Preliminary observations were made utilizing a combination of leucine, at 10^{-3} M concentration, and insulin at 100 micro-units/ml. Coincidently, separate determinations for leucine at 10^{-3} M concentration and insulin at 100 micro-units were obtained. Individually, there was a significant increase in uptake with insulin and a signfiicant decrease in uptake with leucine. The combination of insulin and leucine demonstrated a significant increase in glucose uptake above the non-insulin control while being associated with a significant decrease in glucose uptake as compared with the insulin control. In 21 determinations, the mean difference in glucose uptake of the insulin-leucine combination compared with the non-insulin control was + 2.58 mg glucose/g adipose tissue ($p<.001$). When the combination was compared with the insulin control, the mean difference in glucose uptake was −2.04 mg glucose/g adipose tissue ($p<.001$).

A pH effect was considered as a possible factor in the inhibitory action of leucine upon glucose uptake. Determinations of pH performed upon leucine samples were identical with those of control samples, thereby eliminating pH changes as significantly related to the inhibitory action of leucine.

Previous unpublished studies, utilizing an incubation time of 2 hours, demonstrated statistically significant inhibition of glucose uptake by adipose tissue in the presence of certain amino acids. The magnitude of the effects tended to be less than appeared in the present study utilizing longer incubation times.

Summary. Six of 8 amino acids tested at concentrations of 10^{-2}–10^{-3} M demonstrated significant inhibition of glucose uptake by adipose tissue. This inhibitory action was diminished as amino acid concentration was further decreased.

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The Value of Certain Steroidal Sapogenins in Rations of Fattening Lambs and Cattle. (26378)

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The growth-promoting action for ruminants of diethylstilbestrol and certain other estrogens is well known. However, because of the estrogenic activity of these compounds, there must be close control of the quantities given in practical feeding operations. Combinations of estrogens with androgens or progesterone have been used as growth stimulants, the estrogenic action being diminished by the other hormone so that anabolic action predominates. However, it would be desirable to have available a single, non-estrogenic compound with substantial anabolic action (1).

There are structural similarities between natural animal steroid hormones and certain plant steroids. These plant products are not known to have estrogenic activity, nor have they been evaluated for effect on ruminant productive performance. The present report provides data on growth rate, feed efficiency and carcass quality of fattening steers and lambs fed plant steroidal sapogenins, saponins, and a natural source of unisolated sapogenins. The products tested were smilagenin and smilagenin saponin isolated from *Agave lecheguilla,* ground *Agave lecheguilla*

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TABLE I. Experimental Rations.

	Cattle	Lambs*
Cracked shelled corn	Full fed	
Alfalfa hay/day, lb	3	
Supplement/day, lb	2	
Composition of supplement or complete pellet		
Ground yellow corn		55
Sun-cured alfalfa meal		35
Soybean oil meal	69.05	
Dehydrated alfalfa meal	10.00	
Liquid molasses		5
Dried "	10.00	
Urea	5.00	1
Bone meal		1
Dicalcium phosphate	4.50	
Ground limestone	1.00	
Trace mineral premix	.40	
Vitamin A†	.05	
Bentonite		3
	100.00	100

* The lamb ration was made into 3/8 inch pellets and self-fed.

† Gelatin-stabilized vit. A mix containing 10,000 U.S.P. units of vit. A palmitate/g.

containing smilagenin in the form of a saponin, and sarsasapogenin, tigogenin, diosgenin and hecogenin from commercial sources.

Procedure. Five lamb experiments were conducted with graded levels of smilagenin ranging from 2 to 24 g per ton of total ration. Lamb experiment 3 included a group receiving ground *Agave lecheguilla* (1.6% smilagenin) supplying the equivalent of 8 g smilagenin per ton of ration. Lamb experiment 4 included groups receiving sarsasapogenin, diosgenin and tigogenin at 8 g per ton of ration. Lamb experiment 5 included groups receiving smilagenin saponin at levels of 4, 8 and 16 g per ton, hecogenin at 8 g per ton and diethylstilbestrol at 1.25 g per ton.

In all lamb experiments, 12 animals were allotted per treatment, the controls comprising 2 replicate lots of 12 each. Individual weighings were made every 14 days. Some death losses occurred and the data have been corrected for these losses.

In cattle experiment 1, smilagenin at 20 mg and 100 mg per head was compared with oral stilbestrol at 10 mg per head. In cattle experiment 2, smilagenin at 20 mg per head was compared with stilbestrol at 10 mg per head. The steers were weighed at 28-day intervals. Basal rations for the lamb and cattle trials are presented in Table I.

