LXXXV. A STUDY OF THE HYDROGEN ION CONCENTRATION OF THE POTATO TUBER.

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NUMEROUS investigations have been made on the acidity of plant sap and several methods have been employed. The hydrogen electrode has been used extensively but the time required for the system to reach equilibrium, the alteration of the biological fluid by the passage of hydrogen and the poisoning of the electrode by the plant products are obvious disadvantages of the method. Colorimetric methods are not entirely satisfactory on account of the rapid colour changes which may take place in the sap as a result of enzyme action. With the quinhydrone electrode, the precision of which has been critically examined by Morgan, Lammert and Campbell [1931], equilibrium is attained very rapidly and manipulation is simple. Plant oxidases, however, attack quinol [Chodat, 1910] and, consequently, the ratio quinol:quinone is liable to be altered. In this paper, the results obtained by rapid measurement of the acidity of tuber tissue with a special quinhydrone capillary electrode are described and compared with those obtained by other methods.

METHODS.

(a) *Extraction of sap.* When it was desired to examine the sap, the tubers were washed in cold water and passed through a pulping machine. The mash was filtered through fine linen and the filtrate, which gave essentially the same results as the pulp, was generally employed. The pulped tubers decomposed rapidly so that the sap usually had a red-brown colour which became darker on standing.

(b) *Hydrogen electrode measurements.* A Hildebrand half-cell was used in conjunction with a saturated calomel electrode. About 5 cc. of the plant sap were taken for each determination and equilibrium was reached after approximately 10 minutes. The hydrogen electrode was given a fresh deposit of platinum black after each measurement.

(c) *Quinhydrone electrode measurements.* The quinhydrone electrode consisted of a piece of polished platinum foil which was placed in the potato sap saturated with quinhydrone. Some preliminary experiments showed that at least 0.12 g. of quinhydrone had to be added to each 10 cc. of sap in order to

obtain reproducible results. The procedure adopted was to shake the sap with sufficient quinhydrone for a few seconds, complete the potentiometer circuit with the platinum and saturated calomel electrodes and read the E.M.F. of the cell at once. The whole operation was carried out in less than 1 minute but decomposition of the phenol had already commenced. The platinum foil was washed and heated to redness in an alcohol flame after each measurement and from time to time was also cleaned in hot chromic acid solution.

A comparison of results obtained by methods (b) and (c), included in Table III, shows that in most cases the hydrogen electrode yields slightly higher p_H values. This may be due to some alteration in the sap during the passage of hydrogen or to some error in the application of the quinhydrone half-cell. The fact that the sap, if allowed to stand for about 10 minutes before the addition of the quinhydrone, gave approximately the same value as with the hydrogen electrode, suggests that the time required by the system to reach equilibrium is the important factor.

(d) *Micro-electrode measurements.* Reproducible results were readily obtained with the quinhydrone electrode, but the use of pulp in any method removes the possibility of studying the variation in acidity throughout the tuber. To meet this difficulty a special electrode was constructed [Robertson and Smith, 1930], which enabled measurements to be made at any desired point on a section of the plant tissue. A few results obtained by means of this electrode and by the method described under (c) are given in Table IV. The p_H of the sap was always greater than that observed with the micro-electrode for the tissue, which was probably due to loss of carbon dioxide in the preparation of the pulp. Ingold [1929] observed a reduction of 0.3 p_H unit when the tuber sap was brought into equilibrium with concentrations of carbon dioxide such as have been recorded for the intercellular spaces of the tuber. In addition, the oxidase activity of the disintegrated tissue is very great and the formation of melanin and its intermediate compounds from tyrosine [Raper, 1928] may also reduce the acidity.

Variation in acidity throughout the tuber.

Although some data are available on the hydrogen ion concentration gradients of the sap of plants [Gustafson, 1924], no such observations seem to have been made on the potato tuber. Attention was directed to a study of the changes which take place during the formation, storage and sprouting of the tuber.

(a) *During growth period.* A number of healthy and typical plants was selected and examined at intervals throughout the summer. The tubers were cut along a plane through an eye and the heel. Readings were taken with the micro-electrode at points in the middle and 1–2 mm. below the eye and heel. The results for two varieties given in Table I are typical of a large number of observations and each figure is an average value for four tubers.

