^{D}\) |
| Atrazine + Tembotrione +S        | 900+90              |                                        | 1.3 \(^{C}\)       | 2.3 \(^{C}\)      | 0.0 \(^{B}\)      | 0.1 \(^{C}\)           | 0.1            | 3.4         | 7.0 \(^{DE}\) |
| Paraquat (Protected spray)       | 500                 |                                        | 16.3 \(^{B}\)      | 19.9 \(^{B}\)     | 107.7 \(^{A}\)    | 40.8 \(^{B}\)          | 0.0            | 1.9         | 186.6 \(^{B}\) |
| Untreated weedy check            |                     |                                        | 30.9 \(^{A}\)      | 55.9 \(^{A}\)     | 109.2 \(^{A}\)    | 84.3 \(^{A}\)          | 0.0            | 4.6         | 284.9 \(^{A}\) |
| p-Value                          | <0.0002             | <0.0001                                | <0.0001            | <0.0001           | 0.0806            | 0.8441                 | <0.0001        |

\(^{1}\text{Original values of weed dry weight data were square root transformed } \{\sqrt{(x+1)}\}\text{ before statistical analysis and based on the analysis of the transformed data letter have been assigned to original values for interpretation. Means with at least one letter common within a column are not statistically significant using Fisher's Least Significant Difference at 5\%.} \(^{\ddagger}\text{S}= \text{Cationic surfactant (Leader Mix) at 1000 ml ha}^{-1}.$

The total weed dry weight with atrazine application was 70.2 and 53.4 g m\(^{-2}\) at 3 WAS and 88.4 and 172.1 g m\(^{-2}\) at 6 WAS, during 2013 and 2014, respectively. The total weed dry weight reduction with atrazine application compared to untreated check was 64.3-69.0%. Balyan et al. (1994) also reported effectiveness of post-emergence atrazine against weeds in maize.

Among three HPPD herbicides (topramezone, tembotrione and mesotrione) applied alone, mesotrione 120 g ha\(^{-1}\) was comparatively poor in controlling weeds than topramezone 50 g ha\(^{-1}\) and tembotrione 120 g ha\(^{-1}\). The total weeds dry weights under mesotrione were 43.1 and 251.7 g m\(^{-2}\) at 3 WAS during 2013 and 2014, respectively, whereas at 6 WAS during 2013 and 2014, the respective values were 93.3 and 332.7 g m\(^{-2}\). Based on weed dry weights, the weed control efficiency (WCE) with mesotrione application was poor (67.3 and 31.0%) due to poor control of dominant grass weeds, \(D.\ aegyptium\) and \(D.\ sanguinalis\) as evident from dry weight data presented in Tables 1 to 4. However, mesotrione at 120 g ha\(^{-1}\) applied alone was very effective against broad-leaved weeds namely \(T.\ portulacastrum\) and \(D.\ arvensis\). The earlier findings also reported poor efficacy of mesotrione against grassy weeds but was quite effective for the control of many annual broadleaf weeds (Armel et al. 2003a; Zhang et al. 2013; Chhokar et al. 2019). Zollinger and Ries (2006) also reported that mesotrione gave less yellow foxtail (65%) and common ragweed...
Tank-mix application of \( \text{p-hydroxyphenylpyruvate dioxygenase} \) (\( \text{Ambrosia artemisiifolia} \) L.) (52%) control than tembotrione (88 and 94%) and topramezone (92 and 97%). Also, Kohrt and Sprague (2017b) reported better control of atrazine resistant palmer amaranth (\( \text{Amaranthus palmeri} \)) population with tembotrione and topramezone than mesotrione. In the present studies, topramezone was the most effective against grass weeds infested the experimental plots but had over all less efficacy against \( \text{T. portulacastrum} \) compared to tembotrione, which effectively controlled this weed. In earlier studies, Singh et al. (2012) reported that tembotrione 120 g ha\(^{-1}\) effectively controlled the grass weeds namely \( \text{Echinochloa colona} \) L. and \( \text{Digitaria sanguinalis} \) L. and sedge \( \text{Cyperus rotundus} \) L. Similar results that topramezone controlled a greater range of annual grasses than mesotrione, most notably at the earlier stage of seedling growth have been reported by Soltani et al. (2012). The protected application of paraquat 500 g ha\(^{-1}\) between crop rows recorded poor weed biomass reductions/weed control and only caused 27.2 to 34.5% weed dry weight reduction compared to untreated weedy control. Similarly, 2,4-D-Na was also less effective in the total weed dry weight reductions because of failure to control grass weeds. Application of 2,4-D effectively controlled the broad-leaved weed, \( \text{D. arvensis} \) but was relatively poor against \( \text{T. portulacastrum} \). The tank or ready mix combinations of reduced dose of atrazine 900 g ha\(^{-1}\) with reduced doses of either of the HPPD herbicide (topramezone 37.5 g ha\(^{-1}\) or tembotrione 90 g ha\(^{-1}\) or mesotrione 90 g ha\(^{-1}\)) were superior to the application of solo herbicide treatments.

