tion (accomplished by pouring water into the concentrated sulfuric acid solution), were identified as uric acid through their ultraviolet absorption spectrum[3] (absorption maximum: 280 mμ in 0.1% sodium hydroxide) and certain qualitative properties. The filtrate, together with the second 400-ml extract, was evaporated *in vacuo*, and the residue was washed with 2*N* hydrochloric acid. The properties of the insoluble portion (white crystals and powder) will be described.

The hydrochloric acid solution was evaporated *in vacuo*, water was added, and the solution was then made alkaline with ammonia, the insoluble portions being discarded each time. After final refinement by the addition of charcoal, the green fluorescent solution was submitted to paper chromatography, using the butyl alcohol-acetic acid mixture by Partridge (*6*). The bands were located by their fluorescence (*2*). Since pure xanthopterin was unavailable, paper chromatography of the wings of a pierid (*Eurema etaeta* Boisduval) known to contain xanthopterin was carried out simultaneously. Though Good and Johnson (*2*) have given 0.38 for the R_f value of xanthopterin, we have obtained a value of 0.54. The yellow pigment from *Bombyx mori* gave the same R_f value, and several qualitative properties showed it to be xanthopterin. Riboflavin also gave the same R_f value, but its fluorescence and chemical behavior were different.

It is to be noted that, when left in the sunlight for one day, both the silkworm and pierid extracts acquired a fluorescence of a bright blue tone and instead of the yellowish green band on the paper chromatogram a bright blue band with an R_f value of 0.26 appeared. Similarly, when the paper-chromatographed filter strip of the undecomposed solution was left in the sunlight, the yellowish green band changed into a band with a bright blue fluorescence. The reason for this kind of photolysis of xanthopterin is obscure and apparently has not been described in the literature.

Xanthopterin was also concentrated from the original boiling water extract, according to the method of Crammer (*1*), using liquid phenol. This solution gave a band on the paper chromatogram with a purple fluorescence (R_f: 0.50) corresponding to the pterinlike substance mentioned below, in addition to the band of xanthopterin. One hundred milliliters of water was added to the 2*N* hydrochloric acid-insoluble portion mentioned above, the rather heavy crystals (uric acid) were separated, and the remaining flocky precipitate was reprecipitated several times from boiling water. The filtrate was next dissolved in a small amount of concentrated sulfuric acid and was poured into water dropwise, when white crystals were obtained. The hot aqueous solution gave a strong purple fluorescence (R_f: 0.50).

Thus, xanthopterin, uric acid, and a minute amount of some white crystals have been obtained from the yellow mutant. This white crystal, though closely related to leucopterin, isoxanthopterin, and the purine homologue, is not identical with any of them, and seems probably to be a new pterin. The chemical researches on this substance are being carried out and will be reported subsequently. This substance is obtained in greater amount from skins of the normal type of silkworms.

Jucci (*3*) has stated that the white epidermis pigment is a urate, and Shimizu (*7*), basing his work on this view of Jucci, has made some quantitative studies of the uric acid production by means of the Folins-Denis-Wu reaction. However, since the new pterinlike substance also gives a strongly positive Folins-Denis-Wu reaction, the results of Shimizu should be given further consideration.

We are not yet in a position to discuss the formation of epidermis pigments from a genetic point of view until the chemical structure of this substance is clarified.

[3] The authors are greatly indebted to Prof. K. Yamasaki for the measurements.

References

1. Crammer, J. L. *Nature,* Lond., 1948, **161**, 349.
2. Good, P. M. and Johnson, A. W. *Nature,* Lond., 1949, **163**, 31.
3. Jucci, C. *Proc. sixth internat. Congr. Genet.*, 1932, **1**, 377.
4. Kikkawa, H. *Advances in biology.* Tokyo: Kyoritsu Publishing Co. (in Japanese). 1948, Vol. III. Pp. 36.
5. ———. *Genetics,* 1941, **26**, 587.
6. Partridge, S. M. *Biochem. J.,* 1948, **42**, 238.
7. Shimizu, S. *Rep. sericultural exp. Sta.* (in Japanese), 1943, **11**, 379.

