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# Effect of aureomycin on blood coagulation

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INTRODUCTION:

Since 1945, when Moldovsky and his coworkers first reported a lowering of the clotting and bleeding time in patients receiving penicillin, considerable interest has been shown in the effect of antibiotics and other drugs on coagulation of blood and particularly the implied possibility in this and later reports that the use of certain drugs have increased the occurrance of thrombotic phenomena.

Dr. Alton Ochsner in an address before the International Society of Surgery in New Orleans in 1949, stated, "The incidence of thrombo-embolism in the past two years may be explained in one of two ways. The increase may be more apparent than real, in that diagnosis is more frequent, and because of the increased interest in the condition. It is probably more real than apparent, for at the New Orleans Charity Hospital where the condition has been studied for many years, it is questionable that many cases were missed".

He continued, "An explanation of the increase in recent years may be a result of the routine use of antibiotics in treatment of most, if not all, hospital patients, as a shortening of coagulation time may occur favoring venous thrombosis".

In a letter to the editor of the J.A.M.A. in 1950

Long, after having reviewed the literature on the subject, commented, "The facts are not too clear in relation to experimental observations, at least so far as aureomycin is concerned, are contradictory". He points out that, since World War II, there has been an enormous increase in intravenous therapy and manipulation. We know that prolonged and complicated operative procedures are more common. We also know that reports of thrombo-embolism in medical patients are not more frequent. Long asks, "Is it not desirable that this be carefully studied by the surgeons of this country in order that unassailable data may be obtained on this point?

Surgeons summed up the evidence as follows: "Unfortunately, our knowledge of the effects produced by antibiotics on the coagulation process has progressed very little. Most of the reports in favor of an effect do not convince the critical reader. The matter warrants more thorough investigation".

REVIEW OF THE LITERATURE:

The effect of aureomycin on coagulation of blood is the subject of this dissertation. A review of the literature reveals contradictory reports on this topic.

Ross, Burk, and others in 1948, during some of the early clinical investigation on the toxicity or lack of toxicity of aureomycin, included in their studies several

laboratory tests referable to the hepatic, renal and hematopoetic systems, among which were bleeding time, erythrocyte fragility test, platlet count, and coagulation time. Five children from the Children's Hospital in Washington D. C. were chosen at random as subjects, and they were given aureomycin orally, 5 mgm. per kilo. every two hours for several days. The tests were run before giving the aureomycin, as a control, and then on the third and ninth day after aureomycin had been started. No significant changes were encountered in any of the laboratory tests in these children.

Harned, Cunningham, and others, in an experiment similar in purpose to the one mentioned above, used ten dogs to which they gave aureomycin, 100 mgm. per kilo. orally, for nine to fifteen weeks, in determining the coagulation time before giving the drug and then during the final week that aureomycin was given, no significant difference or even trend in the results were noted. The capillary tube method for measuring coagulation time was used.

Herrill, in a paper on the clinical uses of aureomycin, in referring to other reports in the literature concerning the effect of aureomycin on coagulation, states, "While it is true that slight alterations in coagulation of blood may be noted, this alteration is of no practical significance or importance when the ordinary therapuetically

effective amounts of the drug are employed.

Macht and Farkas used as subjects for their experiments, rabbits, cats, and patients. None of these had had previous medications. All subjects received aureomycin by mouth. The clotting time of whole blood was determined before aureomycin was given and at various intervals afterwards using the Lee White method. In one experiment four rabbits were used. They were tested at one, one and one half, and three hours after the drug was given. The coagulation time was shortened in all cases.

Using a cat, there were no changes in the coagulation time of blood taken repeatedly from the carotid artery before aureomycin was given, but there was progressive diminution in clotting time after administering 200 mgm of aureomycin.

In another experiment the coagulation time before and after one to two 250 mgm capsules were given to fourteen patients was determined. The coagulation times before and after the aureomycin was received, were taken from twenty to forty five minutes apart. In every case, some shortening of coagulation time was produced. Control experiments on human subjects who did not receive aureomycin did not reveal such changes.

