

EFFICACY OF EBDC AND COPPER FUNGICIDES FOR CONTROL OF *CERCOSPORA BETICOLA* IN SUGARBEET

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Integrated disease management practices such as crop rotation, tillage to reduce inoculum levels, and use of tolerant cultivars are critically important for managing *Cercospora* leaf spot (CLS). Nonetheless, no substitute is available for fungicides for interrupting CLS epidemics in growers' fields. With only infrequent exceptions, CLS appears during July for Minnesota and North Dakota growers in the Red River Valley (RRV). Currently, fungicides are used at full rates, rotated for mode of action, and often used in mixtures for best efficacy and to minimize the development of resistance in the pathogen (Khan and Hakk, 2015). Specific mode of action fungicides tend to be at high risk for developing resistance. This is the case for each of the fungicides normally used to control CLS with the frequency of observed resistance rising in growers' fields (Secor et al., 2015). Multi-site fungicides or fungicides that are not target-site specific include EBDC (ethylenebisdithiocarbamate) and copper fungicides (Fungicide Resistance Action Committee, 2015). Their activity is non-systemic, functioning as protectants. For these fungicides, development of resistance is not expected or will take substantial time.

Fungicides are used in mixtures to improve disease control, increase surety of control when fungicide resistance is present and to help delay the development of resistance in the pathogen (Karadimos and Karaoglanidis, 2006). Components of mixtures may be used to advantage when additive or synergistic interactions among fungicides can result in more potent disease control. Alternatively, performance characteristics can be combined to increase effectiveness; for example, curative plus preventative, or systematic plus non-systematic. When mixtures are used for resistance management it is important that at least two components have activity against the pathogen. That is if only one component of a mixture provides control (with the pathogen resistant to the other components), the selection pressure toward development of resistance will be similar to the solo use of the active component.

Mixing a specific mode of action fungicide that is at high risk for developing resistance with a low risk multi-site fungicide is often recognized as a favorite strategy for managing development of fungicide resistance (Fungicide Resistance Action Committee, 2010). Current information regarding the CLS disease control associated with various mancozeb (EBDC) and copper fungicide formulations will facilitate their planned use as the multi-site fungicide component in mixtures.

OBJECTIVES

The trial objective was to determine efficacy of five copper formulations, three mancozeb formulations and a formulation containing both mancozeb and copper for 1) control of CLS disease on sugarbeet and 2) effect on harvestable root yield and quality.

MATERIALS AND METHODS

The trial was established at the University of Minnesota, Northwest Research and Outreach Center (NWROC), Crookston on a Wheatville very sandy loam soil. Primed seed (Xbeet) of the variety Crystal 981RR was planted on April 30. The seeding rate was approximating 100,000 seeds per acre. After emergence, plants were removed to realize a 4.5 inch plant spacing within a row. Liquid starter fertilizer (macro nutrient analysis 10-34-0) was applied in-furrow at 3 gallons per acre. Counter 20G (8 lb per acre) was applied over-the-row for control of root maggot. Root diseases control included both the seed treatments Kabina ST and Tachigaren (45 g.) and a post-emergence broadcast application of Quadris on June 10 to supplement the *Rhizoctonia* crown and root rot (RCRR) control; 15.5 fluid ounces per acre Quadris applied with 11002 flat fan nozzles calibrated to deliver 11.2 gallons of water per acre at 40 psi. Weeds were controlled with three applications of glyphosate; a 4.5 lb active ingredient per gallon glyphosate product was applied at 22 fl ounces per acre on May 28, June 11 and June 29.

The experimental design was a randomized complete block with four replicates. A plot comprised six 35-foot rows spaced 22 inches apart. Plots were inoculated on July 16 and 17 (Fig. 1) with a mixture of talc and ground CLS-infested sugar beet leaves. On July 16, rows one and six were inoculated with inoculum tracing from an infected-leaf collection during October, 2014. On July 17, rows two to five were inoculated with older inoculum from an unknown source. Fungicide treatments were applied within plots to rows 2 through 5 at rates indicated in Table 1 on the dates July 30, August 13, and August 26. A uniform spray pattern was accomplished using a tractor mounted 3-point sprayer with 11002 flat fan nozzles calibrated to deliver 19.8 gallons of water/A at 100 psi.

