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EFFECT OF THE NITROGEN, PHOSPHORUS, POTASSIUM AND MAGNESIUM NUTRITION OF POTATO PLANTS ON THE CONTENT OF FREE AMINO-ACIDS AND ON THE AMINO-ACID COMPOSITION OF THE PROTEIN OF THE TUBERS

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INTRODUCTION

It is a well-known fact that the mineral nutrition of plants may affect the protein and the soluble non-protein nitrogen contents of the tissues. Plants amply supplied with nitrogenous nutrients, for instance, generally have a considerably higher content of protein and in particular a higher content of non-protein nitrogen than those with a poor nitrogen supply. In the case of phosphorus, potassium, and magnesium, deficient plants are usually higher in these nitrogen fractions than normal plants.

In plant-nutrition experiments the number of nitrogenous compounds estimated is often limited; protein and soluble non-protein fractions and, in some cases, the amides asparagine and glutamine, and the amino-acids tyrosine and arginine, are usually determined. Recently developed methods of quantitative chromatography, however, enable a more detailed investigation of the nitrogenous fractions to be made.

Potato tubers have been chosen as the test material in the present investigation for the following reasons: (*a*) they can be stored for a considerable length of time without much alteration in their chemical composition, (*b*) the protein can be obtained in

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a relatively pure state much more easily than in the case of other plant materials, (*c*) a considerable amount of physiological and biochemical data concerning these materials has been collected in this laboratory during the last ten years, (*d*) potato tubers are important from the point of view of human and animal nutrition.

EXPERIMENTAL PROCEDURE

The potato tubers used in the present study were obtained from field experiments in which the plants were grown under widely differing nutritional conditions. For this purpose certified seed potatoes of the varieties Noordeling and Voran were planted on sandy or peaty soils, poor in nitrogen, phosphorus, potassium or magnesium. In some cases the soils used were deficient in more than one nutrient element. The potato plants were dressed with different amounts of the element in which the soil was deficient; the other nutrient elements were mostly supplied in optimum amounts. The tubers were harvested and transported carefully to the laboratory where they were stored in shallow layers in wooden boxes at a temperature varying from 5 to 10°C.

Analytical procedure

Preparation of the sample.

For each analysis longitudinal and transverse slices are cut from 25 tubers in order to obtain a representative sample. These slices are cut up into small pieces and 50 g of the latter are ground in a mortar after the addition of a trace of $NaHSO_3$. The pulp is transferred to a Büchner funnel and washed with water sufficient to give 500 ml of solution. This solution contains the bulk of the protein and the soluble non-protein N-compounds (see Table I). The solution is heated for 5 minutes in a boiling water-bath after the addition of a few drops of acetic acid and a knife point of sodium

TABLE I

Nitrogen contents of filtrate (divided into protein-N and soluble non-protein N) and of residue of ground potato tubers (mg per 10 mg fresh weight)

Protein-N	Soluble non-protein N	Residue-N
32.1	29.1	2.1
33.8	34.7	3.1
35.1	27.3	1.6
29.4	20.9	3.7
23.4	23.3	2.7
20.2	20.7	3.3
19.0	21.1	2.1
18.3	23.8	2.3

chloride. The protein which is precipitated is then separated by centrifugation. The precipitate is washed twice with a very dilute solution of acetic acid and three times with 95 per cent ethanol. The washed protein is transferred to dishes, dried at 37°C and ground in a mortar. It contains approximately 14.5 per cent nitrogen (see Table II), corresponding to a purity of 90%, based on protein of 16% N.

TABLE II

N-content of potato protein		
Manurial treatment	N %	Purity of protein *)
low N	14.65	91.6
high N	14.8	92.5
low P	14.0	87.5
high P	13.85	86.6
low K	14.8	92.5
high K	14.6	91.3

*) Based on protein of 16% N.

The filtrate which contains the amino-acids, the amides and the peptides is concentrated *in vacuo* at a temperature not exceeding 40°C, and made up to 25 ml.

