was recently investigated by Turk, Allison, and Oxman (1962) in guinea-pigs. Their experiments showed that there was little difference in the quantity of virus which multiplied in animals devoid of circulating antibody with delayed hypersensitivity compared with normal animals, and suggested that the spread of vaccinia virus is normally limited by factors, other than antibody and delayed hypersensitivity, among which is interferon. Although vaccinia virus is sensitive to interferon (Andrews, 1961) there is nevertheless no evidence that the immune status which an individual develops after vaccination is due to an increased amount of interferon in his skin. Our failure to influence the progress of the lesions in our patient with interferon was perhaps due to the low titre of interferon available at the time, and interferon should certainly be tried in future cases of progressive vaccinia.

It is remarkable that the foot lesion remained static for nine months while the arm and thumb lesions progressed, yet in spite of an actively spreading edge the early arm lesions healed with little scarring.

The only occasion on which there was any apparent sign of remission in the patient was a few days after the fresh blood transfusion during a period of pyrexia. It is possible that this temporary remission may have been due to the transfer of a delayed type of hypersensitivity by the white cells in the donor's blood. In any event it would seem that in future cases of progressive vaccinia, in addition to providing passive antibody, hypersensitivity to vaccinia might be induced by local injection of disrupted white cells or lymph-node suspensions from recently vaccinated individuals as suggested by Kempe (personal communication). The injection of viable immunologically competent cells may cause runt disease (Nisbet and Heslop, 1962), but this can be avoided by using white-cell extracts, which have been shown to be effective in transferring delayed type hypersensitivity to humans (Lawrence, 1960). Alternatively, sensitization might be induced by injecting vaccinia virus which had been inactivated by ultra-violet radiation or formalin.

We considered the possibility of surgical removal of the lesions in our patient but were dissuaded from this by the very slow progress of the condition, by the fact that the healed areas showed little scarring, and by the hope that the child might eventually overcome the infection.

There was no evidence that *N*-methylisatin β -thiosemicarbazone had any effect in controlling the progress of the disease or the cutaneous lesions, and the drug appeared to be toxic. This compound has caused no abnormalities when subjected to chronic toxicity tests in animals and no ill effects were observed in two adult human volunteers to whom the drug was given (Bauer and Goodwin, 1962), nor in an adult patient with leukaemia who developed progressive vaccinia (Davidson and Hayhoe, 1962).

Summary

The failure of large doses of antivaccinal gamma-globulin or *N*-methylisatin β -thiosemicarbazone to control the progress of infection in a fatal case of progressive vaccinia is described. Possible measures for treating further cases are discussed.

We thank Dr. R. Hall, who looked after this patient initially, and Dr. I. J. Carré for medical care during the first

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Preliminary Communications

Eczema Vaccinatum Treated with *N*-Methylisatin β -Thiosemicarbazone

Much work has been done in recent years on the chemotherapy of the pox virus infections with isatin β -thiosemicarbazone and its derivatives. The compound itself will confer protection against death and development of symptoms in mice infected intracerebrally with 100,000 LD₅₀ of vaccinia virus (Bauer, 1955), and is equally effective against rabbit-pox virus (Bauer and Sheffield, 1959). A number of derivatives have been found with antiviral activity greater than that of isatin β -thiosemicarbazone itself (Bauer and Sadler, 1960a); of these, the *N*-ethyl derivative has been found to be active in mice infected with alastrim virus (Bauer and Sadler, 1960b), and the same compound and a number of others have been found to possess antiviral activity in mice infected with variola major virus (Bauer *et al.*, 1962). The antiviral activities observed have in all cases been sufficiently great to indicate that some of these compounds might be of use in the specific chemotherapy of smallpox and vaccinia infection in man, and in the present communication an account is given of the use of *N*-methylisatin β -thiosemicarbazone (compound 33T57) in the treatment of eczema vaccinatum.

