

THE EFFECT OF THE X-RAY ON THE NODULES OF
VERRUGA PERUANA

By HENRY R. MULLER, M.D., AND JOSEPH R. TYLER

(From the Laboratories of The Rockefeller Institute for Medical Research)

PLATE 1

(Received for publication, October 21, 1929)

In a recent paper from this laboratory¹ it was shown that convalescent serum, taken from monkeys which had recovered from experimental verruga, has two evident effects on the disease as produced in monkeys by the inoculation of cultures of *Bartonella bacilliformis*.

The convalescent serum inhibits the development of verrugous nodules, that is, delays the appearance or slows-up the growth, but it does not prevent their ultimate appearance and their reaching considerable size. On the other hand, it effectively prevents the proliferation and persistence of *Bartonella bacilliformis* in the blood stream. It is hoped that the treatment of cases of Carrion's disease in man may be made more effective through the employment of the blood serum of convalescents from verruga and from Carrion's disease itself.

The curative influence of the X-ray on various skin lesions in man has suggested experiments to ascertain whether the rays, applied therapeutically, would affect the evolution of verruga nodules in monkeys receiving skin inoculations of *Bartonella bacilliformis*. The pathology of the lesions indicated that this was probable. In histological structure the verruga nodule^{2,3,4} is a circumscribed collection of proliferating endothelial cells, arranged in groups and masses along blood and lymph spaces, in a manner resembling an endothelioma. Ordinary inflammatory changes such as exudation of polynuclears and round cells occur in mild degree only. Furthermore, the verruga nodules in spite of their definitely proliferative nature tend to regression ultimately. In this they differ essentially, of course, from the endotheliomata. The time period during which evolution and regression take place can be gauged quite accurately.

With these facts in mind, we undertook two sets of experiments.

In one we wished to observe the effect of a single application of X-ray on the nodules after they had just started to develop in monkeys, and in the other, to see whether the skin of a normal monkey which had received one application of X-ray would fail to develop verruga nodules on later inoculation.

Method

After the monkey had been shaved it was stretched on a board and the chest and abdomen were covered with a lead-rubber mat 1/16 inch thick. The mat was so placed that the nodule to be treated was in the center of a hole in it 3 cm. square. It follows that an area of 9 square cm. in all was exposed to the X-ray, having at its center the nodule to be treated.

An attempt was made to find and use an X-ray dosage sufficient to produce only an erythema of the skin. Four, six and eight minute exposures at a distance of six inches, with a three inch spark gap and 10 M.A. were tested. It was found that sometimes a six or an eight minute exposure produced desquamation and a serous discharge from the area X-rayed, but in the type experiments reported here only erythema occurred.

Treatment of Growing Nodules

Macacus rhesus X-1 was intradermally inoculated with living cultures of *Bartonella bacilliformis*, strain P5, in four places, two on the left side and two on the right side of the abdomen. An inoculation was also made between them into a scarified area on the midline. On the sixth day, nodules 3 to 4 mm. in diameter were distinctly visible and palpable at all the sites of intradermal inoculation (Fig. 1). The following day the two nodules on the left side were treated with X-ray, the left upper nodules for six minutes and the left lower nodule for only four minutes. The nodules on the right side and the scarified area were not exposed to the X-ray, serving as controls. These nodules on the right side progressed steadily; 26 days after inoculation the upper one was 12 mm. and the lower 20 mm. in diameter (Fig. 2). Seventy days after inoculation they had completely regressed and only small scars remained. The nodules on the left side, however, during this period, when those on the right were flourishing, remained stationary in size for about 14 days, and on the 46th day after inoculation had entirely disappeared.

