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ABSTRACT Reducing chemical use for suppressing internal and external parasites of livestock is essential for protecting environmental health. Although plant condensed tannins are known to suppress gastro-intestinal parasites in small ruminants, no research on the effects of tannins on external arthropod populations such as the house fly, *Musca domestica* L., have been conducted. We examined the impact of plant material containing condensed tannins on house fly development. Prairie acacia (*Acacia angustissima* (Mill.), Kuntze variety *hirta* (Nutt.) B.L. Rob.) herbage, panicled tick-clover (*Desmodium paniculatum* (L.) DC.) herbage, and quebracho (*Shinopsis balansae* Engl.) extracts were introduced at rates of 1, 3 or 5% condensed tannins/kg beef cattle, dairy cattle, and goat manure, respectively. In a second experiment, we also introduce purified catechin at 1 or 3% of dairy manure dry matter and measured its impact on house fly development. For the house flies used in these experiments, the following was recorded: percent fly emergence (PFE), average daily gain (ADG), and average fly weight (AFW). No effects ($P > 0.05$) in house fly development were measured in the caprine manure. Prairie acacia (20.9% condensed tannins) had no effect on house flies developing in either bovine manures. Tick clover (4.9% condensed tannins) had a negative effect on all three quantifiable variables of house fly development in the bovine manures, whereas quebracho extract (64.0% condensed tannins) at the 3 and 5% rate reduced fly emergence in beef manure and average daily gain in dairy manure. The application of purified catechin at 3%, but not 1%, reduced fly PFE, ADG, and AFW.

KEY WORDS *Musca domestica*, cows, goats, catechin

House flies, *Musca domestica* L., (Diptera: Muscidae) are serious pests in confined animal facilities because of their ability to transmit over 20 pathogens such as campylobacters (Sukontason et al. 2004). Traditionally, insecticides commonly are applied for suppressing house fly populations, which has in turn selected for resistance to these compounds (Sawicki and Denholm 1984). Not only are older insecticides becoming less effective, the development of "greener" philosophies for suppressing arthropod pests brings to the forefront issues related to insecticide use and resulting risks to human health and the environment (Coats 1994). Because of efforts to protect the environment, interest in developing additional natural products for fly control has grown. Common examples of insecticides derived from plant-toxin materials such as nicotine (Tomizawa and Casida 2005) and pyrethrum have been developed (Balandrin et al. 1985).

Condensed tannins are complex phenolic compounds that vary in quantity and type within different plant species and are associated with protection against insect herbivory (Forkner et al. 2004). Molecular structure, concentration, and occurrence of condensed tannins vary widely among plant species (Lowry et al. 1996). They exist primarily in broadleaf plants and rarely in grasses (Waghorn 2008). Although condensed tannins suppress gastro-intestinal nematodes in vertebrate hosts that consume them (Iqbal et al. 2007), only a few studies examined their effects on external pests such as mosquitoes (Diptera: Culicidae) (Rey et al. 1999a,b). In the case of gastro-intestinal nematodes, their specific mode of action is still under debate, but proposed theories include direct effects such as impairing nematode movement or egg hatching, as well as indirect actions such as precipitating proteins at critical times that affect host nutrition (Min and Hart 2003). For this study, the objective was to determine if treating manure with condensed tannins and feeding it to house fly larvae affected their percent fly emergence (PFE), average daily gain (ADG), and average fly weight (AFW). We looked at multiple sources of manure (dairy cattle, beef cattle, and meat goats), multiple sources of condensed tannins, and a purified condensed tannin applied at two rates.

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Table 1. Nitrogen (N), carbon (C), and condensed tannin concentrations (extractible (E), protein-bound (PB), fiber-bound (FB), and total) of three sources of condensed tannins

Plant source	Percent concn					
			Condensed tannins			
	N	C	Extractible	Protein-bound	Fiber-bound	Total
Quebracho			60.31	1.13	2.54	63.98
Panicled tick-clover	2.09	45.52	2.60	1.99	0.48	5.07
Prairie acacia	2.40	48.50	11.46	6.38	3.03	20.87

Materials and Methods

Manure Collection Procedure. Manure was collected and used as a rearing medium for house flies. Over a period of 7 d, fresh manure was collected from individual goats and steers confined to pens and fed a sorghum-sudan grass [*Sorghum sudanense* (Piper) X *S. bicolor* (L.) Moench] hay with no additional food supplements. Manure from confined cows on a balanced dairy ration was collected at a commercial dairy in Stephenville, TX. All samples were placed in individual plastic bags, labeled with collection information, and placed in a chest freezer set at −20°C.

