Wheat straw placement effects on total phenolic compounds in soil and corn seedling growth

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Opoku, G., Vyn, T. J. and Voroney, R. P. 1997. **Wheat straw placement effects on total phenolic compounds in soil and corn seedling growth**. Can. J. Plant Sci. 77: 301–305. The effects of placement of wheat (*Triticum aestivum* L.) straw on the release of phenolic compounds potentially phytotoxic to corn (*Zea mays* L.) seedling growth were examined in plastic pots under controlled conditions in growth cabinets. The treatments were soil only (control), soil with straw only in the top 2.5 cm, and soil with mixed straw. Each of these treatments was either sown or not sown with corn. Corn radicle length and seedling biomass were significantly reduced, relative to other treatments, when wheat straw was present only in the top layer of soil. Water extracts from soil with surface straw had higher amounts of total phenolic compounds than the other treatments at each of the sampling times. The concentration of phenolic compounds increased with time, with average amounts at 14 and 28 d being 53 and 77%, respectively, of the concentration at 42 d. The presence of corn reduced the amount of phenolic compounds at 14 and 42 d compared with when corn was absent ($P = 0.01$). Water extracts prepared from soil after 42 d from all the treatments were used to investigate germination of corn seeds. Germination of corn seeds and radicle extension were inhibited by water extracts from all the above soils at 42 d compared with the control (deionized water). Coleoptile length was inhibited when concentration of phenolic compounds exceeded 10 ppm. Soil NO_3-N differences among the treatments were insignificant at the various sampling times. This indicated that the observed corn growth differences were mainly due to the presence of phenolic compounds.

Key words: Wheat (winter), straw replacement, phenolic acids, seedling, allelopathy

Opoku, G., Vyn, T. J. et Voroney, R. P. 1997. **Effets du mode d'enfouissement de la paille de blé sur les composés phénoliques totaux dans le sol et sur la croissance du maïs au stade jeune**. Can. J. Plant Sci. 77: 301–305. Nous avons examiné en pots de plastique en enceinte de croissance les effets du placement de la paille de blé (*Triticum aestivum* L.) sur le dégagement de composés phénoliques éventuellement phytotoxiques envers les plantules de maïs (*Zea mays* L.). Les traitements comparés étaient : sol sans paille, sol avec paille seulement dans les 2,5 cm supérieurs et sol mélangé à la paille. Chacun des traitements étaient combinés ou non avec un semis de maïs. La longueur de la radicule et la biomasse de la plantule étaient significativement plus basses dans le traitement avec paille seulement dans la couche supérieure du sol que dans les deux autres traitements. L'extrait aqueux du sol dans le premier traitement contenait de plus fortes quantités de substances phénoliques totales à toutes les dates de prélèvement. Les concentrations de substances phénoliques augmentaient en fonction du temps, donnant à 14 et à 28 jours des quantités moyennes respectives de 53 et 77 % des valeurs mesurées à 42 jours. La présence du maïs dans le sol abaissait les quantités de substances phénologiques à 14 et à 42 j par rapport au sol non cultivé ($P = 0{,}01$). Les extraits aqueux du sol obtenus dans tous les traitements à 42 j ont ensuite servi à examiner la germination du maïs. La germination du grain et l'élongation de la radicule étaient inhibées dans les extraits aqueux de tous les sols par rapport aux sols témoins (eau désionisée). L'élongation du coléoptile était inhibée en présence de concentrations des substances phénoliques excédant 10 ppm. Les concentrations de N nitrique dans le sol étaient sensiblement les mêmes dans tous les traitements aux diverses dates de prélèvement. Les différences de croissance du maïs étaient donc essentiellement dues à la présence des composés phénoliques dans le sol.

Mots clés: Blé d'hiver, enfouissement de la paille, acide phénolique, maïs, plantule, allélopathie

A major problem limiting the acceptance of a no-till system for corn production in rotation with winter wheat in Ontario is that high levels of wheat residues left on the soil surface can decrease corn grain yields. The cause of the yield reduction is not clear, but studies have implicated phytotoxins released from decomposing wheat residues (Purvis 1990; Schreiber 1992; Voroney et al. 1992; Lund et al. 1993). These compounds may have inhibitory or stimulatory effects on plant growth, depending on their concentration in the soil and the sensitivity of the growing crop (Einhellig 1985). The degree of growth inhibition depends on type of residue, quantity, placement and degree of decomposition, microbial activity, nutrient status and other physical parameters (Rice 1984).

