The Colours and Pigments of Flowers with Special Reference to Genetics.

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Investigations, of which the following is some account, have been undertaken with a view to being of assistance in the interpretation of the phenomena observed in the inheritance of flower-colour. An attempt has been made to classify, of necessity roughly, the pigments, more especially those soluble in water, found in flowering plants, and at the same time to ascertain whether there is any connection between the genetic behaviour of pigments and their chemical reactions and constitution. On the basis of this classification it was thought that, at some future time, further investigations might be carried out in greater detail among the various classes of pigments.

This account deals more with yellow pigments than with red; attention was first directed to the yellow group, because of a certain correlation between reds and yellows observed in the inheritance of flower-colour in *Antirrhinum majus*, the relationship suggesting that a greater knowledge of yellows might be useful in classifying the reds. No detailed examination has been made of any one pigment, but merely a general survey of the colouring matters of genera from various natural orders.

A classification in outline of the pigments, other than chlorophyll, found in flowering plants has been given by many authorities,* but perhaps a repetition will not be out of place here, as follows:—

A. Pigments in solution in the cell-sap.†

(1) Soluble red-purple-blue pigments known as "*anthocyanin*"; that this term includes several different classes of pigments seems probable when observations are made as regards their behaviour towards various reagents. The sub-classes will be described later.

(2) Soluble yellow pigments known as "*xantheïn*." Again, various sub-classes, to be described later, may be made according to their reactions towards reagents.

* General classifications and properties of pigments, as far as they are known, are given by Czapek (4) and Zimmermann (13).

† Said to be occasionally precipitated or crystallised out on natural concentration of the cell-sap.

B. Pigments associated with specialised protoplasmic bodies—chromoplastids—the colour in this case being usually yellow, orange-yellow, orange, or orange-red. Insolubility in water appears to be a constant characteristic of this group. Two well-known pigments are included here :—

(1) *Carotin*—a hydrocarbon of definite and characteristic properties. According to Zimmermann (13), it is insoluble in water, almost so in alcohol, slightly soluble in ether, more readily in benzine, and most so in chloroform and carbon bisulphide. It occurs naturally and can be obtained artificially in crystalline form, and is microchemically recognised by certain reagents; with concentrated sulphuric acid it gives an indigo-blue colour, at first momentarily violet, and with iodine a green or greenish-blue colour.

(2) *Xanthin*—again, according to Zimmermann (13), occurs in the plastid in amorphous form. It is insoluble in water, somewhat soluble in ether, chloroform, and benzine, but more so in alcohol. Microchemically xanthin can be recognised by giving with concentrated sulphuric acid a blue colour, at first momentarily green, and with iodine a green colour.

In addition to the above, there appear to be other plastic pigments, which do not give a blue colour with sulphuric acid, but a yellow or brown.

Anthocyanin.

Classification of the soluble red-purple-blue pigments has always been a difficult problem, but there seems to be evidence that anthocyanin is a general term, including several different pigments. The differentiation made here is based on the inheritance of colour in certain genera, on the sequence of flower-colour in cultivated varieties, and finally on the behaviour of red pigments towards chemical reagents.

There are present in most plants colourless or pale yellow substances, soluble in water, but insoluble in ether; with strong acids and alkalis they give a canary-yellow colour, and a similar coloration or most frequently a precipitate of the same colour with basic lead acetate. In some cases, *Eschscholtzia californica* (Courchet (3)), *Argemone grandiflora*, and yellow species of *Viola*, these substances crystallise from extract solutions in needle-shaped crystals aggregated in clusters or spherules; solutions of the crystals from the above genera reduced Fehling's solution slightly, but after prolonged boiling with dilute acid, a deep yellow substance, together with a reduction of Fehling's solution, was obtained, suggesting the glucoside nature of the crystalline bodies. A similar glucoside is probably present in *Narcissus Tazetta* (Bidgood (2)).

The colour reaction with alkalis and acids is most obvious in parts free

from chlorophyll; in unpigmented genera, *Galanthus nivalis*, for instance, or in pigmented (anthocyanic) genera such as many *Umbelliferæ*, with white flowers, the petals turn bright canary yellow with ammonia.