Table 3. Performance of early post-emergence application of HPPD herbicides alone and in combination with atrazine against weeds in maize at 3 WAS 2014

| Herbicide           | Dose ha\(^{-1}\) (g. a.i.) | \( ^{1}\)Weed dry wt. g m\(^{-2}\)  |
|---------------------|---------------------------|------------------------------------|
|                     |                           | \( E. crusgalli \) | \( D. sanguinalis \) | \( D. aegyptium \) | \( T. portulacastrum \) | \( P. niruri \) | \( P. minima \) | Other weeds | Total weeds |
| Atrazine            | 1000                      | 0.3\(^{b}\)         | 19.2                      | 26.1\(^{CD}\)      | 0.8\(^{E}\)         | 0.0\(^{C}\)  | 0.1\(^{b}\)  | 6.9         | 53.4\(^{E}\) |
| Topramezone+S\(^{1}\)| 50                        | 0.0\(^{b}\)         | 0.0                       | 0.2\(^{E}\)        | 197.0\(^{C}\)       | 0.9\(^{B}\)  | 0.0\(^{B}\)  | 4.5         | 202.6\(^{CD}\) |
| Atrazine+Topramezone+S\(^{1}\)| 900+37.5 | 0.0\(^{b}\)         | 0.0                       | 0.2\(^{E}\)        | 1.0\(^{E}\)        | 0.0\(^{C}\)  | 0.0\(^{B}\)  | 8.7         | 9.9\(^{F}\)  |
| Mesotrione+S        | 120                       | 3.3\(^{a}\)        | 42.4                      | 157.2\(^{A}\)      | 26.9\(^{D}\)       | 0.4\(^{BC}\) | 2.0\(^{B}\)  | 19.5        | 251.7\(^{C}\) |
| Atrazine + Mesotrione+S\(^{1}\)| 900+90   | 0.4\(^{b}\)        | 7.3                       | 5.1\(^{DE}\)       | 2.0\(^{E}\)        | 0.0\(^{C}\)  | 0.1\(^{B}\)  | 6.3         | 21.2\(^{F}\) |
| Tembotrione+S       | 120                       | 0.9\(^{c}\)        | 6.9                       | 93.2\(^{AB}\)      | 10.9\(^{DE}\)      | 7.4\(^{A}\) | 7.2\(^{A}\)  | 9.8         | 136.2\(^{D}\) |
| Atrazine + Tembotrione+S\(^{1}\)| 900+90   | 0.5\(^{b}\)        | 0.8                       | 7.8\(^{DE}\)       | 2.1\(^{E}\)        | 0.0\(^{C}\)  | 0.4\(^{B}\)  | 4.6         | 16.1\(^{F}\) |
| Paraquat (Protected spray) | 500          | 2.7\(^{bc}\)       | 5.8                       | 41.4\(^{E}\)       | 369.9\(^{B}\)      | 0.0\(^{C}\)  | 0.3\(^{B}\)  | 2.7         | 422.8\(^{B}\) |
| 2,4-D-Na            | 1000                      | 9.7\(^{a}\)        | 5.9                       | 157.7\(^{AB}\)     | 292.9\(^{B}\)      | 0.0\(^{C}\)  | 1.7\(^{B}\)  | 2.0         | 470.0\(^{B}\) |
| Untreated weedy check | 9.9\(^{a}\)           | 1.6                    | 91.9\(^{B}\)               | 550.3\(^{A}\)    | 0.4\(^{BC}\)      | 1.1\(^{B}\)  | 4.0         | 659.1\(^{A}\) |

\(^{1}\)Original values of weed dry weight data were square root transformed \(\sqrt{(x+1)}\) before statistical analysis and based on the analysis of the transformed data letter have been assigned to original values for interpretation. Means with at least one letter common within a column are not statistically significant using Fisher's Least Significant Difference at 5%. \(^{1}\)S= Cationic surfactant (Leader Mix) at 1000 ml ha\(^{-1}\).
**Table 4.** Performance of early post-emergence application of HPPD herbicides alone and in combination with atrazine against weeds in maize at 6 WAS 2014

| Herbicide                  | Dose ha\(^{-1}\) (g. a.i.) | \(^{1}\)Weed dry wt. g m\(^{-2}\) E. crus-galli | E. colona | D. aegyptium | D. sanguinalis | T. portulacastrum | P. niruri | P. minima | Other weeds | Total weeds |
|----------------------------|----------------------------|-----------------------------------------------|-----------|--------------|---------------|------------------|----------|-----------|------------|-------------|
| Atrazine                   | 1000                       | 8.1 \(^{BC}\)                              | 1.8 \(^{B}\) | 130.9 \(^{BC}\) | 24.5 \(^{AB}\) | 2.7 \(^{C}\)      | 0.0      | 0.2 \(^{D}\) | 4.0        | 172.1 \(^{C}\) |
| Topramezone+S\(^{1}\)      | 50                         | 0.0 \(^{D}\)                              | 0.0 \(^{B}\) | 1.9 \(^{D}\)  | 0.0 \(^{C}\)  | 76.3 \(^{B}\)    | 0.0      | 0.0 \(^{D}\) | 9.5        | 87.8 \(^{D}\)  |
| Atrazine+Topramezone+S     | 900+37.5                   | 0.8 \(^{CD}\)                             | 0.2 \(^{B}\) | 2.5 \(^{D}\)  | 0.7 \(^{C}\)  | 3.5 \(^{D}\)      | 0.0      | 0.0 \(^{D}\) | 7.8        | 15.6 \(^{E}\)  |
| Mesotrione +S              | 120                        | 2.9 \(^{BCD}\)                            | 11.7 \(^{A}\) | 272.5 \(^{A}\) | 30.2 \(^{A}\) | 1.3 \(^{C}\)      | 0.1      | 12.5 \(^{ABC}\) | 1.6        | 332.7 \(^{B}\) |
| Atrazine + Mesotrione +S   | 900+90                     | 6.5 \(^{BCD}\)                            | 0.0 \(^{B}\) | 68.8 \(^{C}\)  | 26.0 \(^{AB}\) | 5.5 \(^{C}\)      | 0.0      | 1.3 \(^{CD}\) | 6.8        | 114.8 \(^{CD}\) |
| Tembotrione+S              | 120                        | 5.5 \(^{BCD}\)                            | 0.0 \(^{B}\) | 241.0 \(^{A}\) | 7.2 \(^{B}\)  | 3.6 \(^{C}\)      | 0.7      | 14.9 \(^{ABC}\) | 5.7        | 278.5 \(^{B}\) |
| Atrazine +Tembotrione+S    | 900+90                     | 7.8 \(^{BCD}\)                            | 0.3 \(^{B}\) | 98.9 \(^{BC}\) | 2.9 \(^{C}\)  | 6.7 \(^{C}\)      | 0.0      | 2.7 \(^{BCD}\) | 6.8        | 117.9 \(^{CD}\) |
| Parquat (Protected spray)  | 500                        | 9.7 \(^{B}\)                             | 2.1 \(^{B}\) | 98.9 \(^{BC}\) | 9.8 \(^{AB}\) | 206.3 \(^{A}\)    | 0.1      | 21.3 \(^{A}\)  | 2.4        | 350.6 \(^{B}\) |
| 2,4-D-Na                   | 1000                       | 61.6 \(^{A}\)                            | 11.3 \(^{A}\) | 341.3 \(^{A}\) | 2.2 \(^{C}\)  | 52.1 \(^{B}\)     | 0.2      | 14.3 \(^{AB}\) | 1.5        | 484.5 \(^{A}\) |
| Untreated weedy check       | 57.7 \(^{A}\)             | 13.4 \(^{A}\)                            | 152.3 \(^{B}\) | 16.0 \(^{ABC}\) | 215.6 \(^{A}\) | 0.4              | 0.05     | 20.3 \(^{A}\)  | 6.1        | 481.8 \(^{A}\) |
| \(p\)-Value                | \(<0.0001\)                | 0.0053                                    | \(<0.0001\) | 0.0139       | \(<0.0001\)  | 0.0785           | 0.0400   | 0.405       | \(<0.0001\) |