Plants Used in Study. Tick clover, *Desmodium paniculatum* (L.) DC. and prairie acacia, *Acacia angustissima* (P. Mill.) Kuntze variety *hirta* were harvested in October 2006 from forage plots located at the Texas AgriLife Research Center, Stephenville, TX. These species were selected because they are known to contain condensed tannins (Pawelek et al. 2007). Mature leaves were harvested from plants grown under identical conditions (Windthorst fine sandy loam, no irrigation or fertilizer); these were washed with tap water to remove sediment, and all stems were removed. Leaves were placed in a forced-air oven at 55°C, dried for 48 h, and then ground through a sheer mill fitted with a 1-mm screen. Material passing through the screen was used for analysis and standard purification (Terrill et al. 1992, Wolfe et al. 2008). Quebracho, *Schinopsis balansae* Engler, (Anacardiaceae) tanning extract with no adjuvants added (Traditional Tanners, Cave Junction, OR) also was used as a treatment because of known levels of condensed tannins and availability (Paolini et al. 2003).

Purified Tannin. For the second experiment, catechin hydrate minimum 98% was purchased from Sigma-Aldrich (St. Louis, MO). Catechin was shipped on dry ice to Texas A&M University. Upon arrival, the compound was stored in a −20°C freezer until treatments were prepared. Catechin was added at 0.5 and 1.5 $g\ kg^{-1}$ to dairy manure, dry matter basis, to achieve a 1 and 3% treatment level, respectively.

Assays. Nitrogen and carbon (C) concentrations in the condensed tannins sources were assayed using an Elementar Vario Macro (Elementar, Mt. Laurel NJ) C/N analyzer (Table 1) to determine if there were differences among manures that might affect the dependent variables measured. Condensed tannins in *A. angustissima, D. paniculatum,* and *S. balansae* were determined (Table 1) using methods described by Terrill et al. (1992). This assay divides total condensed tannins (TCT) into extractable condensed tannins (ECT), protein-bound condensed tannins (PBCT) and fiber-bound condensed tannins (FBCT). Standards were developed for each species as recommended by Wolfe et al. (2008).

Experiment 1: Plant Condensed Tannin Experiment Design. Manure was removed from the freezer and allowed to thaw 24 h before homogenization by hand. Moisture concentration of each homogenized manure type was determined gravimetrically by dry weight. Five 50-g samples of each manure type were placed individually in 236-ml Styrofoam cups, held in an oven at 55°C for 48 h, and then weighed a second time. Initial and final weights were compared with determine initial moisture concentration. Dry matter concentration of manure was as follows: goat 29%; beef 17%; and dairy 27%. Nutrient concentrations were measured at the Texas AgriLife Extension Laboratory (Texas AgriLife Extension 2011) (Table 2).

Ground plant materials from each source and manure type were mixed to achieve 10- (1%), 30- (3%), and 50- (5%) g condensed tannins/kg dry matter manure. No plant material was added to manure samples treated as controls (0%). Distilled water was added to homogenize all mixtures at 300-g dry matter/kg mixture and moisture was subsequently maintained at 78%. Each manure and treatment combination was allocated to four replicates of 100-g dry matter in 500-ml plastic cups (Arriba Scientific, Inc., TX).