The inheritance of colour in *Antirrhinum* (Wheldale (12)) has led to the suggestion that anthocyanin is possibly a compound of such a glucoside-like body with a reddening substance. The original type of *Antirrhinum* has magenta (anthocyanin) flowers.* Loss of the reddening substance, which may be represented by a Mendelian factor (M), gives a variety bearing ivory-white flowers containing no pigment (except in the palate and hairs), but a glucoside-like body giving the reactions with acids, alkalis, and lead acetate described above. Further loss of a substance, again represented by a factor (I), from the glucoside-like body in the superficial cells of the lips gives a yellow xantheïc pigment, and the variety thus bears yellow flowers. Loss of yellow pigment, represented by yet another factor (Y), gives an albino, containing no pigment and no glucoside-like body. Local decomposition produces the same xantheïc pigment on the palate and in hairs on the inner surface of the tube in all varieties except the albino. The albino may carry I or M, or both, since these factors are invisible unless the fundamental colour Y is present. Moreover, the reddening factor can exist with Y, the decomposition product, giving a mixed colour, *i.e.*, crimson. Each variety may breed true or may throw itself and one or more varieties below it in the scale of colour, according as it is homo- or heterozygous in the various factors. Magenta can throw all varieties; crimson can throw yellow and white; ivory, yellow and white; and yellow, white only.

At this point it is interesting, perhaps, to give views regarding the constitution of anthocyanin, based on results obtained from a totally different kind of investigation. Overton (9) found that in plants supplied artificially with excess of sugar and other carbohydrates there is a correlated increase in the production of anthocyanin, and he concludes that the latter, in many cases, is a glucoside compound of a tannic acid. Molisch (8) and Heise (6) are also of the opinion that some red sap-pigments are glucosidal in nature. Tannin is by no means always present in plants containing anthocyanin; the magenta pigment of *Antirrhinum* gives a tannin reaction, but no tannin has been found in the albinos.

A similar range of colour to that found in *Antirrhinum*, *i.e.*, various shades of purple or magenta and crimson, together with ivory, yellow, and sometimes white, also occurs in *Althæa rosea*, *Azalea*, *Dahlia variabilis*, *Dianthus Caryophyllus*, *Helichrysum bracteatum*, *Linaria*, *Nemesia*, *Phlox Drummondii*,

* With yellow pigment locally on the palate and in hairs on the inner surface of the tube.

and *Rosa*. It is probable that the inheritance of flower-colour will be found to be similar in these genera.

The inheritance of colour in *Lathyrus* and *Matthiola* (Bateson, Punnett and Saunders (1)) differs from *Antirrhinum*, in that two factors (C and R) are required to produce colour, and the loss of either gives a white containing only a glucoside-like substance. An additional factor, B, gives the purple varieties.

A similar range of colour to that in *Lathyrus*, *i.e.*, shades of blue or purple and red, together with white, is shown by *Campanula*, *Digitalis purpurea*, *Iberis*, *Lobelia*, *Nemophila insignis*, *Pisum*, and many others; no xantheïc variety occurs in this series.

It is evident, then, that there are two classes of anthocyanin as regards the series of varieties to which each can give rise; in one case the decomposition possibly of the glucoside-like constituent gives a yellow xantheïc form, and in the other case no such decomposition is possible, and no yellow variety exists.

The close relationship between xantheïc pigments, the glucoside-like bodies, from which they may be derived, and the anthocyanin, of which these bodies are themselves possibly constituents, is suggested also by the fact that yellow xantheïc varieties almost always have an anthocyanic type. This connection is well exemplified among the genera of the Compositæ. Usually plastid pigments in addition are present in this order, but these may be disregarded for the moment. Yellow varieties of *Coreopsis*, *Chrysanthemum carinatum*, *Dahlia variabilis*, *Helichrysum bracteatum*, contain xantheïn, while the type has anthocyanin; other genera, *Zinnia elegans*, *Gaillardia*, *Hieracium rubrum*, have anthocyanin of the kind which gives no xantheïc varieties, while, finally, *Calendula officinalis*, *Helianthus annuus*, *Erigèron* spp., and *Senecio* spp. have no xantheïn and no anthocyanin.

When we consider the behaviour of anthocyanin towards such substances as acids, alkalis, certain salts, etc., we find this term includes at least several groups of pigments. Two classes have been described by Weigert (11), and termed "weinrot" and "rübenrot" respectively. The former is soluble in both alcohol and water, giving with basic lead acetate blue-grey or blue-green precipitates, and with concentrated sulphuric acid a bright red colour. It is found in the leaves of *Vitis*, *Ampelopsis quinquefolia*, *Rhus typhina*, *Cornus sanguinea*, and others. The latter, rübenrot, Weigert found in leaves of the *Amarantaceæ* and *Chenopodiaceæ* (*Beta vulgaris*, *Iresine*, *Amaranthus*, *Atriplex*), and in fruits of *Phytolacca decandra*. Though readily soluble in water, the pigment is insoluble in alcohol; with basic lead acetate it gives a red precipitate, with sulphuric acid and with ammonia a deep violet, but with

other bases a yellow colour. He further distinguishes a pigment, "Malven-violett," occurring in the leaves of *Coleus Hero, Perilla nankinensis, Corylus avellana atropurp.*, red *Brassica*, and *Malva* sp. It is present as a compound, which, on the action of acids, produces the weinrot pigment.

