

After-Cooking Discoloration of Potatoes. Iron Content in Relation to Blackening Tendency of Tissue^a

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SUMMARY

Data are presented on the iron contents of stem and bud end, whole and deproteinized, extracts of 41 samples of potatoes representing various degrees of discoloration. The stem end generally contained more iron than the bud end. Statistical analysis of all samples treated as one group revealed a highly significant correlation (1% level) between increasing iron content and increasing degree of after-cooking discoloration. Of the two types of iron studied, "free iron" and "protein iron," the protein iron gave the higher degree of correlation with blackening.

The highly significant correlation between iron values and degree of discoloration was lost in about half the cases when subgroups of the samples were formed according to location grown, crop year, and variety.

The data revealed that the percentage of total iron associated with the protein is higher in the stem end of the potato. Also, the stem-end protein contained considerably more iron than the bud-end protein. The difference in iron content between stem-end protein and bud-end protein showed a highly significant correlation (1% level) with tendency to blacken.

INTRODUCTION AND LITERATURE REVIEW

The after-cooking discoloration problem in potatoes, and its importance, have been reported (Yanovsky, 1955; Hunter *et al.*, 1957; Heisler *et al.*, 1962). It is now generally accepted that the discoloration is due to the formation of a dark-colored complex of ferric iron and an orthodihydric phenol, probably chlorogenic acid, since this is the major compound of this class in the potato (Juil, 1949; Kiermeier and Rickerl, 1955a). Many investigators have attempted to reduce or eliminate after-cooking blackening with metal-chelating agents (Hawkins *et al.*, 1959; Hunsader and Hanning, 1958; Greig and Smith, 1955, 1960; Smith and Davis, 1962). The iron-chlorogenic acid theory of blackening is supported by the success of some of those experiments. Published analytical data

on iron content, however, are contradictory. Robison (1941), working with an acidic extract, found that iron content and after-cooking discoloration were correlated in tubers drawn from the same sample but not in tubers drawn from different samples. Muneta (1959) analyzed an aqueous extract of cooked potato, and found correlation between iron content and blackening. Other workers (Tottingham, 1939; Juil, 1949) found little or no correlation between these two factors. These apparent contradictions are explained by the presence of modifying factors such as pH, citric acid, and phosphoric acid. Hughes and Swain (1962b) studied the effect of citric, orthophosphoric, and malic acids, and of pH on the color of various phenol-iron complexes and concluded that citric acid was the most important of these factors in reducing the intensity of color of the chlorogenic acid-iron complex. They also correlated blackening with the ratio of citric acid to chlorogenic acid: the

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lower the citric acid content the more intense the blackening. This result is supported by potassium data of Heisler *et al.* (1962) in which a correlation was established between low potassium content and tendency to blacken, since it is reasonable to assume that potassium content would be directly proportional to citrate content.

Thus, it can be seen that the iron-chlorogenic acid theory is fairly well established. In an attempt to establish iron further as a prime factor in the blackening mechanism, we have determined the iron content of the stem and bud ends of a large number of samples representing a wide range of discoloration, and made a statistical study of the data. An important consideration in the following treatment of the data is the separation of the total iron into "free iron" and "protein iron."

METHODS

Potatoes. The 41 samples of potatoes used were sent to us as blackening samples from various parts of the country over a three-year period. Most were received from Wisconsin (19) and Michigan (15), with the remainder coming from Pennsylvania (3), Maine (3), and Long Island (1).

Experimental details for obtaining the sample and determining the degree of discoloration were as presented previously (Heisler *et al.*, 1962). For convenience, a brief outline is given here.

Potato sampling. Longitudinal plugs were taken from the stem- and bud-end sections of thoroughly washed and scrubbed potatoes with a no. 15 cork borer. The plugs were cut to a length of approximately $\frac{1}{2}$ in. and then cut in half lengthwise. One half was used for the iron determination, and the other for measurement of degree of discoloration by a reflectance test. The half-cylinders used for the iron determination were adjusted in length so that 26 pieces totaled 100.00 g. Those used for the reflectance measurement were adjusted to total 65.00 g.

