*Ind. Jour. Med. Res.*, 57, 5, May, 1969.

# 6 AZAURIDINE IN SMALLPOX.

S. M. H. JAFFARI*, AND AZHER HUSSAIN†.

(*From the Institute of Medical Sciences and Fever Hospital, Hyderabad.*)

[Received for publication, October 14, 1968.]

## INTRODUCTION.

HAMRE, Bernstein and Donovick (1950) laid the foundations of antiviral chemotherapy, and in 1960 clinical trials of a thiosemicarbasone against vaccinia virus was taken up. Since then, the progress was rapid. Marboran was tried with unsuccessful results. Idoxuridine was introduced with optimism but its toxic reactions curtails it further and free application. M & B 7714, CG 662, wild banana seeds and many other drugs were evaluated with doubtful result. Then Rada *et al.* (1960) and Rada and Blaskovic (1966) reported that 6 Azauridine (AzUR)—an antimetabolite inhibit vaccina virus multiplication. Jaffari *et al.* (1968) observed that the multiplication of variola virus on the chorioallantoic membrane was inhibited in the presence of 6 Azaruidine.

6 Azauridine is non-toxic for most of the human cells (Raskova *et al.*, 1965). Doses as high as 270 mg./kg. body-weight were given to patients for a period of three weeks without any untowards effects.

## MATERIALS AND METHODS.

*Patients.*—The unvaccinated smallpox patients admitted in Fever Hospital during 1967-1968 were taken for study. The unvaccinated patient is defined as one who was never vaccinated in life or said to have been vaccinated but has not visible scars, or to whom vaccination was performed during the incubation period of the disease.

After determination of the vaccinial status, irrespective of the type of smallpox, patients were randomized and the treatment letter A and B of the randomization sheet was allocated. To group 'A' drug was given and the 'B' were on placebo.

Only patient in the early stage of rash were included. No patient in the late stage of vesiculo-pustular eruption was taken for study.

*Drug and dose.*—AzUR—the prepartion Riboazauracil (Spofa, Czechoslovakia) was used. This was supplied to us through the Indian Council of Medical Research, New Delhi, India.

A dose of 100 to 200 mg./kg. body weight was given intravenously in two divided doses at an interval of 12 hours for 3 days. The durg was dissolved in 5 per cent glucose and 50 ml. to 100 ml. was slowly infused.

*Parameters employed in assessment of chemotherapeutic activity* :

1. Case fatality rate with reference to type of smallpox.

*Prof. of Infections Diseases and Superintendent, Fever Hospital, Hyderabad.
†Asstt. Prof. of Infections Diseases and Civil Assistant Surgeon, Fever Hospital, Hyderabad.

2. The survival days after initiation of therapy in fatal cases.
3. The action of the drug on focal eruptions.
4. Its effect on febrile reactions and toxicity.
5. Its role in prevention of complications.
6. Any untoward reactions.

*Analysis of the study.*—No patient of haemorrhagic or flat type survived but an analysis of the survival days indicate that the survival days were over 4·2 in the flat variety, in the drug group (refer Tables II and III). In ordinary type of smallpox, the

TABLE I.

*Details of the two groups A and B.*

| | Group A : | | Group B : | |
|---|---|---|---|---|
| | Attacks. | Deaths. | Attacks. | Deaths. |
| 1. Total number of patients | 69 | 28 | 71 | 32 |
| 2. Details of exclusion from analysis | | | | |
| (*a*) Death within 24 hours | 1 | 1 | 1 | 1 |
| (*b*) Absconded | 1 | *Nil* | 1 | *Nil* |
| (*c*) Late admission randomized but excluded | *Nil* | *Nil* | 1 | *Nil* |
| (*d*) Additional treatments given excluded | .. | .. | 1 | .. |
| Net patients for study | 67 | 27 | 67 | 31 |

TABLE II.