In the present paper, conclusions are based upon the examination of relatively few red pigments, and even these present great complexity. Yet, on the whole, a rough classification may be given as follows :—

1. A purple anthocyanin, which is characterised by giving a deep blue or violet colour with ferrous sulphate and ferric chloride, a green colour with alkalis and a blue or blue-green precipitate with basic lead acetate. Such a pigment is found in the deep purple and crimson varieties of *Dianthus Caryophyllus, Lathyrus odoratus, Phlox Drummondii,* the purple and plum varieties of *Matthiola* (Saunders (1)), purple *Fuchsia*, and in the berries of *Atropa Belladonna* and *Rosa pimpinellifolia.*

2. A purplish-red anthocyanin, corresponding probably to Weigert's weinrot. This pigment does not give the above reaction with iron salts; but with alkalis a green colour and a green precipitate with basic lead acetate. This form occurs in the magenta varieties of *Antirrhinum majus, Dahlia variabilis,* pale magenta and violet varieties of *Phlox Drummondii,* red varieties of *Lathyrus odoratus, Salvia Horminum,* and *Verbena*; also in the crimson *Cheiranthus Cheiri*, in *Tradescantia virginiana* and in *Rhus aromatica.*

3. A red anthocyanin characterised by giving a red precipitate with basic lead acetate. Two sub-groups may be further identified as—(*a*) those giving green colour with alkalis—red, copper, and rose varieties of *Matthiola*, and a red variety of *Delphinium*; (*b*) those giving a reddish yellow colour with alkalis—pink varieties of *Dianthus Caryophyllus,* flesh and terra-cotta varieties of *Matthiola,* certain scarlets of *Lathyrus,* salmon-rose varieties of *Dianthus barbatus, Phlox Drummondii, Verbena,* and the "rose dorée" variety of *Antirrhinum majus.*

The above classes seem to show a gradual diminution in the amount of blueness present as we pass from the very blue purples of *Lathyrus,* through the magentas and blue-reds of *Antirrhinum* and *Lathyrus,* to end finally in such varieties as the "rose dorée" *Antirrhinum,* from which blueness is practically absent. The bluer form is usually the original type and the reds, as derivatives, are probably components of the original anthocyanin.

There is some indication of a connection between the chemical behaviour of the classes and their inheritance. The purple anthocyanin appears to be that form, which, in *Lathyrus* and *Matthiola,* is given when the B factor is present in addition to C and R. The extreme purple is not found in *Antirrhinum*; hence in the latter case, when a blueing factor is present, as in the magenta

type, purplish-red anthocyanin is the ultimate form. The red of *Antirrhinum* is represented by the "rose dorée" type, which finds its parallel in the flesh and copper of *Matthiola*, certain scarlets of *Lathyrus*, and the salmon-rose of *Phlox Drummondii*.

It is of interest to note also that in the reds of *Antirrhinum* and the salmon-rose of *Phlox*,* of which several shades exist, the deeper are dominant to the paler, while in the purples, purple-reds, and magentas of *Lathyrus Matthiola*, *Phlox*, and *Antirrhinum*, the paler shades are dominant to the deeper. As regards shades of one colour, a fuller investigation has been made in *Antirrhinum*, in which genus every shade is a definite zygotic form, and the chemical reactions of these shades are fundamentally similar though differing in degree. The precipitates with basic lead acetate, for instance, are of varying shades of green, yet these remain unaltered on artificial concentration or dilution of the extract. This would appear to indicate that the shade of colour was but an outward indication of some definite organic compound in the sap.

All the red pigments so far described give, with strong sulphuric acid,† bright red and yellow colours, becoming orange when mixed; it seems possible that the red is due to the reddening factor, and the yellow to the glucoside-like constituent of the anthocyanin. With alkalis, the reddening factor of the bluish-red turns blue and the other body yellow, the result being green, sometimes rapidly fading to yellow. Basic lead acetate gives blue-green or green precipitates, due again to the same mixture.

The scarlet pigment of some genera—*Lobelia cardinalis*, *Phaseolus multiflorus*—is again different, in that it gives a bluish colour with alkalis and a red precipitate with basic lead acetate.

