In six experiments carried out on dogs treated with heparin or with procaine block the prothrombin time was measured 10 days after the administration of heparin and after 10 lumbar blocks carried out over 10 days. In all these experiments the prothrombin time was not prolonged.

Conclusions

Many questions connected with the problems tackled remain unsolved; nevertheless, while further investigations are necessary it seems that the following conclusions are justified:

1. The intravenous injection of a 50% solution of sodium salicylate results in thrombosis appearing 6–12 hours later. The first evidence of organization is seen after seven days.

2. The administration of heparin, in amounts of 200 mg. daily, 6–24 hours after the production of thrombosis prevents the development of the thrombotic process.

3. Heparin treatment given three or more days after the production of thrombosis does not prevent its development. Microscopically, symptoms of the fibrinolytic action of heparin are absent.

4. Veins of dogs treated with heparin present in most cases papillomatous proliferations of the endothelium, whereas in dogs treated with procaine block the picture of hyaline degeneration of the intima is prevalent.

5. The application of procaine block less than six hours from the production of the thrombosis prevents the full development of the thrombus, provided the treatment lasts for at least four days.

6. Procaine block applied 24 or more hours after the time of the injection of sodium salicylate solution does not disperse the existing thrombus; nevertheless it accelerates organization of the thrombosis as compared with non-treated animals.

7. The degree of organization of the thrombosis in dogs treated with heparin or with procaine block is approximately the same. The proliferation of endothelial cells is more marked in thrombosis after heparin treatment.

8. The prothrombin time after 10 days of heparin or procaine block shows no significant changes from the normal.

9. The histopathological examinations are not sufficient for the evaluation of dynamic changes of blood supply of the extremity and should be supplemented by the analysis of the phlebographic appearances.

BIBLIOGRAPHY

- Butkiewicz, T. (1947). *Pol. Tyg. lek.*, 2, 1383, 1444, 1489.
 Cummine, H., and Lyons, R. N. (1948). *Brit. J. Surg.*, 35, 337.
 Dryski, J. (1954). *Pol. Tyg. lek.*, 9, 1318.
 Eysholdt, K. G. (1954). *Arch. klin. Chir.*, 277, 455.
 Gottlob, R., and May, R. (1954). *Angiology*, 5, 307.
 Gruca, A. (1928). *Pol. Przegl. chir.*, 1, 182.
 Homans, J., and Zollinger, R. (1929). *Arch. Surg. (Chicago)*, 18, 992.
 Jewell, P., Pilkington, T., and Robinson, B. (1954). *British Medical Journal*, 1, 1013.
 Jorpes, E. (1947). *Pol. Tyg. lek.*, 2, 71.
 Leriche, R., and Kunlin, J. (1934). *Presse méd.*, 42, 1481.
 Nielubowicz, J. (1948). *Pol. Tyg. lek.*, 3, 769, 855.
 Nowicki, St. (1947). *Ibid.*, 2, 936.
 Ochsner, A., and de Bakey, M. (1941). *J. Amer. med. Ass.*, 117, 1915.
 Patey, D. H. (1937). *Surg. Gynec. Obstet.*, 64, 1002.
Proceedings of the First International Conference on Thrombosis and Embolism, edited by Th. Koller and W. R. Merz. Basel, 1954.
 Raeburn, C. (1951). *British Medical Journal*, 2, 517.
 Rappert, E. (1952a). *Wien. klin. Wschr.*, 64, 283.
 — (1952b). *Zbl. Chir.*, 77, 1729.
 Szymanowicz, J., and Woliczko, K. (1935). *Pol. Gaz. lek.*, 14, 854.
 Weltl, J. J., and Breynaert, R. (1951). *Presse méd.*, 59, 518.

A SALICYLSULPHONIC ACID TABLET TEST FOR PROTEIN IN URINE

BY

D. N. BARON, M.D.

Reader in Chemical Pathology, Royal Free Hospital,
London

AND

CELIA OAKLEY, M.B., M.R.C.P.