Results. Weight gains, feed efficiency, dressing per cent and carcass grade data for the lambs are presented in Table II. In Exp. 1, the 4 g of smilagenin per ton of ration had no effect upon weight gains or feed efficiency, but dressing per cent and carcass grade were improved. In Exp. 2 both the 2 and 8 g levels of smilagenin improved weight gains and feed efficiency. The 8 g level exerted a marked effect, bringing about a 32% increase in gain and a 31% improvement in feed efficiency. Carcass grade was significantly improved by both levels of smilagenin, but there was no effect upon dressing per cent. In Exp. 3, no response was noted to the 2 g level of smilagenin, but the 8 and 24 g levels increased gains, the increase being 11% with 24 g. Dressing per cent and grade were not affected. The ground *Agave* plant produced weight gain increases similar to those obtained with 8 g of smilagenin.

In Exp. 4, significant improvements in rate of gain occurred with 4, 8 and 24 g of smilagenin per ton. The 8 g level brought about a 34% increase over the control and a 17% improvement in feed efficiency. The two lower levels of smilagenin improved both dressing per cent and carcass grade. The 8 g of sarsasapogenin produced gains almost identical with those obtained from 8 g of smilagenin. Dressing percent and carcass grades were generally similar to those obtained with the smilagenin treatments. Diosgenin and tigogenin at 8 g per ton did not improve gains over the control and these compounds appeared to be toxic in the early part of the experiment as indicated by feed intake and weight gains.

In Exp. 5 the 4 g and 16 g levels of smilagenin produced comparable improvements in growth, feed efficiency and grade. The 8 g treatment group was not appreciably better than the basal; however it was noted that this supplemented group included one very poor-performing lamb. If this lamb had been eliminated from the data, performance of the 8 g group would have been comparable to that of the other 2 smilagenin-fed

TABLE II. Response of Fattening Lambs (Wethers) to Various Plant Steroids.

Treatment	No. lambs	Avg initial wt, lb	Avg daily gain, lb	Lb feed per lb gain	Dressing, %†	Carcass grade‡
		Exp. 1—53 days				
Basal	23	86.5	.688	6.50	46.81	8.49
+ Smilagenin, 4 g/ton	12	84.7	.633	7.48	48.01	9.17
		Exp. 2—56 days				
Basal	23	70.3	.344	10.38	49.60	8.61
+ Smilagenin, 2 g/ton	10	68.5	.384	8.21	49.46	9.20*
+ " , 8 "	11	69.4	.455	7.14	49.09	9.27*
		Exp. 3—56 days				
Basal	23	71.0	.368	6.84	50.02§	9.82
+ Smilagenin, 2 g/ton	12	71.4	.345	7.36	— §	—
+ " , 8 "	12	69.2	.390	6.23	50.03	9.25
+ " , 24 "	12	69.3	.409	6.11	50.47	9.91
+ Gr. *Agave lechequilla* equiv. to 8 g/smilagenin/ton	12	71.0	.385	7.40	49.98	9.36
		Exp. 4—68 days				
Basal	21	74.0	.320	9.29	44.98	6.84
+ Smilagenin, 4 g/ton	11	73.4	.398*	8.52	45.60	7.82
+ " , 8 "	12	74.6	.430*	7.69	45.54	7.67
+ " , 24 "	12	74.0	.347*	9.16	45.07	6.83
+ Sarsasapogenin, 8 g/ton	12	74.2	.428*	8.29	45.80	7.50
+ Diosgenin, 8 "	11	75.9	.322	9.28	44.48	7.00
+ Tigogenin, 8 "	10	77.4	.304	9.98	44.91	6.90
		Exp. 5—70 days				
Basal	23	65.1	.392	7.38	— ‖	6.80‖
+ Smilagenin, 4 g/ton	11	65.2	.443	7.27	—	8.00
+ " , 8 "	11	66.2	.402	7.35	—	6.75
+ " , 16 "	12	66.7	.440	7.29	—	9.50
+ Smilagenin saponin, 4 g/ton	11	65.8	.403	7.22	—	7.46
+ " " , 8 "	12	64.0	.384	7.51	—	6.75
+ " " , 16 "	12	65.4	.379	7.63	—	6.75
Hecogenin, 8 g/ton	11	66.0	.429	7.23	—	7.46
Diethylstilbestrol, 1.25 g/ton	12	68.0	.379	8.05	—	7.75

* Significant over controls (P = 0.05).

† In Exp. 1 and 3 dressing % based on cold carcass weights and in Exp. 2 and 4 based on warm carcass weights.

‡ 10 = low choice, 9 = high good, 8 = avg good.

§ In the basal groups carcass grade and dressing % available on 12 animals only as 11 were retained for additional experimental work. All 12 lambs on the low level of smilagenin were also retained.