Processing Unit for 35-mm Color Film[1]

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The advantages of 35-mm color film are of such importance that its use is steadily increasing as a means of scientific record. Color recording has its greatest value where a combination of colors and shapes is such that in each separate item changes may exist which are hardly measurable and yet which, taken together, give a very marked visual difference in the total picture. This is of course especially true of pathological changes. At the present time, however, few facilities have been created for the average worker to develop his own films. This may be necessary in cases where the original picture will disappear before commercial development is completed and thus be lost. Having to face such a problem ourselves, and finding no available answer, we were compelled to find its practical solution. As this has been rather satisfactorily accomplished, it occurred to us that others might wish to take advantage of our study.

Very briefly, we may remark that to process color film requires 13 separate operations, one being a reexposure, four being intermediary or final washing. For the commercial operator with adequate volume of work, large and expensive tanks are available, but these are of little value to the ordinary laboratory, as the cost in chemical replacement alone would be prohibitive. For the amateur there are small tanks fitted with spools on which film must be placed. After a few trials with this aggravating and inefficient apparatus, we hastily discarded it. What was needed was an intermediate arrangement that could handle one or two films per day, be economical in operation, fully and evenly expose the film to the processing

[1] Sponsored by The Dr. Geo. W. McFatrich Memorial Ophinjection of solutions A, B, C, and D.

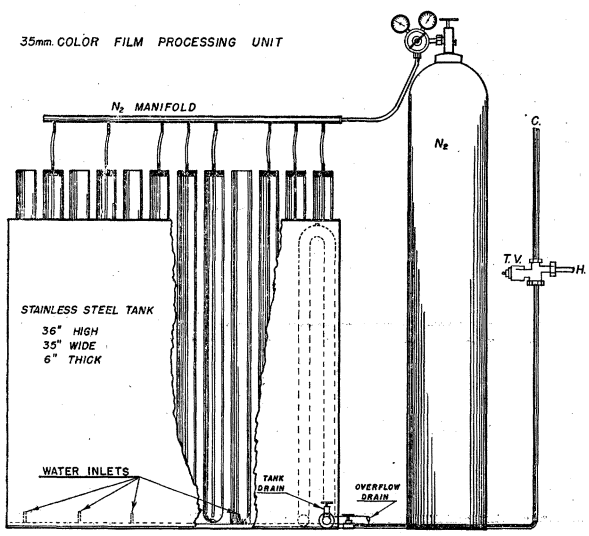

FIG. 1.

fluids, and possess the essential mechanical stirring and temperature control.

With these points in mind we built a tank (Fig. 1) from No. 26 gage stainless steel. Into this was fitted a rack, made by soldering cross bars of ⅛-in. brass rod at appropriate distances to form 12 equal compartments, with a center one to subdivide these into two. There are three layers of rods: one in the middle, the other two 1 in. from the top and 9 in. from the bottom respectively. Those holding the processing tubes are made from 52-mm (I.D.) Pyrex tubing 40 in. long. The tank is of sufficient size to take two rows of 12 tubes, so that two or even more films may be processed together. The ends of the eight processing tubes are sealed off, as we encountered trouble from bungs that work loose, especially with the alkaline solutions. The four wash tubes have a bung at the bottom through which water enters, appropriately led into the bottom of the tank. Water entering the washing tubes leaves simply by spilling over into the tank and from there it leaves by a constant level trap to the drain. The trap inlet, however, is from the bottom of the tank, so that a complete circulation is established. The water entering the wash tubes is connected to both hot and cold water supplies by a thermostatic valve set at 68° F and thus serves to maintain a reasonably constant temperature not only of the wash water but also of the entire processing tank and contained tubes.