Formerly Resident Pathologist, Royal Free Hospital,
London

This communication describes the trial of a commercially available tablet test for protein in urine (albuminuria), which has been designed to take the place of the standard salicylsulphonic acid test. It must be emphasized, because of the common use of the term "albuminuria," that in all circumstances protein present in urine is a mixture of albumin and globulins, with albumin predominating; and this tablet test, the standard salicylsulphonic acid test, and the boiling test all detect every type of protein in urine, not albumin alone. We shall therefore only employ the term "proteinuria." Each tablet ("altest") contains about 25 mg. of salicylsulphonic acid, effervescing agents, and a trace of bromocresol green. A similar tablet called "bumintest" is marketed in the U.S.A. The latter test cannot be recommended for convenience because it entails dissolving the bumintest salicylsulphonic acid tablet in water to make a solution with which to test the urine, and we understand that bumintest is not being introduced into this country.

Method

A series of solutions of protein in urine were prepared using (1) normal human serum, (2) standard albumin solution, (3) urine from a nephrotic patient, at total protein concentrations of approximately 0.01, 0.005, 0.0025, and 0.001 g. per 100 ml. of urine. Each protein concentration was prepared in urines of approximately specific gravities 1002, 1010, and 1020, and separate samples of each were adjusted to approximate pH 5, 6, 7, and 8.

Urines containing Bence Jones protein, a high concentration of urates, and the radio-opaque substances iopanoic acid, diodone, sodium acetoazote, and "biligradin" and "urogratin" were also examined.

Each urine was tested by three methods: (1) standard "boiling test" (Harrison, 1947). (2) Standard salicylsulphonic solution test (about 5 ml. of urine plus about 0.5 ml. of salicylsulphonic acid (=125 mg.) (Harrison, 1947)—called "solution test" below. (3) altest; the method recommended by the manufacturers was found generally satisfactory—called "tablet test" below.

Urine which is cloudy should be filtered before carrying out the tablet test, as with other tests for proteinuria. An "inch" (about 4 ml.) of urine is placed in a test-tube, and one tablet added. When the tablet has dissolved completely (which takes about one minute) the tube is gently shaken, the froth is allowed to clear, and the mixture is examined. (We find, however, that the tablet dissolves more easily if the tube is continuously shaken.) The fluid should be yellow; if it is blue or green this indicates that the mixture is still alkaline, and one or more tablets are added until the final solution is acid and the fluid becomes yellow. The result is read in the same way as other tests for proteinuria: no protein—clear (yellow) solution; protein present—cloudy mixture or white precipitate, depending on the concentration of protein.

Results

The tablet test could regularly detect a protein concentration of 0.005 g. per 100 ml. of urine : at 0.0025 g. per 100 ml. detection of proteinuria was inconstant. It was equally sensitive to the different proteins tested, and throughout the pH and specific gravity ranges which were examined. The tablets appeared to be stable, and tablets which had been stored at room temperature for three months were as effective as at the time the bottle of tablets was first opened. The solution test was also sensitive to 0.005 g. per 100 ml. and usually to 0.0025 g. per 100 ml., and the boiling test was regularly sensitive to 0.0025 g. per 100 ml. while inconstant results were obtained at 0.001 g. per 100 ml.; both these tests were also unaffected by the nature of the protein and the pH or specific gravity of the urine.

The ingenious internal indicator of the tablet test enables successful correction of the pH of alkaline urines. The urines of pH 7 sometimes needed two tablets, and the urines of pH 8 needed two or three tablets. (The solution test adds the equivalent of four tablets when properly performed, and the problem of excessive alkalinity of the urine therefore very rarely arises.) However, minimal turbidity was more difficult to detect in the resulting deeply coloured yellow solution.

Both the tablet test and the solution test gave a positive reaction with Bence Jones protein and a negative reaction with urates in solution.

The radio-opaque substances do not precipitate on boiling. They give a pseudo-positive reaction (deposit of crystals) with the solution test, and a negative reaction with the tablet test (one or two tablets). This is a quantitative and not a qualitative difference, because when four tablets were added to 4 ml. of urine containing these substances precipitation immediately developed.

Fifty consecutive urines, received in the department for routine examination, were tested for protein in parallel by the solution test and tablet test. (Fifteen urines required two tablets, and two urines required three tablets.) Twelve of these contained protein in varying concentration, and identical positive results were given by the solution test and the tablet test.

Discussion

Two previous reports (Bryan, 1956; Whittet, 1956) have briefly described the altest without critically examining its properties.