Determination of discoloration. Taken as the measure of discoloration was the reflectance obtained from a smooth surface (under glass) of cooked mashed potato. The reflectance attachment to the Beckman model B spectrophotometer (no endorsement implied) was used with $MgCO_3$ as standard. In this study, "degree of discoloration" was arbitrarily taken as $(R_B - R_S)/R_S$, where R_B is the reflectance of mash from the bud end, and R_S represents that for the stem end. The more discoloration in the potato, the greater the

difference between R_B and R_S and the lower the value of R_S . Thus, these factors reinforce each other to amplify the value for degree of discoloration, making the system more sensitive in differentiating samples.

Preparation of extracts. The 100.00-g sample of potato tissue (26 half plugs from stem or bud end) was ground for 2 min in 300 ml of iron-free water in a Waring blender (no endorsement implied). The slurry was filtered through Whatman no. 12 paper, the filtrate collected, and the volume measured. Half of it was bottled and set aside. This is referred to herein as "whole extract." The other half was immersed 5 min in a boiling-water bath to coagulate the protein, and then filtered, while hot, through Whatman no. 12 paper. Loss of vapor during heating and filtering was kept to a minimum by stoppering the flask with a ground-glass stopper and by covering the funnel with a watch glass. The filtrate, containing about 1% solids, was labeled "deproteinized extract." The iron contents of both the whole and deproteinized extract were determined.

The resulting samples of whole and deproteinized extracts, though prepared by the same procedure, varied slightly in solids content because of differences inherent in the various lots of potatoes. The exact solids content of each extract was determined by loss of weight after drying. Iron was determined as the quantity present in the whole or deproteinized extract, but it can be related, percentagewise, to the fresh potato weight or to the solids content of the juice.

Although it is recognized that the above procedure is not an exhaustive extraction it was believed sufficient for the purpose of determining differences in the iron content of various samples and between the stem and bud ends of the same sample.

Iron determination. The method of Collins and Diehl (1959) for determining iron in urine was used. This method uses the very sensitive iron reagent, 4,7-diphenyl-1,10-phenanthroline (batho-phenanthroline), a wet digestion with perchloric acid, and an extraction of the colored complex with nitrobenzene. As little as 2 ml of the 1% solids extract can be used. The conditions for the wet digestion had to be changed somewhat from those outlined by Collins and Diehl to accommodate the potato sample.

Two to 6 ml of the whole or deproteinized extract, the amount depending on the concentration of iron expected, was transferred to a 30-ml Kjeldahl flask. The volume was made up to 6 ml with iron-free water, and 1.25 ml concentrated HNO_3 and 0.50 ml of 70% perchloric acid were added. The mixture was digested at low

Table 1. Iron and protein values of potatoes and their relation to after-cooking discoloration.