With regard to natural orders and relationships, as far as these investigations have gone, it appears that the red pigments of the *Papaveraceæ* differ from others. Also those of the allied orders *Amarantaceæ*, *Nyctaginaceæ*, *Phytolaccaceæ*, and *Portulacaceæ* (included in the *Centrospermæ* by Engler), form an isolated group giving reactions essentially different from any hitherto described.

Of the *Papaveraceæ*, the red pigment of *Glaucium phœniceum* and *Papaver Rhœas* gives a purple colour with basic lead acetate.

The red pigment of *Amaranthus* (and other genera of the same order according to Weigert (11)) is characterised by its insolubility in alcohol. With concentrated sulphuric acid it gives a purple colour, with ammonia

* I am indebted to Miss Killby for this information.

† Reactions are best seen by dropping acid on to the pigment on a white porcelain plate. Mixtures of colours are thus more readily detected than in bulk in solution.

a reddish colour, but with other bases a clear yellow, and with basic lead acetate an orange-red precipitate. In the stems and berries of *Phytolacca decandra,* the magenta and crimson flowers of *Mirabilis Jalapa* and *Portulaca grandiflora,* two pigments apparently exist. One, insoluble in alcohol but soluble in water to a magenta solution, gives reactions on the whole similar to those given by the red pigment of *Amaranthus.* The other pigment is soluble in alcohol to a crimson solution, which gives a yellow colour with acids and alkalis and a reddish precipitate with basic lead acetate.

Overton (9) is also of the opinion that anthocyanin contains several classes of pigments, of which he gives—(1) the *Amarantaceæ* and *Beta vulgaris,* (2) *Papaver Rhœas* and other species of *Papaveraceæ,* (3) *Tradescantia discolor** and other *Commelinaceæ.*

The alcoholic solution of the red pigment in some genera is colourless, the colour returning on evaporation or on addition of acid. In other cases, again, the alcoholic solution is as deeply coloured as the flowers. It is possible that these phenomena may indicate a difference in the nature of the pigments.

Lastly, it might be well to mention that anthocyanin is said to occur in solid and crystalline states in the cell. Many instances are cited by Mölisch (8) in his work on crystalline anthocyanin.

Xantheïn.

Xantheïn, like anthocyanin, includes at least several pigments varying in their reactions towards acids and alkalis. They may be classified as follows :—

1. Those giving a deeper yellow, orange, or orange-red colour with acids and alkalis and similarly coloured precipitates with basic lead acetate. Such pigments are found in yellow varieties of *Althæa rosea, Antirrhinum majus, Calceolaria, Coreopsis, Dahlia variabilis, Dianthus Caryophyllus, Helichrysum bracteatum, Phlox Drummondii* and *Tagetes signata.*

2. Those in which the yellow colour becomes paler with acids and alkalis and basic lead acetate gives, as a rule, no precipitate or a precipitate of the same colour as the pigment. Such is the case in *Mirabilis Jalapa, Montbretia* sp., *Nemesia strumosa, Papaver nudicaule,* and *Portulaca grandiflora.*

3. Those in which the yellow colour remains unaltered in the presence of acids and alkalis, and basic lead acetate gives a yellow precipitate, as in *Mesembryanthemum pomeridianum, Verbascum.*

If xantheïc pigments are derivatives of anthocyanin, the dissimilarity of the former among themselves strengthens the view that the reds, from which they are derived, are also dissimilar.

* I have not been able to detect so far any divergence in this case.

Albinism.

It has been suggested that there are two forms of anthocyanin giving respectively two colour series, one containing a yellow xantheïc variety, the other not. Whites occur in both, and it seems probable that the term albinism should be used in a different sense when applied to each of the two series.

The extract from most white flowers (also from flowers coloured only with plastid pigments, when these have been removed), gives a canary colour with strong acids and alkalis, as stated previously. Without exception, as far as observations have gone, whites of genera having no yellow sap type, have given this yellow colour-reaction. These whites may, without hesitation, be declared to be recessive to the red-purple-blue types, and they are albinos as regards anthocyanin.

On the other hand, in the case of *Antirrhinum*, *Azalea* and *Phlox Drummondii*,* belonging to the series giving yellow sap-colour, whites exist which do not give the same yellow colour-reaction. Moreover, these whites are recessive to yellow in *Antirrhinum* and *Phlox*, and are albinos as regards both anthocyanin and xantheïn. It is the ivory in this series which contains the glucoside-like body, and gives the yellow colour-reaction.