\(^{1}\)Original values of weed dry weight data were square root transformed \{\(\sqrt{(x+1)}\)\} before statistical analysis and based on the analysis of the transformed data letter have been assigned to original values for interpretation. Means with at least one letter common within a column are not statistically significant using Fisher's Least Significant Difference at 5%. \(^{1}\)S= Cationic surfactant (Leader Mix) at 1000 ml ha\(^{-1}\).

In comparison to untreated control, these combinations provided 75.5-99.7% reduction in dry weights of weeds. All the herbicide combinations treatments were quite effective in reducing the dry weight of weeds during 2013 at both stages and at 3 WAS during 2014 and these treatments did not differ significantly among themselves. Whereas, during 2014 at 6 WAS, atrazine + topramezone combination was superior in overall weed control. The topramezone + atrazine caused consistent reduction of 96.8-99.5% in total weed dry weight during both the years of studies. Similarly, Bollman et al. (2008) while comparing the efficacy of 4-hydroxyphenyl pyruvate dioxygenase (HPPD)-inhibiting herbicides (mesotrione, tembotrione, and topramezone) applied POST alone or mixed with atrazine found greater giant foxtail control with tembotrione or topramezone than mesotrione alone or mixed with atrazine. Overall, the combinations of HPPD herbicides and atrazine were more effective in reducing the total weed dry weight compared to their alone application. The weed control efficiency (WCE) with application of HPPD herbicides and atrazine combinations was 97.5-99.0%, during 2013 and 2014, it was 75.5-96.8% (Table 5). The lower WCE during 2014 compared to 2013 was due to slightly advanced stage of \(T.\) portulacastrum and \(D.\) aegyptium at the time of herbicide application (20 DAS). Among HPPD herbicides, mesotrione 120 g ha\(^{-1}\) had the lowest WCE (31.0-67.3%) and post atrazine 1000 g ha\(^{-1}\) recorded WCE of 64.3-69.0%.
The better weed control efficacy in tank mix combinations compared to alone application shows synergism of these herbicides when mixed with atrazine.

Table 5. Performance of early post-emergence application of herbicides against weeds in maize.

| Herbicide               | Dose ha$^{-1}$ (g a.i.) | Weed control efficiency (WCE) % at 6 WAS | Grain Yield, q ha$^{-1}$ |
|-------------------------|-------------------------|----------------------------------------|-------------------------|
|                         | 2013    | 2014 | 2013 | 2014 | 2013 | 2014 | 2013 | 2014 |
| Atrazine                |         |      |      |      |      |      |      |      |
| 1000                    | 69.0    | 64.3 | 74.16 | AB  | 65.28 | BC  |         |      |
| Topramezone+S$^\dagger$ | 50      |      | 95.6 | 81.8 | 79.03 | AB  | 68.50 | B     |
| Atrazine+Topramezone+S  | 900+37.5|      | 98.1 | 96.8 | 82.33 | A   | 80.30 | A     |
| Mesotrione+S            | 120     |      | 67.3 | 31.0 | 73.83 | AB  | 59.82 | B     |
| Atrazine + Mesotrione+S | 900+90 |      | 99.0 | 76.2 | 79.00 | AB  | 79.63 | A     |
| Tembotrione+S           | 120     |      | 94.1 | 42.2 | 77.10 | AB  | 67.87 | B     |
| Atrazine +Tembotrione+S | 900+90 |      | 97.5 | 75.5 | 79.59 | AB  | 78.84 | A     |
| Paraquat (Protected spray) | 500   |      | 34.5 | 27.2 | 70.22 | B   | 55.64 | C     |
| 2,4-D-Na                | 1000    |      | -    | 0.0  | -     |     | 43.48 | D     |
| Untreated weedy check   | -       |      | 0.0  | 0.0  | 56.41 | C   | 42.21 | D     |

$^\dagger$S= Cationic surfactant (Leader Mix) at 1000 ml ha$^{-1}$.