A study by Borner (1960) indicated that cold water extracts of wheat residues contained phenolic compounds which were toxic to plant growth. Some of these compounds were ferulic acid, p-coumaric acid, vanillic acid and p-hydroxybenzoic acid. Guenzi and McCalla (1966) found several phenols, including p-coumaric and ferulic acids in residues from oats, wheat, sorghum, and corn, and in soils under these crops.

Evidence for the hypothesis that phytotoxic compounds released by previous crop residues are responsible for reduced yields in a no-till system in Ontario is lacking because attempts to isolate these compounds in no-till soils have not been successful (Farquharson 1991; Wanniarachchi 1993). The objectives of our study were to determine (i) whether the placement of wheat straw affects the release of phenolic compounds, and (ii) whether the compounds released are phytotoxic to corn seedling growth.

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MATERIALS AND METHODS

The experiment was conducted in pots in controlled growth cabinets with a 16-h photoperiod and day/night air temperatures adjusted to approximately 15/8°C at 70% relative humidity. The light intensity was approximately 300 µmol m^{-2} s^{-1}. There were six treatments arranged in a randomized complete block design with six replications. The treatments were (i) soil only (control), (ii) soil with wheat straw present only in the top 2.5 cm (surface), (iii) soil mixed with wheat straw (mixed), and each of these treatments was either sown or not sown with corn. Because destructive sampling was involved, all pots with corn were in the same growth cabinet.

The soil, which was obtained from the Elora Research Station, Elora, Ontario, was classified as a Maryhill silt loam (Orthic Humic Gleysol). It contained 30% sand, 55% silt, 15% clay, and 4% organic matter and was placed in 19-cm-diameter by 20-cm-high plastic pots. Winter wheat (cv. Harus) straw (chopped straw on the ground) was collected from an experimental field at the Elora Research Station, Elora, Ontario, in the spring of 1994 and ground to pass through a 2-mm sieve. Twenty grams of the ground straw, equivalent to a field application rate of 5 Mg ha^{-1}, were either added to the surface 2.5 cm depth (treatment ii) or mixed thoroughly throughout the plastic pot (treatment iii). These treatments were selected to replicate till and no-till situations in the growth cabinet. A controlled release fertilizer (Nitricote total, 18–6–8) was applied 5 d before planting to provide 150 kg ha^{-1} of nitrogen. All plastic pots were watered uniformly and kept in the growth cabinet for 5 d before five corn seeds (hybrid Pioneer 3905) were sown at 5-cm soil depth. The plants were thinned to three seedlings per plastic pot after emergence. Watering occurred at 3-d intervals using deionized water.

Corn seedling radicle length from each of three plants per pot was measured at 7, 10, and 14 d after planting. Corn seedling dry weights were also determined at 28 and 42 d after being dried to a constant weight at 80°C. Soil samples were collected at 14, 28, and 42 d after planting (0–5 cm soil depth), and kept in a freezer until preparation for extraction of NO_3-N and phenolic compounds. Nitrate-nitrogen was extracted with 2 M KCl (Maynard and Kalra 1993) and determined using the TRAACS 800 analyzer (Alfa-Laval, AB., Stockholm, Sweden). Extractions for phenolic compounds were made with 50 g of air dried soil and 50 mL of deionized water. Each mixture was shaken on a rotary shaker for 45 min in a 250-mL polypropylene centrifuge bottle. The supernatant was filtered through Whatman #4 filter paper. The filtered extracts were kept frozen until analysis. The analytical procedure for measuring the total phenolics was similar to that of Tel and Covert (1992) using the Technicon Autoanalyzer (AA II) equipped with a 720 mm interference filter. Standard solutions of tannic acid, Folin-Denis reagent, and EDTA were used. Six plastic pots from each corn treatment were sampled at 7 d and also at 10 d without replacement, and another six pots from each of the treatments were sampled at 14, 28, and 42 d without replacement. Each sampling time was replicated six times. The whole experiment was also replicated twice (i.e. conducted in two cycles).

