ACTIVATED charcoal has been used as a purifying agent for many years. Holt and Holz (1963) reviewed the use of charcoal to absorb poisons and other materials in man in which they list many organic and inorganic chemicals which are effectively absorbed by activated charcoal. Decker, Combs and Corby (1968) studied the relative adsorbency by activated charcoal of a wide variety of drugs and other chemicals often found in the home. Wilson, Cook and Emery (1968) reported that activated plant charcoal could increase the elimination of residues of the chlorinated hydrocarbon pesticide, dieldrin, in the dairy cow to more than twice the natural rate. Smalley, Crookshank and Radeleff (1971) reported that activated charcoal in a diet containing the organophosphate insecticide, ronnel, reduced the residue levels in the omental fat to 10% of that in sheep not receiving the charcoal.

One problem to be considered in the continued use of activated charcoal in the feed is that not only will it adsorb poisons but possibly enzymes, amino acids, and other nutrients as well. This could seriously affect the health of the animal. This study reports the effect of orally-administered activated charcoal of plant origin and of charcoal and the organophosphate insecticide, ronnel, over an 84-day period upon the well being of sheep as measured by changes in blood serum composition, urine excretion, weight, feed efficiency and microscopic studies of tissue.

Experimental Procedure

Thirty-six commercial medium-wool lambs were used. The sheep were examined, identified and treated for internal parasites. Three ewes and 3 wethers were randomly allotted to each of the six treatment groups as follows:

Group I —Control, basal diet
Group II —Charcoal added to the diet
Group III—Basal diet, drenched twice weekly with charcoal in a slurry
Group IV—Ronnel added to the diet
Group V —Charcoal and ronnel added to the diet
Group VI—Ronnel added to the diet and drenched twice weekly with charcoal slurry.

The basal diet for the first 9 weeks consisted of 30% ground sorghum grain, 55% cottonseed hulls, 10% cottonseed meal and 5% molasses. During the last 22 days, the grain content was increased to 37.5% and cottonseed meal to 12.5% with a compensating decrease in cottonseed hulls. At the end of the 84-day period, those animals continued on the charcoal diet were fed the initial diet.

The period of continuous administration of ronnel (84 days) was chosen to approximate the length of time lambs would remain in the feedlot. Crystalline ronnel, (O-O-dimethyl-O-2,4,5-trichlorophenyl) phosphorothioate, 92.5% purity (maximum purity available from the manufacturer, Dow Chemical Company, Midland, Michigan) was used. It was added to the diet at the level of approximately 1,000 ppm by dissolving 50 g of ronnel in 200 ml chromatographic grade acetone and spraying the solution over 45.4 kg of diet spread over a plastic sheet. The acetone was allowed to evaporate at room temperature; then the diet was thoroughly mixed. Activated charcoal (Darco S-51, Atlas Chemical Industries, Inc., Wilmington, Delaware) was added at the level of 5% of the total diet. The charcoal slurry was prepared by mixing 150 g charcoal with approximately 700 ml water and was administered with a drenching gun. The slurry in each drench was calculated to afford approximately 50% of the level of charcoal consumed in the diet each week.

Blood samples and 24-hr. urine samples were taken from each animal immediately
prior to the start of the treatment, at the midpoint and at the conclusion of the feeding period but prior to the removal of either charcoal or ronnel from the diets.

Necropsies were performed on all subjects which succumbed to urolithiasis during the course of the experiments and from two animals from each treatment group at the end of the test period. Tissues were taken, fixed in buffered 10% formalin, embedded, cut and stained with hematoxylin and eosin.

The animals were individually weighed every 7 days and feed consumption by treatment groups was recorded. Daily gain, feed consumption and feed efficiency were determined on a group basis for each treatment.

The blood was collected in plastic tubes, allowed to clot and the serum separated by double centrifugation. The serum obtained was stored in glass screw-capped vials at \(-10^\circ\text{C}\) until analyzed. The 24-hr. urine samples were collected with a small amount of thymol in the container to control bacterial action. Generally, the urine analysis was completed the same day the sample was collected. If analysis could not be completed that day, analyses for the nitrogen containing components were completed, and the urine frozen and stored at \(-10^\circ\text{C}\) until the analyses could be completed.

Blood serum and urine were analyzed for total calcium, copper, iron, magnesium, potassium, sodium and zinc using standard atomic absorption spectrophotometric technics. Inorganic phosphorus, creatinine, uric acid and urea nitrogen levels were determined using N-methods and a single channel Technicon AutoAnalyzer. In addition, the AutoAnalyzer was used to determine the alkaline phosphatase and total serum protein levels in the blood serum. Urine volume to the nearest 5 ml was measured with a graduated cylinder and urine pH determined with a pH meter.

As part of a separate study of tissue residue levels, omentectomies were made periodically on these animals. These results have been reported elsewhere (Smally et al., 1971).

Results and Discussion

While there was considerable variation among the individual animals in a particular test group, no significant differences, based on mean values and standard deviation, were found between groups in any of the parameters measured in the blood serum or 24-hr. urine samples. Approximately 25% of the animals starting the test were continued for a total of 6 months on the diet containing the charcoal plus a small amount of ammonium chloride to control the formation of urinary calculi. At the end of this time, these animals did not show visible signs of any nutritional deficiency or pathology. Blood serum samples did not vary appreciably in composition from those previously taken. Histopathological examination of these sheep at the end of the 6-month period did not show any significant differences in gross or microscopic examination. It does not appear that the oral administration of activated charcoal, either by drench or by addition to the diet, or the addition of ronnel to the diet alone or with charcoal had any effect upon the blood serum composition or urinary excretion of the parameters measured.