Bollman et al. (2008) also reported that common lambsquarters (Chenopodium album L.), velvetleaf (Abutilon theophrasti Medik.), and common ragweed (Ambrosia artemisiifolia L.) were controlled by 98% or greater with the HPPD-inhibiting herbicides (mesotrione, tembotrione, and topramezone) when mixed with atrazine. Similar findings of improved weed control by the addition of atrazine with tembotrione, mesotrione and topramezone have also been reported earlier (Williams et al. 2011; Kohrt and Sprague, 2017b). Maize grain yield was significantly influenced (<0.0001) by weed control treatments (Table 5). The uncontrolled weed competition throughout the season led to the lowest grain yield of 56.41 and 42.21 q ha$^{-1}$ during 2013 and 2014, respectively. Compared to untreated weedy control, the post-emergence application of atrazine 1000 g ha$^{-1}$ recorded significant improved grain yields due to better weed control. Among three HPPD herbicides, mesotrione had the lowest maize grain yield, whereas statistically similar yield levels were recorded with topramezone and tembotrione applications. However, grain yields under various mixtures of HPPD herbicides with atrazine 900 g ha$^{-1}$ applied as post-emergence were statistically similar but significantly better than the applications of mesotrione 120 g ha$^{-1}$, 2,4-D 1000 g ha$^{-1}$ and atrazine 1000 g ha$^{-1}$. The better yields observed with herbicide mixture treatments compared to solo applications were due to better control of broad spectrum weeds (grass and broad-leaved).
Comparative performance of pre and post-emergence atrazine and its combinations with topramezone and tembotrione against weeds

In another experiment, as compared to untreated weedy check treatment, the applications of atrazine alone (pre or post-emergence) or its post-emergent combinations with HPPD herbicides (tembotrione and topramezone) reduced the weed dry weight at 3 and 6 WAS (Figure 1).

Figure 1. Comparative performance of pre and post-emergence atrazine and its post-emergent tank-mixture with either topramezone or tembotrione against weeds in maize. Vertical error bars above means represent ±SEM. Means data are of 9 observations from three field evaluations except weed dry weight at 6 WAS consisted of 6 observations.

Between the pre and post- applications of atrazine 1000 g ha⁻¹, post (15-18 DAS) application was better in reducing the weed dry weight at both the stages (Figure 1). The post atrazine application accumulated 51.7 and 43.9% lower weed dry weights (135.2 and 217.6 g m⁻²) compared to pre-emergence atrazine application (280.1 and 388.1 g m⁻²) at 3 and 6 WAS, respectively. Whereas, in comparison to untreated weedy control, the post atrazine application reduced the weed dry weight by 61.8 and 57.2%, respectively at 3 and 6 WAS. Balyan et al. (1994) also reported superiority of post-emergence atrazine (0.25-0.50 kg ha⁻¹) at 7 or 14 DAS over pre-emergence application for control of horsepurslane (T. portulacastrum), jungle rice (E. colonum) and Digera arvensis. As for better activity of pre-emergence herbicide application, the rainfall or sufficient moisture is required, so environmental factors affect the efficacy of pre-emergence herbicide. Moreover, the
Tank-mix application of p-hydroxyphenylpyruvate dioxygenase ...

The performance of pre-emergence herbicides is also not good in some of the situations such as the adoption of no-till techniques in maize production (Zhang et al. 2013). Whereas, post-emergence application is less affected by environmental factors (Tapia et al. 1997). Additionally, some times higher rates for pre-emergence applications of weak acid herbicides such as mesotrione are required due to their adsorption by soil organic matter. The addition of HPPD herbicide (tembotrione or topramezone) with atrazine drastically reduced (91.9-95.4%) the weed dry weights compared to untreated weedy check. As a result of reduction in weed dry weight, the maize grain yield improved and maximum yield was obtained with tank mixture of atrazine with tembotrione (80.1 q ha\(^{-1}\)) followed by atrazine plus topramezone at 900 + 40 g ha\(^{-1}\) (78.7 q ha\(^{-1}\)). Also, between the two atrazine application timing treatments, the maize yield was better with post-atrazine (64.3 q ha\(^{-1}\)) as compared to pre-emergence atrazine (40.9 q ha\(^{-1}\)) due to improved weed control. Whereas, the maize yield was higher in tank-mix herbicide treatments compared to sole atrazine applied as either pre- or post-emergent. Similarly various researchers had reported poor weed control with pre-emergence atrazine as compared with post-emergence application of tembotrione 110-120 g ha\(^{-1}\) (Singh et al. 2012) or tembotrione in combination with reduced dose of atrazine (Recker et al. 2015). Janak and Grichar (2016) also observed higher corn yield when herbicide treatments consisted of mixtures of more than one active ingredient as compared to a single active ingredient.

The importance of using atrazine as a tank-mix with other herbicides particularly the HPPD herbicides is due to the improved herbicide efficacy, increased spectrum of weed control (Bollman et al. 2008) and reduced risk of evolving herbicide resistance even with the application of lower amount of total active ingredients (Zhang et al. 1995). This has been shown by earlier research (Woodyard et al. 2009b; Walsh et al. 2012) that even, the addition of lower dose of mesotrione to atrazine improves the control of atrazine resistant velvetleaf (*Abutilon theophrasti* Medik.) and wild radish (*Raphanus raphanistrum* L.), demonstrating the synergistic herbicide interactions in overcoming the target-site herbicide resistance mechanism. Previous research have also reported that HPPD herbicides provide an opportunity to control triazine resistant weeds, like common lambsquarters (Bollman et al. 2006) and in combination with atrazine effectively control glyphosate resistant weeds (Sutton et al. 2002; Vyn et al. 2006; Kumar and Jha, 2015), such as, Palmer amaranth (*Amaranthus palmeri* S. Wats.) and kochia (*Kochia scoparia* (L.) Schrad.).