Sample	Variety	$\frac{R_p-R_s}{R_s}$	Iron in micrograms/ml of extract ^b						Iron assoc. with Protein (%) ^d						Protein (% in extract ^b) ^e						Iron based on Protein (%) ^f					
			Total iron ^a			Free iron ^b			Protein iron ^c			Iron assoc. with Protein (%) ^d			Protein (% in extract ^b) ^e			Iron based on Protein (%) ^f								
			Stem	Bud	Stem-Bud	Stem	Bud	Stem-Bud	Stem	Bud	Stem-Bud	Stem	Bud	Stem-Bud	Stem	Bud	Stem-Bud	Stem	Bud	Stem-Bud	Stem	Bud	Stem-Bud			
59-23	Wisc. Antigo	0.697	1.91	1.19	0.72	1.04	1.01	0.03	0.87	0.17	0.70	45.5	14.3	31.2	0.18	0.23	-0.05	.048	.007	.041						
59-24	Wisc. Red Lasoda	0.695	2.83	1.01	1.82	1.15	0.71	0.44	1.68	0.30	1.38	59.4	30.7	28.7	0.15	0.18	-0.03	.112	.017	.085						
59-19	Wisc. Katahdin	0.623	2.43	1.26	1.17	0.75	0.68	0.07	1.60	0.57	1.03	68.1	46.1	22.0	0.13	0.15	-0.02	.123	.041	.082						
59-20	Wisc. Kennebec	0.572	2.84	1.09	1.75	1.01	0.56	0.45	1.83	0.53	1.30	64.4	49.1	15.3	0.21	0.24	-0.03	.087	.023	.064						
59-25	Wisc. Red Lasoda	0.522	1.51	0.81	0.70	1.09	0.55	0.54	0.42	0.26	0.16	27.8	32.1	-4.3	0.15	0.18	-0.03	.028	.014	.014						
59-15	Wisc. Early Gem	0.519	1.66	1.21	0.45	0.41	0.34	0.07	1.25	0.87	0.38	75.3	71.4	3.9	0.14	0.19	-0.05	.089	.046	.043						
61-1	Mich. Ontario	0.516	0.33	0.32	0.01	0.05	0.13	-0.08	0.28	0.19	0.09	84.8	60.0	24.8	0.13	0.11	0.02	.022	.017	.005						
61-3	Mich. Huron	0.503	1.27	0.51	0.76	0.80	0.34	0.46	0.47	0.17	0.30	37.1	33.1	4.0	0.13	0.15	-0.02	.036	.011	.025						
59-22	Wisc. Antigo	0.497	1.58	1.12	0.46	0.84	0.86	-0.02	0.74	0.30	0.44	46.8	24.1	22.7	0.15	0.19	-0.04	.049	.014	.035						
59-18	Wisc. Katahdin	0.475	3.03	1.49	1.54	1.28	0.84	0.44	1.71	0.63	1.08	57.2	42.9	14.3	0.22	0.22	0	.078	.031	.047						
60-1	Pa. Merrimack	0.469	1.10	0.66	0.44	0.19	0.17	0.02	0.91	0.48	0.43	81.8	69.2	12.6	0.19	0.23	-0.04	.047	.020	.027						
60-2	Pa. Merrimack	0.449	0.95	0.54	0.41	0.20	0.22	-0.02	0.75	0.32	0.43	78.6	59.2	19.4	0.22	0.28	-0.06	.034	.011	.023						
61-2	Mich. Ontario	0.434	0.47	0.39	0.08	0.12	0.21	-0.09	0.35	0.18	0.17	74.2	45.7	28.5	0.10	0.10	0	.035	.018	.017						
60-3	Pa. Merrimack	0.421	1.43	0.74	0.69	0.15	0.19	-0.04	1.28	0.55	0.73	89.7	74.3	15.4	0.25	0.28	-0.03	.051	.020	.031						
60-5	Wisc. Antigo	0.405	0.97	0.60	0.37	0.24	0.29	-0.05	0.73	0.31	0.42	75.5	52.0	23.5	0.12	0.12	0	.061	.025	.036						
61-5	Wisc. Kennebec	0.396	2.23	0.69	1.54	1.84	0.28	1.56	0.39	0.42	-0.03	17.6	60.0	-42.4	0.23	0.15	0.08	.017	.028	-.011						
60-4	Me. Katahdin	0.332	0.99	0.49	0.50	0.45	0.33	0.12	0.54	0.16	0.38	54.8	32.3	22.5	0.21	0.18	0.03	.027	.009	.018						
60-7	Wisc. Katahdin	0.332	0.72	0.59	0.13	0.38	0.27	0.11	0.34	0.32	0.02	47.0	54.3	-7.3	0.12	0.10	0.02	.027	.031	-.004						
59-16	Wisc. Early Gem	0.322	1.41	0.73	0.68	0.52	0.23	0.29	0.89	0.50	0.39	63.1	68.5	-5.4	0.16	0.16	0	.056	.031	.025						
59-13	Mich. Huron	0.306	1.19	1.04	0.15	0.76	0.56	0.20	0.43	0.48	-0.05	36.1	45.6	-9.5	0.20	0.24	-0.04	.022	.020	.002						
61-6	Wisc. Katahdin	0.269	1.08	0.49	0.59	0.83	0.24	0.59	0.24	0.25	-0.01	22.6	51.0	-28.4	0.06	0.04	0.02	.040	.063	-.023						

Table 1. Iron and protein values of potatoes and their relation to after-cooking discoloration (concluded).