To obtain a larger amount of protein it was found more convenient to use a separate sample of 200 g tuber tissue. This was macerated in a Waring blendor and treated as described above.

Estimation of protein and soluble non-protein nitrogen.

Estimations of the nitrogen contained in these fractions are carried out on separate samples of 10 g tuber tissue. These are ground and separated from starch and other cell residues by filtration on a Büchner funnel. The protein of the filtrate is precipitated as described above. The nitrogen of the latter plus that of the cell residues is calculated as protein nitrogen, that of the filtrate from the heat coagulum as soluble non-protein nitrogen.

Hydrolysis of the protein.

The protein is hydrolysed in 6*N* HCl for 16–18 hours (500 mg of protein in 25 ml redistilled HCl). After hydrolysis the hydrochloric acid is removed by repeated distillation *in vacuo* (5–6 times). The HCl should be removed as completely as possible to prevent the formation of interfering amounts of NaCl when this solution is brought up to pH 6.0 with NaOH. After the addition of NaOH the solution is filtered and made up to 50 ml.

The protein hydrolysates and the protein-free solutions were analysed for amino-acids by the quantitative paper chromatographic method of Thompson and Steward [12] [13]. This method will be described in some detail.

Quantitative paper chromatography of amino-acids.

The solution to be analysed should contain approximately 4 mg of amino-acid-N per ml. For a complete analysis of the amino-acids two different dilutions of this solution are used containing (*a*) 50 and (*b*) 100–200 μg of N per 0.05 ml, respectively. The dilute solution (*a*) is used for those amino-acids which, in large concentrations, may overlap (aspartic acid and glutamic acid, serine and glycine, alanine and threonine). Solution (*b*) is used for all other amino-acids. A second aliquot of 0.05 ml of (*b*) is used for the S-containing amino-acids (see below). 0.1 ml of (*b*) is used for the estimation of those amino-acids which occur in very small amounts or which give a weak colour with ninhydrin (lysine, tyrosine, proline).

The sample is applied to a sheet of filter paper at a point 8 cm from top and side of the paper and is confined to a circle 1–1.5 cm in diameter (drops of 0.01 ml are used). To support the paper a watch glass is placed below the circle during the operation. To promote the drying of the drops, heat from an electric lamp is used; this may be done as long as the spot is moist. After drying, mostly for 5 minutes, the paper is transferred to the cabinet in which the chromatography (descending method) is carried out. A 2-directional procedure was used first with phenol-water and then with collidine-lutidine-water as the moving phases. The cabinet used contains three troughs (see Fig. 55b in [15]); in each trough two sheets of filter paper may be placed. The papers are held in the troughs by a heavy glass rod in such a way that the sample lies approximately 3.5 cm beyond the edge of the trough. Dishes with phenol-water (2 : 3) are placed at the bottom of the cabinet to allow the papers and the atmosphere to equilibrate with this solvent before adding the latter to the troughs (approximately 8 hours).

80 ml of phenol-water (3 : 1, pH 5–5.5) are then added to the troughs through a small hole in the top of the cabinet which is closed by a stopper. The phenol solution travels downwards through the paper, carrying the different amino-acids at different rates. When the solvent has moved for approximately 45 cm, *i.e.* when it has reached a distance of approximately 1–2 cm from the lower edge (36 hours at 20°C) the paper is removed from the cabinet and allowed to dry at room temperature in an air stream for about 20 hours. A margin of 2 cm containing the phenol front is cut from the paper. The paper is then transferred to a second cabinet in which a similar procedure is carried out with collidine-lutidine-water (1 : 3 : 3, pH 8.0) in a direction at right angles to the first. As with the phenol treatment the second cabinet contains dishes with the solvent to ensure equilibrium between paper and atmosphere. After equilibrium has been reached (4–6 hours) 80 ml of the collidine-lutidine-water mixture are added to the troughs and the second phase of the chromatography is begun. After 20–24 h at 20°C when the solvent has moved for approximately 35 cm the procedure is stopped and the paper is again dried in an air stream (about 16 hours).

