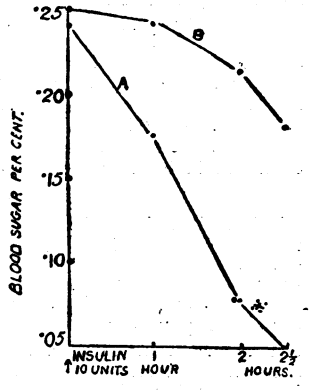
a machine boat against high compression, and in exercise on a sliding seat. Each form of exercise was indulged in vigorously in turn until the groups of muscles used would respond no more. It is a tribute to insulin that this violent exercise could be carried out without undue fatigue, for three years ago the mere ascent of stairs made the patient's legs ache with exhaustion.

Time. Minutes.	A. Insulin + Exercise.	B. Insulin, no Exercise.
2 p.m.	240 Insulin 10 units	254 Insulin 10 units
20	245	—
30	—	242
45	219	—
60	175	239
90	116	230
120	73*	211
150 4.30 p.m.	51*.	181



The effect of 10 units of insulin on diabetic blood sugar. A, with exercise; B, no exercise.

* Symptoms of hypoglycaemia.

The figures give striking proof of the effect of exercise in increasing insulin action. The difference on the two days is far greater than I had anticipated, and it is of interest to record what happened after these experiments. After B the usual 8 units were taken before the evening meal (20 grams of carbohydrate), and nothing unusual occurred either that evening or the next day. Immediately after A, at 4.30 p.m. 20 grams of glucose were given to relieve the symptoms of hypoglycaemia, and no further exercise was taken. In spite of that, symptoms recurred at 6.15 p.m., and a further 10 grams of glucose were taken. No insulin was administered before the usual evening meal, and the usual insulin and breakfast were taken next morning. Hypoglycaemia occurred at 11.30 a.m. in spite of the extra 30 grams of glucose and the omission of one dose of insulin. This can only be explained by the effect of exercise in burning up and depleting the usual carbohydrate stores of the body and upsetting the usual balance of diet and insulin next day.

It may be of interest and some practical importance to note the long time in experiment B before insulin had any appreciable effect on the blood sugar. In some former experiments[4] I found that even in large doses insulin never reduced the fasting blood sugar level more than 40 mg. per cent. in the first one and a half hours, but Murray Lyon[5] found that it usually dropped about 100 mg. in the first two hours. It is therefore perfectly safe, and may be deemed advantageous, to give a severe diabetic with a raised fasting blood sugar (0.016 per cent.) his insulin one or even one and a half hours before a meal with a view to lowering his blood sugar before the carbohydrate of his meal raises it.

It is not proposed to discuss here the questions that arise regarding the relative effect of insulin in storing or burning sugar in the body—questions on which other exercise experiments to be reported later have thrown some light. In the meantime it seems clear that muscular activity greatly enhances the action of insulin.

Conclusions and Practical Application.

I. The immediate effect of exercise in increasing the fall of blood sugar caused by insulin is very great. This occurs only during the maximum period of insulin activity—that is, from one to four hours after injection. Severe and prolonged exercise and half the usual dose of insulin may produce as great a fall of blood sugar as the usual dose without exercise.

II. Besides the immediate fall of blood sugar, exercise causes insulin to burn more carbohydrate than usual and to deplete the body stores of carbohydrate, as the above clinical cases and the aftermath of experiment A show. For this reason the dose of insulin succeeding exercise has often an unusually powerful effect in producing hypoglycaemia.

III. If the diet is kept constant, an increase of exercise over days and weeks allows a reduction of insulin to be made. This is one of the reasons why the dose of insulin can often be reduced early in treatment when patients leave hospital and adopt a more active existence. If this reduction is not made symptoms of hypoglycaemia appear. When the added exercise is omitted the insulin must be raised again.

IV. It should be the object of all treatment to enable a diabetic to lead a normal and varied life, and to forget all about his disease except at meal times. Accordingly allowances must be made for the changing conditions that may arise in his life. Patients should and do easily learn to reduce their insulin before unaccustomed exercise or activity, or they must take vigorous exercise only at times when the effect of their injections has worn over. Even after exercise, when the usual carbohydrate stores have been partially depleted, it is usually advisable to reduce the next dose of insulin. If exercise is increased over days or weeks it is obviously more physiological to increase the diet instead of, or as well as, reducing the insulin. The increased calorific output must ultimately be balanced by an increased intake, otherwise weight and energy will be lost. A diet which supplies 30 calories per kilogram of body weight may be a sufficient "maintenance" diet for a sedentary life, but 40 calories may not be too much merely to maintain the requirements of a more active existence.

