but, if so, I had completely forgotten it. Any one interested in our subject should not fail to look it up, as it is by far the most careful and detailed description of the mysterious aerial sounds of Yellowstone Park which has thus far been given by any one.

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

A RAPID IRON HAEMATOXYLIN TECHNIQUE

THERE seems to have been little if any work done aiming to secure a stock solution of haematoxylin which would remain indefinitely, or at least for a long time, in an entirely unoxidized condition. Such a solution would appear to be useful, especially in view of the fact that unoxidized haematoxylin solutions can be oxidized almost instantaneously by the addition of appropriate substances.[1,2,3] This line of procedure seemed to the writer to offer promising possibilities. If one could start with a totally unoxidized solution it would seem possible to devise a technique that would permit the stain to reach the tissue sections at the peak of the staining power, thus making possible a maximum of action in a minimum of time. The duration of the staining power of the solution would be of minor importance, providing that the method of preparing the ready stain were simple and rapid.

A series of experiments with the above considerations in mind has resulted in a technique which has been used with success on a considerable variety of tissues, following fixation in corrosive acetic, Bouin's, Carnoy's No. 1 or Zenker's solution. The formulae for the necessary stock solutions are as follows:

Mordant	50 per cent. alcohol	20 cc
	Ferric chloride	1 grm
	Glacial acetic	2 cc
Stock Haematoxylin Solution	Absolute alcohol	20 cc
	Sodium hydrosulphite	0.2 grm
	Distilled water	5 drops
	Haematoxylin crystals	1 grm

In the stock haematoxylin solution only a slight amount of the sodium hydrosulphite ($Na_2S_2O_4$) will dissolve.[4] It is necessary that an excess be added, so that at all times there will be some crystals in the bottle. The haematoxylin crystals should be the light brown, not the dark product.[5] After the ingredients have been mixed the bottle should be stoppered and solution facilitated by shaking. This completes the preparation of a very powerful stock solution which will keep without oxidation for a long time. Such a solution, which has been kept in ordinary daylight for over a year, still retains its original light amber color and shows no sign of oxidation. Tests made at intervals show that its staining power is unimpaired.

The mordant and stain may be used separately or combined. Staining may be accomplished either by flooding the slides or by immersion in staining jars. This procedure is not only useful for sections but also has a somewhat more restricted use for staining in bulk. The capabilities and limitations of this technique are being investigated further. At present it may be said that flooding sectioned material affixed to slides, first with the mordant and then with the stain, gives the best results. It has been found most convenient to keep both mordant and ready stain in dropping bottles.

Before giving the general outline of procedure it is necessary to describe the method of preparing the ready stain. To five cc of tap water in a dropping bottle are added five drops of the stock haematoxylin solution, followed by one drop of ammonium hydroxide. The solution is ready for use within thirty seconds and will retain vigorous staining power for about four hours. The substitution of 95 per cent. alcohol for water in the preparation of the ready stain increases the life of the solution so that it will stain satisfactorily for five days or even longer. The alcoholic solution, however, will not stain vigorously until it has set about twenty minutes after preparation. If the five drops of stock solution are added to ten drops of tap water, and a drop of ammonium hydroxide then added, oxidation proceeds rapidly. After thirty seconds five cc of 95 per cent. alcohol may be added. The stain is then ready for immediate use and has a considerably longer life than the aqueous ready stain. This last modification, then, combines the immediate readiness of the first solution and the longer life of the second.

Sections, in paraffin ribbons, are affixed to slides by the albumen and water method and are allowed to dry into contact. Then follows the usual proce-

[1] Mayer, Paul, 1891, "Ueber das Färben mit Hämatoxylin," Mitth. Zool. Stat. Neapel, Bd. 10, Heft 1, S. 170–186.

[2] Unna, P. G., 1892, "Ueber die Reifung unsere Farbstoffe," *Zeit. wiss. Mikr.*, Bd. 8, S. 475–487.

[3] Piazza, C., 1912, "L'invecchiamento rapido delle soluzioni ematossiliniche," *Zeit. wiss. Mikr.*, Bd. 29, S. 69–71.

[4] Sodium hydrosulphite used was that manufactured by E. I. du Pont de Nemours and Co., Inc.