Water extracts from each treatment obtained after 42 d were used to test their effect on germination of corn seeds (hybrid Pioneer 3905). Ten kernels were placed in a circle, with the micropyle end oriented towards the centre, on Whatman #42 filter paper in a petri dish (110 × 15 mm). A second filter paper was placed over the seeds and 10 mL of extract was added by pipette, soaking both filter papers. The lid was placed on the dish and sealed with parafilm. Deionized water was added to control dishes. Each dish was then placed in a germination cabinet set at 25°C in light and a relative humidity of 95–100%. Percentage of seeds that germinated was determined after 4 d. The lengths of the coleoptiles and radicles of the seedlings in each dish were recorded after 3, 4, and 5 d. At 5 d, the dry weight of all the roots in each dish was recorded. Seven dishes (one from each treatment) were sampled each time without replacement. Each sampling time was replicated six times. The whole germination experiment was repeated twice (two cycles).

Each of these experiments was analyzed separately as a randomized complete block design. When there was a cycle by treatment interaction, analyses were done separately for each cycle. All treatment means were compared using the protected least significant difference (LSD) at 5% probability level. Arcsine transformation was performed on all percentage data, but since the transformation did not significantly alter the conclusions, all means are reported as untransformed values. Orthogonal single degree of freedom contrasts were performed using SAS procedures (SAS Institute, Inc. 1987).

RESULTS

Effect of Straw on Seedling Growth

Corn seedlings grown in soil mixed with straw resulted in longer radicle lengths at 7, 10, and 14 d as compared with seedlings grown in soil with surface straw (Table 1). Radicle lengths of seedlings grown in soil with surface straw were reduced by 40% after 7 d and by 30% after 10 and 14 d. Differences between seedlings grown in soil mixed with straw and soil only were insignificant at all the sampling times. Corn seedlings grown in soil mixed with straw and in soil only treatments tended to have greater biomass after 28 and 42 d as compared with those grown in soil with surface straw (Table 1). The amount of extractable NO_3-N ranged between 20 and 25 mg kg^{-1} for the 6-wk period in both cycles and differences among the treatments at each of the sampling times were insignificant. Nitrogen was therefore not limiting in this study, but corn plants grown in the surface straw treatments appeared weak, pale, and yellowish compared with those in the other treatments.

Effect of Straw on Extractable Phenolic Compounds

Wheat straw placement and corn sowing significantly affected the amounts of water-extractable total phenolic compounds. Soil with surface straw was higher in total phenolics at the various times after sowing corn in both cycles, relative to the other treatments (Table 2). The soil-only treatments (with or without corn) resulted in the least

Table 1. Effect of wheat straw placement on corn seedling radicle length and corn biomass at different times after sowing averaged over cycles

Straw placement	Radicle lengths 7 d	10 d	14 d	Corn seedling biomass 28 d	42 d
	(mm)			(g plant^{-1})	
Soil only	21.7*a*	55.8*a*	95.6*a*	0.24*ab*	0.46*a*
Surface straw	13.4*b*	41.1*b*	66.5*b*	0.17*b*	0.31*b*
Mixed straw	22.2*a*	57.5*a*	94.9*a*	0.26*a*	0.50*a*

*a,b*Within columns means followed by the same letter are not significantly different at $P = 0.05$.

amounts of phenolic compounds at all the sampling times. The total amount of phenolics in cycle 2 was higher than in cycle 1 by at least 30% at all the sampling times. When averaged over both cycles and corn treatments (sown, not sown), soil with surface straw were higher in phenolic compounds at 42 d (11.5 mg kg^{-1}) compared with soil only (5.56 mg kg^{-1}) and soil mixed with straw (6.81 mg kg^{-1}). Total phenolics in soil mixed with straw was not significantly different from soil only treatments in most cases (Table 2). Soil sown with corn tended to have lower amounts of phenolics as compared with soil without corn in both cycles; the reduction was significant at 14 and 42 d (Table 2).

Germination Studies in Petri Dishes

Coleoptile lengths were either unaffected or very slightly stimulated at the various times in cycle 1 by water extracts (Table 3). The least stimulation was associated with extracts from surface straw treatments and, by 5 d, coleoptile lengths were only 100 and 101% of control (deionized water) as compared with a maximum stimulation of 5% for soil alone with corn. In cycle 2, surface straw inhibited coleoptile length by 3 to 17% compared with soil alone or when straw was thoroughly mixed with soil (Table 3). Coleoptile length was either unaffected or slightly inhibited (by 2%) with extracts from soil only or soil with mixed straw treatments. Differences among all the treatments in cycle 2 were insignificant at 3 d. By 5 d, significant differences existed between surface straw (with or without corn) and all the other treatments.