References.

[1] Allen, F. M.: *Total Dietary Regulation in the Treatment of Diabetes*, 1919, p. 408. [2] Lawrence, R. D.: *The Diabetic Life*, 1925, p. 87. [3] Hetzel, K. S.: British Medical Journal, January 17th, 1925, p. 102. [4] Lawrence, R. D.: Ibid., March 22nd, 1924, p. 516. [5] Murray Lyon: *Lancet*, July 26th, 1924.

THE KASTLE-MEYER TEST FOR THE DETECTION OF BLOOD

CONSIDERED FROM THE MEDICO-LEGAL ASPECT.

BY

JOHN GLAISTER, JUN., M.B., CH.B.,

ASSISTANT TO THE PROFESSOR OF FORENSIC MEDICINE, UNIVERSITY OF GLASGOW.

Some time ago my attention was directed to the Kastle-Meyer test for the detection of occult blood present in faeces. Its technique and rationale are described by R. Goiffon in the *Manuel de Coprologie Clinique*. He there states "that the test is an extremely delicate one, with the result that interpretable traces of blood are rendered definitely possible clinically, and, further, that by its use the causes of important errors yielded by certain other tests for blood are practically eliminated." After careful perusal of the article it was thought by the writer that the test might prove of considerable value in the laboratory examination of blood stains from the medico-legal viewpoint. The results obtained from experimental work with the tests are regarded as interesting and important to the medical jurist.

Reagents.—Two chemical solutions are required for the test, one being hydrogen peroxide of strength twenty volumes; the other is composed of a mixture of 2 grams of phenolphthalein, 20 grams of potassium hydrate, and distilled water in sufficient quantity to produce 100 c.cm. of the reagent. These three ingredients are boiled, and during the process 10 to 30 grams of powdered zinc are added. Boiling is continued until the solution becomes colourless, the colour disappearing usually after about ten minutes. The reagent when prepared in this manner will remain effective for long periods if a small quantity of the powdered zinc is left deposited at the foot of the reagent bottle to ensure reduction. The technique of the test consists in adding about 10 to 20 drops of the Kastle-Meyer reagent to the suspected blood-stained surface, thereafter adding a few drops of fresh hydrogen peroxide. If blood be present a deep permanganate colour develops almost immediately.

Rationale.—According to Goiffon the rationale of the test lies in the facts that the phenolphthalein in the reagent is

reduced into phthalein by the zinc and is regenerated by the oxygen liberated from the hydrogen peroxide by the haemoglobin present. The solution being alkaline in reaction, the indicator readily permits of the development of the reddish colour.

Method of Applying the Test.—I have found by experiment that the test can be carried out successfully either with a solution of the blood extracted from the stain, when placed in a test tube, or with the stain placed in a watch-glass and a few drops of distilled water or 0.83 per cent. saline applied to moisten it. In using the watch-glass method the stain need not be left in contact with the distilled water or saline for more than a few seconds, the reaction to the test being given almost immediately after the small quantity of diluent has been added. This is of importance in medico-legal work, as the usual tests—Day's or Schönbein's test and others—require soakage of the blood-stained material for some hours before the testing chemicals are added if reliable results are to be obtained. In adopting the watch-glass method it was found preferable to add a few drops of the reagent and only two drops of hydrogen peroxide, as when more of the peroxide was added the reaction seemed to be delayed, and was not infrequently masked by the development of a copious dense foam; further, excess of peroxide produced a milky hue in the fluid, and when the colour reaction did appear it was retained only for a short period, whereas if but two drops were added the colour reaction developed quickly and remained for several hours or longer. It appears from experiment that distilled water is a more satisfactory diluent than normal saline for the medico-legal application of the test, as a quicker and more intense colour reaction is produced.

Delicacy of the Test.—A positive reaction with the Kastle-Meyer test was obtained with human blood a year old extracted with saline from filter paper on which it had been dried, even when in a dilution of 1 in 212,000. A similar blood-stain diluted to 1 in 800,000 with distilled water yielded an immediate and positive reaction. It should be noted that although a positive result was obtained with these strengths of dilution and would be given in much higher dilutions, it was not considered necessary to conduct further tests, as the delicacy of the reaction was thought to have been fully established from a medico-legal aspect. As the result of noting carefully the colour reactions when using dilutions of blood from 1 in 10 upwards, it was found that the extent of the colour obtained on adding the reagents is quantitative for the blood present only within wide limits.