Whites giving no colour-reaction have not yet been observed in the other genera mentioned in the anthocyanic-xantheïc series, though relatively few types have been examined. It is possible that the true albino, as contrasted with ivory, is rare in commercial samples, since the albino type in *Antirrhinum* and *Phlox* has been found to set poor seed unless fertilised artificially.

What appears at first an exception to this view is the case of *Mirabilis Jalapa*. Here we find a range of colour similar to that in *Antirrhinum*, *i.e.*, shades of magenta and crimson, together with deep and pale yellow and white. The white (when it does not carry a reddening factor) is recessive to yellow,† and yet gives a colour-reaction with ammonia, etc. The explanation lies in the fact that both the yellow and red pigments in *Mirabilis* (see p. 50) are of an entirely different nature from those in *Antirrhinum* and *Phlox*. For the same reason, the inheritance in *Portulaca grandiflora* will, if worked out, doubtless prove to be similar to *Mirabilis*.

Shull (10) also gives the case of *Verbascum Blattaria*, in which a very

* I am indebted to Miss Killby for the information that in *Phlox Drummondii* the ivory type may throw both the yellow and the albino.

† I am indebted to Miss Marryat for this information from results obtained in cross-breeding of *Mirabilis*.

pale form, perhaps an albino, is recessive to the full yellow. He also states that Correns found white *Polemonium cœruleum* to be dominant to yellow *Polemonium flavum*. If the former contained a glucoside-like body, and the latter a xantheïc pigment, the result might be analogous to *Phlox*. As material of these genera was not available, the pigments have not been investigated, and consequently this conjecture must remain unverified for the present.

Plastid Pigments.

The plastid pigments, carotin and xanthin, are well-known substances, of which the properties and characteristics have been investigated. Both may be present in the same plastid, when the colour is orange-yellow, orange or orange-red, and this condition is very widely distributed; or xanthin only may be present, when the colour is yellow. In the orange-yellow or red type the loss of the power to produce carotin in the plant may give rise to a lemon-yellow variety. This is the case in flowers of *Argemone grandiflora*, *Calendula officinalis*, *Tagetes signata*, *Tropæolum majus*, and probably *Cheiranthus Cheiri* and *Salpiglossis grandiflora*.

In other cultivated genera, where the type contains xanthin, this pigment appears to give rise to paler yellow varieties containing derivative plastid pigments, probably decomposition products of xanthin, and giving a yellow or brown colour with strong sulphuric acid. At present these derivatives have not been thoroughly examined. They are found in the pale yellow varieties of *Helianthemum* spp., *Chrysanthemum carinatum*, in the autumn cultivated *Chrysanthemum*, and in *Zinnia elegans*.

There is evidence from cross-breeding in *Cheiranthus* and *Tropæolum** that presence of carotin is dominant to its absence, that is the orange-yellow variety is dominant to the lemon-yellow.

The plastid pigment in cream varieties of *Lathyrus odoratus*, *Matthiola*, *Rosa*, and *Eschscholtzia caniculata rosea*, is again different from carotin and xanthin as regards its chemical reactions. There is evidence from cross-breeding in *Lathyrus* and *Matthiola* (Bateson and Saunders (1)) that cream plastid pigment is recessive to its absence, *i.e.*, colourless plastid.

Combinations of Soluble and Plastid Pigments.

Anthocyanin and plastid pigments are frequently found together in plants. When the red sap occurs with plastids containing both carotin and xanthin, the resulting colour is some shade of brown, crimson, or orange-red; with plastids containing xanthin only, or some derivative product of xanthin the

* I am indebted to Miss Saunders for information regarding *Tropæolum*.

resulting colour is maroon, purple, or salmon-pink. Hence we find in cultivated genera containing plastid pigments and anthocyanin a colour series brown, crimson, or orange, purple, magenta, or salmon-pink, deep yellow and pale yellow.

Such is the case in *Cheiranthus Cheiri*, *Chrysanthemum* spp., *Helianthemum* spp., *Salpiglossis grandiflora*, *Tagetes signata*, *Tropæolum majus*, and *Zinnia* spp.

This series differs from the anthocyanic-xantheïc series in one respect; in the former the type is crimson and the purple or magenta is the derivative, whereas in the latter the purple or magenta is the type, while crimson is a derivative. These two contrasting series cannot be better exemplified than by the two indigenous genera, *Antirrhinum* and *Cheiranthus*, and their cultivated varieties. The wild *Cheiranthus* is deep yellow tinged with brown; cultivation has produced from the original, a pale yellow type, to which the addition of anthocyanin gives purple. The wild *Antirrhinum* is magenta, which, on loss of some constituent, has given a yellow xantheïc type, and this gives, further, in presence of the reddening substance, a crimson.

