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**Field Studies to Evaluate Potential Differences between Bt and non-Bt Corn Residue**

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**Introduction**

Anecdotal information has continued to circulate within the agricultural community that some Bt corn hybrids may be resistant to degradation or be less desirable for forage. Implement manufacturers and dealers now market improved or alternative tillage machinery to specifically deal with “tough Bt corn residue”.

An initial report on Bt corn stalk composition indicated that the Bt corn may have higher lignin content (Saxena and Stotzky 2001). If so, that would explain the slower degradation rate. However, subsequent studies of both residue composition and litter decomposition under laboratory conditions have produced ambiguous results (Flores et al. 2005; Hopkins and Gregorich 2003; Jung and Sheaffer 2004). Effects not directly associated with the function of the inserted gene (Bt), but ostensibly related to the genetic engineering event, are termed “potential unintended effects”. Confirmed mechanisms for such effects remain elusive.

Adoption of insect-resistant corn hybrids incorporating a gene(s) producing modified Bt endotoxins may have positive benefits on soil and water conservation due to reduced pesticide

application and tillage. However, if the increase in Bt corn acreage is accompanied by increasing amounts of recalcitrant residue, there could be other potential outcomes. Increasingly aggressive tillage would intensify soil erosion and negatively affect carbon/nutrient cycles. The deposition of additional organic matter with longer residence times to the soil may provide carbon sequestration benefits, or paradoxically, increase the mineralization of soil carbon pools. Other effects might be expected with respect to the residue’s suitability for forage or biofuels production.

Given the continuing reports of corn residue persistence and the conflicting data reported in the literature, we conducted a series of studies of corn residue (Bt and non-Bt corn hybrids) composition and decomposition.

**Methods**

The decomposition rates of above-ground biomass from different corn hybrids were evaluated using the litter bag technique (Harmon et al. 1999). Subsamples of dried corn residue were placed in nylon mesh litter bags and buried so the top of the vertically placed bag was five cm below the soil surface. At subsequent intervals, blocks of litter bags were excavated and the percent residue remaining was calculated from the difference between the initial and ending weights of dry organic mass, corrected for the organic matter content of the soil entrained in each bag (ash weight x % soil organic matter). All decomposition studies were performed in the Barnes clay loam soil at the Eastern South Dakota Soil and Water Research Farm (ESDSWRF), Brookings, SD.

Compositional properties (% cellulose, hemicellulose, lignin) were determined on the residues by standard fiber analyses procedures. The mechanical (three-point bending) strength of corn stalks was determined using an Instron compression tester.

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Study #1. The decomposition rates for four hybrids with varying Bt genetics from seed one manufacturer were determined. The four hybrids were (i) DKC60-15 (non-Bt, base genetics), (ii) DKC60-16 (Bt, corn borer), (iii) DKC60-12 (Bt, corn rootworm), and (iv) DKC60-14 (stacked Bt, borer + rootworm). Residue was collected from replicate plots cultivated at the Dakota Lakes Field Station where no insect pressure was reported in 2004 (Dwayne Beck, personal communication). The residue was dried and chopped. Sixty-three litter bags were prepared for each of the four hybrids for a total of 252 litter bags (3.2-mm mesh) that were buried into incorporated spring wheat stubble November 15, 2004. Blocks of 36 bags were excavated at these intervals following burial: 134 d, 168 d, 217 d, 262 d, and 337 d, 559 d, and 658 d. The last retrieval date (658 d) for Study 1 litterbags was September 5, 2006.

Study #2a (Field). Residue from stacked (rootworm and corn borer) Bt and non-Bt hybrids were used from two seed manufacturers: Cropland 344 and 344CRW/Bt; DKC4626 (conventional) and 4625 (stacked Bt). These hybrids were cultivated in replicated plots in 2005 at the ESDSWRF with little insect pressure. Litter bags (240; 3.2 mm mesh) containing dried, chopped residue were buried within rows of non-Bt (Cropland 344) and Bt corn (Cropland 344CRW/Bt) to examine the potential for rhizosphere exudates containing the modified Bt endotoxin to influence decomposition rates. These litterbags were buried June 6, 2006 and sets of bags (80) were excavated on August 21, November 15 of 2006, and June 25 of 2007. The corn plants surrounding the litter bag study were allowed to reach maturity, the ears were hand-harvested, and the plants were left standing to senesce for the remainder of the study.

Study #2b (Greenhouse). The decomposition of cellulose filters was examined in the soil surrounding greenhouse grown Bt and non-Bt corn hybrids. Twelve plants each of CL344 and CL344CRW/Bt were raised to near maturity (R4 growth stage), killed by withholding water, and then allowed to senesce. During the growth and senescence phases, sets of cellulose filters in mini-litterbags (1-mm mesh) were removed from the potting soil for mass loss determination.

Study #3. The potential for corn rootworm damage of non-protected corn to influence residue decomposition rates was examined in this study. Above-ground biomass

from Cropland 344 and 344CRW was harvested from replicated plots at ESDSWRF with a strong natural infestation of western corn rootworm. Sixty litterbags (3.2-mm mesh; 30 litterbags for each residue) containing dried, chopped residue were buried into incorporated soybean stubble on November 8, 2006. Sets of 15 bags for each hybrid were excavated on July 2 and November 14 of 2007. Independent measures of rootworm larval abundance were made on the plots where the corn was grown.