‖ Dressing % figures were not available in Exp. 5 and grade values were based on live grade evaluation.

groups. Smilagenin saponin at the 4 g level produced only slightly better performance than the control and the consistently depressed performance obtained with each increment suggests that the saponin form of steroidal sapogenins is not well tolerated. Hecogenin at 8 g produced improvements in growth and feed efficiency comparable to those with the several levels of smilagenin. Stilbestrol did not enhance performance in this experiment.

Results of the 2 cattle experiments are presented in Table III. In the first, an increase of 0.27 lb in average daily gain followed feeding of 20 mg smilagenin and an increase of 0.15 lb followed 10 mg of stilbestrol. Feed efficiency was improved by 20 mg of smilagenin and dressing per cent was improved by both levels of smilagenin and by stilbestrol. Carcass grade appeared to be improved by stilbestrol.

In cattle experiment 2, average daily gains were increased 0.17 lb by 20 mg smilagenin and 0.31 lb by 10 mg stilbestrol. Feed efficiencies were increased by both compounds. Dressing per cent appeared to be lowered by the diethylstilbestrol. Carcass grade was improved markedly by the smilagenin and a

TABLE III. Response of Fattening Steers to Smilagenin and Diethylstilbestrol.

Treatment	No. steers	Avg initial wt, lb	Avg daily gain, lb	Lb feed per lb gain	Dressing, %*	Carcass grade†
		Exp. 1—98 days				
Basal	9	924.6	2.14	12.02	60.67	10.00
Smilagenin, 20 mg/head	8	951.5	2.41	11.44	61.67	9.88
" ,100 "	8	941.6	2.21	12.29	61.98	9.88
Diethylstilbestrol, 10 mg/head	8	920.5	2.29	11.73	61.92	10.62
		Exp. 2—139 days				
Basal	8	677.1	2.57	9.56	57.28	7.88
Smilagenin, 20 mg/head	8	680.1	2.74	8.68	57.14	8.75
Diethylstilbestrol, 10 mg/head	8	674.8	2.88	8.88	56.60	8.25

* Based on cold carcass wt.

† 10 = low choice, 9 = high good.

slight improvement was noted in the stilbestrol group.

Discussion. In the lamb trials, the 8 g level of smilagenin appeared to be most consistent in increasing weight gains. The average increase obtained with this level in 4 trials was 17.7% above controls and average feed efficiency increase was 16.2%. This level supplied approximately 12 mg smilagenin per lamb per day. Nearly identical growth responses (34%) were obtained with 8 g levels of sarsasapogenin and smilagenin (one comparison-experiment 4). In general, sarsasapogenin appeared to increase dressing per cent and improve grade. Hecogenin was comparable to smilagenin in improving growth (Exp. 5) and appeared to improve feed efficiencies and grade. Diosgenin and tigogenin did not improve performance (Exp. 4).

Measurement of the bulbo-urethral glands in lamb experiments 3 and 4 did not reveal an enlargement from plant steroid feeding as is noted with lambs fed diethylstilbestrol(1). Mouse uterine assays in this laboratory did not indicate estrogenic activity in smilagenin.

In the 2 cattle experiments the 20 mg daily feeding level of smilagenin improved gain by an average of 9.6% as compared with 9.5% for 10 mg stilbestrol.

No basis is immediately available to explain the mechanisms whereby smilagenin, sarsasapogenin or hecogenin bring about more rapid gains. Smilagenin and sarsasapogen are related in that smilagenin can be obtained by acid isomerization of sarsasapogenin(2). The mechanism of the growth-stimulating action of the plant steroids may be due to an anabolic action similar to that of stilbestrol but wtihout the undesirable estrogenic effects.

These studies indicate that smilagenin and certain other plant steroids may be of practical use in production of fat lambs and steers. Additional study is needed before the full significance of these findings can be assessed.

Summary. 1. Smilagenin fed to lambs at levels of 2, 4, 8 and 24 g per ton of complete feed improved growth rate and feed efficiency. The most consistent results were obtained with 8 g per ton, or approximately 12 mg per lamb per day. Dressing per cent and carcass grade tended to be improved by addition of smilagenin. Ground *Agave lecheguilla* containing smilagenin in the saponin form appeared to be quantitatively similar to the free sapogenin; however higher levels of the isolated saponin depressed performance. Sarsasapogenin or hecogenin at 8 g per ton in separate tests exerted a growth effect comparable to that from smilagenin. Diosgenin and tigogenin were not beneficial under conditions tested. 2. In 2 trials, steers fed 20 mg of smilagenin daily showed weight gains similar to those of animals receiving 10 mg stilbestrol daily. In one trial, 100 mg of smilagenin was not as effective as 20 mg.

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