Also, Elmore et al. (2013) studied the combinations of HPPD-inhibiting herbicides (mesotrione and topramezone) with the PSII-inhibiting herbicide amicarbazone and observed that mesotrione and amicarbazone combination was synergistic for annual bluegrass (*Poa annua* L.) control but
topramezone plus amicarbazone was not. Similarly, POST-applied atrazine plus HPPD-inhibiting herbicides efficacies showed the synergistic interactions on many weed species (Bollman et al. 2008; Jhala et al. 2014). Similarly, Williams et al. (2011) reported that atrazine improved the tembotrione efficacy with consistent better weed control compared to alone tembotrione. In other studies, tembotrione provided differential control of annual grass weed species (Bollman et al. 2008; Williams et al. 2011) and in such situations; its applications in combinations with other herbicides are beneficial in term of broaden weed control spectrum and reduced production costs (Damalas, 2004). Earlier, Janak and Grichar (2016) also observed consistent annual grass weeds control including barnyardgrass (E. crus-galli), browntop panicum (Panicum fasciculatum L.), Texas millet (Urochloa texana L.), and sprawling signalgrass (Brachiaria reptans L.) by three different group of herbicides combinations (S-metolachlor + atrazine + mesotrione) than one active ingredient. So, in order to widen the weed control spectrum, it is imperative to use combination of herbicides having different mode of actions. Further, herbicides having independent modes of action when used in sequence, rotation, or as mixtures, generally help in delaying the evolution of resistance (Wrubel and Gressel, 1994; Beckie et al. 2001) and the tank or ready mixtures may delay the resistance evolution longer than rotations (Diggle et al. 2003) or sequential application.

Further, to improve the efficacy of these synergistic combinations of herbicides; the role of some adjuvants may also be investigated. As in earlier studies, nitrogen (N) enhanced the crabgrass (Digitaria spp.) control with mesotrione and topramezone (Elmore et al. 2012; Beck et al. 2017). Beck et al. (2017) were of the view that growers can pair nitrogen and post-mesotrione applications together or place in closed proximity for better weed control as integration of N and mesotrione causes more bleaching and necrosis. The ammonium sulphate (AMS) addition also enhanced the mesotrione efficacy (Devkota et al. 2016). Adjuvant MSO plus fertilizer origin adjuvants (AMN, UAN) are typically applied with HPPD-inhibitor herbicides (tembotrione or mesotrione), which substantially improve the weed control (Young et al. 2007). Similarly, Idziak and Woznica (2014) also observed the benefit of adjuvants and reported satisfactory weed control in maize even with the reduced rates of tembotrione, when applied along with adjuvants (methylated seed oil and ammonium nitrate). Therefore, further studies are needed to examine the role of adjuvant in relation to herbicide combination and weed management.

**Johnsongrass (S. halepense) infestation as affected by crop sequence**

Johnsongrass (Sorghum halepense (L.) Pers.) is a noxious perennial grass weed produced by rhizomes and is one among the ten worst weeds interfering in the crop production. Most of the maize herbicides are not effective against this common and troublesome weed in maize production.
(Damalas and Eleftherohorinos, 2001). In upland situation, it is a severe problem particularly in maize and if not controlled can drastically reduce the maize yield. It can reproduce via seeds and rhizomes. In a continuous maize-wheat system, its population was significantly high (27481 panicles ha⁻¹) compared to puddle rice-wheat sequence (no emergence of *S. halepense*) (Figure 2). Puddled (wet tillage) rice had water stagnation, which completely inhibited its emergence might be due to loss of viability of its seeds and rhizomes. In earlier studies (Chhokar et al. 2014) also, it has been reported that many upland weeds fail to establish under water stagnation or anaerobic conditions in rice fields. Since, chemical control of this weed is difficult and therefore, adoption of crop rotation can be an effective strategy for its control. The fields having problem of *S. halepense*, maize can be replaced with puddled rice for 2-3 seasons to contain the problem of *S. halepense* in subsequent maize crop. In addition to this non-chemical strategy of crop rotation, for johnsongrass control, ALS inhibiting herbicides such as rimsulfuron at 10 g ha⁻¹ have been found very effective (Damalas and Eleftherohorinos, 2001). However, contrary to HPPD herbicides and atrazine synergistic combination responses, many researchers (Damalas and Eleftherohorinos, 2001; Kaastra et al. 2008; Damalas et al. 2015) have reported two way antagonistic interactions between HPPD and ALS inhibiting herbicides, which are weed species specific. While, Schuster et al. (2008) showed the decreased efficacy of sulfonylurea herbicides (nicosulfuron and foramsulfuron) on green foxtail (*Setaria viridis* (L.) Beauv.), yellow foxtail (*Setaria pumila* (Poir.) Roemer & J.A. Schultes), and shattercane (*Sorghum bicolor* (L.) Moench ssp. arundinaceum (Desv.) de Wet & Harlan) with the addition of mesotrione to sulfonylurea herbicides. Nevertheless, Kaastra et al. (2008) observed that this antagonism can be overcome by addition of atrazine, but differing, Damalas and Eleftherohorinos (2001) reported reduced johnsongrass control by ALS inhibiting herbicides (rimsulfuron and primisulfuron), when atrazine was added as tank mixture. Contrary, such negative interactions between tembotrione and ALS-inhibiting herbicides were not observed by Damalas et al. (2017) particularly for the control of rhizomatous *S. halepense*. These results highlight the need to further evaluate the responses of combinations of various other herbicide groups for effective weed control in maize.
Figure 2. Effect of crop sequence on *Sorghum halepanse* abundance. Vertical error bars above means represent ± SEM. Means are based on 9 observations from three field experiments and significantly differed at P=0.004.

