EFFECTS OF SPRING HERBICIDE TREATMENTS ON WINTER WHEAT GROWTH AND GRAIN YIELD*

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Herbicides provide a low-cost solution for protecting crops from significant yield losses. If weed infestations are below damage thresholds, however, then herbicide application is unnecessary and can even lead to yield loss. A small-plot field trial was conducted to examine the effect of herbicides on winter wheat yields. Weeds were removed manually from the trial area before herbicide application. Twenty-four treatments were tested in four replications. Treatment 1 consisted of an untreated weed-free control, whereas the other treatments comprised applications of the following herbicides and their combinations: metsulfuron-methyl + tribenuron-methyl (4.95 + 9.99 g ha^{-1}), pinoxaden (30 g ha^{-1}), fluroxypyr (175 g ha^{-1}), and clopyralid (120 g ha^{-1}). Water (250 l ha^{-1}) or a urea-ammonium nitrate fertilizer solution (UAN, 120.5 l ha^{-1}) was used as the herbicide carrier. Crop injury 30 days after treatment and yield loss were recorded. Results showed minor crop injury by herbicides and their combinations when applied without UAN and moderate injury caused by UAN in combination with herbicides. Yield losses reached 5.3% and 4.3% in those treatments where all of the tested herbicides were applied with and without UAN, respectively. The effect of all treatments on crop yield was, however, statistically insignificant ($P = 0.934$).

weed control; herbicides; phytotoxicity; cereals; yield loss

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INTRODUCTION

Herbicides are an indispensable aid in weed management for modern, large-scale farming. They provide low-cost solutions for protecting crops from significant yield losses. If weed infestations in fields are below damage thresholds, however, herbicide application is unnecessary and can even lead to yield loss (Ritter et al., 2008). Site-specific weed management (SSWM) methods enable spatially variable treatment of weed populations according to actual weed abundance, thus offering the opportunity for herbicide savings and diminishing environmental impact while potentially also decreasing crop injuries caused by herbicides.

Cereals crops are particularly suitable for SSWM, because they can tolerate relatively high weed infestation compared to, for example, row crops. High potential for cost savings and decreased risk of yield loss therefore exist when accurate weed-mapping and site-specific herbicide application are used. For example, Rew et al. (1996) reported the potential reduction in herbicide use from patch spraying of *Elytrigia repens* (L.) Nevski to be as much as 97% compared with whole-field application. Nordmeyer (2006) found in five cereal fields that herbicide treatment was needed for grass weeds on 39% of the area, for *Galium aparine* on 49%, and for other broadleaved species on 44%. Hamouz et al. (2013) calculated herbicide savings in winter wheat ranging from 15.6 to 100% according to the herbicide and application thresholds used. Although herbicide savings alone would not be sufficient to justify the increased costs of weed-mapping, if potential injuries to crops also are taken into account, then the profitability of SSWM may be considerably higher.

The main mechanism of herbicide selectivity consists in the differential metabolism between weeds and crops (Devine et al., 1993; Cole, 1994). Herbicide detoxification is achieved by enzymes which carry out two major functions. First, they alter the chemical structure of the herbicide into that of a biologically inert compound, and second, they invert the react-