Table I. *Variation of tuber p_H with stage of growth.*

Variety	Date of sampling ...	30. vi. 30	21. vii. 30	16. viii. 30	30. ix. 30
Duke of York	"Rose" end	6.19	6.04	5.90	5.65
	Centre	6.20	6.08	5.75	5.71
	"Heel" end	5.53	5.90	5.85	5.73
Epicure	"Rose" end	6.08	6.13	5.81	5.68
	Centre	6.06	6.09	5.88	5.75
	"Heel" end	5.78	5.82	5.80	5.80

The first readings were made with tubers weighing 5–15 g. and it was observed that the underground stem attaching the tuber to the parent plant had approximately the same p_H as the heel of the tuber. The final sampling was made after the plants were completely ripe, the haulms having withered away some time previously. The results show that all parts of the tuber, except the heel, become more acid as the plant ripens. The p_H of the heel rises quickly and then falls off gradually. There is a p_H gradient across the young tuber, the heel being more acid than the rose end. In the mature tuber the reverse holds good. This change coincides with the physiological development of the tuber, for in the early stages of growth the active part is at the heel where nutrients are received from the plant, whereas in the mature state this process has ceased and the eyes develop rapidly.

(b) *During rest period.* A large number of tubers was examined between the time of harvesting and the appearance of sprouts. Observations were made at eleven points on a section of each tuber, both cortex and medulla being represented. In the majority of cases the rose end was slightly more acid than the heel end, the centre being intermediate. The values given in Table I for the last sampling are typical in this respect. In many tubers, however, no difference was noted between the rose and heel ends, while in a few isolated cases the heel was slightly more acid than the rose end. Generally, the average value for all the points approximated closely to that for the centre of the tuber.

(c) *During sprouting period.* In March a number of dormant tubers was exposed to daylight and allowed to sprout. At certain intervals a few tubers were cut down through a sprout and eye to the heel and the acidity was determined at a number of positions across the section. Tubers of nine varieties were examined and all yielded similar results. In Table II the average values for three tubers of the variety Duke of York are given.

Table II. *Variation of p_H during sprout formation.*

Position on tuber or sprout	20. iii. 30	16. iv. 30	15. v. 30	24. vi. 30
Growing tip of sprout	4.28	4.30	4.38	4.51
Middle of sprout	4.45	4.55	4.62	4.90
Tuber end of sprout	4.25	4.36	4.54	5.09
Surface of "eye"	4.58	4.73	5.70	5.85
2 mm. below surface of "eye"	5.31	5.60	5.86	—
5 mm. below surface of "eye"	5.61	5.67	5.83	—
10 mm. below surface of "eye"	5.75	5.74	5.86	—
Middle of tuber	5.74	5.71	5.78	5.80
"Heel" end of tuber	5.80	5.85	5.86	5.88

The following is a description of the sprouts:

20. iii. 30.—Sprouts soft and light green, and about 0.5 cm. long.

16. iv. 30.—Sprouts hard and dark green, 0.5–1.5 cm. long.

15. v. 30.—Sprouts thicker, 2.0–2.5 cm. long, small leaves forming.

24. vi. 30.—Sprouts very thick and hard, 3 cm. long, small leaves and shoots formed. Tuber soft.

When sprouts appear, the region round the active eyes is very acid, but gradually becomes less acid as the sprouting progresses until June, when the p_H is practically uniform throughout the tuber. The growing tip of the sprout retains its relatively high acidity throughout the growth of the plant; in a young field plant it had p_H 4.46 while in an older plant the p_H was 4.65.

These variations in p_H through the tuber are to be expected if consideration is given to the different functions of the various parts. The actively growing parts are usually more acid than the fully developed members. On the other hand, the acidity of the tuber increases with maturity, which may be accounted for by the fact that, according to Appleman and Miller [1926], the protein in the young tuber gives place to non-protein- and amino-nitrogen in the mature tuber.

Factors influencing the p_H value of the mature tuber.

(a) *Environment.* Tubers of six common varieties, grown at seven places in Midlothian, were examined immediately after harvesting. The altitudes of the different places varied from 100 to 600 feet above sea level; the different soils varied in texture from sandy loams to clay loams and in p_H from 4.9 to 7.3. There was no obvious relationship, however, between environment and the acidity of the tuber. The maximum variations of individual values for all sources were as follows: Duke of York, 5.70–5.89; Epicure, 5.70–5.85; Great Scot, 5.65–6.11; Golden Wonder, 5.70–5.85; Majestic, 5.50–5.73; Ally, 5.67–5.84.

In order to investigate more fully the effect of soil reaction upon the acidity of the tuber, a crop of variety King George, grown on a series of specially treated plots on a soil of p_H 4.0, was examined. The p_H of the tubers was measured by methods (b) and (c) and the results are summarised in Table III.

Table III. *Relation between p_H of soil and tuber sap.*

p_H of soil	No. of plots	p_H of tuber sap	
		Method (b)	Method (c)
4.0–4.5	4	5.69	5.66
4.5–5.0	11	5.68	5.64
5.0–5.5	11	5.66	5.63
5.5–6.0	5	5.69	5.63
6.0–6.5	3	5.68	5.63
6.5–7.0	2	5.62	5.68

In 1929, five plots were laid down on a soil having p_H 5.5. Two plots were made more acid by treatment with flowers of sulphur and two more alkaline by treatment with calcium hydroxide. Two varieties were grown on each plot.