**Pot experiments: Tank mix interactions of HPPD herbicides with atrazine against grass weed species**

Three HPPD herbicides (mesotrione, topramezone and tembotrione) and atrazine either alone or in combinations at graded doses were evaluated against three grass species (Tables 6 and 7). Among three HPPD herbicides, mesotrione was inferior in controlling grass species compared to topramezone and tembotrione. Also, atrazine applied alone was poor against *Digitaria sanguinalis*. In first set of herbicide treatments evaluation, application of atrazine at 500 and 1000 g ha$^{-1}$ exhibited respective fresh biomass reductions in comparison to untreated check of 84.2 and 100% of *E. crus-galli*, 53.1 and 100% of *E. colona* and 1.2 and 3.3% of *D. sanguinalis*. While, tembotrione at 55 g ha$^{-1}$ gave biomass reductions of *E. crus-galli*, *E. colona* and *D. sanguinalis* to the extent of 98.5, 64.0 and 86.1%, respectively. Moreover, the highest rate of tembotrione 110 g ha$^{-1}$ provided control in term of biomass reduction of these weeds in the range of 96 to 100%. In combination, tembotrione + atrazine at 55 + 500 and 110 + 1000 g ha$^{-1}$ provided complete kill of all the three grass species. The expected controls (Colby’s values) were also in the same range (Table 6).
Table 6. Grass weed biomass reduction (% control) by the application of HPPD herbicides alone and in combination with atrazine.

| Treatments       | Herbicide dose g ha\(^{-1}\) | E. crus-galli | E. colona | Digitaria sanguinalis |
|------------------|------------------------------|---------------|-----------|-----------------------|
|                  | Observed control (%)         | Expected control (%) | Significance p=0.05 | Observed control (%) | Expected control (%) | Significance p=0.05 | Observed control (%) | Expected control (%) | Significance at p=0.05 |
| Control          | 0.0 D                        | 0.0 E         |           |                       |                       |                   |                       |                       |                       |
| Atrazine         | 500                          | 84.2 B        |           |                       | 53.1 C               | 1.2 E              |                       |                       |                       |
| Atrazine         | 1000                         | 100.0 A       |           |                       | 100.0 A              | 3.3 E              |                       |                       |                       |
| Tembotrione+St   | 55                           | 98.5 A        |           |                       | 64.0 B               | 86.1 B             |                       |                       |                       |
| Tembotrione+St   | 110                          | 99.7 A        |           |                       | 100.0 A              | 96.0 A             |                       |                       |                       |
| Tembo+Atra+St    | 55+500                       | 100.0 A       | 99.8      | S                     | 100.0 A              | 81.6 S             | 100.0 A               | 86.3 S                | S                     |
| Tembo+Atra+St    | 110+1000                     | 100.0 A       | 100       | NS                    | 100.0 A              | 100 NS             | 100.0 A               | 96.1 NS                | NS                    |
| Topramezone+St   | 25                           | 100.0 A       |           |                       | 100.0 A              | 100.0 A            |                       |                       |                       |
| Topramezone+St   | 50                           | 100.0 A       |           |                       | 100.0 A              | 100.0 A            |                       |                       |                       |
| Topra+Atra+St    | 25+500                       | 100.0 A       | 100       | NS                    | 100.0 A              | 100 NS             | 100.0 A               | 100 NS                | NS                    |
| Topra+Atra+St    | 50+1000                      | 100.0 A       | 100       | NS                    | 100.0 A              | 100 NS             | 100.0 A               | 100 NS                | NS                    |
| Mesotrione+St    | 60                           | 60.6 C        |           |                       | 29.7 D               | 54.8 D             |                       |                       |                       |
| Mesotrione+St    | 120                          | 80.1 B        |           |                       | 58.0 BC              | 72.5 C             |                       |                       |                       |
| Meso+Atra+St     | 60+500                       | 99.6 A        | 93.8      | S                     | 100.0 A              | 66.9 S             | 100.0 A               | 55.4 S                | S                     |
| Meso+Atra+St     | 120+1000                     | 99.7 A        | 100       | NS                    | 100.0 A              | 100 NS             | 100.0 A               | 73.6 S                | S                     |

p-Value <0.0001 <0.0001 <0.0001

LSD (0.05) 4.36 10.64 7.21

Abbreviations: St= cationic surfactant, Leader-mix at 1000 ml ha\(^{-1}\); Atra= Atrazine; Tembo= Tembotrione; Meso= Mesotrione; Topra= Topramezone; NS= not significant and S= significant. The expected values are based on Colby’s equation \[E = (X+Y) - (XY)/100\] and ‘NS’ indicates the expected value was not different than the observed value.

Alone application of topramezone at 25 and 50 g ha\(^{-1}\) or in combination with atrazine as 25 + 500 or 50 + 1000 g ha\(^{-1}\) also caused hundred per cent kill of these grass weed species. The expected values were similar as that of the observed values (Table 6). Mesotrione applied at 60 and 120 g ha\(^{-1}\) had 61 and 80%; 30 and 58%; and 55 and 73%, control of E. crus-galli, E. colona and D. sanguinalis, respectively. Nevertheless, mesotrione plus atrazine combinations (60 + 500 and 120 + 1000 g ha\(^{-1}\)) recorded 99.6 to 100% control/biomass reduction of the three grass weeds. The expected control values for E. crus-galli, E. colona and D. sanguinalis with mesotrione plus atrazine applications at 60 + 500 g ha\(^{-1}\) were 93.8, 66.9 and 55.4%, respectively, indicating the synergistic interactions. Similarly, with application of mesotrione plus atrazine at 120 + 1000 g ha\(^{-1}\), the observed control of D. sanguinalis was 26.4% higher than expected control value of 73.6%.
Table 7. Grass weed biomass reduction (% control) by the application of HPPD herbicides alone and in combination with atrazine.