Experiments with the Test.

In an endeavour to exclude a fallacious positive reaction with certain substances in the absence of haemoglobin, ninety materials were tested at random with the Kastle-Meyer reagent, but in no instance was a true positive reaction exhibited. In most cases where chemical fluids were used the solution was about 20 per cent. in strength, which was regarded as being stronger than a dilution likely to be obtained from a stain composed of such chemicals.

List of the Substances Tested.

Rust, urine, perspiration, seminal fluid, leucorrhoeal discharge, albumin, saliva, human milk, cow's milk, soap solution, red ink, healthy faeces, colostrum, bile salt (sodium taurocholate), aniline dyes (namely, chlorazol fast red, aqueous fuchsin, neutral red, methyl red), pitch, rubber solution, wheat, starch, arrowroot, tapioca, barley, rice, rye, oats, sago, bean, maize, potato, manganese chloride, barium chloride, mercurous chloride, mercuric chloride, phenylhydrazine hydrochloride, aluminium chloride, lithium chloride, ferric chloride, ammonium chloride, ammonium phosphate, calcium chloride, calcium carbonate, magnesium chloride, magnesium sulphate, magnesium carbonate, magnesium oxide, ferrous sulphate, cobalt sulphate, calcium sulphate, magnesium sulphate, aniline oil, sodium bisulphate, sodium acetate, sodium nitrite, sodium nitrate, sodium citrate, ammonium sulphocyanide, ammonium iron alum, silver nitrate, lead nitrate, sulphanilic acid, gallic acid, citric acid, pyrogallic acid, carbolic acid, chromic acid, tannic acid, naphthylamine acetate, azobenzol, ortol (photographic developer), copper acetate, copper subacetate, copper sulphate, potassium bromide, potassium bromate, potassium cyanide, potassium ferrocyanide, potassium ferricyanide, potassium nitrate, potassium chromate, potassium iodide, potassium chloride, logwood, bromine, annatto hyspanica (colouring material for milk), calcium fluoride, calcium hydrate, and iodine.

In the case of iodine it was found that on adding the reagents necessary for the test a dark red coloration with an orange tinge was produced. This colour, however, did not resemble the permanganate colour which the test produces when blood is present. A positive reaction when once seen is not readily confused with other reddish colorations. If a few drops of a solution of one part in three of sulphuric acid be first added to the iodine solution a brownish colour is produced on the addition of the Kastle-Meyer reagents in the absence of blood.

Tests were made with cochineal, using the Kastle-Meyer reagents, because it was thought that the initial colour of this substance might readily mask a positive reaction when blood was present. This surmise was substantiated. It was found, however, that if a solution of blood be added to one of cochineal the latter loses its red colour and becomes orange on the addition of the testing reagents, whereas if they are added to a solution of cochineal which does not contain blood its initial red colour is intensified. On the addition of testing reagents to cochineal containing blood to which a few drops of a solution of one part in three of sulphuric acid had been added, a pale yellowish-green cloudy appearance was exhibited, whereas when blood was absent a milky orange colour was produced. If the testing reagents be added to a solution of cochineal containing blood to which a few drops of one part in three alkaline solution have been added, a very faint clear straw colour is produced, whereas if blood be absent a milky orange colour is noted.

Certain substances contained in the foregoing list—for example, potassium ferrocyanide, sodium nitrite, sodium nitrate, and potassium iodide—occasionally yielded a very faint pink colour on the addition of the testing reagents when blood was absent. The coloration alluded to was so faint that it should never be confused with a positive reaction by anyone who had used the Kastle-Meyer test previously. Goiffon has stated that it is not necessary to note the late colour reactions, or the very faint colorations, on account of the delicacy of the test, with which statement I am in complete agreement. If blood be present the extent of the coloration is well marked. In order to make assurance doubly sure in these cases, if three drops of one part in three of sulphuric acid be added to the substance under test before adding the testing reagents, a pink or reddish colour will not appear in the absence of blood. It is evident that the likelihood of encountering any of the last mentioned chemicals in a blood stain is very remote.