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ibility and polarity of the herbicide so that it can be removed from the cytoplasm and stored in vacuoles or bound to the cell walls (Cobb, Reade, 2010). The metabolism of herbicides is usually classified into four main phases, as described by such authors as Yuan et al. (2007). Generally in these processes, the sequential reactions of several enzymes are required. The actions most commonly involved are oxidations by P450s and hydrolyses by carboxylesterases (phase I reactions) (Kreuz et al., 1996) as well as conjugations catalyzed by glutathione-*S*-transferases (GST) or UDP-dependent glycosyltransferases (phase II) (Dixon et al., 2009). Further detoxification involves transporting the conjugated molecule into the vacuole or extracellular space by active transport (phase III) and further degradation with P450S, GSTs, glycosyltransferases and other enzymes (phase IV). It seems that in wheat GSTs predominate in detoxification processes (Dixon et al., 1998). Cummins et al. (2001) found a large number of diverse proteins in wheat capable of hydrolyzing herbicide esters and that these activities differed from those in competing grass weeds. Devine et al. (1993) determined that herbicide selectivity is derived not only from differences in metabolism but also from differences among plants in interception and uptake of herbicides, in sensitivity of the target enzyme (e.g. site of action), and in tolerating product phytotoxicity. Moreover, this selectivity can be affected by product characteristics, application timing, use of safeners, and other factors. The actual phytotoxic effect of herbicides or their combinations on a crop is therefore the result of numerous internal and external influencing factors.

There is evidence of some negative effect of herbicides on cereal crops. Wagner (2004) found that applying the herbicide imazamethabenz at the full rate (i.e. 1 × the label rate) caused some injury to the wheat crop compared to a lower herbicide rate (75% of the full rate). Dastgheib et al. (1994) found that shoot dry weight of some wheat cultivars was significantly reduced by applying 15 g of chlorsulfuron 43 days after treatment. Decrease in grain yield was significant only for a 60 g dose. Wells (2008a) reported 3% wheat crop injury and 2% grain yield loss after treatment with florasulam + clopyralid (5 + 30 g ha^{-1}) and 5% yield loss for a combination of metosulam, clopyralid, and MCPA herbicides. Wells (2008b) reported 2, 1, and 3% yield losses after applying pyroxsulam (30 g ha^{-1}), mesosulfuron (20 g ha^{-1}), and iodosulfuron (20 g ha^{-1}), respectively, compared to an untreated weed-free control. Rengel, Wheal (1997) demonstrated that chlorsulfuron herbicide can decrease growth of fine roots and micronutrient uptake in some wheat genotypes. Sikkema et al. (2007) reported visible injury to a wheat crop of as much as 5.7% when evaluated 42 days after treatment by an herbicide combination of dicamba, MCPA, and mecoprop. Kong et al. (2009) found that grain yield was reduced due to mesosulfuron application without safener by 2.7–10.1% for varieties of hard winter wheat and by 1.6–6.6% for soft winter wheat. Actual loss depended on wheat variety and treatment date. Ritter et al. (2008) reported 0.71 t ha^{-1} yield loss in winter wheat caused by treatment with 1250 g of isoproturon.

On the other hand, Geminiani et al. (2008) observed no yield reductions in various cultivars of soft wheat after application of the following post-emergence herbicides and their combinations: iodosulfuron, fenoxaprop-P-ethyl, mefenpyr-diethyl, mesosulfuron-methyl, tribenuron-methyl, clodinafop-propargyl, cloquintocet-mexyl, and florasulam. Only temporary symptoms of phytotoxicity were occasionally recorded. Pasquer et al. (2006) found that herbicides affect gene expression in crops. Compounds could trigger a systemic acquired resistance-like response and might therefore even be beneficial for crops in their responding against pathogen attack. When applied in low doses, herbicides may even have stimulatory effect on the crop, known as hormesis (e.g. Cedergreen, 2008; Belz et al., 2011).

The present work is focused on determining the crop response to herbicides and their combinations which had been used for experimental SSWM in a previous study (Hamouz et al., 2013) when applied to winter wheat.

MATERIAL AND METHODS

A small-plot field trial was conducted near Kolín (Central Bohemia, 49.999´N, 15.166´E) during 2014 to examine the effects of herbicides on winter wheat yields. A nearly weed-free area of 60 × 85 m with a visually homogeneous wheat stand was selected in an ordinary wheat field during early spring. Mean annual air temperature at the experimental site is 9.0°C and mean annual precipitation is 560 mm. Elevation is 320 m a.s.l. The soil type is modal Greyzem on loess. Crop growth stage was 22 on the BBCH scale. Sporadic weed infestation had occurred by BBCH stages 12–13 and it had negligible competitive effect on the crop during autumn and winter. Weeds were carefully removed by hoe immediately after selection of the trial area. Occasional weeds germinating later during the vegetation were also removed manually. In this way, a completely weed-free crop was ensured for herbicide testing.