CASE REPORT

The patient is a boy born on July 7, 1961, who had suffered from infantile eczema since the age of 6 weeks; at the age of 4½ months he was seen by a consultant dermatologist (Dr. J. B. MacMillan), and on treatment with a steroid-containing cream the lesions cleared. He was not vaccinated, but on January 18, 1962, both parents were successfully vaccinated, the mother having a primary vaccination, the father a secondary vaccinia with the immune accelerated reaction. On January 30 two small pustules appeared on the baby's scalp; these were irritable, and the parents applied some of the steroid cream previously prescribed for the eczema. Within three days typical vaccinia lesions appeared all over the scalp and became

almost confluent, with spread on to the face and to a less extent on to the shoulders and upper and lower limbs. Massive oedema of the scalp was a very marked feature at this stage. The distribution of the lesions was typical of the eczematide dispersion.

The child's general condition was extremely grave, with persistent pyrexia, prostration, and toxæmia. Routine blood investigations and plasma protein estimations with electrophoresis were all normal. Vaccinia virus was isolated from the pustular fluid by Dr. J. D. Abbott, of the Public Health Laboratory, Manchester.

Treatment with antibiotics and vaccinia hyperimmune gamma-globulin did not have any beneficial effect: the lesions continued to extend and new lesions developed and the condition remained very grave. On February 13 treatment with 33T57 was begun; the drug was administered orally, 1 g. daily in divided doses of 250 mg. every six hours. The pustules continued to spread and a few new lesions erupted until February 15 and 16, when there was a sudden and dramatic improvement in the child's general condition. This change was accompanied by marked regression of the rash and subsidence of the oedema of the scalp: no further pustules developed. The hitherto purulent lesions dried, became crusted, and healed with little or no scarring.

On February 20, at a time when severe gastroenteritis was occurring in the same ward, severe and persistent diarrhoea and vomiting set in and the drug was discontinued. Unfortunately these symptoms persisted, the condition and nutritional state deteriorated, and the child became very ill and remained so. Continuous intravenous medication and alimentation were necessary, and it was not until March 12 that these symptoms abated. The child has now recovered.

DETECTION AND MEASUREMENT OF 33T57 IN PLASMA

One millilitre of a sample of heparinized venous blood taken on the second day of treatment with 33T57 (which had haemolysed somewhat in transit) was acidified with 0.1 ml. of 2 N HCl and shaken for 30 seconds with

Fig. 1.—Extinction curve of a toluene extract of human plasma (full circles) and of the same plasma to which had been added 33T57 at 10 μ M (open circles).

1 ml. of toluene. After centrifuging for 20 minutes at 2,500 g the extinction of the toluene layer was measured at intervals from 340 to 440 m μ with the Unicam SP 500 spectrophotometer fitted with micro-cell attachment (10 mm. light-path). Of the toluene solution 0.5 ml. was then basified with 0.1 ml. of 0.05 M methylbenzethonium hydroxide in ethanol and a second series of measurements made.

Toluene extracts of human plasma made in this way absorbed very little light at wavelengths above 350 m μ (Fig. 1): extraction of plasma to which had been added a small amount of 33T57 led to an almost quantitative recovery of the drug and to a solution having a characteristic extinction curve with a peak at about 375 m μ (Fig. 1). Basification of this extract resulted in a spectral shift, so that the peak was now found near 400 m μ . An extract of the patient's plasma had a very similar extinction curve in the 340 to 390 m μ region (Fig. 2), and on basification showed that spectral shift characteristic of extracts of plasma containing the compound.

Fig. 2.—Extinction curve of a toluene extract of the patient's plasma before (open circles) and after (full circles) basification.

From the height of the two peaks in Fig. 2 it was calculated that the plasma sample contained drug at a concentration of about 8 μ M (190 μ g./100 ml.), a level which had been found effective in tissue-culture experiments (Sheffield, 1962). This figure cannot be exact, for comparison of Figs. 1 and 2 shows that the shape of the patient's curve at wavelengths above 390 m μ differed from the "standard" curve: preliminary experiments have suggested that this was in part due to the presence of haemolysis. Moreover, the curve established after basification had a rather sharper peak (although at the same wavelength) than the standard curve. Similar effects have been observed in samples taken from experimental animals dosed orally with 33T57 and may suggest there to be present in the circulation a product of the drug that has undergone some small metabolic change.