Each cup was inoculated with 50 house fly eggs obtained from the Knipling-Bushland U.S. Livestock Insects Research Laboratory (Kerrville, TX). The eggs

Table 2. Mineral concentrations in manure used in experiment based on dry weight

Manure	%						ppm			
	N	P	K	Ca	Mg	Na	Zn	Fe	Cu	Mn
Goat	1.456	0.300	0.195	1.091	0.444	0.224	81	590	19	137
Beef	1.685	0.441	0.234	1.313	1.120	0.230	93	1,727	16	107
Dairy	1.507	0.368	0.823	1.612	0.577	0.577	144	2,066	25	164

were placed on moistened 2.54-cm^2 black cotton cloth and placed in the middle of each cup containing manure or manure/plant mixtures. Cups then were covered with C-fold towels (State Chemical, Stephenville, TX) secured with a rubber band and placed in a Percival rearing chamber (V-model, Boone IA) set at 27°C, 60–70% RH, and a photoperiod of 14:10 (L:D) h. Egg hatch was recorded 24 h after initiation of the experiment. Percent survivorship to the adult stage was determined by dividing number to emerge as adults by number to successfully hatch. Days to emergence, number of emergence, and average weight of all emerging flies were recorded daily in each individual cup, which was considered an experimental unit. Resulting adults were allowed to move into a sweep net which was then stored in a −20°C freezer for ≈20 min, which euthanized the flies. The flies then were removed and weighed individually with an Adventure-Pro AV64 Ohaus scale (Ohous, Pine Brook, NJ). Percent fly emergence (PFE), average daily gain (ADG), and average fly weight (AFW) was determined for flies emerging from each replicate of each treatment.

Experiment 2: Catechin Experiment Design. Manure was handled as in experiment 1. Catechin extract and dairy manure were mixed to achieve 10-g (1%) and 30- (3%) g condensed tannins/kg dry matter of manure. The control did not have any catechin added to the manure. Each manure and treatment combination had four replicates of 50-g dry matter in 0.47-liter clear plastic cups. Moisture content of the manure was maintained at the same level as in experiment 1. Both treatment and control cups were inoculated with 30 house fly eggs by using procedures described in the first experiment. The eggs were placed on moistened 2.54-cm^2 black cotton cloth and placed in the middle of each cup containing manure or manure and catechin mixture. Number of eggs hatched, total emergence, and average weight of all emerging flies were recorded for each cup and considered an experimental unit. Voucher specimens for both experiments have been placed in the arthropod collection at the Agrilife Research Center, Stephenville, TX.

Statistical Analyses. Percentage data were normalized using an arcsine transformed before analysis. PFE, ADG, and AFW were analyzed using PROC Mixed (SAS Institute 1998). In Experiment 1, two-way interaction between condensed tannin source and percent was detected ($P < 0.05$) so data are presented and discusses within this interaction. A Least Significant Difference (least significant difference [LSD]) test (SAS Institute 1998) was used to separate multiple means where appropriate. Analyses of results were considered significant at $P < 0.05$ so specific P values are not presented.

Results and Discussion

Experiment 1. Interactions were detected among manure type, condensed tannin source, and condensed tannin concentration when measuring percent adult house fly emergence, weight of emerging adults, and ADG. Data are therefore presented by condensed tannins source and concentration within manure type. In addition, no effect of condensed tannins was measured in goat manure. No clear reason is apparent for this because moisture levels and nutrient concentrations were similar for all three manures, with the exception of Fe, which was lower in the goat manure than in the other two (Table 2), and initial moisture was much lower than in the other two sources of manure. The reasons for failure of this effectiveness in goat manure need further research and may provide insight into how these additions suppress house fly development in bovine manure.

Table 3. Percent fly emergence, avg daily gain (ADG) and avg adult fly weight (AFW) of house flies (n = 4 cups with 50 eggs each) raised on dairy manure homogenized with plant material at different concentrations of condensed tannins in a growth chamber at 27°C and a photoperiod of 16:8 (L:D) h