Sample	Variety	Iron in micrograms/ml of extract ^b												Iron assoc. with Protein (%) ^d						Protein (% in extract ^b) ^e						Iron based on Protein (%) ^f													
		R_B, R_S^a			Total iron ^a			Free iron ^b			Protein iron ^c			Stem			Bud			Stem			Bud			Stem			Bud										
		Stem	Bud	Stem-Bud	Stem	Bud	Stem-Bud	Stem	Bud	Stem-Bud	Stem	Bud	Stem-Bud	Stem	Bud	Stem-Bud	Stem	Bud	Stem-Bud	Stem	Bud	Stem-Bud	Stem	Bud	Stem-Bud	Stem	Bud	Stem-Bud											
59-17	Wisc. Ontario	0.259	1.22	1.26	-0.04	0.61	0.72	-0.11	0.61	0.53	0.08	50.0	42.1	7.9	0.10	0.15	-0.05	0.61	0.35	0.26	0.259	1.22	1.26	-0.04	0.61	0.72	-0.11	0.61	0.53	0.08	50.0	42.1	7.9	0.10	0.15	-0.05	0.61	0.35	0.26
61-7	Me. Kennebec	0.256	0.83	0.46	0.37	0.20	0.14	0.06	0.63	0.32	0.31	75.8	68.7	7.1	0.22	0.16	0.06	0.28	0.20	0.08	0.256	0.83	0.46	0.37	0.20	0.14	0.06	0.63	0.32	0.31	75.8	68.7	7.1	0.22	0.16	0.06	0.28	0.20	0.08
60-6	Wisc. Red Lasoda	0.239	1.13	0.42	0.71	0.50	0.25	0.25	0.63	0.17	0.46	56.0	41.4	14.6	0.15	0.09	0.06	0.43	0.19	0.24	0.239	1.13	0.42	0.71	0.50	0.25	0.25	0.63	0.17	0.46	56.0	41.4	14.6	0.15	0.09	0.06	0.43	0.19	0.24
60-16	Mich. Huron	0.235	0.99	0.69	0.30	0.22	0.32	-0.10	0.77	0.37	0.40	77.9	53.4	24.5	0.20	0.19	0.01	0.38	0.20	0.18	0.235	0.99	0.69	0.30	0.22	0.32	-0.10	0.77	0.37	0.40	77.9	53.4	24.5	0.20	0.19	0.01	0.38	0.20	0.18
60-11	Mich. Ontario	0.204	0.75	0.62	0.13	0.09	0.22	-0.13	0.66	0.40	0.26	88.0	64.7	23.3	0.15	0.06	0.09	0.45	0.65	0.020	0.204	0.75	0.62	0.13	0.09	0.22	-0.13	0.66	0.40	0.26	88.0	64.7	23.3	0.15	0.06	0.09	0.45	0.65	0.020
60-12	Mich. Ontario	0.183	0.67	0.78	-0.11	0.20	0.27	0.07	0.48	0.52	-0.04	70.8	66.1	4.7	0.10	0.08	0.02	0.49	0.63	0.014	0.183	0.67	0.78	-0.11	0.20	0.27	0.07	0.48	0.52	-0.04	70.8	66.1	4.7	0.10	0.08	0.02	0.49	0.63	0.014
61-4	L. I. Katahdin	0.162	0.59	0.47	0.12	0.25	0.34	-0.09	0.34	0.13	0.21	58.2	26.8	31.4	0.14	0.15	-0.01	0.25	0.08	0.017	0.162	0.59	0.47	0.12	0.25	0.34	-0.09	0.34	0.13	0.21	58.2	26.8	31.4	0.14	0.15	-0.01	0.25	0.08	0.017
59-14	Mich. Cherokee	0.150	1.10	1.00	0.10	0.53	0.56	-0.03	0.57	0.44	0.13	51.8	43.4	8.4	0.25	0.33	-0.08	0.23	0.13	0.10	0.150	1.10	1.00	0.10	0.53	0.56	-0.03	0.57	0.44	0.13	51.8	43.4	8.4	0.25	0.33	-0.08	0.23	0.13	0.10
60-9	Wisc. Kennebec	0.134	1.53	0.56	0.97	0.85	0.24	0.61	0.67	0.32	0.35	44.1	57.9	-13.8	0.16	0.11	-0.05	0.43	0.29	0.14	0.134	1.53	0.56	0.97	0.85	0.24	0.61	0.67	0.32	0.35	44.1	57.9	-13.8	0.16	0.11	-0.05	0.43	0.29	0.14