The trial area was split into an array of 24 × 4 plots, each 40 m^2 in size (20 × 2 m) and with 0.2 m plot separation. A total of 96 plots allowed for testing 24 treatments in four replications. Distribution of treatments across all blocks was randomized. Treatment 1 consisted of an untreated weed-free control, whereas the other treatments comprised applications of the following herbicides and their combinations: metsul-

Table 1: Specification of herbicide and carrier combinations in individual treatments, phytotoxicity estimations and crop yields

Treatment	Herbicide				Mean phytotoxicity estimation (%)	Mean crop yield (kg ha^{-1})	Standard deviation for yield (kg ha^{-1})	Yield relative to treatment 1 (%)
	metsulfuron-ethyl + tribenuron-methyl	pinoxaden	fluroxypyr	clopyralid				
1	-	-	-	-	0	9149	143	100.0
2	+	-	-	-	0	8979	298	98.1
3	-	+	-	-	0.75	8957	324	97.9
4	-	-	+	-	1.25	9028	285	98.7
5	-	-	-	+	1.5	9000	430	98.4
6	+	+	-	-	0.75	8946	419	97.8
7	+	-	+	-	0	9122	428	99.7
8	+	-	-	+	1.25	9028	466	98.7
9	-	+	+	-	0	9077	518	99.2
10	-	+	-	+	1.75	8864	229	96.9
11	-	-	+	+	1.5	9181	192	100.3
12	+	+	+	-	0.75	9231	308	100.9
13	+	+	-	+	1.5	9025	474	98.6
14	+	-	+	+	1.25	9095	489	99.4
15	-	+	+	+	1.5	9037	526	98.8
16	+	+	+	+	1.75	8756	479	95.7
17	- (U)**	-	-	-	1.5	9005	390	98.4
18	+ (U)*	-	-	-	1.25	9307	269	101.7
19	-	+ (U)*	-	-	2	9103	364	99.5
20	-	-	+ (U)*	-	1.75	8951	250	97.8
21	-	-	-	+ (U)*	3.25***	8764	332	95.8
22	+	-	+ (U)*	-	2.25	8969	172	98.0
23	-	+ (U)*	-	+	3.5***	8876	623	97.0
24	+	+	+ (U)*	+	4***	8668	584	94.7

[*] For herbicide applications marked with (U), liquid UAN fertilizer was used as the carrier instead of water.

[**] In treatment 17, only liquid UAN fertilizer was applied without herbicides.

[***] Significant difference at probability level α = 0.05

furon-methyl + tribenuron-methyl (4.95 + 9.99 g ha^{-1}), pinoxaden (30 g ha^{-1}), fluroxypyr (175 g ha^{-1}), and clopyralid (120 g ha^{-1}). All applied rates correspond with recommended rates in the Czech Republic. Water (250 l ha^{-1}) or a urea-ammonium nitrate solution (UAN; 39% N w/v., 120.5 l ha^{-1}) was used as the herbicide carrier. All treatments are summarized in Table 1. Metsulfuron-methyl + tribenuron-methyl and fluroxypyr were applied on 10 April, 2014 with an interval of 2 h at the crop growth stage 29 on the BBCH scale. Pinoxaden and clopyralid were applied on 13 April, 2014 with the same interval at BBCH stage 31. Herbicides were applied with a small-plot wheel sprayer equipped with a compressed-air spraying system and speedometer. Weather conditions were cloudy and generally suitable for post-emergence herbicide application. Air temperatures were 10°C and 16°C for the first and second treatment dates, respectively.