Plant Species	% Condensed tannins	% Emergence[a]	ADG (mg)[b]	AFW (mg)[c]
Prairie acacia	0	38.3^{a}	1.34^{a}	13.4^{a}
	1	37.3^{a}	1.28^{a}	14.1^{a}
	3	42.3^{a}	1.28^{a}	12.8^{a}
	5	40.7^{a}	1.21^{a}	13.3^{a}
Panicled tick-clover	0	38.3^{a}	1.34^{a}	13.4^{g}
	1	37.0^{a}	1.33^{a}	14.7^{g}
	3	23.0^{b}	0.47^{b}	5.2^{h}
	5	0.0^{c}	0.00^{c}	0.0^{f}
Quebracho	0	38.3^{a}	1.34^{a}	13.4^{xy}
	1	37.3^{a}	1.33^{a}	14.6^{y}
	3	29.0^{a}	1.22^{b}	13.4^{xy}
	5	33.7^{a}	1.08^{c}	11.9^{x}

[a] Plant species by percent condensed tannin interaction detected (P = 0.003). Pooled SE = 4.6. Values within column for each plant species with different letters (a, b, c) differ according to multiple range least significant difference analysis.

[b] Plant species by percent condensed tannin interaction detected (P<0.001). Pooled SE = 0.04. Values within column for each plant species with different letters (a, b, c) differ according to multiple range least significant difference analysis.

[c] Plant species by percent condensed tannin interaction detected (P<0.001). Pooled SE = 0.55. Values within column for each plant species with different letters (a, b, c) differ according to multiple range least significant difference analysis.

Fly emergence for the dairy manure control was close to 38% and decreased with the addition of 3 and 5% condensed tannins from panicled tick-clover to 23 and 0%, respectively, but was unchanged by the addition of either prairie acacia or quebracho (Table 3). Both panicled tick-clover and quebracho at 3 and 5% condensed tannins caused a decline in ADG which, in the case of panicled tick-clover, lowered AFW by over 60 at the 3% condensed tannins level. Differences in the effectiveness of condensed tannins sources indicate that molecular weights and their binding capabilities (Lowry et al. 1996) may be a factor in house fly suppression either via direct physical interference of larvae (Molan et al. 2003) or through neutralization of nutrient sources such as N (Makkar 2003).

The general pattern observed in dairy manure was evident in beef manure as well (Table 4), because prairie acacia condensed tannins had no effect on house fly parameters, whereas high rates of panicled tick-clover lowered all parameters. The addition of

Table 4. Percent fly emergence, average daily gain (ADG), and average adult fly weight (AFW) of house flies (n = 4; 50 larvae/n) raised on beef manure homogenized with plant material at different concentrations of condensed tannins in a growth chamber at 27°C and a photoperiod of 16:8 (L:D) h

Plant species	% Condensed tannins	% Fly emergence	ADG (mg)	AFW (mg)
Prairie acacia	0	22.3[a]	0.47[a]	5.2[a]
	1	18.7[a]	0.46[a]	5.1[a]
	3	22.7[a]	0.65[a]	7.1[a]
	5	26.7[a]	0.61[a]	6.7[a]
Panicled tick-clover	0	22.3[b]	0.47[a]	5.2[a]
	1	34.7[a]	0.53[a]	5.8[a]
	3	19.0[c]	0.23[b]	2.3[b]
	5	0.0[d]	0.00[b]	0.0[c]
Quebracho	0	22.3[a]	0.47[a]	5.2[a]
	1	27.7[a]	0.51[a]	5.6[a]
	3	9.7[b]	0.44[a]	4.8[a]
	5	6.0[b]	0.42[a]	4.7[a]

Within each column, for tannin concentrations within each plant species, different letters indicate significant difference ($P < 0.05$.)

quebracho to manure, however, did not affect ADG or AFW but lowered PFE by over 56% at 3 and 5% condensed tannins compared with the control. As evidenced by the negative effects of quebracho extract and panicled tick-clover plant material on house fly development, condensed tannins appeared to have the potential to be used as an alternative to insecticide application. Our observations suggest some but not all condensed tannins incorporated into manure at higher concentrations negatively affect house fly development in bovine manure. Our study also suggests the use of quebracho, a rich supply of condensed tannins, as a potential source for insect control.

