capacitation of the sweat glands, indicated that the process of normal transudation through the treated skin surface was not markedly disturbed. This is in conformity with the findings of Pinson (8) in man and Shelley and Hurley (10) in man and the dog. McDowell et al. (7) also reported that the moisture output from rabbit skin, which is devoid of sweat glands, was essentially unaffected when the same technique employed in this study was utilized. These results support the hypothesis that the major portion of the substance available for evaporation from the skin surface of cattle under thermal stress comes from the sweat glands.

Acknowledgment

These investigations were financed in part by a grant made by the United States Department of Agriculture, Agricultural Research Service, under Public Law 480.

B. C. Joshi
R. E. McDowell
and
D. P. Sadhu
Indian Veterinary Research Institute
Izatnagar, U. P., India
and
Cornell University, Ithaca, New York

References

(9) Shelley, W. B. 1954. Personal communication.

Composition of Skin Secretions from Three Indian Breeds of Cattle Under Thermal Stress

Abstract

Six 12- to 24-month-old heifers from each of the three breeds, Hariana, Tharparkar, and Gir were used to determine composition of skin surface secretions produced under hot, humid conditions (dry bulb temperature, 40.5 C and wet bulb, 35.0 C). The secretions were collected from shaved areas on the rump by absorbing the secretions in pieces of filter paper placed over the body surface. The color ranged from dark gray to brown, indicating the presence of the pigment, melanin. All samples gave uniformly high alkaline reactions (average pH 7.8). The fluids were hypotonic to blood plasma but contained inorganic phosphorus, total and nonprotein nitrogen, reducing sugars, and lactic acid in concentrations much higher than values reported for whole blood or plasma. The results show that similar to horses, cattle sweat has high contents of total protein nitrogen and inorganic salt and a relatively high urea content. The low chloride secretion indicates that under hot conditions cattle do not have a need for large amounts of salt replacement in the diet. On the other hand, the high losses of total protein nitrogen could be a factor in the poor performance of cattle under thermal stress. The composition of the secretions from the three Indian breeds agrees with that found for European breeds of cattle and horses.

The possibilities of the loss of salt in sweat is extremely important, secondary in its physiological effects only to the loss of water. Since it has become recognized that sweating plays an important role in promoting heat loss from cattle under thermal stress, it is important that a knowledge of the possible imbalances which may be brought about is appreciated. There is some evidence that the sweat of Bos taurus
cattle contains no measurable chlorides (7, 13). Since the native cattle of India developed in field environments likely to promote sweating more frequently than the usual habitat for Bos taurus breeds, the current series of investigations were undertaken to determine if Zebu breeds had different characteristic skin secretions from Bos taurus under thermal stress.

Experimental Procedures

Skin surface secretions were collected from six heifers each of the Hariana, Gir, and Tharparkar breeds of Indian cattle. The animals ranged in age from 12 to 24 months. On the rump of each heifer an area resembling an inverted triangle with rounded apex approximately 15 by 25 cm was shaved smoothly and thoroughly cleaned with several washings of soap and tap water followed by distilled water. Cotton swabs soaked with distilled water were gently rubbed over the shaved areas until the cotton showed no discoloration. When the sites were dry, they were partially covered with pieces of sterilized ash-free filter paper. The filter paper was then covered with six ml clear polyethylene. The edges of the plastic coverings were sealed to the shaved skin with rubberized cement to form a closed chamber around the filter paper.

After preparation of the test sites the heifers were exposed, six at a time, to hot, humid conditions (dry bulb temperature, 40.5 C, wet bulb temperature, 35.0 C) in a psychrometric chamber for a period of six to seven hours to induce thermal sweating. During the exposure period, feed and water were withdrawn. At the end of the exposure the filter paper patches were removed. The absorbed secretions were squeezed into clean sterilized test tubes and transferred to a refrigerator for further processing. Due to the small quantities that could be collected from a single patch, a composite sample was made from all six animals of the same breed exposed on the same day. The samples were diluted and aliquots prepared for the various analyses.