*Analysis of the study in relation to the type of smallpox.*

| | Group A : | | | Group B : | | |
|---|---|---|---|---|---|---|
| | Attacks. | Deaths. | CFR. | Attacks. | Deaths. | CFR. |
| Haemorrhagic | 7 | 7 | 100 | 6 | 6 | 100 |
| Flat | 11 | 11 | 100 | 4 | 4 | 100 |
| Ordinary | 49 | 9 | 18·37 | 57 | 21 | 36·8 |

case fatality is less than 55 per cent of the control group. The fatality difference in the two groups is 18·43 per cent, the standard error is 8·45 per cent and as the CFR difference in drug group exceeds twice that of the standard error, this difference is reckoned as statistically significant (Table II). The difference in survival days is 4·4 days (Table III) in the drug group. It appears that the drug has a favourable effect in this type of smallpox.

TABLE III.

*Mean survival days of the deceased patients after initiation of therapy.*

| Types. | Group A : | | Group B : | |
|---|---|---|---|---|
| | Number of patients. | S. days. | Number of patients. | S. days. |
| Haemorrhagic | 7 | 8·3 | 6 | 7·7 |
| Flat | 11 | 7·9 | 4 | 3·7 |
| Ordinary | 9 | 10·3 | 21 | 5·9 |

S. days = Survival days.

The drug has a moderating effect on focal eruptions. In a few patients, the lesions dried up earlier and have not passed the usual expected stages of the rash. In others, the rash reached the scabbing stage earlier. The effect of the drug was significant in all stages of the rash, in enhancing the progress of the lesions to the scabbing stage (Table IV). Over 50 per cent of the placebo group failed to reach the scabbing stage (Table V).

TABLE IV.

*Mean and standard deviation days from the initial stage of focal eruption to initial scabbing.*

| Stage of eruption. | Group A : | | | Group B : | | | Difference in two groups : | | Remarks. |
|---|---|---|---|---|---|---|---|---|---|
| | Number of patients. | Mean days. | S.D. days. | Number of patients. | Mean days. | S.D. days. | Difference in mean days. | S.D. | |
| Papular | 23 | 6·1 | 1·57 | 11 | 10·4 | 3·45 | 4·3 | 0·09 | Significant |
| Papulo-vesicular | 3 | 6·0 | 1·414 | 1 | 9·0 | .. | .. | .. | Not tested |
| Vesicular | 16 | 4·8 | 1·88 | 19 | 7·3 | 2·62 | 2·5 | 0·764 | Significant |
| Vesiculo-pustular | 13 | 4·0 | 1·494 | 11 | 5·3 | 1·513 | 1·3 | 0·616 | Significant |

S.E. = Standard Error. S.D. = Standard Deviation.

TABLE V.

*Analysis of patients who have not reached the scabbing stage.*

| | Group A. | Group B. |
|---|---|---|
| 1. Patients under study | 67 | 67 |
| 2. Number not reached scabbing | 12 | 25 |
| 3. Per cent of the total not reached scabbing | 17·9 | 37·3 |

The drug has an ameliorating effect on fever. The response and the mean febrile days were shorter in the drug group than in the placebo. No case was afebrile in placebo group but 14·3 per cent of the cases in the drug group touched normal at the pustular stage and over 50 per cent were running normal temperature at the scabbing stage, whereas 37·96 per cent were having normal temperature in the placebo group (Tables VI and VII) at the scabbing stage.

TABLE VI.

*Duration of temperature in alive non-complicated patients of ordinary variety of smallpox considered in relation to the initial stage of focal cruption.*

| Stage of rash. | Number of patients. | Group A mean days. | Number of patients. | Group B mean days. | Difference in mean days. |
|---|---|---|---|---|---|
| Papular | 11 | 6·9 | 9 | 10·6 | 3·7 |
| Vesicular | 13 | 4·8 | 16 | 9·0 | 4·2 |
| Vesiculo pustular | 4 | 5·5 | 4 | 11·0 | 5·5 |

TABLE VII.

*The range of temperature at the pustular and scabbing stage.*

| RANGE OF TEMPERATURE : | 102 AND OVER. | | 99°—110° F. | | NORMAL. | |
|---|---|---|---|---|---|---|
| | Group. | | Group. | | Group. | |
| | A. | B. | A. | B. | A. | B. |
| A. *Pustular stage* | | | | | | |
| a. Number of patients | 7 | 11 | 17 | 18 | 4 | *Nil* |
| b. Per cent of the total | 25·0 | 37·9 | 60·7 | 62·1 | 14·3 | *Nil* |
| B. *Scabbing stage* | | | | | | |
| a. Number of patients | 2 | 3 | 10 | 15 | 16 | 11 |
| b. Per cent of total | 7·14 | 10·34 | 35·7 | 51·70 | 51·70 | 37·96 |

Major complications were not seen except multiple abscesses and boils in both the groups. A single case developed encephalitis in the control group. No untoward reactions were observed in the patients on drug.