60-10	Wisc. Ontario	0.128	0.89	0.80	0.09	0.33	0.35	-0.02	0.56	0.45	0.11	62.7	56.6	6.1	0.07	0.09	-0.02	0.76	0.48	0.28	0.128	0.89	0.80	0.09	0.33	0.35	-0.02	0.56	0.45	0.11	62.7	56.6	6.1	0.07	0.09	-0.02	0.76	0.48	0.28
60-13	Mich. Ontario	0.124	0.90	0.68	0.22	0.23	0.29	-0.06	0.67	0.39	0.28	74.2	56.8	7.4	0.12	0.10	0.02	0.57	0.40	0.17	0.124	0.90	0.68	0.22	0.23	0.29	-0.06	0.67	0.39	0.28	74.2	56.8	7.4	0.12	0.10	0.02	0.57	0.40	0.17
60-19	Mich. Ontario	0.095	0.87	0.65	0.22	0.18	0.28	-0.10	0.69	0.37	0.32	79.8	56.7	23.1	0.11	0.09	0.02	0.62	0.40	0.22	0.095	0.87	0.65	0.22	0.18	0.28	-0.10	0.69	0.37	0.32	79.8	56.7	23.1	0.11	0.09	0.02	0.62	0.40	0.22
59-12	Mich. Manota	0.069	0.74	0.75	-0.01	0.30	0.49	-0.19	0.44	0.26	0.18	59.5	44.7	14.8	0.16	0.21	-0.05	0.28	0.16	0.16	0.069	0.74	0.75	-0.01	0.30	0.49	-0.19	0.44	0.26	0.18	59.5	44.7	14.8	0.16	0.21	-0.05	0.28	0.16	0.16
60-8	Wisc. Early Gem	0.052	0.81	0.53	0.28	0.25	0.13	0.12	0.56	0.40	0.16	69.7	76.1	-6.6	0.06	0.11	-0.05	0.88	0.36	0.52	0.052	0.81	0.53	0.28	0.25	0.13	0.12	0.56	0.40	0.16	69.7	76.1	-6.6	0.06	0.11	-0.05	0.88	0.36	0.52
61-8	Me. Katahdin	0.052	0.77	0.52	0.25	0.26	0.19	0.07	0.51	0.33	0.18	69.3	63.3	6.0	0.05	0.05	0	0.64	0.41	0.23	0.052	0.77	0.52	0.25	0.26	0.19	0.07	0.51	0.33	0.18	69.3	63.3	6.0	0.05	0.05	0	0.64	0.41	0.23
60-14	Mich. ?	0.015	0.77	0.44	0.33	0.38	0.26	0.12	0.39	0.18	0.21	50.7	41.8	8.9	0.13	0.10	0.03	0.31	0.18	0.13	0.015	0.77	0.44	0.33	0.38	0.26	0.12	0.39	0.18	0.21	50.7	41.8	8.9	0.13	0.10	0.03	0.31	0.18	0.13
60-17	Mich. Cherokee	0.010	1.36	0.89	0.47	0.24	0.43	-0.19	1.13	0.47	0.66	82.6	52.2	30.4	0.20	0.22	-0.02	0.56	0.21	0.35	0.010	1.36	0.89	0.47	0.24	0.43	-0.19	1.13	0.47	0.66	82.6	52.2	30.4	0.20	0.22	-0.02	0.56	0.21	0.35
60-18	Mich. Russet Rural	0	0.74	0.56	0.18	0.16	0.21	-0.05	0.58	0.36	0.22	78.5	63.5	15.0	0.11	0.16	-0.05	0.54	0.23	0.31	0	0.74	0.56	0.18	0.16	0.21	-0.05	0.58	0.36	0.22	78.5	63.5	15.0	0.11	0.16	-0.05	0.54	0.23	0.31
59-21	Wisc. Kennebec	0	1.44	0.82	0.62	0.33	0.38	-0.05	1.11	0.43	0.68	77.1	52.4	24.7	0.20	0.24	-0.04	0.56	0.18	0.38	0	1.44	0.82	0.62	0.33	0.38	-0.05	1.11	0.43	0.68	77.1	52.4	24.7	0.20	0.24	-0.04	0.56	0.18	0.38
60-15	Mich. Russet Rural	0	0.47	0.55	-0.08	0.17	0.24	-0.07	0.30	0.31	-0.01	64.7	55.9	8.8	0.10	0.09	0.01	0.31	0.034	0.003	0	0.47	0.55	-0.08	0.17	0.24	-0.07	0.30	0.31	-0.01	64.7	55.9	8.8	0.10	0.09	0.01	0.31	0.034	0.003