Macacus rhesus X-2 was injected intradermally in six places on the abdominal wall, 3 on each side, with cultures of *Bartonella bacilliformis*, strain 2. A scarified area on the midline was also inoculated. Ten days later each intradermal site of inoculation showed a nodule 4 mm. in diameter (Fig. 3). The next day the nodules on the left side were treated with X-ray, 8 minutes for the upper nodule, 6 minutes for the middle nodule and 4 minutes for the lower nodule. The nodules on the right side and the scarified area were not treated and served as controls. On this,

the untreated side, the nodules were 15 mm. in diameter 34 days after inoculation (Fig. 4), and had completely regressed 63 days after inoculation. The nodules on the left, the treated side, remained stationary for about 10 days after treatment, with the surrounding skin slightly hyperemic and edematous. They then gradually regressed and disappeared (Fig. 4).

Treatment of the Skin Prior to Inoculation

In *Macacus rhesus* X-5 the shaven skin of the anterior surface on the left side was exposed to the X-ray for 6 minutes, while the right side remained untreated and served as control. Then, 4 days later, two intradermal inoculations of cultures of *Bartonella bacilliformis*, strain 4, were made on each side of the animal; i.e., two on the treated and two on the untreated side. The nodules on the right, the untreated side, showed the usual progress. On the 46th day after inoculation they were two large pedunculated masses, each 20 mm. in diameter (Fig. 6), and 42 days later they had entirely disappeared. On the left, or treated side, there was marked retardation of the development of the nodules. Eight days after inoculation, they were just palpable and much smaller than those on the right which were red, prominent and 5 mm. across. On the 23rd day the nodules on the right were 10 mm. and 5 mm. respectively, and those on the left had entirely disappeared.

SUMMARY AND CONCLUSIONS

The supposition that X-ray would affect the developing and the developed verruga nodules experimentally induced in the monkey, has proved correct. The experiments show that the early verruga nodules when exposed to a single, properly graduated dose of X-ray producing merely erythema of the skin, are inhibited in their evolution. Moreover, the skin of *Macacus rhesus* monkeys is modified by a single erythema dose of X-ray in such a way that infection of it with *Bartonella bacilliformis* is rendered more difficult.

These results are sufficiently striking to warrant the trial of X-ray in suitably guarded doses in the treatment of verruga nodules in man. Should the employment of convalescent serum influence the course of Carrion's disease favorably and the use of X-ray bring about a more certain and rapid devolution of verruga nodules in man, two practically applicable therapeutic measures will have been provided for the treatment of the two forms of the human disease.

REFERENCES

1. Noguchi, H., Muller, H. R., Tilden, E. B., and Tyler, J. R., *J. Exp. Med.*, 1929, 50, 355.
2. da Rocha-Lima, H., *Verhandl. d. deut. path. Gesells.*, 1913, 16, 409.
3. Noguchi, H., *J. Exp. Med.*, 1927, 45, 455.
4. Noguchi, H., Muller, H. R., Tilden, E. B., and Tyler, J. R., *J. Exp. Med.*, 1929, 50, 455.

EXPLANATION OF PLATE 1

FIG. 1. *Macacus rhesus* X-1, showing condition of verruga nodules 7 days after inoculation, before treatment with X-ray.