COMMENT

Vaccinia in a child of this age and of such extent and severity carries with it a very unfavourable prognosis, and until the drug was administered this child appeared to be no exception. The failure of the natural defences and of the hyperimmune gamma-globulin to control the condition and the rapid and dramatic improvement three days after starting therapy with *N*-methylisatin β -thiosemicarbazone (Compound 33T57) suggest that the drug was responsible for the resolution of the lesions. The diarrhoea may have been a complication of treatment with this drug, but as at the time there were other patients in the ward suffering from a severe form of gastroenteritis, cross-infection from one of these cannot be excluded.

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WINSTON TURNER, M.D., M.R.C.P., D.C.H.,
Consultant Paediatrician, Burnley and District Hospitals.
D. J. BAUER, M.A., Ph.D., M.B., B.Chir.,

R. H. NIMMO-SMITH, M.A., D.Phil., M.B., Ch.B.,
Wellcome Laboratories of Tropical Medicine, London.

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Medical Memoranda

Spontaneous Rupture of Spleen Infiltrated by Secondary Carcinoma

Spontaneous rupture of an enlarged, congested, or friable spleen has been well documented, but rarely in association with a carcinoma of the colon. Ellerker (1955) described a case where a carcinoma of the splenic flexure had become adherent to the splenic veins, resulting in thrombosis and septic infarcts. The case here described is apparently unique in that the only demonstrable cause of the splenomegaly was diffuse infiltration by a signet-ring-celled carcinoma, histologically identical with a primary tumour of the pelvic colon.

CASE REPORT

A 53-year-old woman was admitted to hospital on January 12, 1957, complaining of colicky lower abdominal pain, loss of weight, and the passage of blood and slime per rectum. The only abnormal physical sign was blood on the glove finger after rectal examination. A carcinoma was seen on sigmoidoscopy, but biopsy showed non-specific chronic inflammatory changes only. A barium enema on January 30 showed no lesion. However, it was felt that a laparotomy was essential, and on February 6 this confirmed a carcinoma of the pelvic colon.

Sections of this tumour showed it to be a mucus-secreting, signet-ring-celled carcinoma (Fig. 1). Three lymph nodes at the level of the primary showed secondary invasion. The patient's recovery was uneventful.

On January 25, 1958, she was readmitted in a shocked state, semicomatose, and hypotensive, with a 12-hour history

of epigastric pain and vomiting. Examination revealed a flaccid abdomen and an easily palpable tender spleen.

Resuscitative measures (intravenous dextran and nor-adrenaline) restored the blood-pressure to 120/80 and she recovered sufficiently to give a lucid history. Two weeks previously she had had an upper respiratory illness from

FIG. 1.—Section of primary from pelvic colon showing infiltration of submucosa by signet-ring-celled carcinoma. (Haematoxylin and eosin.)

which she had completely recovered. She denied loss of weight, trauma to the abdomen, malarial infection, or sore throat. Two hours later she suddenly collapsed and died.

Necropsy Report.—The peritoneal cavity contained 2 pints (1,140 ml.) of clotted blood. The spleen was much enlarged, firm, and friable, measured 16 by 13 by 8 cm., and had a deep irregular rupture, 3 cm. in length, on the lateral surface. Both the splenic artery and vein were externally normal and their lumina patent. There were no infarcts or subcapsular haematomas. A few para-aortic lymph nodes were apparently involved by secondary growth, but they in no way encroached on the lumen of the splenic artery and vein. There were no splenic aneurysms or arterio-venous anomalies and no portal or splenic vein thrombosis. No bruising of the abdominal wall or perisplenic adhesions were seen. All other viscera were pale but normal.

Examination of Spleen.—There was no obvious carcinomatous infiltration, and all the hilar vessels and their branches were patent. Blocks taken from the hilum, perimeter, and central portions of the body showed diffuse infiltration of the splenic pulp with typical (mucus-secreting) signet-ring-celled carcinoma (Fig. 2).

Blocks taken from the enlarged para-aortic lymph nodes showed no microscopical evidence of secondary tumour.

FIG. 2.—Section of spleen showing diffuse infiltration by mucus-secreting signet-ring-celled carcinoma. (Haematoxylin and eosin.)