Extracts from soil with surface straw (with or without corn) significantly reduced corn germination compared to the other treatments (Table 4). There was no difference in corn germination among extracts from soil only and soil mixed with straw treatments. Extracts from all the treatments inhibited corn radicle length at 3, 4 and 5 d averaged over cycles (Table 4). Corn radicle length was inhibited by 12 to 16% at 3 d, but differences among the treatments were insignificant. At 4 and 5 d, radicle inhibition by extracts from soil with surface straw (without corn) was significantly more than with the other treatments. Averaged over cycles, extracts from the surface straw only treatment resulted in the lowest root dry weight relative to the other treatments (Table 4). By 5 d, root dry weight for surface straw was 70%, while that of surface straw with corn was 75%, compared with 85% or more for all other treatments (expressed as percent of control).

Table 2. Effect of wheat straw placement on the amount of total phenolics at different times after planting in cycles 1 and 2

Treatment	Days after planting 14 d	28 d	42 d
Cycle 1		(mg kg^{-1})	
Soil only + corn	2.61*b*	4.68*bc*	5.15*d*
Surface straw + corn	3.02*a*	5.11*b*	7.93*b*
Mixed straw + corn	2.88*b*	4.78*bc*	6.41*c*
Soil only	2.96*b*	4.05*c*	5.59*cd*
Surface straw	4.30*a*	6.50*a*	9.46*a*
Mixed straw	3.19*b*	4.11*c*	7.11*b*
	Levels of significance for orthogonal contrasts		
Surface vs. soil	**	**	**
Surface vs. mixed	*	**	**
Corn vs. no corn	**	NS	**
Cycle 2			
Soil only + corn	2.22*d*	4.49*b*	5.52*c*
Surface straw + corn	6.02*bc*	8.57*a*	12.68*b*
Mixed straw + corn	4.06*cd*	4.98*b*	6.48*c*
Soil only	4.38*bcd*	4.80*b*	5.97*c*
Surface straw	8.87*a*	8.99*a*	16.03*a*
Mixed straw	6.85*ab*	5.06*b*	7.21*c*
	Levels of significance for orthogonal contrasts		
Surface vs. soil	**	**	**
Surface vs. mixed	*	**	**
Corn vs. no corn	**	NS	**

*a–d*Within column means followed by the same letter are not significantly different at $P = 0.05$.
*, **Significant at $P = 0.05$ and $P = 0.01$, respectively. NS = not significant ($P = 0.05$).

Table 3. Influence of water extracts of soil on coleoptile length of corn seeds in cycles 1 and 2 expressed as percent of control

Treatment	Coleoptile length 3 d	4 d	5 d
Cycle 1		(Percent of control)	
Soil only + Corn	100*a*	101*a*	105*a*
Surface straw + Corn	99*a*	101*a*	101*ab*
Mixed straw + Corn	100*a*	103*a*	103*ab*
Soil only	100*a*	101*a*	103*ab*
Surface straw	99*a*	100*a*	100*b*
Mixed straw	99*a*	100*a*	104*ab*
Cycle 2			
Soil only + Corn	100*a*	99*a*	98*a*
Surface straw + Corn	97*a*	89*ab*	84*b*
Mixed straw + Corn	102*a*	100*a*	100*a*
Soil only	100*a*	98*a*	98*a*
Surface straw	97*a*	85*b*	81*b*
Mixed straw	99*a*	98*a*	100*a*

*a,b*Within column means followed by the same letter are not significantly different at $P = 0.05$.

DISCUSSION

Our results suggest that phenolic compounds released from wheat straw have a significant allelopathic effect on corn growth. Placement of the straw on the soil surface resulted in more extractable phenolic compounds in the top 5 cm than the other treatments. Phenolic compounds present in the straw are likely to be leached into the soil with higher amounts associated with surface straw treatments due to higher concentration of straw within the top layer of soil relative to the other treatments. There are reports that wheat

Table 4. Influence of water extracts of soil on corn germination, root dry weight and radicle length averaged over cycles expressed as percent of control