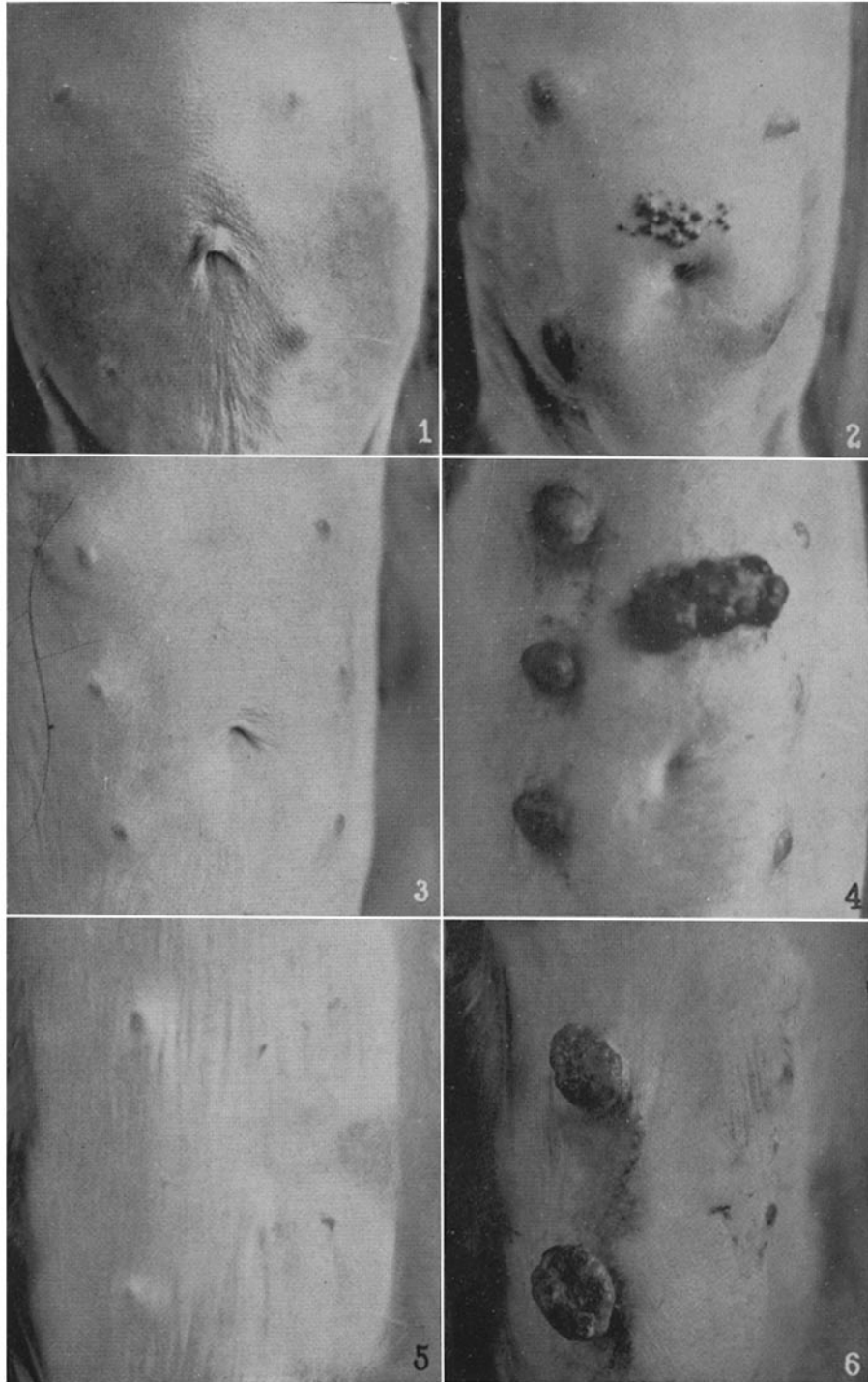
FIG. 2. Same animal 21 days after X-ray treatment. The two nodules on the left side are considerably smaller than the two untreated nodules on the right. Scarified area in the center is untreated.

FIG. 3. *Macacus rhesus* X-2. The six nodules 11 days after inoculation and before X-ray treatment.

FIG. 4. The same animal 23 days after X-ray treatment. The three nodules on the right side of the animal and the scarified area are strongly positive. The three treated nodules on the left side are very much smaller.

FIG. 5. *Macacus rhesus* X-5. The skin of the left side was X-rayed 4 days before inoculation. Picture taken 10 days after inoculation, shows nodules slightly smaller on left side than on right.

FIG. 6. Same animal, 47 days after inoculation, with two large masses on the right side (untreated), but only two small nodules on the left side (treated).



(Muller and Tyler: Effect of X-ray on verruga peruana nodules)