[5] Light brown haematoxylin crystals manufactured by McAndrews and Forbes were used and gave entire satisfaction.

dure, involving the removal of paraffin and the running of the slides down the graded alcohols to water. A slide is then flooded with a few drops of mordant, used full strength. After five minutes the slide is rinsed for a few seconds in tap water and flooded with ready stain. Staining begins at once and is complete in ten minutes or less. After rinsing the slide is differentiated in 0.1 to 0.4 per cent. hydrochloric acid in water. When differentiation has reached the desired point the slide is transferred without rinsing to a jar of water containing one or two drops of ammonium hydroxide. As soon as the blue color has appeared in the sections the slide is removed, counterstained with erythrosin, passed up the graded alcohols to xylol and mounted in balsam. The final mount shows nuclear material in deep blue and cytoplasmic material in pink. The results are comparable to those obtained with the use of Delafield's haematoxylin, followed by erythrosin.

The color of the ready stain gives a clue as to the color of the nuclear material in the final preparation. Freshly prepared aqueous ready stain is deep purple in color, with a strong tint of blue. It will stain nuclear material a deep blue. After about an hour the ready stain has lost most of its blue tint and is a red orange in color. It will then stain nuclear material a bluish black. Still older stain will give a black nuclear stain. In other words, as oxidation of the ready stain proceeds the color changes from brilliant blue-purple to purple to orange-red to orange brown, and the color of the nuclear stain secured progresses from brilliant blue to blue-black to black. When the stain has reached the orange brown stage its staining power is largely gone and its use will give an undesirable yellow-brown nuclear staining but little stronger than the color imparted to the cytoplasm. The black nuclear stain resulting from the use of the orange-red ready stain is comparable to that secured by the use of Heidenhain's iron haematoxylin.

The ammonium hydroxide used as a "bluing" agent has the most powerful action on the color of sections stained with fresh stain, and least with those stained with the old stain. While there is a considerable amount of haematoxylin dissolved in the ammonia of the ready stain the nuclear stain resulting will be distinctly blue; as more and more of the haematoxylin becomes converted into haematein-ammonia and further oxidation products the resulting nuclear stain tends more and more toward black.

The best way to secure a blue nuclear stain is to use a freshly prepared ready stain. If an aqueous one, it should be used for this purpose within a half hour; if an alcoholic solution, within eight or ten hours. There are several ways in which to secure a black nuclear stain. First, use old ready stains; second, add two or three drops of ammonia in the preparation of the ready stain; third, flood slides with either fresh or old ready stain, and add a drop of dilute ammonium hydroxide to each slide. In any case the desired results will have been secured when the stain flooded on the slide has become orange brown in color. Contrary to expectations, the use of ammonium hydroxide as described in these three variations does not appear to injure the tissues. The third method is, perhaps, the easiest and best. As soon as the flooded stain has become orange brown in color the slide may be rinsed in tap water, counterstained, dehydrated, cleared and mounted in balsam. It must be admitted that there is still some question as to the permanence of the stain, but a number of slides stained by the methods given above show no signs of fading, although exposed for several months to ordinary daylight. Further study is being made along these lines and results will be reported at the appropriate time.

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SPECIAL ARTICLES

THE EFFECT OF PRESSURE ON THE MAGNETIZATION OF MAGNETITE

AMONG the interesting properties of magnetite, $FeO.Fe_2O_3$, is that of being oxidizable to Fe_2O_3 without loss of its ferro-magnetism and without a change in its cubic crystal structure. A closer study of this phenomenon and of hysteresis in magnetite reveals the following three facts which clearly suggest that hydrostatic pressure should increase the permeability:

(1) Magnetite oxidized to Fe_2O_3 is even more magnetic than before oxidation.[1] In one sample the maximum permeability increased 16 per cent. and the position of the maximum was shifted from a magnetizing field of 85 to 65 gausses.

(2) On annealing magnetite at 1000° C. the permeability maximum is shifted from 85 gausses to about 350.[2] Since annealing implies the removal of strains, the shift from 85 gausses to 65 produced by oxidation means that any existing strains were then increased.

(3) A consideration of the inter-atomic distances in magnetite[3] shows that the free unoccupied radial distance is 0.55Å, whereas the oxygen radius is usually found to be 0.65Å. The oxygen which enters on oxida-

[1] *Phil. Mag.*, 1925, 1, 403.
[2] *Phil. Mag.*, in press.
[3] *Phil. Mag.*, 1925, 1, 406.