CLS severity was rated using a modification of a 1 to 10 scale described by Jones and Windels (1991). For the modified scale 0 equals no disease, 1 indicating a 0.1% disease severity (the observation of 1 to 5 spots per leaf), 4 approximates a level expected to result in economic loss and 9 equals necrotic leaves with regrowth initiating. Rows 2 to 5 were assessed for CLS disease severity on August 7, 10, 19, and 27, and on September 18 and 25. CLS scores were transformed according to the equation $\log^{(\text{score} + 1)}$. The transformed values were subjected to analysis of variance using SAS (SAS Institute, Cary, NC). Reported means are the means from the analyses restored to the original scale using the equation $10^{(\text{mean})} - 1$.

Row 3 and 4 in each plot were mechanically defoliated and harvested on September 28. Weight data for root yield per plot was collected. A sample of 10 representative roots from each plot was analyzed for quality traits at the American Crystal Sugar Company Quality Tare Laboratory, East Grand Forks, MN.

Yield and quality data were subjected to analysis of variance using SAS (SAS Institute, Cary, NC). The traits analyzed are as follows:

- Field weight - Tare adjusted field weight equals pounds of roots harvested from rows 3 and 4 and reduced by 1.6 %, the average tare for this trial.
- Tons/Acre – the product of tare adjusted field weight and 0.169714.
- Sugar - percent sugar reported from analyses at the American Crystal Sugar Company Quality Tare Laboratory.
- SLM - percent of sugar lost to molasses reported from analyses at the American Crystal Sugar Company Quality Tare Laboratory.
- Recovered sugar per ton (RST) – (Sugar % minus SLM %) X 20.
- Recovered sugar per acre (RSA) – the product of RST and tons/A.

RESULTS

Planting was on April 30; seed went into warm moist soil that had only a moderate level of moisture in the lower soil strata. As a reference, the top 60 inches of soils held only 5.37 inches of water on April 18, only 45 % of field capacity (above 100 % gravity and drainage can move water). Emergence appeared uniform. Precipitation for May and June was 2.56 and 3.70 inches respectively, largely equivalent to their 30-year averages of 2.96 and 3.74. July precipitation was above the 30-year average; i.e., 4.96 compared with 3.08 inches. Soil moisture levels rose as a result, but to only 70 % of field capacity. Levels dropped below 45 % by August and with only 1.0 inches of precipitation in August and 0.27 inches in September, levels approached 40 % by harvest. Only very few observations of RRCCR-disease-affected plants were noted; disease severity was judged too low to warrant data collection.

DIV values determined from the number of hours with relatively high humidity and temperature indicated favorable conditions for CLS disease development during July 12 to 19 (Cercospora Daily Infection Values reported by the North Dakota Agriculture Weather Network). Plots were inoculated on July 16 and 17 to use these environmental conditions to advantage. CLS lesions appeared on leaves in rows 1 and 6 within seven to 10 days. Inoculum applied to rows 2 to 5 was at least a two years older and apparently less vigorous. Only a modest number of lesions developed in these rows relative to rows 1 and 6. Fourteen days were judged moderately conducive and only two days conducive to CLS development during the balance of July and August.

Table 1. Nine protectant fungicides were evaluated for efficacy against the foliar-disease pathogen *Cercospora beticola*. July 30 was the first application date, with repeated applications on August 13 and 26. Fungicide applications included Preference non-ionic surfactant at 0.13 % v/v. Entries 1 and 11 are an untreated and treated check, respectively.