^a "Degree of discoloration" where R_B is reflectance of cooked bud tissue and R_S reflectance of stem tissue.

^b Extract contains approximately 1% total solids.

heat until the appearance of white (perchloric) fumes; then it was heated for 10 more min and cooled. The sides of the flask were washed with iron-free water, then heat was applied (to boiling) to dissolve the precipitate and remove Cl_2 . Contents of the flask were then transferred while still hot to a 125-ml separatory funnel, using about 8 ml of iron-free wash water. Two ml of 10% $\text{NH}_2\text{OH}\cdot\text{HCl}$ (aqueous solution, iron-free) and 5 ml of bathophenanthroline (0.001M in 50% ethanol, iron-free) were added. A piece of congo-red indicator paper was placed in the funnel, and NH_4OH (iron-free) was added dropwise until the paper turned red. Five ml of buffer (4M NaAc-4M HAc) and 4 ml nitrobenzene were pipetted into the funnel and shaken vigorously. The nitrobenzene layer (lower) was collected in a 10-ml volumetric flask. Extraction was continued with 2 more portions (2 ml each) of nitrobenzene, and volume was made to mark with absolute ethanol. The colored solution was transferred to a spectrophotometer tube and read at 538 $\text{m}\mu$. Iron content was obtained from a standard curve made using 2-6 ml of a standard ferrous iron solution (1.00 $\mu\text{g}/\text{ml}$).

RESULTS AND DISCUSSION

Forty-one samples, representing a wide range of after-cooking discoloration, were analyzed for iron content. The stem- and bud-end samples of whole and deproteinized extracts were studied. Table 1 lists the samples in order of decreasing degree of discoloration, giving the iron content of the whole extract (total Fe) and the deproteinized extract (free Fe), and, by difference, the protein Fe. The stem end generally contains more iron than the bud end. For example, the stem end is higher in 37 of the 41 samples of whole extract, and in 36 of the samples when considering the protein iron. With the deproteinized extract, however, there is no clear-cut differentiation of stem and bud ends in iron content. Since the blackening occurs only at the stem end, the above facts indicate that the protein iron is more likely involved in the after-cooking blackening than the free iron.

Another interesting aspect of the data is that the range of values is greater for the stem end than for the bud end, i.e., there is more variation in the stem-end iron content between samples. This fact is illustrated in Table 2.

Since only the stem end blackens, the bud

Table 2. Range of iron values ($\mu\text{g}/\text{ml}$).

