

chiasmata. Bivalents also sometimes separate completely into 40 monads at metaphase I. Following numerous tests it was discovered that these abnormalities were correlated with the use of 3 X 5 filing cards to support tassels of Zea whose stalks had been incised. In response to inquiries to the manufacturers of the cards as to changes in their manufacturing procedure (since the cards had been previously and often used without effect) we received samples of two different kinds of modified starch used by them for sizing in their cards. Starch from one of these samples (which had been treated with ethylene oxide) consistently produced the abnormalities in several different maize stocks. We have now isolated the active substance in crystalline form in small quantity. It is water soluble and unsuitable for gas chromatograph or mass spectrograph analysis without modification. We are now hoping to obtain the biologically active, modified starch in sufficient supply for conventional chemical analysis and are planning studies of the mechanism of its action.

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1. Pigments in hydrolyzed methanol extracts of maize tissues.

Anthocyanidins.

Seah and Styles (M.G.C.N.L. 43:183-184) reported that seven anthocyanidin spots were present on thin-layer chromatograms of the hydrolysates of pigments extracted from various pigmented Pr strains of W22. Although cyanidin was the predominant pigment, lesser amounts of pelargonidin, peonidin, and four other unknown anthocyanidin-like pigments were also found. One of the unknowns was an orange pigment, and this has now been characterized as luteolinidin (3-deoxycyanidin). The colours, R_f values, and spectral data of the other three pigments and of two other pigments since found are shown in Table 1. Their properties do not match those of any of the previously reported

Table 1. Properties of five anthocyanidin pigments.

Spot No.	Colour		Rf x 100		λ max (m μ) MeOH, HCl	$E_{440}/E_{max.}$ x100	$\Delta\lambda$ (m μ) AlCl ₃ in MeOH
	Visible	U.V.	Forestal	FHW ¹			
5.	Purple	Dull red	74(7) ²	48(2)	275,544 (6)	38 (6)	40 (3)
6.	Purple	Dull red	84(2)	54(2)	265,546 (6)	48 (6)	34 (4)
7.	Purple	Dull red	82(2)	52(1)	274,539 (3)	55 (3)	39 (2)
8.	Purple	Dull red	36(5)	9(1)	275,540 (6)	35 (6)	39 (3)
9.	Purple	Dull red	39(5)		538 (2)	32 (2)	33 (1)

¹Formic acid - conc. HCl - water (5:2:3)

²Figures in parentheses indicate number of chromatograms or spectra from which mean values shown were derived.

anthocyanidins. The fact that they undergo a bathochromic shift in the presence of aluminum ion indicates that they have at least one catechol group in their structure. It is not known whether they exist in the plant as glycosides, or whether they are a product of the conversion of some leuco substance. We have good reason to believe, however, that they are not artifacts.

Luteolinidin (3-deoxycyanidin)

Luteolinidin can be isolated from hydrolyzed extracts of anthers, silks, tassel glumes, and in some cases leaf sheaths of pigmented Pr plants. It can also be found, in approximately the same amount, in r^s b plants with no other detectable anthocyanidins, and in a₂ plants that have only trace amounts of cyanidin resulting from the conversion of leucoanthocyanidin. It is present in the same amounts also in pr plants, even in the anthers where the predominant anthocyanidin is pelargonidin. No apigeninidin (3-deoxypelargonidin) is found in pr anthers as might be expected if the 3-deoxy anthocyanidins were under the control of the Pr gene. From this evidence, therefore, it would seem that the pathway to the 3-deoxyanthocyanidins may be separate from the pathway to the other anthocyanidins. That the two pathways may not be completely independent is shown by the complete absence of all anthocyanidins (3-deoxy anthocyanidins included) in a₁ plants. Reddy's proposed gene action sequence for the pathway to anthocyanin in maize places the A₁ gene after R, but before A₂. This presumably means that the Pr, R, and A₂ actions are required only for the pathway to the common anthocyanins, whereas the A₁ action is required for both pathways.

Probable origin of luteolinidin

Luteolinidin is present in moderate amounts in the hydrolyzed extracts from silks of all genotypes tested except a₁. Chromatography of unhydrolyzed silk extracts does not yield an orange spot as expected from a luteolinidin glycoside, but a test for the presence of luteoforol (leucoluteolinidin) is positive. Thus it appears that the silks, and probably other tissues also, contain luteoforol which converts to luteolinidin upon hydrolysis of the methanol extracts.

Probable origin of bz₁ pigment

Hydrolyzed extracts of bz₁ silks yield a relatively large luteolinidin spot, but no other anthocyanidins. When bz₁ silks are extracted in 1% HCl in MeOH prior to their emergence from the leaf sheath, the extract is light green and tests positive for luteoforol. After the extract has been in the refrigerator for a period of time (two or three weeks), it becomes orange-brown. When chromatographed, a brown pigment is obtained that behaves in the same manner as the brown pigment which is obtained by extracting bronze coloured tissues of mature bz₁ plants. It seems probable, therefore, that the brown coloured bz₁ pigment is a phlobaphene formed largely if not solely from luteoforol.

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2. Repression of anthocyanin pigmentation in young seedlings by Pl.

We have repeatedly observed that when our W22 r^g B pl and r^g B Pl strains are grown together under the same conditions, the r^g B pl seedlings are always the first to become pigmented. At the second or third internode stages, plants of the two genotypes are virtually indistinguishable, and it is not until the fourth or fifth internode stages that the r^g B Pl strains are clearly darker than the r^g B pl. We have recently compared O.D. readings of extracts from several Pl and pl strains, and have found that Pl not only represses B pigment in the seedlings, but also pigment conditioned by an R^r factor (specifically, R^r Ecuador 1172). The effect is most marked at the first internode stage, and becomes progressively less as the seedling matures. In one experiment, measuring only the first internode plus the ligule region of the first leaf, significantly lower O.D. readings were obtained from r^g B Pl plants as compared to r^g B pl plants in samples taken every day for a period of 10 days. In another group of seedlings at the early first leaf stage, Ecuador R^r B^b pl seedlings were strongly pigmented in contrast to Ecuador R^r B^b Pl seedlings, which had little or no pigment and were indistinguishable from r^g B^b Pl and r^g B^b pl plants of the same stage. The Pl seedlings did develop pigment later, but differences were still measurable even at the