Tests conducted with lochia obtained at the sixth day of the puerperium and blood-stained pus yielded positive results. The age of the blood stain under examination does not apparently impede the reaction, as markedly positive results were obtained with the test on applying it to a blood stain fifteen years old, as well as to portions of bones some thirty years old. It would appear that heat insufficient in intensity to char or scorch when applied to a blood stain does not prevent a positive reaction being obtained, since it was found that blood stains upon cloth kept for a year and then submitted to a temperature of 160° C. for one and a half hours, and to higher temperatures for varying periods, readily produced a positive reaction when a few drops of distilled water were added and the reagents applied. It should be noted, however, that if the heat to which the stain is submitted is sufficiently great to produce charring a positive reaction will not be exhibited. Scorching vitiates the test to the extent of producing only a faintly positive reaction, not sufficiently marked to warrant it being termed positive from a medico-legal aspect. Blood-stained cloth kept for a year, then boiled for an hour in 200 c.cm. of distilled water, produced a positive result both with the stain and the water in which it had been boiled. Similar stains boiled with 1 gram of Castile soap powder yielded a positive reaction when the soapy solution was tested, although the stained cloth gave a negative result.

The amount of blood present in the solution apparently matters little so far as the production of a positive reaction is concerned, because the smallest speck of blood upon filter paper revealed a strong reaction. If the blood solution under test be heated before the reagents are added the colour reaction is expedited, being almost instantaneous, but the solution quickly becomes milky, more especially if

the stained cloth is present. If the solution be heated after the reagents have been added, a clear ruby-red colour is quickly given. It is a wise precaution to remove the stained cloth before adding the reagents if the material be dyed a deep reddish hue, lest the dye coming into solution impart a pinkish colour, simulating a positive reaction when blood is absent. Stains composed of pig, ox, and sheep blood were also tested, but no difference in reaction from that of human blood of the same age was revealed.

Conclusions.

From my experiments with the test applied medico-legally, it would appear that it is very delicate and reliable in the detection of haemoglobin. It is apparently a worthy substitute for Day's test, which possesses may fallacies, requires fresh reagents, frequently fails to act when only small quantities of blood are present, and in certain cases gives only a faintly positive reaction, creating doubt in the mind of the examiner whether such should be regarded as positive or negative. With the Kastle-Meyer test there are no "border-line" results. The reaction is well marked, is practically immediate, and time is economized in not having to await soakage of the stains before applying the test, if the watch-glass method is utilized. This becomes an important consideration when there are many productions to be examined. The age of the stain does not apparently affect the delicacy of the test. It must not be forgotten, however, that, having regard to the extreme delicacy of the test, the greatest care must be taken to ensure absolute cleanliness of all vessels used in the medico-legal examination of the stains, in order to exclude rigidly extraneous contamination of the vessels as the result of a previous examination. The apparatus should be boiled, the test tubes and watch-glasses carefully scrubbed with a suitable brush and thereafter wiped with gauze.

The results of the foregoing experiments indicate that the test is one of superior value for the detection medico-legally of haemoglobin. The test should be used in conjunction with the microscopic examination of stains for the detection of the presence of mammalian blood corpuscles, and the precipitin or serological test in determining the source of the blood, provided the examiner has by experience of the latter test acquired sufficient knowledge to determine in an accurate manner the results obtained.

LITERATURE.

J. H. Kastle and Norman Roberts: *Tests for Pus and Blood.* From the Division of Chemistry, Hygienic Laboratory, U.S. Public Health and Marine Hospital Service, Washington, D.C. (See *Journal of Biological Chemistry*, vol. 6, 1909.)

Bulletin No. 51: Variations in the Peroxidase Activity of the Blood in Health and Disease, by J. H. Kastle and H. L. Amoss. Hygienic Laboratory, U.S. Public Health and Marine Hospital Service, Washington, D.C.

R. Goiffon: *Manuel de Coprologie Clinique*, p. 78 et seq., Paris, 1921. (Translation by writer.)

Dorland's *Medical Dictionary*, 1925, "Meyer's reagent."

MALDEVELOPMENT OF OESOPHAGUS.

BY

RAYMUND THERON, M.D.LOND., L.R.C.P., M.R.C.S.,

HONORARY ASSISTANT MEDICAL OFFICER, MATERNITY HOME, BLOEMFONTEIN.

A MARRIED woman was under my care recently during her second pregnancy. The first child is a healthy boy, aged 3, and is said to have weighed 9¾ lb. at birth. She was not a robust-looking woman, and suffered from a series of bad colds with a little bronchitis throughout the eighth month of pregnancy. Towards the end of December she developed hydramnios, so that it became almost impossible to make out the lie of the child. She complained a great deal of thirst. The urine contained no albumin or sugar.