Winter wheat variety Magister was sown in October 2013. Ploughing and seedbed preparation had been performed before seeding. Calcium ammonium nitrate (CAN; 27% N) was used as nitrogen fertilizer in three separate applications. Total N applied in spring was 128 kg ha^{-1}. The second CAN application was skipped in treatments 17–24 to compensate precisely for the nitrogen applied in the UAN fertilizer. Other spring operations, such as application of prothioconazole (93.75 g ha^{-1}) + tebuconazole (93.75 g ha^{-1}) fungicide, were performed uniformly for the entire experimental field.

Phytotoxic effect was evaluated visually in relation to the untreated plot 30 days after treatment (DAT). Trial plots were harvested using a small-plot combine harvester and grain weight and moisture were measured immediately after harvest. Grain yield was *standardized* for a *moisture* content of *12*%. One-way ANOVA

Table 2. Effect of individual factors on the wheat yield - univariate tests of significance (standard error of the estimate = 375.04)

Effect	Sum of Squares	*F*	*P*-value
Intercept	6.545E + 09	46532.93	0.0000
Carrier	2.041E + 05	1.45	0.2315
Metsulfuron-ethyl + tribenuron-methyl	0.000E – 01	0.00	1.0000
Pinoxaden	1.365E + 05	0.97	0.3272
Fluroxypyr	1.459E + 02	0.00	0.9744
Clopyralid	3.674E + 05	2.61	0.1096
Error	1.266E + 07		

with Duncan's post hoc test and main-effects ANOVA were used in the statistical analyses. All statistical analyses were performed using STATISTICA software (Version 12, 2013).

RESULTS

Herbicide effect estimation performed 30 DAT revealed negligible visible injury in herbicide treatments 2–16, where no UAN had been applied. Small differences in the height of the crop stand were observed only in these treatments. Mean values of crop injury ranged between 0 and 1.75%. In treatments 17–24, slight chlorosis was also visible, but only treatments 21 (UAN + clopyralid), 23 (UAN + pinoxaden + clopyralid), and 24 (UAN + all herbicides) were found to be significantly different from the untreated control. Percentage estimated crop injury for all treatments is shown in Table 1.

Mean grain yield of winter wheat varied from 8668 kg ha^{-1} to 9307 kg ha^{-1} among treatments. Treatment 1 (untreated control) provided a mean yield of 9149 kg ha^{-1}. The highest yield (9307 kg ha^{-1}) was achieved by treatment 18 where metsulfuron-ethyl + tribenuron-methyl with UAN were applied. The lowest gain yield was found for treatments 16 and 24, where all herbicides were applied. Treatments with UAN application showed mean yield of 8955 kg ha^{-1}, whereas other treatments provided a mean of 9030 kg ha^{-1}. One-way ANOVA showed no statistically significant differences in crop yield (P = 0.934). Therefore, no further analyses of differences between individual treatments were warranted. Crop yields for all treatments are summarized in Table 1 and Fig. 1.

Main-effects ANOVA revealed no significant effect on crop yield at a significance level of α = 0.05. The nearest to significant value of P = 0.110 was found for clopyralid. Significance values for all analyzed effects are summarized in Table 2.

DISCUSSION

Crop injury caused by herbicides was very low at 30 DAT in single herbicide treatments and did not exceed 1.75%. This result was expected, as only herbicides registered for use in wheat were applied and it is in accordance with findings from other studies. For example, Hofer et al. (2006) reported 1.5% average phytotoxicity after pinoxaden treatment at the recommended rate. Time of assessment was not specified in their work, however. Crop injury caused by herbicide combinations was slightly higher in some cases, but it was still negligible as subjectively assessed by growth reduction.

