Experimental Production of Squamous-Cell Carcinomas in the Skin of Chickens*

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Squamous-cell carcinomas, hemangiomas, fibromas, and papillomas have occurred in the skin of ducks following the local application of a 0.25 per cent acetone solution of methylcholanthrene daily for a period of 1 month (5, 6). The spontaneous disappearance of a high per cent of these neoplasms in the duck has proved to be a feature of considerable interest (7). Attempts have been made infrequently to produce neoplasms in fowls by the local application of carcinogens to the skin. Hartwell (2), in 1951, cited four experiments in which the skin of fowls was treated with 3,4-benzpyrene. In one the skin of pigeons was used, and one spinocellular epithelial neoplasm developed. In three experiments the skin of fifteen chickens was treated, and one bird developed an epithelioma.

The spontaneous occurrence of carcinomas in the skin of fowls is apparently infrequent. Olson and Bullis (4) reported one case of a squamous-cell carcinoma in the skin of a chicken. Feldman and Olson (1), in 1944, cited only a few previously reported cases in their study of neoplastic diseases in chickens. These authors stated "In the cases we have observed the neoplasms have remained localized, and in the cases reported previously by others, metastasis has rarely been demonstrated."

In this study methylcholanthrene was applied to the skin of chickens, and squamous-cell carcinomas developed.

METHODS AND MATERIALS

The chickens were 2-day-old Rhode Island Reds obtained from a commercial hatchery. They were kept for 42 days in the laboratory, at which time a 0.25 per cent solution of methylcholanthrene in acetone was first applied locally to the skin beneath the right wing of eighteen chickens. This carcinogen was applied daily for 1 month thereafter. It was dropped onto the skin, and approximately 1–2 ml. was used daily per bird. Acetone was applied in a similar manner to six control chickens.

All chickens were kept in a battery and observed frequently. Chick Startera and Growena (Purina) and water were available ad libitum. The neoplasms that developed in the treated areas of skin were observed macroscopically, and many were photographed at intervals. Some of the lesions were excised for histological study; they were fixed in a 4.0 per cent solution of formaldehyde. Paraffin sections were prepared and stained routinely with hematoxylin and phloxine. A few of the chickens with tumors were sacrificed and autopsied. This material was also studied histologically. The majority of the neoplasms were allowed to regress completely, and the birds were then sacrificed and autopsied.

RESULTS

Six chickens did not show any gross changes in the skin and were discarded on the 86th day following the last application of acetone. During the time the chickens were treated with the acetone solution of methylcholanthrene, a few developed local areas of superficial ulceration. The methylcholanthrene was discontinued for several days. These ulcerated areas completely healed within 30 days, and the feathers continued to grow.

Eight of the eighteen methylcholanthrene-treated chickens did not develop any neoplasms and were discarded on the 86th day after the last application of the carcinogen. Approximately 60 days after administration of the carcinogenic agent was discontinued, papillomas and focal areas of ulceration began to appear in the treated areas of skin. These lesions continued to appear for an additional 40 days. One chicken, however, developed new lesions in the treated areas of skin 5 months after the last application of methylcholanthrene. The papillomas were usually small. Histologically, they showed the typical characteristics of such a lesion. The focal areas of ulceration...
tion were usually round or oval, their margins were slightly indurated, and the base was granular. They varied in size from 0.5 to 1.5 cm. (Fig. 1). Five such lesions on four different chickens were removed for histological study. One chicken, #589, with six tumors was also sacrificed, and the lesions were studied histologically. The base of these ulcers showed a wide zone of squamous epithelial cells (Figs. 3, 4, and 5). Intercellular bridges and epithelial pearls were present (Figs. 4, 7, and 10). The epithelial cells varied widely in size and shape, as well as in their staining reaction (Figs. 8 and 9). In some of these ulcers extensive degenerative changes had occurred, while in others the tumor cells were well preserved. Necrosis and leukocytic infiltration were conspicuous in some of the lesions. Usually, these ulcers were also infiltrated with a large number of mononuclear cells that resembled lymphocytes and plasma cells. Large groups of lymphocyte-like cells were present about the base of some of these carcinomatous ulcers (Figs. 3 and 7).

Five chickens had from one to three ulcerated areas, varying in size from 0.5 to 1.5 cm. in diameter and 1 to 3 mm. in depth. Each lesion progressively healed spontaneously within 20–60 days. When these chickens were sacrificed 1–2 months later, there was no evidence of either local recurrence or metastasis.