Feed consumption, average daily gain and feed efficiency by treatment groups are given in table 1. Although the feed consumption, daily gain and feed efficiency figures are low for finishing sheep, the basal diet was not designed for optimum growth but to insure deposition of omental fat. The animals were under considerable stress due to the frequent omentectomies required for the tissue residue study, the drenching and the other sample collecting and diagnostic practices. With the exception of Group IV, ronnel only, during weeks 4 through 9, the weight changes of the groups were remarkably parallel and reflected the stress situations equally. During weeks 4 through 9, Group IV showed a slight weight loss while the other groups showed a small gain. Prior to and after this period, the weight changes approximated that of Group I, Control.

The addition of either charcoal or ronnel to the diet reduced feed consumption when compared to the control group. However, the addition of charcoal by drench had little effect upon feed consumption (Group I vs. III and IV vs. VI). The addition of both charcoal and ronnel to the diet had little effect upon feed consumption over that of charcoal alone (Group II vs. V). A similar situation was found when charcoal by drench was added to a ronnel containing diet (Group IV vs. VI). No consistent pattern was observed other than the reduction of both the average daily gain and feed efficiency when charcoal, ronnel or both were added to the basal diet.

While the basal diet was known to be calcudogenic, it was designed primarily to insure adequate fat deposition for the omentectomies
TABLE 1. AVERAGE FEED CONSUMPTION, DAILY GAIN AND FEED EFFICIENCY OF SHEEP FED CHARCOAL, RONNEL OR RONNEL AND CHARCOAL

<table>
<thead>
<tr>
<th>Group</th>
<th>Ronnel</th>
<th>Charcoal</th>
<th>Feed consumption kg/hd/day</th>
<th>Gain kg/hd/day</th>
<th>Feed efficiency kg feed/kg gain</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>0</td>
<td>0</td>
<td>1.91</td>
<td>0.13</td>
<td>14.7</td>
</tr>
<tr>
<td>II</td>
<td>0</td>
<td>5% in diet</td>
<td>1.60</td>
<td>0.08</td>
<td>20.0</td>
</tr>
<tr>
<td>III</td>
<td>0</td>
<td>drench (twice weekly)</td>
<td>1.85</td>
<td>0.11</td>
<td>16.8</td>
</tr>
<tr>
<td>IV</td>
<td>1000 ppm</td>
<td>0</td>
<td>1.43</td>
<td>0.07</td>
<td>20.4</td>
</tr>
<tr>
<td>V</td>
<td>1000 ppm</td>
<td>5% in diet</td>
<td>1.56</td>
<td>0.10</td>
<td>15.6</td>
</tr>
<tr>
<td>VI</td>
<td>1000 ppm</td>
<td>drench (twice weekly)</td>
<td>1.36</td>
<td>0.08</td>
<td>17.0</td>
</tr>
</tbody>
</table>

required for the residue study and incidentally to determine the effect of the regimens on calculogenesis. Several cases of renal calculi developed with the first case 75 days after the test was started. Since uroliths were found in wethers from each treatment group and from a completely unrelated group on the same basal diet, it appeared that neither charcoal nor ronnel had any significant effect on their formation.

Although the sheep in the two groups (II and V) receiving finely powdered charcoal in the diet quickly became covered by a charcoal dust and necropsy later showed the presence of charcoal particles in the lungs, there was no instance of a clinical pneumonia. The wool was not affected by breaks or lack of fiber growth, and the charcoal easily scoured off.

Lesions observed at necropsy were confined to the lungs and to the mediastinal lymph nodes of the sheep receiving charcoal mixed into the diet. As seen in figure 1, carbon par-

Figure 1. Carbon particles in peribronchial connective tissue in lung of sheep on a diet containing 5% activated charcoal. 625×.
Figure 2. Distribution of carbon particles in parenchyma of a mediastinal lymph node of sheep on a diet containing 5% activated charcoal. 475X.

ticles were diffusely distributed throughout the lungs but tended to concentrate near the bronchi and bronchioles. Microscopically, there appeared to be an increased activity of the mucous glands in the bronchi. Some particles of carbon had been phagocytized. There was no increase in reticuloendothelial cells. The alveolar walls were slightly thickened and increased connective tissue was evident in areas adjacent to the heavier concentrations of charcoal. No inflammatory cells or exudate were noted in any of the lungs. Microscopic examination (figure 2) showed that the lymph nodes draining the lungs were black with carbon particles evident throughout the parenchyma.

Summary

The addition of activated charcoal of plant origin at the level of 5% of the total diet, as biweekly drench, or in combination with the organophosphate insecticide, ronnel, at the level of 1,000 ppm in the diet did not significantly affect the blood serum level or urinary excretion of total calcium, copper, iron, magnesium, inorganic phosphorus, potassium, sodium, zinc, creatinine, uric acid and urea nitrogen. In addition no changes were found in the serum alkaline phosphatase and total protein levels nor in urine pH or 24-hr. volume. While the addition of either charcoal or ronnel to the diet decreased both feed intake and feed efficiency, combining the two did not produce a further decrease. Drenching with charcoal did not affect feed intake. While animals fed charcoal in the diet showed the presence of charcoal particles in the lungs upon necropsy, there was no instance of a clinical pneumonia due to inhalation of particles. At necropsy, extensive charcoal deposits were noted in the lungs and mediastinal lymph nodes. However, no inflammatory cells or exudate were noted in any of the lungs examined.

Literature Cited