Product name	Formulation	Active ingredient	Rate per acre	pH [†]	Cu [‡]
1. Untreated check		Untreated	-	-	-
2. Manzate Max	flowable concentrate	mancozeb 37.0 %	1.6 qt	7.3	-
3. Koverall	water dispersible granule	mancozeb 75.0 %	2 lb	7.9	-
4. Roper DF Rainshield	dry flowable	mancozeb 75.0 %	2 lb	7.6	-
5. ManKocide	dry flowable	mancozeb 15.0 % + copper hydroxide 46.1 %	4.3 lb	8.9	30 %
6. Badge SC	suspension concentrate	copper oxychloride 17.6 % + copper hydroxide 16.4 %	4.6 pt	8.8	20 %
7. Kocide 3000 SL	dry flowable	copper hydroxide 46.1 %	2 lb	9.1	30 %
8. Cuprofix Ultra 40 Disperss DF	dry flowable	copper sulfate basic 71.1 %	3 lb	7.9	40 %
9. ChampION++	water dispersible granule	copper hydroxide 46.1 %	2 lb	8.9	30 %
10. Mastercop	aqueous solution	copper sulfate pentahydrate 21.46%	1.5 pt	5.6	5.4%
11. Topsin + Super Tin/ Eminent VP + Super Tin/ Headline	Liquid flowable + flowable/ micro emulsion + flowable/ suspension concentrate	thiophanate methyl + triphenyltin hydroxide / tetraconazole + triphenyltin hydroxide / pyraclostrobin	10 oz + 6 oz/ 7.6 oz + 6 oz/ 9 oz	- - -	- - -

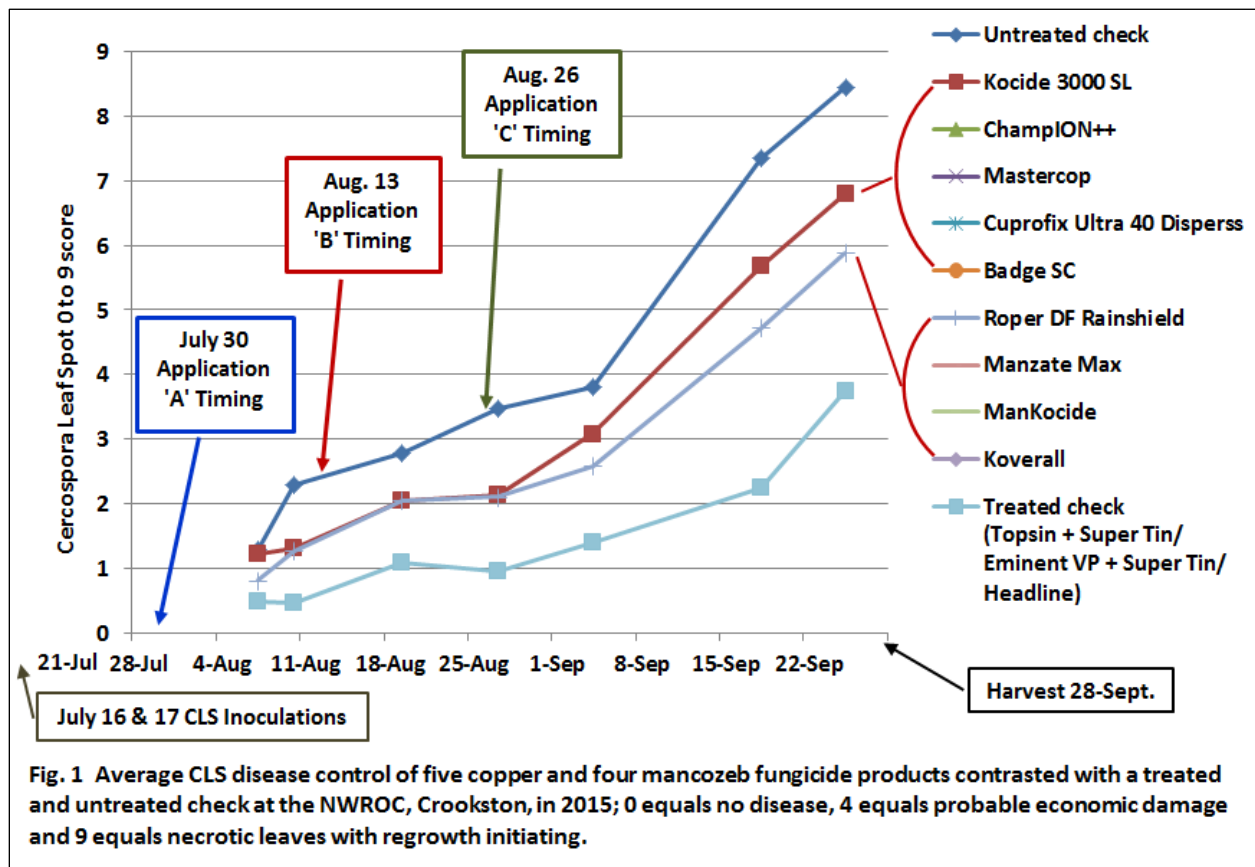
[†] Average pH of spray solution when product was mixed with 8.29 pH well water; values were not determined for the treated check.