The authors conclude from their findings -"There is undoubtedly a definite shortening of clotting time noted at the height of antibiotic therapy. While ordinarily nature provides a wide mechanism for prevention of thrombo-

embolic accidents, still the coagulatory apparatus may be considered as in a metastable state, so that sudden physiological disturbances might precipitate thrombo-embolic accidents. Hence suitable prophylactic measures by use of anticoagulant drugs may be instituted".

Waisbren and Glick measured the coagulation time of eleven patients just before and just after infusions of aureomycin. 0.5 gm of aureomycin was mixed with 500 cc of isotonic saline and given intravenously in 45 - 60 minutes. The three tube Lee White coagulation time was used and all tests were performed by the same technician. Blood was taken from one arm and tested just before the drug was administered, and blood for the second test was taken from the other arm immediately after the solution had been given. The results showed ten of the eleven patients had shortened coagulation time and one patient showed no change. The authors postulate that the cause for this phenomenon may be explained on the basis of an antiheparin effect of aureomycin. As increasing amounts of aureomycin were added to serum and plasma in vitro, the amount of heparin decreased. In the presence of 100 - 200 mg of aureomycin per cc of serum the concentration was approximately halved. Less than 20 mg of aureomycin per cc did not seem to have an effect on the concentration of heparin as measured by the method used. (The Jaques, Mankhouse, Stewart method of heparin analysis).

In an experiment by Lasser, Versakos, and Loewe, thirteen patients of the older age groups on the medical wards were selected as subjects. Five were given a single oral dose of 0.5 gm of aureomycin after which a Lee White coagulation time was run at two to four hours after taking the capsules. The patients were then given 0.5 gm more aureomycin and retested in six hours. Control tests were provided by doing coagulation times on the day preceding, and again just before the first aureomycin capsule was administered. The other eight patients were given 0.5 gm of aureomycin every six hours for three to ten days. The conclusions were that in both single dose and multiple dose groups, the coagulation times showed no significant changes, all values being within limits of variability of the pretreatment controls. There were no thrombo-embolic episodes observed in any of the patients during or following the investigation. The conclusion was that this experiment did not confirm the coagulative property of aureomycin.

Sharpse and Wright gave aureomycin alone to thirty patients, some orally and some intravenously. Lee White, three tube coagulation time were run before giving aureo-mycin and at six, twelve, twenty four, and forty eight hours after having been started on aureomycin. Each patient received 0.5 gm twice a day. Pretreatment clotting times varied greatly. In addition, aureomycin was given to over

one thousand patients at Harlem Hospital with only one case of thrombo-embolism. At the same time there were several who were on no antibiotic therapy who developed various thrombo-embolic manifestations. One patient with multiple myeloma received 3 gm of aureomycin a day for nine months with no untoward effects. The results of the above mentioned experiment showed slight changes in clotting times which alternated between slight increases to slight decreases. During the first twelve hours, the majority of the patients showed a slight increase in the coagulation time. After twenty four hours the coagulation times were about the same as before the antibiotic was given or slightly decreased. After forty eight hours there was again a slight increase. These coagulation times varied in individual cases, however, the variations were slight in magnitude and not considered sufficient clinically to contraindicate the therapeutic use of aureomycin.

Galt and Hunter administered aureomycin to five healthy male adults. An oral dose of 0.5 gm was given before each meal and at bedtime, a total of 2 gm per day, for at least seven days, and in some cases ten days. Clotting times by the three tube Lee White method using ordinary glass tubes revealed a prolongation above that of the control period of the average clotting time, during the period of the experiment, in four out of five patients. The clotting time was measured to the nearest thirty second

interval during the control and treatment period. Little significance is attributed to these findings by the authors. They point out the limitations of the Lee White test and the small number of subjects used in this experiment, but they believe that the subject should be further evaluated.

The limited evidence contributing to our knowledge of the effect of aureomycin on the coagulation of blood is obviously contradictory. Therefore, the following experiment was undertaken with the object of studying, first hand, the effect of aureomycin on coagulation, and perhaps to add to our information on this subject.