Study #4. The potential for European Corn Borer (ECB) damage of non-protected corn to influence residue decomposition rates was examined in this study. Using hybrids from three manufacturer-maturity series (CL44, DKC46, DKC50/51), above-ground biomass from hybrids for each manufacturer-maturity series with no Bt gene (base genetics), Bt active against European Corn Borer, and Bt active against corn rootworms were harvested from replicated plots at ESDSWRF. In 2006, there was a strong natural infestation of ECB at this site. For each of the hybrids, whole stalks were collected and ECB damage assessed using the number of tunnels and total length of tunnels in the stalks. For the decomposition portion of the study, whole stalk segments were used to allow proper consideration of the stalk damage to the residue decomposition. Because whole stalk sections were used, a larger 13-mm mesh was used for the litterbags that permitted access to most soil invertebrates. Two-hundred litterbags were buried into incorporated soybean stubble and removed after about 1 year of burial.

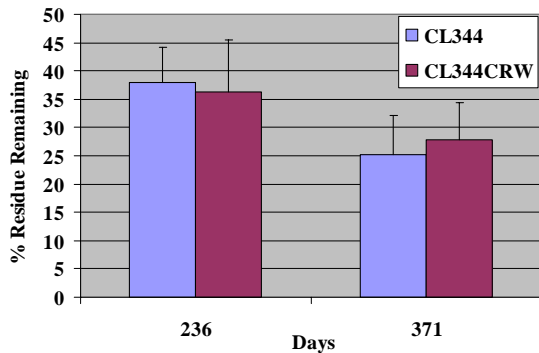
## **Results and Discussion**

Study #1. Through about 262 days, decomposition rates were fairly linear, and by the one-year mark when decomposition rates were slowing, 75% - 80% of the dry weight had been lost from the litter bags. During the second year of the study, little additional weight loss was recorded. Calculated litter half-lives (for the period up to and including 262 d) were approximately 200 days and decomposition rate constants (zero order) were about  $-0.25 \text{ day}^{-1}$  for all four hybrids. There were no significant differences (1-way ANOVA) in the amounts of residue remaining among the four corn hybrids ( $n = 9$  for each hybrid) at any sampling period. No significant differences in the lignin content for these hybrids were observed. These findings have been published (Lehman et al. 2008a).

Study #2. No significant differences in residue decomposition rates were observed

among the four hybrids. Curiously, the litter bags from all four hybrids decomposed faster in the burial zone of the CL344CRW/Bt compared to CL344. During the replicated greenhouse study, no significant differences in the decomposition of cellulose filters were observed in pots containing CL344CRW/Bt compared to CL344. No compositional differences in the residues from the four hybrids were detected. No significant differences in the mechanical strength of individual stalk internode sections were detected among the four hybrids. All data from Study #2 have been published (Lehman et al. 2008b).

Study #3. No significant differences in the rate of decomposition of chopped residues were detected between the rootworm-damaged CL344 and the rootworm-protected CL344CRW hybrids at intervals of either 236 or 371 days of burial in the soil (Fig. 1).



**Figure 1.** Decomposition of rootworm damaged CL344 and rootworm-protected CL344 CRW.

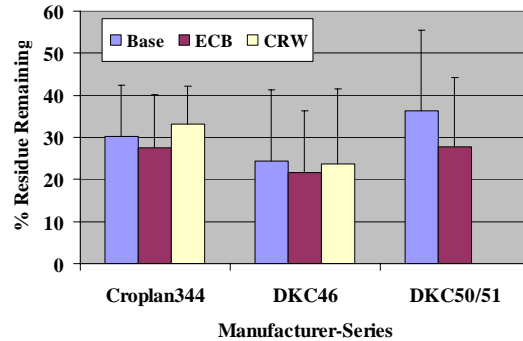
No differences in above-ground biomass were found between the two hybrids (6 plots each).

Study #4. The assessments of ECB damage to the stalks showed that ECB protected hybrids had far less damage than non-protected hybrids. However, stalk sections from the ECB-protected hybrids did not decompose more slowly than their corresponding non-protected isolines (Fig. 2).

**Conclusions**

Our approach for studies #1 - #3 using chopped litter in buried litter bags to simulate the effect of chopping and tillage operations on the residue showed that Bt-containing hybrids decomposed at rates similar to their non-Bt near isolate under these conditions. When ECB-damaged residues were tested using whole stalk sections (Study #4), Bt-protected stalk sections did not decompose more slowly than the non-

protected sections. No differences in residue composition or stalk mechanical strength were found among hybrids that could be related to the presence of Bt genes.



**Figure 2.** Decomposition of stalk internode section number two from ECB-protected and non-protected hybrids under ECB pressure.

It is possible that the apparent persistence of Bt corn residue observed by producers may be related to the overall amount of residue produced, regardless of residue compositional qualities. No consistent differences in silage yield between Bt and non-Bt hybrids were reported by Folmer et al. (2002). Similarly, Mungai et al. (2005) found no differences between Bt and non-Bt hybrids with respect to above- or below-ground biomass in a two year study, nor did we find differences in above-ground biomass in our study #3. Yields and residue amounts have been increasing for decades (Johnson et al. 2006), and the latest increase may simply have co-occurred with the introduction of Bt-containing corn hybrids

It has also been brought to our attention by producers that Bt hybrids tend to stay green longer and therefore would be tougher to cut at harvest. While that greenness at harvest does not appear to translate into differences in residue decomposition over longer terms, it may contribute to the perception of “resistant Bt residue”.

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