	Stem	Bud
Total Fe	0.33-3.03	0.32-1.49
Free Fe	0.05-1.84	0.13-1.01
Protein Fe	0.24-1.83	0.13-0.87

end of each sample can be considered a control and the iron value for the difference between the two should be related to the tendency to blacken. For this reason this value is included in Table 1.

A statistical study was made of the iron data in Table 1 by the linear regression method, and an analysis of variance test determined that the relation of degree of discoloration to iron content was highly significant (1% level) in 7 of the 9 cases tested. Table 3 summarizes the results.

Table 3. Summary of statistical study.

	"F" values		
	Stem	Bud	Stem minus bud
Total Fe	20.5**	7.4**	17.8**
Free Fe	13.9**	10.5**	3.9
Protein Fe	9.6**	0.04	12.7**
Fe assoc. with protein	1.4	1.7	<1
Protein Fe content	3.0	1.6	2.0
of protein	2.6	2.3	7.7**

** Significant at 1% level.

Again, considering the fact that the stem end blackens and the bud end does not, correlation of tendency to blacken should be obtained with stem-end and stem-end minus bud-end iron values. This situation exists for the protein iron. It is not surprising, however, that this ideal relation is not obtained in every case, since, as stated in the introduction, other factors are involved in the blackening mechanism.

The significant correlation between iron values and degree of discoloration was lost in some cases when subgroups of the samples were formed according to location grown, crop year, and variety. Table 4 summarizes the results of this statistical analysis of the subgroups. This same situation was encountered when studying the potassium data (Heisler *et al.*, 1962) and raised

Table 4. Statistical analysis of subgroups.

Subgroups	Correlation of stem end total Fe values with discoloration	
	Not significant	Significant
Crop year		
1959		X
1960	X	
1961	X	
Location Grown		
Wisconsin		X
Michigan	X	
Variety		
Antigo		X ^a
Red LaSoda		X ^a
Early Gem		X ^a
Ontario	X	
Katahdin		X
Kennebec	X	
Huron		X ^a
Merrimac	X ^a	

^a By observation (not enough samples to warrant a statistical analysis).

a question regarding the validity of the significance found between discoloration and iron content for the data considered as a whole. However, when the complexity of the blackening mechanism is considered, it is not surprising that significant correlation is not obtained in every case. Hughes and Swain (1962a) stated that the relative importance of any one factor probably varies even from potato to potato.

An interesting fact brought out by the data was the relatively large percentage of iron associated with the protein. Immediately the question arose: Is the iron precipitated as a part of the protein molecule or simply removed by occlusion along with the protein? Levitt and Todd (1952) reported that as much as one-third of the total iron was associated with the protein, and their data indicated that the iron was chemically bound to the protein. Dialysis experiments at our laboratory also point to a chemical combination of the iron and the protein. The percentage of total iron associated with the protein is listed in Table 1. It is readily seen that the percentage of iron associated with the protein is considerably higher in the stem end than in the bud end. This is true in 33 of the 41 samples. In an attempt to find correlation between degree of blackening and the percentage of iron

associated with the protein, a linear regression was fitted to the data. An analysis of variance test, however, determined that this relationship was not significant (see Table 3).

Since such a large percentage of the iron is associated with the protein it was thought advisable to study the protein values. A rough estimation of the amount of protein present was obtained by taking the difference in the soluble solids content of the extract before and after precipitation of the protein. The protein values obtained in this manner are listed in Table 1. There was no significant difference in the protein contents of the stem and bud ends, and a statistical study of the data showed no significant correlation with blackening (see Table 3). Going a step farther, the percentage of iron based on protein was calculated (Table 1). These data demonstrate that the stem-end protein contains a considerably higher percentage of iron than the bud-end protein. This is true in 35 of the 41 samples. A statistical study of the data showed no significant correlation with tendency to blacken when considering the stem- or bud-end values. However, a highly significant correlation (1% level) was obtained with the stem-end minus bud-end difference values (see Table 3).

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