The samples were examined for color, hydrogen ion concentration, and quantitative analyses of some inorganic and organic constituents. A Beckman pH meter was used for determining the hydrogen ion concentration. The total protein nitrogen was estimated by the micro-Kjeldahl and distillation method described by Hawk et al. (6), using boric acid with Tashiros indicator. Deproteinized filtrate was used in analyses for the remaining constituents. For deproteinization a modification of the method of Folin-Wu (6) for determination of nonprotein nitrogen was employed. Instead of using 1 ml distilled with 7 ml of water, 5 ml of the aliquot were used. Into this was put 3 ml of distilled water, to make up the same volume. One milliliter of 10% sodium tungstate solution was added. While slowly shaking, 1 ml of 2/3 N sulfuric acid also was incorporated. The flask was stoppered and well shaken, then allowed to stand for ten minutes. The mixture was filtered by double Whatman filter paper (no. 40) to obtain a clear filtrate. The nonprotein nitrogen fraction was determined by micro-Kjeldahl and distillation method using 2% boric acid containing bromoresol green indicator. The chloride concentration was determined by the Whitehorn method (17) and lactic acid recorded photometrically by the method of Barker and Summerson (1). For reducing sugar, the procedure adopted was that advocated by Umbreit et al. (14) using anthrone reagent. Inorganic phosphorus content was derived by the method of Fiske and Subbarow (5). Limitations on laboratory facilities prevented consideration of analyses for other elements.

Results and Discussion

The collected secretions ranged from dark gray to deep brown in color. This is in contrast to the usual absence of color thermally induced sweat of man, but similar to that reported by Mc Dowell et al. (7) for Jerseys and Holsteins and for the horse by Smith (12). Addition of a few drops of ferric chloride to the secretion gave a typical gray precipitate which dissolved when excess ferric chloride was added. According to the work of Van Jerksack-Pollack as described by Hawk et al. (6), such a reaction indicated the pigment melanin was the major source of the initial coloration. The melanin might have been excreted along with other cytoplastic inclusions of the sweat gland epithelial cells. The incidence of colored apocrine secretion (chromadrosis) has also been observed in other species. For instance, Shelley and Hurley (11) described the blood sweating in the hippopotamus as an example of secretion of pigments by the sweat glands themselves.

The pH values and means for certain inorganic and organic constituents are presented in Table 1. The secretions were uniformly alkaline in reaction, due either to the increased elimination of basic substances in the sweat or because of loss of CO2 (along with other volatile substances) when the fluid was exposed to air during and after collection. CO2 has been found to be given off from the skin in appreciable amounts during sweating, particularly in man (8). It influences the reaction of sweat by its presence either as carbonic acid or as bicarbonate. Bacteriological decomposition of urea, found to be present in unsecreted sweat, Weiner and Hellman (18), might be yet another cause of increased alkalinity, this being accompanied by production of ammonia. Pemberton and Cruoner (9) pointed out that initial sweat was usually more acid than that secreted later, if conditions favorable for sweating were maintained. This change in reaction was assumed as being of compensatory nature, to check systematic alkalosis which would develop during
prolonged exposure to heat. The significance of such a high alkaline reaction of sweat is difficult to explain, unless it is to keep the proteins in solution, as has been suggested by Smith (12). The high pH values agree, however, with those of Taneja (13) for cattle and those for horses by Smith (12).

The presence of inorganic phosphorus may represent, among other things, an increased phosphatase activity in the body with consequent increase in its concentration and circulation in blood. Blineoe and Brody (2) suggested the phosphoric acid concentration in blood might in some way be related to the tolerance of cattle to heat, as high levels seemed to be associated with high adaptability to extreme temperature. Burge and Blineoe (3) noticed that at air temperatures above 30 C there was an increase in the level of blood inorganic phosphorus in Jerseys and Holsteins. VanLandingham et al. (15) also noticed a higher blood phosphorus level in cows during summer and early fall than during winter and spring. The high inorganic phosphorus level in our study may, therefore, result from increased indigenous production in response to thermal stress. The difference in the concentration of inorganic phosphorus in the thermogenic sweat of man and that observed in this study could be due to differences in mode of secretion. In man, the sweat gland cellular structure does not participate in composing its secretion.

Such high total nitrogen contents (Table 1) could be of great practical significance. Animals of these three breeds had high sweating rates under artificially controlled thermal stress environments (unpublished data). If such losses occurred continuously or during long seasons of adverse climatic conditions, the debilitating influences could contribute to the usual observed reduction in performance by cattle. Loss of general muscle tone was observed by Smith (12) in horses sweating freely for long periods. Portions of epithelium may have adhered to the filter paper and made some contribution to the protein portion. Nevertheless, total protein nitrogen is similar to that reported by McDowell et al. (7) for Jerseys exposed to 35 C and is within the ranges reported by Ritter (10) and Smith (12) for sweating horses.

The high concentration of total nonprotein nitrogen fraction indicates the magnitude of the role played by sweating as a source of clearance of various waste products from the body. These levels agree with that reported for man by Weiner et al. (16) and for horses by Ritter (10).

Although the concentrations of chloride varied among breeds and exposure periods (Table 1), all the values were much lower than the 440 to 550 mg/100 ml NaCl given for whole blood of bovines (4). Such differences signify active participation of the sweat glands in formation of the secretions collected. The low values for chlorides agree with the findings of Taneja (13) in European breeds and support the conclusion of Yeates (19) that there is little need for salt replacement in the diet of cattle when exposed to high temperatures.