Three chickens developed typical papillomas. One was removed, and it had the characteristic microscopic features of such a tumor. Usually only one or two papillomas occurred in a single bird. They were small—3–5 mm. in diameter. Only one reached a height of 1.0 cm. These papillomas ultimately disappeared spontaneously. Three chickens had several papillary-like lesions in the skin that could not be diagnosed grossly as either a benign or malignant lesion. These also spontaneously regressed.

The process of spontaneous regression of these ulcerated lesions is shown in Figures 1 and 2. Grossly, these regressing lesions were identical with those ulcers removed and showed the histological features consistent with the diagnosis of squamous-cell carcinoma. The rate of regression varied. Some completely regressed within a period of 3 weeks, while others slowly regressed over a period of 6 weeks.

DISCUSSION

The skin of five chickens from the group of eighteen treated daily for 30 days with a 0.25 per cent solution of methylcholanthrene developed one or more ulcerative lesions that showed histologically the characteristics of a squamous-cell carcinoma. No metastasis occurred. Five other chickens with similar lesions had a complete and spontaneous regression of the lesion. The mechanism by which this regression occurs is unknown. It is of interest, however, to observe the extensive degeneration of the epithelial cells and to note the accumulation of reticulo-endothelial cells in and about the periphery of these neoplasms. Whether this cellular response is "cause or effect" is difficult to state at this time. It is of interest to find a similar cellular reaction about squamous-cell carcinomas and hemangiomas in the skin of white Pekin ducks treated with methylcholanthrene that likewise spontaneously regressed (5, 7). A similar cellular reaction has been observed in the spontaneously developing and regressing hemangiomas in chickens (8).

Some may question the diagnosis of squamous-cell carcinomas in these chickens, since none of the tumors metastasized and all spontaneously regressed. Histologically, these tumors, in our opinion, have all the characteristics of a malignant neoplasm, as shown in the group of illustrations. We do not think that these squamous cells...
Fig. 7.—Chicken #584. Squamous cells and epithelial pearls are present in base of this ulcer. Note their extension into adjacent fatty stroma. Frequently lymphocytes are present at base of these neoplastic lesions. H&E X 140.

Fig. 8.—Chicken #620. This tumor is characterized by sheets of epithelial cells extending down into the underlying stroma. H&E X 140.

Fig. 9.—Chicken #620. Variations in size, shape, and staining reaction occur in these tumor cells. Mitotic cells are frequent. H&E X 855.

Fig. 10.—Chicken #620. Some of the squamous epithelial cells show intercellular bridges. H&E X 1425.
at the base of the lesion represent a pseudo-epitheliomatous hyperplasia. They are interesting in view of the studies of Rous and associates (8, 9) with tarring of rabbits. These investigators observed that “tarring itself will provide the needed encouragement, but when it is left off the growths disappear unless other factors act to maintain them, as e.g. chronic pathological changes in the supporting tissue, or inflammation due to bacterial infection or to crowding” (5). These tumors in the chicken followed by several weeks the last application of methylcholanthrene, and apparently the only injury that might have occurred would have been the plucking of feathers. In these neoplastic lesions in the chicken there were no macroscopic changes to suggest a preceding benign type of lesion, such as has been described in rabbits (9). Rous and Kidd (9) state “When the chemical agents commonly termed carcinogenic are applied to the skin of man, the rabbit, or the mouse, they nearly always elicit benign growths some while before cancer appears, and the latter frequently takes origin from one or another of them.” Could it be that a neoplastic growth, such as these in the chicken, requires some “factor” to keep the cells progressively developing, while the absence of such a “factor” results in spontaneous regression of the growth?

It is of interest to find that methylcholanthrene, when applied locally to the skin of young Rhode Island Red chickens, produces primarily a squamous-cell carcinoma, while the same carcinogen applied to the skin of white Pekin ducks produces primarily hemangiomas. The absence of a lymphatic system in the skin of fowls, like that in mammals, may be a significant factor affecting the development, regression, and spread of malignant tumors.

SUMMARY

A histological squamous-cell carcinoma occurred in the skin of five chickens in a group of eighteen treated with an acetone solution of methylcholanthrene. Five other chickens, similarly treated, developed one or more ulcerative lesions macroscopically identical to those studied histologically and referred to above. These latter lesions spontaneously regressed within a period of 2–8 weeks.

REFERENCES

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