| Treatments                  | Herbicide dose g ha\(^{-1}\) | Digitaria sanguinalis | Dactyloctenium aegyptium |
|-----------------------------|------------------------------|-----------------------|--------------------------|
|                             |                              | Observed control (%)  | Expected control (%) at p=0.05 | Significance | Observed control (%) | Expected control (%) at p=0.05 | Significance |
| Control                     | -                            | 0.0 F                 | 0.0 H                     |             | 30.6 F               | 48.3 E                     |             |
| Atrazine                    | 125                          | 3.7 EF                | 48.3 E                    |             | 14.4 E               | 90.6 B                     |             |
| Atrazine                    | 250                          | 4.8 EF                | 48.3 E                    |             | 27.5 D               | 98.3 A                     |             |
| Atrazine                    | 500                          | 14.4 E                | 90.6 B                    |             |                      |                            |             |
| Atrazine                    | 1000                         | 27.5 D                | 98.3 A                    |             |                      |                            |             |
| Tembotrione + St            | 13.75                        | 51.4 C                | 67.7 D                    |             |                      |                            |             |
| Tembotrione + St            | 27.5                         | 82.5 B                | 97.3 AB                   |             |                      |                            |             |
| Tembotrione + St            | 55                           | 9.6 EF                | 28.1 F                    |             |                      |                            |             |
| Tembotrione + St            | 110                          | 100.0 A               | 100.0 A                   |             |                      |                            |             |
| Atra + Tembo + St           | 125 + 13.75                  | 97.1 AB               | 100.0 A                   | S           | 99.5 A               | 77.6 S                     |             |
| Atra + Tembo + St           | 250 + 27.5                   | 100.0 A               | 99.7 A                    | S           | 99.7 A               | 98.7 NS                    |             |
| Atra + Tembo + St           | 500 + 55                     | 100.0 A               | 100.0 A                   | NS          | 100.0 A              | 100.0 NS                   |             |
| Atra + Tembo + St           | 110 + 1000                   | 100.0 A               | 100.0 A                   | NS          | 100.0 A              | 100.0 NS                   |             |
| Mesotrione + St             | 15                           | 1.5 F                 | 14.1 G                    |             |                      |                            |             |
| Mesotrione + St             | 30                           | 9.6 EF                | 28.1 F                    |             |                      |                            |             |
| Mesotrione + St             | 60                           | 53.1 C                | 49.6 E                    |             |                      |                            |             |
| Mesotrione + St             | 120                          | 84.4 B                | 82.2 C                    |             |                      |                            |             |
| Atra + Meso + St            | 125 + 15                     | 26.8 D                | 47.1 E                    | S           | 40.1 S               |                            |             |
| Atra + Meso + St            | 250 + 30                     | 57.9 C                | 67.8 D                    | S           | 62.7 S               |                            |             |
| Atra + Meso + St            | 500 + 60                     | 97.7 A                | 100.0 A                   | NS          | 95.2 NS              |                            |             |
| Atra + Meso + St            | 1000 + 120                   | 100.0 A               | 100.0 A                   | NS          | 99.8 NS              |                            |             |
| Topramezone + St            | 6.25                         | 93.9 AB               | 99.3 A                    |             |                      |                            |             |
| Topramezone + St            | 12.5                         | 97.47 A               | 100.0 A                   |             |                      |                            |             |
| Topramezone + S             | 25                           | 100.0 A               | 100.0 A                   |             |                      |                            |             |
| Topramezone + S             | 50                           | 100.0 A               | 100.0 A                   |             |                      |                            |             |
| Atra + Topra + St           | 125 + 6.25                   | 100.0 A               | 100.0 A                   | NS          | 99.5 NS              |                            |             |
| Atra + Topra + St           | 250 + 12.5                   | 100.0 A               | 100.0 A                   | NS          | 100.0 NS             |                            |             |
| Atra + Topra + St           | 500 + 25                     | 100.0 A               | 100.0 A                   | NS          | 100.0 NS             |                            |             |
| Atra + Topra + St           | 1000 + 50                    | 100.0 A               | 100.0 A                   | NS          | 100.0 NS             |                            |             |
| p-value                     | <0.0001                      | 6.92                  | 6.92                      |             |                      |                            |             |
| LSD (0.05)                  | 11.92                        | 6.92                  | 6.92                      |             |                      |                            |             |

Abbreviations: St= cationic surfactant at 1000 ml ha\(^{-1}\); Atra= Atrazine; Tembo= Tembotrione; Meso= Mesotrione; Topra= Topramezone; NS= not significant and S= significant. The expected values are based on Colby’s equation \[E = \frac{(X+Y) - (XY)/100}{100}\] and when the expected value was not different than the observed value then denoted by ‘NS’ and when significant denoted by ‘S’.