The concentration of total reducing sugars (Table 1) corresponds to the significant amounts of glycogen found in thermally induced secretions of Red Sindhi-Jersey crosses reported by McDowell et al. (7). If under hot conditions sweating in cattle is an active phenomenon, the transformation of glucose or lactic acid would

### Table 1. Composition of pooled samples of skin secretions collected from Hariana, Tharparkar, and Gir heifers at temperatures of 40.5 C dry bulb and 35.0 C wet bulb.

<table>
<thead>
<tr>
<th>No. obs.*</th>
<th>pH</th>
<th>Inorganic P</th>
<th>Total protein nitrogen (mg/100 ml)</th>
<th>Non-protein nitrogen (g/100 ml)</th>
<th>Chlorides (mg/100 ml)</th>
<th>Total reducing sugar (mg/100 ml)</th>
<th>Lactic acid (mg/100 ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Hariana</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>7.6</td>
<td>15.3</td>
<td>3.8</td>
<td>311.8</td>
<td>100</td>
<td>70</td>
<td>130.4</td>
</tr>
<tr>
<td>2</td>
<td>7.7</td>
<td>14.6</td>
<td>2.5</td>
<td>280.6</td>
<td>100</td>
<td>60</td>
<td>46.4</td>
</tr>
<tr>
<td>3</td>
<td>7.7</td>
<td>13.8</td>
<td>3.0</td>
<td>300.9</td>
<td>135</td>
<td>64</td>
<td>80.3</td>
</tr>
<tr>
<td><strong>Tharparkar</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>7.6</td>
<td>16.2</td>
<td>2.6</td>
<td>150.9</td>
<td>150</td>
<td>89</td>
<td>116.0</td>
</tr>
<tr>
<td>2</td>
<td>7.6</td>
<td>15.7</td>
<td>3.2</td>
<td>201.3</td>
<td>166</td>
<td>48</td>
<td>50.5</td>
</tr>
<tr>
<td>3</td>
<td>7.9</td>
<td>2.9</td>
<td>111</td>
<td></td>
<td></td>
<td>49</td>
<td>46.2</td>
</tr>
<tr>
<td>4</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>160</td>
<td>56</td>
</tr>
<tr>
<td><strong>Gir</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>7.6</td>
<td>14.7</td>
<td>3.0</td>
<td>125.8</td>
<td>250</td>
<td>80</td>
<td>100.0</td>
</tr>
<tr>
<td>2</td>
<td>7.8</td>
<td>14.7</td>
<td>2.8</td>
<td>475.7</td>
<td>100</td>
<td>68</td>
<td>102.1</td>
</tr>
<tr>
<td>3</td>
<td>8.3</td>
<td>3.0</td>
<td>502.0</td>
<td></td>
<td>200</td>
<td>100</td>
<td>100.5</td>
</tr>
<tr>
<td>4</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>381.7</td>
<td>270</td>
</tr>
<tr>
<td>5</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>283</td>
<td></td>
</tr>
<tr>
<td><strong>Avg</strong></td>
<td>7.8</td>
<td>15.0</td>
<td>3.0</td>
<td>303.4</td>
<td>169</td>
<td>65</td>
<td>85.6</td>
</tr>
</tbody>
</table>

* Each observation represents aliquot for six animals tested the same day.

J. Dairy Science Vol. 51, No. 6
provide a source of energy for the production and secretion of strongly hypotonic sweat. A prerequisite for such a theory is that lactic acid must be actually synthesized in the sweat gland and not merely cleared from blood plasma. The high concentration of lactic acid in the secretions obtained (Table 1) as compared to the 5 to 20 mg/100 ml given for whole blood of cattle (4) confirms the validity of this hypothesis. The small amount of glycogen in the sweat glands of Ayrshires found by Yang (18) under low temperatures, further suggests participation of the sweat glands in the secretions produced under thermal stress.

During the period the secretions remained on the body surface before samples were collected, some water and possibly salts may have been reabsorbed. This could account for high values for some constituents recorded in this study. However, the general agreement with other reports for cattle indicates the values are reasonable and also that the composition of sweat from breeds originating in India is similar to that for European breeds.

In this study differences among breeds were nonsignificant.

Acknowledgment

These investigations were financed in part by a grant made by the United States Department of Agriculture, Agricultural Research Service, under Public Law 480.

B. C. JOSHI
H. B. JOSHI
R. E. McDOWELL
and
D. P. SADHU

Indian Veterinary Research Institute
Izatnagar, U. P. India
and
Cornell University, Ithaca, New York

References