Experiment 2. To confirm the impact of condensed tannins on house fly development, we applied purified catechin at 1 and 3% to dairy manure samples (Table 5). The purified condensed tannins impacted house fly survival and development. However, house fly response was dictated by treatment concentration. The 1% application showed no effect on PFE (F = 1.01; df = 1, 6); ADG (F = 0.00; df = 1, 6); or AFW (F = 0.01; df = 1, 6). However, the 3% treatment impacted PFE (F = 46.18; df = 1, 6); ADG (F = 36.64; df = 1, 6); and AFW (F = 30.85; df = 1,6).

Table 5. Percent fly emergence, avg daily gain (ADG) and average adult fly wt (AFW) of house flies (n = 4 cups with 30 eggs) raised on dairy manure homogenized with catechin at 1 and 3% concentration in a growth chamber at 27°C and a photoperiod of 14:10 (L:D) h

Catechin concn (%)	% fly emergence[a]	ADG (mg)[b]	AFW (mg)[c]
0	88.4[a]	1.00[a]	12.1[a]
1	80.2[b]	1.02[a]	12.4[a]
0	91.2[a]	0.89[a]	8.91[a]
3	20.6[b]	0.26[b]	2.93[b]

[a] Effect of Catechin concn. Pooled SE = 7.7. Values within column with different letters (a, b) differ according to multiple range least significant difference analysis.
[b] Effect of Catechin concentration. Pooled SE = 0.16. Values within column with different letters (a, b) differ according to multiple range least significant difference analysis.
[c] Effect of Catechin concentration. Pooled SE = 1.88. Values within column with different letters (a, b) differ according to multiple range least significant difference analysis.

Condensed tannins, such as catechin, could impact the microflora associated with manure. This impact on the microflora might explain the high house fly mortality observed in the 3% treatment as house flies use bacteria in manure as nutrition (Zurek et al. 2000). An alternate hypothesis to consider is that bacteria also are competing with other consumers for resources (Janzen 1977). The same competition could be occurring between house fly larvae and bacteria in manure. While house flies can survive with a select group of bacteria species present in the manure, the presence of condensed tannins might favor microbial species that are deleterious to house fly larvae.

Although these ecological interactions remain to be determined, we believe catechin, as well as other condensed tannins, might be developed as green approaches for pest control in confined animal facilities. We believe these compounds could be included in animal diets or applied in solution form directly into manure at targeted sites in confined animal facilities, thus reducing associated house fly emergence. Targeting specific sites would reduce the amount of compound being applied as well. The addition of condensed tannins as a means of fly control also would diversify the options available for producers. Although we did not examine it, the application of these compounds to manure also could reduce house fly attraction and oviposition as well.

Further research is needed to identify how condensed tannins suppress house fly development in ruminant manure. Our research was not designed to verify which molecular weights were most effective, and if condensed tannins interfere with nutrients before these reach the larvae, or directly interfere with larvae motility or nutrient ingestion. The catechin trial, in our opinion, confirmed that condensed tannins are a primary factor, but neither trial identified the mechanisms involved. Trials using condensed tannins sources with neutralized condensed tannins also will allay any doubts as to whether factors other than condensed tannins come into play.

Future research also should examine the relationship between different pure condensed tannins extracts and various other external arthropod populations. In addition, the incorporation of condensed tannins into ruminant diets and subsequent excretion in manure may have different results than the direct addition of condensed tannins to excreted manure. As vectors for pathogens and a persistent annoyance to livestock and humans, house fly control by means of condensed tannins needs to be considered in future integrated pest management (IPM) programs.

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References Cited

Balandrin, M. F., J. A. Klocke, E. S. Wurtele, and W. H. Bollinger. 1985. Natural plant chemicals: sources of industrial and medicinal materials. Science 228: 1154–1160.

Coats, J. R. 1994. Risks from natural versus synthetic insecticides. Annu. Rev. Entomol. 39: 489–515.

Forkner, R. E., R. J. Marquis, and J. T. Lill. 2004. Feeny revisited: condensed tannins as anti-herbivory defences in leaf-chewing herbivore communities of *Quercus.* Ecol. Entomol. 29: 174–187.