At term labour started with the sudden rupture of membranes and escape of a large quantity of liquor amnii. On my arrival I found the cord in the vagina; the vertex presented and the cervix admitted only two fingers; labour pains were absent. Under anaesthesia I replaced the cord, which was pulsating rather slowly, put on a tight binder, and raised the foot of the bed. Pains came on a few hours later, and about six hours after rupture of the membranes a male child, weighing only 5¾ lb., was born. There was a great deal of mucus in its mouth and it was rather blue, but soon recovered. Delivery of a normal placenta caused no trouble.

The next day I was told that the baby had vomited a lot of mucus and most of the water it had been given to drink. Although it was rather a lean and undersized infant it cried vigorously, and at first there seemed to be nothing much wrong with it. The reports as to the nature of the vomiting were contradictory, but by the third day it was apparent that the child was not keeping much down. As yet it had passed nothing but meconium. I noticed that, after a drink of water, it seemed to choke and soon regurgitated, rather than vomited, all of the water, mixed with mucus. The abdomen was sunken a little, but was otherwise normal. No peristalsis was noticed and no tumour could be felt. A catheter failed to pass more than about 4½ inches from the mouth down the oesophagus. A diverticulum or an impervious oesophagus was therefore diagnosed, and it was realized that no operation was possible that would benefit such a puny infant.

The next day it was reported that some flatus had been passed, besides a little more meconium. *X*-ray examination showed that an opaque bougie passed down the oesophagus only as far as the level of the fourth dorsal vertebra, and that a barium feed filled up a little pyriform sac at the same level and did not get into the stomach. In the stomach there appeared to be a bubble of air.

It was therefore concluded that we had that type of malformation of the oesophagus which is clearly described by Professor Arthur Keith in his *Human Embryology and Morphology* (p. 250). He states that it is not uncommon. The paratracheal or upper part of the oesophagus, which is developed together with the trachea from the hinder part of the primitive pharynx, ends blindly, while the retrotracheal or lower part, which arises from the primitive alimentary canal, opens from the trachea and is covered by non-striated muscle. This, of course, explained the presence of air in the stomach and the passage of flatus.

The child lived six and a half days, gradually becoming weaker, passing fair quantities of urine, and crying only at long intervals for a feed. It suckled well and could swallow about two or three drachms easily, and sometimes kept it down nearly an hour. Invariably, however, the feed, mixed with mucus, was regurgitated, and occasionally nearly choked the child.

Post-mortem examination exactly confirmed the diagnosis. The oesophagus ended abruptly with a slight dilatation just above the level of the bifurcation of the trachea, having no connexion with the latter at that point. At the same level, in the middle line posteriorly, a narrow tube with a lumen of about 1 mm. opened from the trachea and gradually widened until it joined the cardiac end of the stomach. The stomach and duodenum were normal. The first six inches of the jejunum was collapsed, while the rest of the intestines were somewhat distended with air and contained a little meconium. The bases of both lungs were oedematous. No further abnormalities were discovered. The emaciation was great.

This malformation may, of course, be "not uncommon" as congenital abnormalities go, but luckily it is rarely met with in practice. It is interesting from several points of view.

First, the baby continued to pass a good deal of urine, although the urine was rather highly coloured. Probably, therefore, the pharynx and oesophagus were able to absorb some fluid, as no rectal salines were given.

Secondly, it is proved that a newborn infant, and a puny one at that, can exist for six and a half days on absolute starvation.

Thirdly, this case may have some bearing on the theories as to the purpose and fate of the liquor amnii. That the liquor amnii consists largely of foetal urine is, of course, no longer believed. It is generally supposed that the liquor is secreted by the amnion. That the foetus does swallow some of it is apparently admitted by all authorities, since lanugo hairs can often be demonstrated in the meconium. In this case there was a large excess of liquor amnii, well over a gallon being discharged, and the question arises whether the excess of liquor might not have been a result of the foetus being unable to swallow and absorb the liquor. It seems quite reasonable to believe that normally the foetus swallows liquor amnii, deriving fluid from it wherewith to dilute the waste products of its metabolism and excrete these by way of the placenta.

Although the mother's general condition was not very good during the last few months of pregnancy she remained fairly well nourished. Her condition, therefore, could have had little to do with the undersized child she produced. The placenta macroscopically presented no abnormalities. Again, excessive intrauterine pressure on the placenta came into play only during the last few weeks of pregnancy, so that that could not have had much to do with the small size and insufficient nourishment of the infant.

It seems likely, therefore, that normally the swallowing of liquor amnii is an essential part of the foetal metabolism—not, indeed, for the solids contained in this fluid, but merely to provide an internal fluid supply to the baby.