The negative effect of UAN alone as well as in combinations with herbicides found in this study is relatively common in practice and often reported in the literature. Stahlman et al. (1997) reported that urea-ammonium nitrate (112 l ha^{-1}) alone or as an her-

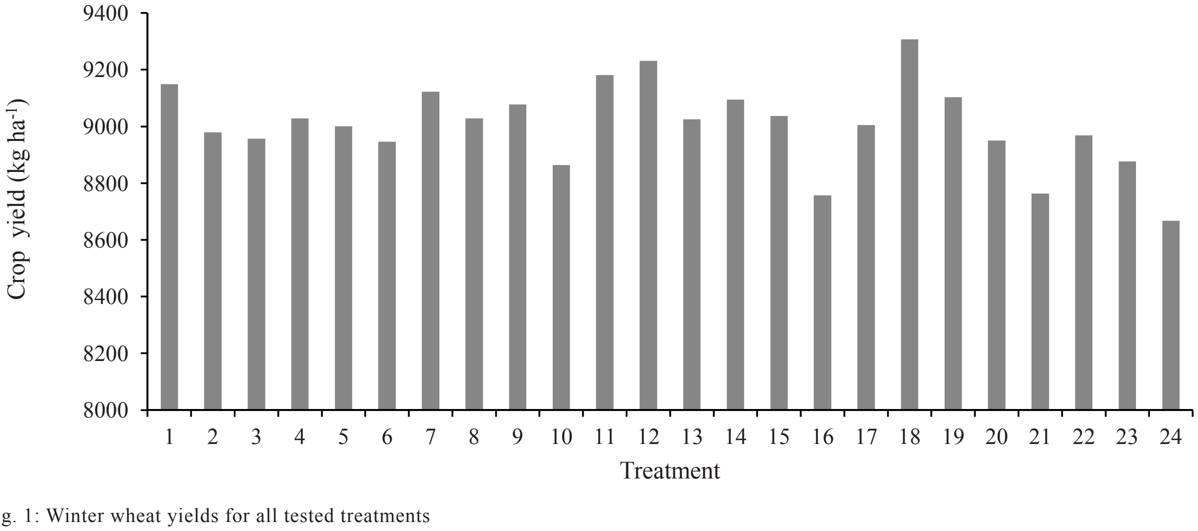

Fig. 1: Winter wheat yields for all tested treatments

bicide carrier caused moderate to severe foliar injury of winter wheat, and especially when it was combined with triasulfuron and 2,4-D herbicides. This injury was most evident 4–7 DAT and disappeared within 2–3 weeks. Sosnoskie et al. (2009) noted severe wheat injury (up to 40%) when UAN was combined with mesosulfuron application. However, wheat injury did not exceed 8% when UAN was applied at least 14 days after herbicide treatment.

Although yield differences among treatments reached a maximum of 639.6 kg ha^{-1}, they were not significant at $\alpha = 0.05$. The lack of significance of the results was partially caused by high intra-group variance, which may be accounted for in soil variation. Despite this, some clear tendencies are traceable in the acquired data. Both treatments within which all herbicides were applied (16, 24) showed the lowest grain yields. The possible negative effect of clopyralid herbicide found in this study can be compared with the findings of O'Sullivan, Kossatz (1984). They reported statistically significant yield losses in wheat when clopyralid was sprayed at rates exceeding 300 g ha^{-1}. No effect of the carrier (UAN, water) on crop yield was evident (Table 2) despite the aforementioned visible crop injury. This is also a common finding in the literature (e.g. Stahlman et al., 1997) and farming practice. Some yield loss is more probable, however, if UAN is combined with several herbicides or is applied at a time close to herbicide treatments.

CONCLUSION

Based on the presented results and the results of other studies, it can be expected that SSWM not only will provide herbicide savings, but it also will help decrease the negative impact of herbicides on cereal crops – especially in the cases when multiple herbicides need to be applied to a crop or when some herbicides are applied using a UAN carrier. These economic effects may compensate for the expensive and time-consuming weed-mapping procedure needed for reliable SSWM. The achieved results cannot be fully generalized, however, because influences from such other factors as crop variety and weather conditions can be expected.

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