Iqbal, Z., M. Sarwar, A. Jabbar, S. Ahmed, M. Nisa, M. S. Sajid, M. N. Khan, K. A. Mufti, and M. Yasen. 2007. Direct and indirect anthelmintic effects of condensed tannins in sheep. Vet. Parasitol. 144: 125–131.

Janzen, D. H. 1977. Why fruits rot, seeds mold, and meat spoils. Am. Nat. 111: 691–713.

Lowry, J. B., C. S. McSweeney, and B. Palmer. 1996. Changing perceptions of the effects of plants phenolics on nutrient supply in the ruminant. Aust. J. Agric. Res. 47: 829–842.

Makkar, H.P.S. 2003. Effects and fate of tannins in ruminant animals, adaptation to tannins, and strategies to overcome detrimental effects of feeding tannin-rich feeds. Small Rum. Res. 49: 241–256.

Min, B. R., and S. P. Hart. 2003. Tannins for suppression of internal parasites. J. Anim. Sci. (E. Suppl. 2) E102–109.

Molan, A. L., L. P. Meagher, P. A. Spencer, and S. Sivakumaran. 2003. Effect of flavan-3-ols on in vitro egg hatching, larval development and viability of infective larvae of *Trichostrongylus colubriformis.* Int. J. Parasitol. 33: 1691–1698.

Paolini, V., J. P. Bergeaud, C. Grisez, F. Prevot, Ph. Dorchies, and H. Hoste. 2003. Effects of condensed tannins on goats experimentally infected with *Haemonchus contortus.* Vet. Parasitol. 113: 253–261.

Pawelek, D. L., J. P. Muir, B. D. Lambert, and R. D. Wittie. 2007. In *sacco* rumen disappearance of condensed tannins, fiber, and nitrogen from herbaceous native Texas legumes in goats. Anim. Feed Sci. Technol. 140: 225–240.

Rey, D., M. P. Pautou, and J. C. Meyran. 1999a. Differential sensitivity of mosquito taxa to vegetable tannins. J. Chem. Ecol. 25: 537–548.

Rey, D., M. P. Pautou, and J. C. Meyran. 1999b. Histopathological effects of tannic acid on the midgut epithelium of some aquatic Diptera larvae. J. Invertebr. Pathol. 73: 173–181.

SAS Institute. 1998. SAS/STAT user's guide, version 6, 4th ed. SAS Institute, Cary, NC.

Sawicki, R. M., and I. Denholm. 1984. Adaption of insects to insecticides. Ciba Found. Symp. 102: 152–166.

Sukontason, K. L., N. Boonchu, K. Sukontason, and W. Choochote. 2004. Effects of eucalyptol on house fly (Diptera: Muscidae) and blow fly (Diptera: Calliphoridae). Rev. do Instituto de Medicina Tropical de São Paulo 46: 97–101.

Terrill, T. H., A. M. Rowan, G. B. Douglas, and T. N. Barry. 1992. Determination of extractable and bound condensed tannin concentrations in forage plants, protein concentrate meals, and cereal grains. J. Sci. Food Agric. 58: 321–329.

Texas AgriLife Extension. 2011. Soil, water and forage testing laboratory. Methods and methods reference. (http://soiltesting.tamu.edu/webpages/swftlmethods1209.html).

Tomizawa, M., and J. E. Casida. 2005. Neonicotinoid insecticide toxicology: mechanisms of selective action. Annu. Rev. Pharmacol. Toxicol. 45: 247–268.

Waghorn, G. 2008. Beneficial and detrimental effects of dietary condensed tannins for sustainable sheep and goat production-progress and challenges. Anim. Feed Sci. Technol. 147: 116–139.

Wolfe, M. R., T. H. Terrill, and J. P. Muir. 2008. Drying method and origin of standard affect condensed tannin (CT) concentrations in perennial herbaceous legumes using simplified butanol-HCl CT analysis. J. Sci. Food Agric. 88: 1060–1067.

Zurek, L., C. Schal, and D. W. Watson. 2000. Diversity and contribution of the intestinal bacterial community to the development of *Musca domestica* (Diptera: Muscidae) larvae. J. Med. Entomol. 37: 924–928.

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