In another set of herbicide treatments studies, where additional treatments of reduced rates of atrazine (125 and 250 g ha\(^{-1}\)), mesotrione (15 and 30 g ha\(^{-1}\)), tembotrione (13.75 and 27.5 g ha\(^{-1}\)) and topramezone (6.25 and 12.5 g ha\(^{-1}\)) alone and in combinations were included (Table 7). The
test species were *D. sanguinalis* and *D. aegyptium*. The application of atrazine at 125, 250, 500 and 1000 g ha\(^{-1}\) gave control of *D. sanguinalis* as 3.7, 4.8, 14.4 and 27.5%, respectively, indicating very poor efficacy of atrazine against this weed species. Whereas, the *D. aegyptium* control with application of atrazine at 125, 250, 500 and 1000 g ha\(^{-1}\) was 30.6, 48.3, 90.6 and 98.3%, respectively. The control levels of *D. sanguinalis* enhanced (51.4 to 100%) as the rate of tembotrione increased from 13.75 to 110 g ha\(^{-1}\) but between two higher rates (55 and 110 g ha\(^{-1}\)), the control did not differ. Whereas, tembotrione at 27.5, 55 and 110 g ha\(^{-1}\) provided control levels of *D. aegyptium* as 97.3, 100 and 100%, respectively, which among themselves were not statistically different. However, these three rates were significantly superior to the lowest rate of tembotrione 13.75 g ha\(^{-1}\) (67.7%) for *D. aegyptium* control. The control of both the grass weeds was improved with tank-mix application of atrazine and tembotrione and the control level of either of the species did not differ among the four rates of combinations (Table 7). The control levels achieved with applications of lower rates of atrazine plus tembotrione combinations (125 + 13.75 g ha\(^{-1}\) and 250 + 27.5 g ha\(^{-1}\)) were significantly superior to what were expected using Colby’s equation (53.4 and 82.7%) for *D. sanguinalis*. Whereas, for *D. aegyptium*, the observed control (99.5%) was superior to expected control (77.6%) only at lowest dose of combination (125 + 13.75 g ha\(^{-1}\)). Also, the acceptable level of control of both the grass weed species (82-84%) was only observed at the highest rate of mesotrione applied alone at 120 g ha\(^{-1}\). However, in combinations, atrazine + mesotrione caused better control of *D. sanguinalis* and *D. aegyptium* as expected using Colby’s equation. The application of topramezone at graded doses provided the excellent control of both the grass species (≥ 93%) and the observed control of each of the species was statistically in the same range among four rates of applications. When topramezone was tank mixed with atrazine, even at the lowest rate (125 + 6.25 g ha\(^{-1}\)) caused complete kill of both the species and the observed control of *D. sanguinalis* was significantly better as what was expected using Colby’s equation (93.9%). The results of the present pot studies indicate the notable synergistic interactions between atrazine and HPPD herbicides, when applied at reduced rates. Colby (1967) already explained that analyzing for herbicide interactions is better when the herbicides are applied alone at a dose that provides approximately 50% control. Riley and Shaw (1988) and Scott *et al.* (1998) also showed that synergy is more likely when applied at reduced rates and herbicide interactions can vary for two herbicides when mixed at low rates, compared with high rates.

The field and pot studies have clearly indicated the synergistic responses of HPPD herbicides and atrazine combinations leading to improved weed control particularly of grasses. In earlier studies also, the synergistic interactions between atrazine and HPPD-inhibiting herbicides for
improved control of many weeds have been reported (Abendroth et al. 2006; Whaley et al. 2006; Bollman et al. 2008; Woodyard et al. 2009a; Jhala et al. 2014; Kohrt et al. 2017b; Chhokar et al. 2019). Armel et al. (2003a) found better weed control, when mesotrione, tank mixed with atrazine or acetochlor compared to sole mesotrione. Mesotrione and atrazine mixtures increased the development of necrotic tissues compared to mesotrione application alone having the slower rate of bleaching symptoms (Armel et al. 2005). Whereas, Kohrt and Sprague (2017b) and Williams et al. (2011) reported additive responses when atrazine was applied with tolypyralate and topramezone and they are of the view that joint activity in the form of synergism occurs more readily with the triketones compared with the benzopyrazoles. Usually herbicides are mixed to broaden spectrum of activity or improve control of other species. In all the field studies, consistent weed control and maize yields were obtained with two way herbicide combinations. Earlier research has also reported, consistent weed control and maize yield with the usage of two way (Kohrt and Sprague, 2017a; Williams et al. 2011) and three-way (Whaley et al. 2009) herbicide mixtures, as well as, two pass (pre followed by post emergence herbicides having minimum of two effective herbicide site of action) of herbicides. In addition, the use of two herbicides with different sites of action in mixture is an effective tactic to delay the onset of resistance (Norsworthy et al. 2012; Bagavathiannan et al. 2014). Kohrt and Sprague (2017a) have observed that HPPD herbicide, mesotrione in combination with other herbicides effectively control the three-way herbicide-resistant palmer amaranth population. Additionally, the improved weed control with synergistic herbicide combinations, the lower herbicide rates might allow increased profits for growers along with reduced risk of injury to current and succeeding susceptible crops (Blackshaw et al. 2006).

**Conclusion**

The results of these studies indicated that atrazine applied as post-emergence is more effective than pre-emergence application. Among three HPPD herbicides, topramezone and tembotrione were better than mesotrione in controlling weeds and recording better maize yield. However, the tank or ready-mix combinations of atrazine with either topramezone or tembotrione or mesotrione were similar but better than their solo application in term of weed control and producing maize yield. Therefore, atrazine and HPPD herbicides combinations can be used for effective broad-spectrum (grass and broadleaf) weed kill in maize and combinations are significantly better than the standard recommendation of atrazine 1000 g ha⁻¹. Also, weed management strategies involving herbicide combinations with different sites of action, reduce the herbicide selection pressure and delay the evolution of herbicide resistance in weeds (Beckie and Reboud, 2009; Norsworthy et al. 2012). Therefore, the results of present studies have practical implications because HPPD
herbicides in combinations with atrazine besides providing broad-spectrum weed control along with high crop safety will further help in herbicide resistance management. However, for long term sustainable and economic weed management and crop production, proper attention needs to be paid towards integration of non-chemical weed control practices (cultural and mechanical) coupled with effective herbicide combinations having diverse modes of actions.

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Conflicts of Interest

The authors declare no conflict of interest.

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