METHODS;

A. Selection of Subjects:

Twenty patients from the wards of University Hospital were used as subjects for this experiment. They were selected on the basis of their being as physiologically normal as possible. The criteria for 'physiological normality' were as follows:

(1) Pre-operative patients; that is, patients who were scheduled to undergo herriorrhaphies, hemorrhoidectomies, hysterectomies for fibroids, colporrhaphies, etc.

(2) Medical patients who were not diagnosed as having any metabolic diseases such as diabetis, blood dyscrasias, endocrine disorders of any kind or carcinoma.

(3) No patient was used who had less than 12 gm of hemoglobin.

(4) No patient was used who had been on any previous medications.

B. Procedure:

The twenty patients were divided into two groups, an experimental group, and a control group.

(1) Experimental group: (10 patients)

A Lee White coagulation time was run on each of the patients immediately before receiving aureomycin. Then 0.5 gm of aureomycin were mixed with 250 cc of isotonic saline and administered intravenously. A 21 gauge needle was used and the entire solution was run in over a 35 -45 minute period. Immediately after the intravenous needle was withdrawn, a sample of venous blood was taken from the opposite arm and another Lee White coagulation time was run. This procedure is after the technique of Waisbren and Glick. The aureomycin in the blood, when the second sample is withdrawn is ostensibly at its highest level at that time.

The aureomycin is given intravenously in order to by-pass the gastro-intestinal tract, thus obviating any possible effect that aureomycin taken by mouth would have on the bacterial

flora of the gut, the synthesis of vitamin K, and hence the prothrombin level in the blood.

(2) Control Group: (10 patients)

The procedure was the same as with the experimental group except that 250 cc of isotonic saline without aureomycin was used. The technique for running the Lee White coagulation time in this experiment was as follows:

Three cc of venous blood are withdrawn, and one cc is placed in each of three small tubes (Wasserman tubes) which are thoroughly clean and have been rinsed out with 0.85% saline solution. While one tube is being examined at thirty second intervals, the other two tubes are not disturbed and serve as checks on the end points observed in the other tubes. Errors in technique and agitation tend to hasten coagulation. Vena-puncture must be made initially and without trauma or else another vein is selected and a new puncture made. Any admixture of tissue juices would also hasten coagulation. Normally by this method, the coagulation time ranges from six to fifteen minutes.

RESULTS:

The results of this experiment are presented in table form as follows:

EXPERIMENTAL GROUP

| Patient | Sex | Diagnosis | Hemo-Globin | Coag. Time before Soln. | Coag. Time after Soln. | Diff. |
|---|---|---|---|---|---|---|
| N.C. | M | Hernia | 14.8g | 5.0 Min. | 6.5 Min. | + 1.5 |
| S.S. | M | Hemorrhoids | 14.0g | 8.0 Min. | 11.0 Min. | + 3.0 |
| M.H. | F | Adenoma Breast | 14.2g | 9.5 Min. | 11.5 Min. | + 2.0 |
| F.W. | M | Hernia | 13.6g | 11.5 Min. | 11.0 Min. | - 0.5 |
| I.B. | F | Prolapsed Uterus | 13.2g | 8.0 Min. | 9.5 Min. | + 1.5 |
| W.M. | F | Ovarian Cyst | 12.8g | 11.0 Min. | 12.0 Min. | + 1.0 |
| I.S. | F | Uterine Fibroid | 12.7g | 9.0 Min. | 8.5 Min. | - 0.5 |
| H.S. | F | Hemorrhoids | 13.2g | 3.5 Min. | 7.0 Min. | + 3.5 |
| R.V. | M | Hernia | 16.3g | 9.0 Min. | 7.5 Min. | - 1.5 |
| E.D. | F | Rectocele | 14.0g | 8.0 Min. | 10.5 Min. | + 1.5 |