In autumn, samples of tubers were lifted, washed and dried and then examined as follows. Ten tubers were selected at random from each sample and cut through the rose and heel ends. The acidity was determined at various points on the section by means of the micro-electrode. The small injuries made by the electrode were then cut out, the tubers were pulped and the p_H of the sap was determined by method (c). The average results are given in Table IV, the p_H values of the soil being those obtained during September.

Table IV. *Relation between p_H of soil and tuber.*

Plot	p_H of soil	Average p_H of tubers			
		Duke of York		Great Scot	
		(d)	(c)	(d)	(c)
A	8.1	5.70	5.77	5.68	5.73
B	7.5	5.68	5.75	5.69	5.73
C	5.5	5.70	5.74	5.69	5.70
D	5.1	5.70	5.73	5.68	5.74
E	5.0	5.71	5.77	5.68	5.72

It is obvious from the above results that the p_H of the tuber is not influenced by that of the soil. Hoagland and Davis [1925] have obtained similar results for other plants grown in culture solutions, but other workers, for example Haas [1920], have found a certain relationship between the acidities of soil and plant sap.

(b) *Variety*. In addition to the p_H figures already given for different varieties, the following were obtained: Arran Consul, 5.64; Field Marshal, 5.74; Kerr's Pink, 5.75; King Edward, 5.95. The differences between varieties were thus not significantly greater than the variations found among tubers of the same variety.

(c) *Disease*. A number of tubers affected by various diseases was collected and compared with normal tubers of the same variety. The influence of virus infection was also studied. The results are presented in Table V.

Table V. *The effect of disease upon the p_H of tubers.*

Variety	Nature of infection	Normal tuber	Affected tuber	
			Healthy part	Diseased part
Great Scot	Corky scab: <i>Spongospora subterranea</i> Lagerh.	5.70	5.65	4.35
Duke of York	Common scab: <i>Actinomyces scabies</i> (Thaxt.) Güssow	5.75	5.70	4.58
Epicure	Sprain: <i>Bacterium rubefaciens</i>	5.73	—	5.60
Epicure	Blackleg: <i>Bacillus atrosepticus</i> van Hall	5.73	5.68	5.65
Duke of York	Blight (fresh): <i>Phytophthora infestans</i> (Mont.) De By.	5.75	5.75	5.83
Duke of York	Blight (old)	5.75	5.70	5.38
Duke of York	Wart disease: <i>Synchytrium endobioticum</i> (Schilb.) Perceval	5.75	5.65	5.02
Ally	Mosaic	5.80		5.70
	Crinkle	5.80		5.60
	Leaf Roll	5.80		5.85
Arran Comrade	Mosaic	5.64		5.47
	Leaf Roll	5.64		5.70

In diseased tubers a very acid region was observed around the affected part. In mild forms the disease did not seem to affect the rest of the tuber but in severe cases a slight increase in acidity was noted throughout the tuber. The slight decrease in acidity due to Leaf Roll and the large increase due to Wart Disease confirm results reported by Boas [1919] and Weiss and Harvey [1921] respectively. Severe forms of mosaic, such as crinkle, gave rise to an increase in acidity.

(d) *Storage.* A number of tubers of each of six varieties was stored in three different ways, viz.: (1) in a pit in the open, (2) in a cool, well-ventilated storehouse, (3) in a refrigerator maintained at 2°. After a few months the average p_H values were as follows:

Original value (October)	...	...	...	...	5.76
After storage (2)	...	...	...	...	5.84
„ (1) immediately	...	...	...	...	5.49
„ (1) after 24 hours at room temperature					5.88
„ (3) immediately	...	...	...	...	5.39
„ (3) after 24 hours at room temperature					5.83

The tubers from the pit and refrigerator were more acid than before storage, but, on being allowed to stand at room temperature for a day, acquired a p_H slightly higher than the original. This was probably due to increased respiratory activity of the tissue immediately after removal from such storage conditions.

SUMMARY.

The hydrogen and quinhydrone electrodes have been used for p_H measurements of the pulp of the potato (*Solanum tuberosum*), and a micro-electrode enabled measurements to be made at different points of the tissue.

The acidity is not uniform throughout the tuber but depends upon the function of the different parts at different periods in the life of the plant. In the early stages of development the underground stem and the heel end of the tuber are most acid and the average acidity of the tuber increases with maturity. In the dormant state the acidity does not vary much in different parts of the tuber but when sprouting begins the active eyes are most acid.

The acidity of the tuber is independent of environment and is not influenced by large variations in soil acidity.

The differences in acidity due to variety and storage are not significant.

Comparatively large changes in acidity are associated with certain diseases.

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