Possible users of the tablet test are general practitioners who have to test occasional specimens of urine in their patient's home or in their consulting-room, or nurses, students, or laboratory technicians who have to test larger numbers of urine specimens on frequent occasions.

The tablet test can be recommended for general practitioners. It is sufficiently sensitive for all clinical purposes. It is simpler and much more convenient than the boiling test, though it is less sensitive than this test. The great convenience to the practitioner of having to carry a bottle of small tablets rather than a bottle of acid liquid reagent outweighs, in our view, the slight disadvantages of the tablet test when compared with the solution test.

However, for frequent use in a laboratory or side room we consider that the tablet test does not replace the salicylsulphonic acid solution test. Although the tablets are inexpensive, the salicylsulphonic acid solution (which is also stable) can be prepared in bulk by a hospital at less than half the price of the tablets. The tablet test is slightly slower, because of the time necessary to dissolve the tablets and disperse the froth which otherwise interferes with the reading. The tablet test does not seem to be quite as sensitive, probably because the colour of the indicator makes it less easy to read; this could probably largely be remedied by reducing the quantity of bromocresol green. When the solution test is done, using the proper technique, enough salicylsulphonic acid is added to counteract all but the grossest alkalinity.

We must deplore the manufacturer's name for the tablet, altest, because it perpetuates the "albuminuria" fallacy.

Summary

A new manufactured tablet (altest) has been compared with the boiling test and the salicylsulphonic acid solution test. It can detect a concentration of 0.005 g. of protein per 100 ml. of urine throughout the physiological range of urine pH and specific gravity, and the results of the tablet test are generally comparable to those of the salicylsulphonic acid solution test, but are slightly less sensitive than the results of the boiling test.

The tablets contain an indicator which shows when additional tablets need be added to ensure acidity of the solution. The advantages and disadvantages of the new test are discussed.

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REFERENCES

- Bryan, G. (1956). *Brit. overseas Pharm.*, **109**, 120.
Harrison, G. A. (1947). *Chemical Methods in Clinical Medicine*, 3rd ed. Churchill, London.
Whittet, T. D. (1956). *Pharm. J.*, **177**, 19.

Medical Memorandum

Marfan's Syndrome

A small number of patients with Marfan's syndrome (Marfan, 1896) have come to necropsy (Reynolds, 1950; Moses, 1951; Marvel and Genovese, 1951; Whittaker and Sheehan, 1954; McKusick, 1955). A further case, in which Marfan's syndrome was associated with a patent ductus arteriosus and aortic aneurysm from cystic medionecrosis of the aortic wall, is here reported.

CASE REPORT

A married woman aged 32 was admitted to hospital on December 22, 1954. At 18 months of age she had nephritis and an enlarged heart. When 5 years old she was said to have valvular heart disease. She was dyspnoeic as a child, and later had winter bronchitis. From the age of 18 she had oedema of the ankles. Since 1950 she had had attacks of gripping pain in the left chest which radiated into the left arm and occurred both at rest and on exercise. In 1952 she collapsed with a sudden attack of pain in the back and fullness in the chest. After recovery she was breathless on slight exertion and had persistent ankle oedema and attacks of palpitation. For the last six months she had nocturnal dyspnoea accompanied by sweating and palpitations and a gripping retrosternal pain radiating into the left arm. Two weeks before admission a pleuritic pain developed in the middle and lower thoracic regions of the back with dyspnoea, cough, and increased ankle swelling. These improved slightly with digitalis and rest, but her condition again deteriorated.

Examination showed her to be thin, orthopnoeic, and cyanosed. She had arachnodactyly; height 68½ in. (1.74 m.); palate highly arched. No changes in eyes, and no other skeletal deformities. She was in congestive heart failure; pulse regular, rapid, and collapsing; blood pressure 140/40 mm. Hg; femoral pulses palpable. Bed shook with each heart-beat; visible arterial pulsation in neck. Cardiac impulse moved anterior chest wall. Apex beat in 9th left intercostal space in posterior axillary line. Systolic pulsation felt over most of praecordium. In left axilla between 5th and 9th intercostal spaces a loud rumbling diastolic murmur and a blowing pansystolic murmur heard. Below left clavicle, in pulmonary area, and down left sternal