Above the tank is an appropriately arranged manifold of copper tubing with 16 outlets, one for each processing tube. Each outlet is simply connected by rubber tubing to a 41-in. length of 1- to 2-mm Pyrex capillary tubing that goes through the bung to the bottom of the process tube. Simple screw clamps are on each supply. These tubes pass through the bung eccentrically, so that when in use they can be twisted backwards clear out of the way of the film, but still conveniently rest on the edge of the process tube. To this arrangement is connected a tank of nitrogen, which by bubbling from the bottom of the process tubes effectively stirs the contents and has the not-looked-for advantage of keeping the uptake of incidental oxygen to a minimal value. In consequence of this, developers remain remarkably clean and fresh, but they are replaced regularly at the end of each two-week period, since their cost is small in view of the danger of losing a valuable record.

In order to facilitate the actual operation we have also an overhead gear consisting of a ¼-in. brass rod supported on brackets on which a small overhead pulley runs the full length of the tank. Over the pulley is a cord on one end of which is a stainless steel photo clamp (Ansco), and on the other a suitable brass ring, which is prevented by a stop from passing too far over the pulley. (The first part of the process is in absolute darkness and equipment must be readily handled by feel.) Situated directly opposite the correct tube in the series is a floodlight arranged on a suitable folding bracket which, when pulled forward, is exactly the correct distance from the suspended film. This completes the setup. All that is now needed is a vociferous time-interval clock with handles suitable for operation in the dark.

The actual processing is very simple. First, the water is turned on and its temperature is checked. The nitrogen commences bubbling through the first three baths (performed in total darkness). The light is turned off, and the film is fastened by one end to the overhead gear. At the other end is fastened a similar stainless steel clamp loaded with 20 g of bar solder, to keep the film from waving too freely even with excessive bubbling. The film is lowered into the first developing tube, and the clock is turned on. At the proper time, the film is pulled from the tube by the ring and lowered into the second tube, and the timing is repeated. After the processing in the third tube, lights may be turned on, and the remainder of the process may be carried on in ordinary light. At the appropriate time the film is reexposed. This is accomplished by hauling it clear from the tube, which is so arranged that it is exactly opposite the floodlight. Finally, after complete processing and washing, the film, still weighted, is hung up to dry.

It will be noted that the fingers never touch the film, except at the discarded ends, at any stage. We have had practically no trouble with spotting from any cause, and no trouble from uneven development or accidental contamination, although we have used this method for processing several hundred films. It may also be noted that after the dark stage, nitrogen is passed into each tube as needed, thus minimizing waste. On a regular output of one film a day the cost is little more than rent of the cylinder. Finally, when the process is over the bungs are replaced in the process tubes to keep out oxygen and dust.

Appropriate timing and technical details are omitted, since they must be acquired for the particular variety of film used. At this time the only reliable source of suitable film is Ansco, with which we have had satisfactory results. We must warn those inexperienced in color work to pay great attention to details, above all to temperature control, for the film is far less tolerant than the good-tempered material now available for black and white.

We also feel that, as film batches vary somewhat, abso-

lute color rendering is not yet really practical. Experiments should be so planned that reliance is placed on comparison of adjoining material on the same film, with no attempt to set up absolute color values. With these limitations, our experience has shown us that the advantages of color film are such as to render it a necessity for certain types of work. We have attempted color reproduction on paper, which for some purposes would be most useful, but at present we must report adversely. Color rendering is quite markedly diminished and the cost in time, of processing especially, is practically prohibitive except for limited use.

Growth Regulators Prolong the Bloom of Oriental Flowering Cherries and Dogwood

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Considerable interest has been shown in the progress of experiments with growth-regulator (hormone) sprays to prolong the blossom display of the Oriental Japanese, or flowering cherries, Yoshino, *Prunus yedoensis* Matsum; Akebono, *P. yedoensis*; Kwanzan, *P. serrulata* Lindl.; and Shiro-fugen, *P. serrulata*, in Washington, D. C. These experiments have been conducted during the past two seasons (1948 and 1949) and have been extended to include a number of other woody ornamentals. Thus far the treatment has proved most effective on Oriental cherries and white flowering dogwood, *Cornus florida* L.