Straw placement	Germination	Root dry wt.	Radicle length		
	4 d	5 d	3 d	4 d	5 d
	(Percent of control)				
Soil only + corn	95*a*	89*a*	87*a*	88*a*	88*a*
Surface straw + corn	75*b*	83*bc*	88*a*	83*a*	75*b*
Mixed straw + corn	95*a*	86*ab*	86*a*	84*a*	85*a*
Soil only	95*a*	84*ab*	88*a*	87*a*	86*a*
Surface straw	65*c*	78*c*	84*a*	74*b*	70*c*
Mixed straw	95*a*	84*ab*	85*a*	84*a*	89*a*

*a–c*Within column means followed by the same letter are not significantly different at $P = 0.05$.

straw contains phenolic compounds (Borner 1960; Guenzi and McCalla 1966), and these compounds have shown phytotoxicity to corn seedlings (Guenzi et al. 1967; Janovicek et al. 1997). Results similar to those of our study have been obtained by other researchers. Corn root and seedling growth were reduced by 25 to 45% and 47 to 62%, respectively, when grown in water-soluble substances extracted from wheat straw (Guenzi and McCalla 1962). Guenzi et al. (1967) also reported inhibition of corn seedling growth with water extracts of weathered wheat straw after 4 wk of decomposition. Norstadt and McCalla (1968) showed that amending soils with wheat straw was associated with slower rates of corn seedling growth.

The soil used in our study contained 4% organic matter. Therefore, phenolic compounds obtained from the soil only treatment might have resulted from the breakdown of the soil organic matter (Wang et al. 1967; Waller et al. 1987). Soils with corn resulted in lower concentrations of phenolic compounds because these compounds might have been absorbed by the corn roots.

Phenolic compounds can exhibit both stimulatory and inhibitory properties, depending on their concentration in the soil solution and on the growing crop (Rice 1984; Einhellig 1985). Radicle length and root dry weight were inhibited by water extracts in our laboratory study but coleoptile length was unaffected or slightly stimulated (up to a maximum of 5%). The concentration of phenolics in our extracts was sufficient to inhibit radicle growth, but either stimulated or did not affect coleoptile growth since these compounds exhibit both stimulatory and inhibitory properties. Our findings also support results of others. Guenzi et al. (1967) reported that water extracts of wheat seeds inhibited corn root growth by 22%, but stimulated coleoptile growth by 13% while extracts of the hulls inhibited both root and shoot growth by 25 and 26%, respectively. Wanniarachchi (1993) also observed that coleoptile length of wheat was similar to the control after germination for 3 d with extracts of canola roots and stems which had incubated for 21 d. In the same study, coleoptile lengths of barley were 10% longer than the control. Treatments with surface straw inhibited coleoptile length in cycle 2, mainly due to the presence of higher amounts of phenolic compounds relative to the amounts in cycle 1.

The inhibition of root and shoot growth could be due to the ability of phenolic compounds to inhibit gibberellin and indoleacetic acid function (Geissman and Phinney 1972), cell division (Avers and Godwin 1956), and nutrient uptake (Qasem and Hill 1989). Water extracts of certain plant species caused darkening of apical meristerms and cells in elongation regions (Patrick and Koch 1958). The inhibitory effect of phenolic compounds on germination and radicle length in petri-dishes suggests that such an effect could take place early in the germination process and affect the entire phase of seedling growth. The wheat straw used in our study had been over-wintered in the field and was partially decomposed. Therefore, the amount of phenolics released might not be as high as from fresh or undecomposed straw. Voroney et al. (1992) observed that green and fresh plant materials had a greater potential for accumulation of phenolic compounds than weathered and more mature plant material.

The phytotoxic effect of wheat straw on corn seedling growth has been demonstrated in the present study. Delayed corn growth and reduced yields in reduced- and no-till systems following winter wheat could be attributed to phenolic compounds released into the seedbed during decomposition of wheat straw. This has been shown by corn response in soil with surface straw (replicating no-till) and soil mixed with straw (replicating till conditions). In order to overcome the constraints to the adoption of no-till systems, strategies need to be developed to overcome the damaging effects of wheat straw. Removing or reducing the amount of straw in the row-zone would eliminate the source of this inhibition. Further research is needed to examine the presence of other compounds in soil-water extracts phytotoxic to corn seedlings. The analytical procedure used in our study was unable to identify the various phenolic acids in the soil extracts. A more sensitive analytical procedure should be developed to make the separation and identification of low amounts of these compounds possible.

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