Stress should be laid, in connection with colour, on the conception of the pigmentation of a plant as a whole. The power to produce colour is the property of every cell of a pigmented plant; frequently the flowers are white or show but little colour in plants which are really pigmented, as, for example, *Solanum nigrum*, *Geranium Robertianum*, var. *album*. In a plant having red, purple, or blue flowers, anthocyanin may invariably be detected in the vegetative parts, such as cotyledons, under surfaces of leaves, wounded or exposed areas, etc. The diffusion of colour throughout the plant is manifested in the correlation so frequently found between fruit- and seed-colour on the one hand and flower-colour on the other. De Vries (5) gives as examples the green-flowered variety of *Atropa Belladonna* and the white-flowered variety of *Daphne Mezereum* with yellow fruits; also the white-flowered *Linum* with yellow seeds as contrasted with the brown seeds of the blue variety. The colour and pattern of seed-coat in *Matthiola* (Bateson and Saunders (1)) and Pisum (Lock (7)) is also correlated with flower-colour in the same way.

Method for Examination of Pigments.

The material to be examined is ground very finely with powdered glass in a mortar, extracted with methylated spirit and filtered. If from the colour of the residue, or from a microscopic examination, the presence of carotin be suspected, a further extraction is made with benzine or chloroform. The alcohol extract contains the pigments soluble in water and such plastid

pigments as are soluble in alcohol (chiefly xanthin). After evaporation to dryness on a water-bath, the xanthin is separated by ether from the xantheïc and anthocyanic pigments. The chloroform extract contains both xanthin and carotin; the latter can be washed free from xanthin by means of alcohol.

Details for Genera of Various Natural Orders.

The following particulars chiefly concern yellows, the reds having been dealt with in detail previously.

Aizoaceæ.

Mesembryanthemum pomeridianum has a xantheïc pigment, unaffected by acids and alkalis and precipitated by basic lead acetate.

Amaryllidaceæ.

Alstrœmeria aurantiaca has plastids containing carotin and xanthin; anthocyanin is present in addition.

Caryophyllaceæ.

Dianthus Caryophyllus, the parent form of the Carnation, shows the anthocyanic-xantheïc series. The yellow pigment is intensified to orange yellow by acids and alkalis. Magentas and crimsons are probably produced by the addition of red sap to ivory and yellow respectively. Ivory gives the yellow colour-reaction with acids and alkalis, but whether it is dominant to yellow, and whether an albino, recessive to yellow, exists has not yet been ascertained.

Cistaceæ.

Varieties of *Helianthemum vulgare* show the anthocyanin-xanthin series and pale yellows, which are derivative plastid pigments, probably from xanthin. The type is crimson, *i.e.* anthocyanin on xanthin, and the magenta and pink types are due to the same sap-colour on the pale yellow derivative forms.

Compositæ.

Calendula officinalis exists in two varieties, orange and lemon yellow; in the former the colour is due to plastids containing carotin and xanthin, in the latter xanthin only. There is apparently no anthocyanic form.

Chrysanthemum carinatum shows the anthocyanic-xantheïc-xanthin series with pale yellows containing derivative products of xanthin, and whites giving the yellow colour-reaction with acids and alkalis, and containing colourless plastids. Anthocyanin on whites, pale yellows, and deep yellows gives the usual magentas and crimsons. In the yellows, from which antho-

cyanin is absent, a xantheïc pigment exists, which gives an orange colour with acids and alkalis and an orange precipitate with basic lead acetate.

The autumnal cultivated forms of *Chrysanthemum* resemble the above species, except that the anthocyanin appears to be of the kind which does not give a xantheïc derivative.

Coreopsis Drummondii has a brown patch of anthocyanin-containing cells at the base of the ray-florets. The orange-yellow of the florets is due to plastids containing carotin and xanthin. A xantheïc pigment, probably left as the anthocyanin retreats to the base of the florets, is also present in their upper portions; it turns orange-red with acids and alkalis and is precipitated by basic lead acetate as an orange precipitate.

Dahlia variabilis shows the anthocyanic-xantheïc series. The yellow turns a brilliant orange colour with acids and alkalis, and is precipitated as a deep orange-red precipitate by basic lead acetate. The ivory gives the yellow colour-reaction. Magentas and purples are anthocyanin on ivory, and crimsons are anthocyanin with xantheïn.

Gaillardia spp. have usually orange-yellow ray-florets, with anthocyanin at the base. The orange-yellow is due to plastids containing carotin and xanthin. There appears to be no xantheïc derivative.