CONTROL GROUP

| Patient | Sex | Diagnosis | Hemo-Globin | Coag. Time before Soln. | Coag. Time after Soln. | Diff. |
|---|---|---|---|---|---|---|
| W.A. | M | B.P.H. | 12.8g | 10.0 Min. | 8.0 Min. | - 2.0 |
| J.R. | M | B.P.H. | 12.5g | 7.0 Min. | 8.5 Min. | + 1.5 |
| P.S. | F | Cystocele | 12.8g | 11.0 Min. | 10.5 Min. | - 0.5 |
| C.M. | F | Uterine Fibroid | 12.4g | 6.5 Min. | 7.5 Min. | + 1.0 |
| M.F. | F | Cystocele | 13.8g | 8.0 Min. | 6.0 Min. | - 2.0 |
| M.S. | F | Cystocele | 13.6g | 13.5 Min. | 14.5 Min. | + 1.0 |
| H.S. | M | Hernia | 12.5g | 12.5 Min. | 9.5 Min. | - 3.0 |
| F.T. | F | Hemorrhoids | 12.8g | 6.5 Min. | 7.5 Min. | + 1.0 |
| L.T. | F | Prolapsed Uterus | 14.0g | 14.5 Min. | 14.0 Min. | - 0.5 |
| E.J. | M | Rectal Fissure | 14.8g | 15.5 Min. | 15.0 Min. | - 0.5 |

DISCUSSION:

In evaluating the differences between the coagulation times before and after receiving aureomycin and saline in the experimental group, as compared with the control group who received saline alone, it is evident that in both groups the differences are quite small. In the experimental group, seven of the ten subjects showed a slight prolongation of the coagulation time and three subjects showed a slight shortening. In the control group, four coagulation times were slightly prolonged and six shortened.

The amplitude of changes in clotting times of the two groups appear quite comparable. Although one subject in the experimental group, H. S., showed an increase in coagulation time of 3.5 minutes, the coagulation before aureomycin was administered was 3.5 minutes. This was probably a result of error in technique, for this coagulation time is the smallest value in all forty tests performed and is less than the lower limits of normal for the Lee White test.

If one is allowed to assume that the intravenous administration of 250 cc of isotonic saline alone has no effect on coagulation, it then seems possible that the small variations in coagulation time in both groups are simply the result of experimental error and/ or the normal variation of results inherent in the Lee White test.

Although there was a slight tendency toward prolongation of coagulation in the group which received aureomycin,

it must be considered that the tendency is minimal and that the two groups of subjects are small. Therefore, it would not be feasible to submit the data to statistical analysis in order to derive the significance of the differences between the two groups.

This experiment was designed to measure the immediate effect of aureomycin on the coagulation of blood. Perhaps different results would have been obtained had the coagulation times been taken on patients who had received aureomycin over longer periods of time. However, the findings in this experiment do not support those of Waisbren and Glick who did their work under similar conditions and observed shortening of coagulation times in ten of eleven subjects. Furthermore this experiment does not lend support to the thesis that the use of aureomycin may contribute to the incidence of thrombo-embolic phenomena.

It is also well to remember that the validity of the conclusions drawn from the results of an experiment is greatly dependent upon the accuracy and sensitivity of the methods used to measure the necessary variables in the experiment. The Lee White test measures only gross alterations in the coagulative mechanism, unfortunately. Perhaps when a more sensitive method for measuring the coagulation time of blood is devised, different results will be obtainable.

SUMMARY AND CONCLUSION:

In order to test the immediate effect of intravenous aureomycin on the coagulation of blood, 0.5 gm of aureomycin in 250 cc of isotonic saline was given intravenously to ten patients. 250 cc of isotonic saline alone was given to a similar group of ten patients. Lee White coagulation times were determined immediately before and immediately after receiving the solutions and the differences between the coagulation times for each patient were determined. A comparison of the differences in each group showed no appreciable change in coagulation time, either as a result of receiving aureomycin in saline or saline alone. The only difference between the two groups was a questionable slight tendency toward prolongation of the coagulation times in the group which received aureomycin. This tendency could not be interpreted as a significant difference.

It is concluded, therefore, that under the conditions of this experiment, aureomycin has little or no effect on the coagulation of blood.

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