[‡] Metallic Copper Equivalent.

Infected leaves were collected from rows 1 and 6 at the end of the growing season. Fungicide sensitivity was evaluated at the Plant Diagnostic Lab, NDSU Plant Pathology. Isolates were sensitive to Tin (triphenyl tin hydroxide), Inspire (difenoconazole) and Headline (pyraclostrobin); highly sensitive to Eminent (tetraconazole), but relatively resistant to Topsin (thiophanate methyl). Spore germination on media amended with Topsin (5 µ/ml) was 27 %; this is compared to 0 % for media amended with Tin (1.0 µ/ml) and 58 % germination for the untreated media.

Harvest data is summarized in Table 2. Weather and growing conditions provided for an average of 25.6 tons per acre. This is less than the 27.7 tons per acre estimate for American Crystal Sugar growers (the NWROC research farm is located in this grower region). The lower yield level is likely related to the droughty environmental conditions and an earlier harvest date on the NWROC research farm.

The five copper formulations, three mancozeb formulations and a formulation containing both mancozeb and copper reduced CLS disease relative to the untreated check, with control levels intermediate to the untreated check and a treated check (Tin plus Topsin/ Eminent plus Tin/ Headline). The mancozeb fungicides demonstrated significantly better CLS control than the coppers (p=0.001). Specifically, average CLS scores for the mancozeb fungicides approached 5.9 late in September (Fig. 1). The average for the coppers approached 6.8 with 8.5 for the untreated check and 3.8 for the treated. Significant differences for CLS control were demonstrated among the mancozeb fungicides and among the coppers (Table 2). Tons per acre root yield and recovered sugar per acre was lowest for the untreated check, although differences are not statistically significant.



DISCUSSION

Mancozeb and copper formulations might be best characterized as multi-site inhibitors. They are non-systemic, preventive fungicides that form a protectant barrier on the plant surface inhibiting pathogen development prior to penetration into the tissue (Gisi and Sierotzki, 2008). They interact with biochemical pathways in the pathogen in a non-specific manner, affecting many biochemical steps. One result is for enzymes to be inactivated leading to a

Table 2. Performance averages for Cercospora leaf spot (CLS) score on September 25 and for six root yield and quality traits.