Gazania splendens has orange-yellow ray-florets with a dark basal patch of anthocyanin cells. Orange-yellow is again due to carotin and xanthin.

Helianthus annuus has plastids containing xanthin; a pale yellow variety exists in which the plastids probably contain some derivative product of xanthin.

Helichrysum bracteatum shows the anthocyanic-xantheïc series. The white variety gives the yellow colour-reaction with acids and alkalis. The yellow contains a xantheïc pigment, which gives an intense orange colour with acids and alkalis, and a similarly coloured precipitate with basic lead acetate. Magentas and crimsons are due to anthocyanin on white and yellow respectively.

Hieracium rubrum has anthocyanin in addition to plastids containing carotin and xanthin.

Picris pauciflorus, Senecio Jacobæa, and *Taraxacum officinale* have only plastids containing carotin and xanthin.

Tagetes signata shows the anthocyanin-xantheïn-plastid series. There is an orange-yellow variety with plastids containing carotin and xanthin, and a lemon-yellow variety with plastids containing xanthin only; both yellows have in addition a xantheïc pigment derived from anthocyanin; it is intensified in colour by acids and alkalis, and gives an orange precipitate with basic lead acetate. Anthocyanin on the deep yellow gives brown; on the lemon-yellow maroon.

Zinnia elegans shows the anthocyanin-xanthin series, with pale yellow derivative plastids from xanthin. Anthocyanin gives the usual magentas, pinks, crimsons, and orange on the pale and deep yellows.

Cruciferæ.

Brassica sinapis has plastids containing xanthin.

Cheiranthus Cheiri has two yellow varieties, a deep and a pale; the former has plastids containing both carotin and xanthin. It is probable that the pale yellow contains xanthin only, though it has not been tested. Anthocyanin gives the crimson or brown on the deep yellow, and the purple varieties on pale yellow respectively.

Matthiola shows the anthocyanin series with purples, reds, and whites, the latter having colourless plastids; anthocyanin may also exist with plastids containing a "cream" pigment differing from both carotin and xanthin.

Cucurbitaceæ.

Cucurbita Pepo has plastids containing carotin and xanthin.

Fumariaceæ.

Corydalis lutea appears to have plastids containing xanthin and a yellow xantheïc pigment precipitated by basic lead acetate as an orange-yellow precipitate.

Hypericaceæ.

Hypericum Hookerianum has plastids containing xanthin.

Leguminosæ.

Coronilla viminalis has plastids containing carotin and xanthin; some anthocyanin is also present.

Lathyrus odoratus shows the anthocyanin range, purple, red, and white. Also a "cream" plastid pigment similar to that in *Matthiola.*

Spartium junceum has plastids containing carotin (?) and xanthin.

Liliaceæ.

A variety of *Lilium tigrinum* was found to contain plastids and anthocyanin. From the plastids a brick-red pigment was extracted giving a blue colour with sulphuric acid, though no purple was detected; it is probably a form of carotin.

Linaceæ.

Linum flavum has plastids containing carotin and xanthin.

Loasaceæ.

Bartonia aurea has plastids containing carotin and xanthin.

Malvaceæ.

Althæa rosea shows the anthocyanic-xantheïc series. The yellow pigment is intensified in colour by acids and alkalis and is precipitated by basic lead acetate as a brownish-orange precipitate. The ivory gives the yellow colour-reaction. It has not yet been ascertained whether the yellow is recessive to ivory nor whether an albino exists. Anthocyanin gives various purples, mauves, magentas, pinks, crimsons, and orange, according as it is present on an ivory or on a yellow ground.

Nyctaginaceæ.

Mirabilis Jalapa shows the anthocyanic-xantheïc series. The yellow pigment becomes paler with acids and alkalis, and is not precipitated by lead acetate. The white variety gives the yellow colour-reaction, but is recessive to yellow. The red pigment, as already stated, differs from most other forms of anthocyanin.

Onagraceæ.

Œnothera Lamarckiana has plastids containing xanthin and, in addition, a pale yellow xantheïc pigment intensified to orange-yellow by acids and alkalis.

Papaveraceæ.

Argemone grandiflora exists in three varieties, deep and pale yellow and white. The deep yellow is due to plastids containing carotin and xanthin; in addition, a crystalline glucoside, similar to that in *Eschscholtzia,* is present in the sap. The pale yellow appears to contain xanthin and the glucoside, and the white gives the yellow colour-reaction.