In 1948, individual branches on trees of the single-flowered Yoshino variety and small whole trees of the double-flowered Kwanzan cherry were used. Both varieties responded to sprays of α-naphthaleneacetic acid and β-naphthaoxyacetic acid at concentrations of 5, 10, 20, 40, and 80 ppm. Frequently 25% of the petals of sprayed flowers remained attached 4 to 10 days longer than the unsprayed. Foliage of Yoshino and Kwanzan was injured by sprays containing 40 and 80 ppm, but no injury was evident at the lower concentrations. A spray mixture containing 64 ppm of indolebutyric acid and 16 ppm of β-naphthoxyacetic acid was also effective in retarding petal fall of Yoshino, but proportionate mixtures of this combination at one-half and lower concentrations were ineffective. Another compound, *p*-chlorophenoxyacetic acid, was ineffective in retarding petal fall of the Shiro-fugen cherry at spray concentrations 5 to 20 ppm, whereas naphthaleneacetic acid at the same concentrations was quite effective.

Branches of Yoshino that were sprayed retained their petals 4 to 7 days longer than comparable unsprayed branches. During this period the sprayed petals gradually developed a more intense pink pigmentation than did unsprayed ones and the majority of the treated petals appeared fresh until they fell from the tree.

Young Kwanzan trees held their flowers for 7 to 10 days longer than unsprayed ones. The double flowers of this variety are borne on long, slender flower stalks which allow them to be whipped back and forth by the wind, thus causing some discoloration and shedding of the petals. At a distance of 10 to 15 ft the mass floral display was nevertheless fairly attractive during most of the period that the flowers remained attached, even though the petals were injured by the wind.

In another experiment, naphthaleneacetic acid spray (10 ppm) was applied at intervals during the blossoming period. The results of this test indicated that the chemical was most effective when applied as the trees came into full bloom. Treatments applied to flower buds just prior to opening were ineffective.

Over 400 Japanese flowering cherry trees were treated with the growth-regulator sprays in experiments conducted during 1949. In the Tidal Basin planting, some Yoshino and Akebono trees retained 35% to 80% of their blossoms 3 to 7 days longer than unsprayed trees, but others showed little or no response. A detailed check on the effect of naphthaleneacetic acid (10 ppm) applied to 65 large Yoshino trees on March 29–30 showed an average of 20% of the flowers present April 11, 13 days after full bloom, in comparison with 3% remaining on ten comparable unsprayed trees at this time. Direct comparison between naphthaleneacetic acid and β-naphthoxyacetic acid showed that the former compound was the more effective.

Kwanzan flowers responded more consistently to 10 ppm naphthaleneacetic acid than did the Yoshino and Akebono varieties. Thirteen days after treatment, an average of 23% of the flowers was found present on 32 large Kwanzan trees, in comparison with 4% on nine comparable untreated trees.

In addition to Japanese cherry, tests have been conducted on other spring-flowering ornamental plants. White flowering dogwood has shown a definite response in two years of testing. During the 1948 season two compounds, α-naphthaleneacetic acid and *p*-chlorophenoxyacetic acid, were applied to individual branches at 10- and 20-ppm spray concentrations. The petal-like flower bracts of dogwood remained attached 4 to 6 days longer when sprayed with either concentration of these compounds than when left unsprayed. Further tests made in 1949 indicate that *p*-chlorophenoxyacetic acid is much more effective in prolonging dogwood blossom display than is naphthaleneacetic acid; but the former compound tends to deform the young leaves at spray concentrations as low as 5 ppm. For this reason additional experimentation with sprays of lower concentration than 5 ppm is necessary to evaluate the practicability of using *p*-chlorophenoxyacetic acid on dogwood.

In tests conducted thus far, the flowers of a number of other kinds of plants have not shown a definite reduction in rate of petal fall when sprayed with α-naphthaleneacetic acid, *p*-chlorophenoxyacetic acid, or β-naphthoxyacetic acid in concentrations of 10 and 20 ppm. Among the plants used were azalea, aronia, American and Asiatic crab apples, flowering almond, flowering quince, redbud, bridal wreath spirea, lilac, star magnolia, and saucer magnolia.