## DISCUSSION.

6 AzUR is a pyrimidine analogue. It interferes in the synthesis of pyrimidine neucleotides and co-enzyme. It inhibits the decarboxylation of orotidylic acid, and prevent the formation of uridylic acid and nucleic acid, which are essential for final synthesis of the virus particles (Butto *et al.*, 1965 ; Skoda and Sorm, 1959).

The controlled trial revealed that the 6 AzUR has no effect on the haemorrhagic and flat type but in the ordinary type of smallpox, it lowered the lethality by 50 per cent. This effect is significant.

The survival days in all the types were encouraging. It exceeded over 4 days in the drug than control group.

The temperature responded well and this response was more appreciable in the pustular stage of the rash. The drug also exerted a significant effect on focal eruption and the scabbing stage reached early in those patient who were on 6 AzUR. No immediate toxic effects were seen.

*Suggestion.*—Further trials are warranted with this drug and they are in progress. One major disadvantage was that it had to be given by parenteral route. The availability of an oral drug will ease the administration of the therapy.

## SUMMARY.

On 140 unvaccinated smallpox patients of variable ages and type, 6 Azauridine, an antimetabolite was valued for its chemotherapeutic effects. A total dose of 100 to 200 mg./kg.-was given as an intravenous drip in two divided doses for 3 days. No effect on the mortality rate of haemorrhagic and flat was observed but 50 per cent reduction in the fatality rate was recorded in ordinary type of smallpox. The survival days in the two fatal types were encouraging. The flat type patients under drug therapy survived for over 4 days. The toxicity was also less marked in the drug group. The effect on temperature at pustular and scabbing stage was satisfactory. No toxic reaction were seen.

The authors' thanks are due to the Indian Council of Medical Research, New Delhi, under whose auspices the trials were taken up. Their guidance was of immence value. They are grateful to Government of Andhra Pradesh, Director Medical and Health Services and Principal, Osmania Medical College for permitting them to undertake trials in this hospital. The authors are greatly indebted to nursing staff, Medical Officers and House Surgeons whose assistance was of great value. Mr. Hashmi, who helped in maintenance of records, deserves their thanks.

## REFERENCES.

BUTTO, A.S., ISREAL, M.C.G., and WILKINSON, J.E. (1965) Hypocholesterolaemia and orotic aciduria during treatment with 6-Azauridine. *Brit. Med. Jour.*, **1**, 522-554.

HAMRE, D., BERNSTEIN, J.S., and DONOVICK, H. (1950) 'Activity of P-aminobenzaldehyde 3-thiosemicarbazone on vaccina virus in the chick embryo and in the mouse. *Proc. Soc. Exp. Pri. Med.*, **73**, 275.

RADA, B., BLASKOVIC, D., SORM, F., and SKODA, J. (1960) The inhibitory effect of 6-azauracil riboside on the multiplication of vaccinia virus. *Experientia* (Basel), **16**, 487.

RADA, B., and BLASKOVIC, D. (1966) ... Inhibition of vaccinia virus multiplication *in vitro* by 6-azauracil riboside. *Acta Virol.*, **10**, 1-9.

RAŠKOVA, H., JANKU, I., KRSIAK, M., NOVOTHY, J., VOLICER, L., and SEMETOMA, R., (1965) The reciprocal influence of clinical and animal observations. Evaluation of new drugs in man. *Czechoslovak Med. Press.*, 181-188.

ŠKODA, J., and SORM, F. (1959)... .. The accumulation of orotic acid, uracil and hypoxanthin by *Escherichia coli* in the presence of 6-azauracil and the biosynthesis of 6-azauridylic acid. *Czechosl. Chem. Commun.*, **24**, 1331-1337.