Eschscholtzia californica has plastids containing both carotin and xanthin; sometimes the margin or outer half of the petals is yellow or the orange petal is striped with yellow. Examined microscopically the yellow colour of the streaks and margin is seen to be due to yellow plastids, containing, undoubtedly, xanthin only. The orange portions contain orange plastids, carotin being present in addition in these. The sap contains a glucoside (the soluble yellow pigment of Courchet (3)), crystallising in spherules of needles. *E. Caniculata rosea* is cream tinged with pink. Plastids are present containing only a little xanthin; most of the plastid pigment appears to be similar to that in cream *Matthiola.* The glucoside is also

present, though in smaller quantity than in the orange species. The pink tinge is due to anthocyanin.

Glaucium luteum has plastids containing carotin and xanthin.

G. phœniceum, an orange-red species, has anthocyanin in addition.

Papaver nudicaule exists in three varieties, orange, yellow and ivory-white. The pigment appears to be of the xantheïc type, which becomes paler on addition of acids and alkalis, and is not precipitated by lead acetate.

Polemoniaceæ.

Phlox Drummondii has already been considered. The xantheïc pigment is deepened by acids and alkalis and precipitated by basic lead acetate as a deep yellow precipitate. The ivory gives the yellow colour-reaction, and the albino, which does not give this reaction, is recessive to yellow.

Ranunculaceæ.

Ranunculus spp. have plastids containing carotin and xanthin.

Rosaceæ.

Potentilla fruticosa has plastids containing xanthin.

Cream *Rosa* spp. contain a plastid pigment similar to that in cream *Matthiola*.

Scrophulariaceæ.

Antirrhinum has already been fully described.

Calceolaria spp. contain a xantheïc pigment intensified in colour by acids and alkalis, and precipitated by basic lead acetate as an orange-brown precipitate. In some cases plastids containing xanthin are also present.

Varieties of yellow *Nemesia strumosa* have a xantheïc pigment, which becomes paler on treatment by acids and alkalis.

Verbascum Lychnites has a xantheïc pigment giving a blue colour with sulphuric acid, fading to yellow, a yellow colour with alkalis, and a yellow precipitate with basic lead acetate.

Solanaceæ.

Hyoscyamus Chloranthus, a yellow species devoid of purple veining, has plastids containing xanthin.

Physalis Alkekengi has plastids containing carotin and xanthin in the orange calyx.

Salpiglossis grandiflora shows the anthocyanin-plastid series. The deep yellow is due to plastids containing carotin and xanthin; the pale yellow

probably contains xanthin only. Various purples and crimsons are given on the addition of anthocyanin.

Tropæolaceæ.

Tropæolum majus shows the anthocyanin-plastid series. In the pale yellow variety the plastids contain only xanthin, in the deep yellows carotin and xanthin. Anthocyanin may be present in addition at the base of the petals or diffused throughout the flower. Various concentrations of anthocyanin on pale and deep yellow give orange-red, salmon-red, crimson, etc.

T. canariense has plastids containing xanthin only.

T. speciosum contains anthocyanin.

Violaceæ.

Viola tricolor shows the anthocyanin-plastid series. The deep yellow has plastids containing carotin and xanthin, and in addition a crystalline glucoside, similar to that found in *Eschscholtzia*. A paler yellow variety appeared to contain the glucoside only.

Summary of Results.

1. "Anthocyanin," the term used in connection with the red sap-colour in plants, includes several pigments differing as regards their inheritance, the colours to which they give rise in variation, and their behaviour towards chemical reagents.

2. The colours of the varieties arising from an "anthocyanic" type may be regarded as components of the original "anthocyanin"; the type, conversely, may be supposed to lose its components (which are expressible as Mendelian factors) in succession, thus giving rise to a series of colour variations.

3. Broadly speaking, there are two series of colour variations, one containing a "xantheïc" derivative, as, for example, in *Antirrhinum majus*, the other no such derivative, as in *Lathyrus odoratus*.

4. Albinism, in the first series, is a lack of both "anthocyanin" and "xantheïn"; in the second series of "anthocyanin" only.

5. "Xantheïn," a term used in connection with yellow sap-colour, includes several different pigments. This is to be expected if the view that "xantheïn" is a derivative of "anthocyanin" be accepted.

6. There is evidence, as far as investigations have gone, of a correlation between the behaviour of pigments in genetics and their reactions towards chemical reagents.

7. In the case of plastid pigments, the type may contain carotin, xanthin, or both. Varieties arise in some cases from loss of power to

produce carotin, or in others from loss probably of some of the constituents of xanthin.

8. "Anthocyanin" may exist together with plastid pigments in the type, in which case derivative products of both forms of pigmentation are found among the varieties.

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