Treatments	CLS score [†]	Field weight	Tons /A	RSA	RST	Sugar	SLM [†]
	0 - 9 [‡]	lb.	tons	lb./acre	lb./ton	%	%
1. Untreated check	8.21 ^a	144.7	24.55	7733	315	17.25	1.50 ^{ab}
2. Manzate Max	5.67 ^{ef}	155.0	26.30	8352	317	17.33	1.49 ^{ab}
3. Koverall	5.37 ^f	147.0	24.95	7855	316	17.43	1.64 ^a
4. Roper DF Rainshield	6.49 ^{bcd}	154.7	26.26	8483	323	17.58	1.41 ^{ab}
5. ManKocide	5.99 ^{def}	154.9	26.28	8515	326	17.78	1.50 ^{ab}
6. Badge SC	6.24 ^{cdef}	152.3	25.84	8146	315	17.18	1.44 ^{ab}
7. Kocide 3000 SL	7.28 ^{ab}	154.4	26.20	8575	328	17.78	1.39 ^b
8. Cuprofix Ultra 40 Disperss	6.39 ^{bcd}	145.8	24.74	7810	316	17.35	1.56 ^{ab}
9. ChampION++	7.18 ^{abc}	147.4	25.01	8172	327	17.80	1.44 ^{ab}
10. Mastercop [§]	6.69 ^{bcd}	-	-	-	-	-	-
11. Treated check	3.82 ^g	153.1	25.99	8446	325	17.65	1.39 ^b
Mean	6.34	150.9	25.61	8209	321	17.51	1.48

[†] Means within the same column sharing the same superscript letter do not differ significantly.

[‡] CLS disease was scored on a 0 to 9 scale using a modification of a 1 to 10 scale described by Jones and Windels (1991): 0 equals no disease, 1 indicating a 0.1% severity (an observation of 1 to 5 spots per leaf), 4 approximates a level expected to result in economic loss and 9 equals necrotic leaves with regrowth initiating.

[§] No yield and quality data available.

general disruption of metabolism and cell integrity. Based on the multi-site mode of action, resistance to such inhibitors has never developed and is unlikely to appear.

Manzate Max, Koverall, and Roper DF Rainshield all contain the active ingredient mancozeb, a coordination product of zinc ion and manganese ethylene bisdithiocarbamate. Mancozeb products differ for percent active ingredient and whether they were formulated as a flowable concentrate, dry flowable or dispersible granule (Table 1). In contrast, copper fungicides have been developed utilizing several different chemical compounds. In general terms, the copper compounds in fungicides have relatively low solubility in water (Gisi and Sierotzki, 2008). On the leaf, copper ions are gradually released from the compounds, usually with each leaf wetting. The free copper ions denature proteins, thereby destroying enzymes and in turn kill living cells. Effectiveness of a fungicide is often expected to be correlated with percent elemental copper and how the product was prepared. The potency of a copper fungicide, which effectively is the concentration of free copper ions, might be adjusted by whether the product is ground fine or less fine and the percent of copper.

Copper ion concentration also needs to be limited to avoid any entrance into plant tissue that would lead to plant injury (Zitter and Rosenberger, 2013). Copper compounds differ for relative solubility. The more soluble compounds release copper ions at a higher concentration. Hydroxides are more soluble than oxychlorides which are more soluble than tribasic copper sulphates and cuprous. Solubility also increases under acidic conditions. Copper sprays will become more phytotoxic if they are applied in an acidic solution. Often copper products are formulated

to be almost insoluble in water at pH 7.0. As the pH of water decreases the solubility of a copper fungicide increases and more copper ions are released. If a water carrier is too acidic excessive amounts of copper ions could be produced which may cause damage to plant tissues.

The copper fungicide products evaluated in this trial were not created equal. Badge SC, Kocide 3000 SL, Cuprofix Ultra 40 Dispers DF, ChampION++, and Mastercop differ for the copper compounds utilized, percent metallic copper equivalent, and the spray solution pH when mixed with a water carrier (Table 1). Each product might be judged as a unique strategy for delivering a control concentration of copper ions to the surface of plant tissues. Mastercop appears most distinct based on a low percent of copper and the low pH in solution. Nonetheless, Mastercop provided CLS control on par with the other copper products. The pH of the NWROC well-water carrier was 8.29; likely water with a pH closer to 7.0 is more commonly used for fungicide applications. Utilizing a more acidic water source could easily alter and may increase the level of CLS control from these copper fungicides. This could potentially change how the five rank for disease control. A more acidic water source would also increase the risk of plant tissue damage. Further research is needed that looks at how mixtures involving various copper formulations perform in spray equipment and for disease control.

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