Development of the chromatogram: reaction of the

amino-acids on the paper with ninhydrin. For this purpose the chromatogram is sprayed evenly with a solution of 1% ninhydrin in 95% ethanol containing 2% of a collidine-lutidine mixture (1 : 3) (*ca* 30–35 ml per sheet of filter paper 35 by 45 cm *). A hand sprayer activated by compressed CO_2 is used. An excess of the solvent may cause the coloured spots to spread and should therefore be avoided.

The sheets are then transferred to the colour development cabinet which is a modification of the apparatus of T h o m p s o n *et al.* (see Fig. 3 in [12]). It consists of a tank 54 by 34 by 60 cm in which the chromatograms are heated under ethanol-saturated anaerobic conditions. The bottom of the tank contains a layer of ethanol. Three perforated pipes connected to a metal cylinder containing carbon dioxide are immersed in this layer so that a flow of ethanol-saturated CO_2 passes across the paper sheets. The tank has accomodation for three sheets. A perforated shield above the surface of the ethanol prevents splashing of the papers. The papers containing the amino-acids are hung vertically in the tank with the initial spot at the lower end. In order to keep the atmosphere saturated with ethanol two filter papers hang down along the walls of the tank from shallow troughs at the top. These filter papers dip into the ethanol at the bottom of the tank; the troughs also contain ethanol. The tank is contained in an outer cabinet with a constant-temperature control.

Under these conditions an optimum colour development was found to take place within 20–25 minutes at 60°C. The sheets are then dried for 10 minutes in air. No oxidation of the coloured compound was found to occur during drying.

The coloured spots are cut out and extracted with 10 ml 50% ethanol (by volume) for 30 minutes after being cut into pieces of 0.5 by 2 cm. The intensity of the extracted blue (or yellow-brown) compound is measured in a colorimeter at 570 mμ (blue compound) or 330 mμ (yellow-brown compound).

From each sheet blanks of similar size are cut out and also extracted. When the extractions are made immediately after completion of the colour reaction the blank value is low; it increases after 1–2 hours.

R e m a r k s.

P a p e r. Whatman no. 1 filter paper, "for chromatography", 45 by 55 cm, was used throughout the investigation. The paper was not washed before use.

P u r i f i c a t i o n o f s o l v e n t s. Phenol as well as collidine and lutidine were redistilled before use. The latter two products were purchased from N.V. Utrechtse Asphaltfabriek (The Hague) and distilled at 168°C (collidine) and between 153–157°C (lutidine).

S t a n d a r d c u r v e s. As different amino-acids on paper react in different ways with ninhydrin, standard curves for each amino-acid have to be made. For this purpose 20, 30, 40, 50 and 60 μg respectively of each

*) In addition to the removal of margins of 2 cm containing the phenol and collidine-lutidine fronts, respectively, strips of 8 cm are cut from the opposite sides of the sheets before treatment with ninhydrin.

amino-acid are chromatographed and their colour estimated as described above. Two standard solutions are prepared, one containing aspartic acid, glycine and threonine and the other glutamic acid, serine and alanine. The remaining amino-acids may be in either solution. The pH of the standard solutions is adjusted to pH 6.0. The standard solutions are analysed in three or four replicates.

Internal standard. As a check on the reproducibility of the values, a so-called internal standard is included in each case. For this purpose 40 μg of alanine are placed 13 cm from the top and side of each sheet used. After chromatography it takes a position which is not occupied by other amino-acids. In the case of H_2O_2-treated samples it can not be used in this position as it then overlaps the position of methionine sulphone.

When the internal standard deviates considerably from its standard value the sheet is discarded. Corrections are not made.

Sulphur-containing amino-acids. The sulphur-containing amino-acids, methionine, cystine and related compounds have to be oxidized to methionine sulphone and cysteic acid respectively in order to obtain distinct spots on the chromatogram. This can be done by adding 0.02 ml

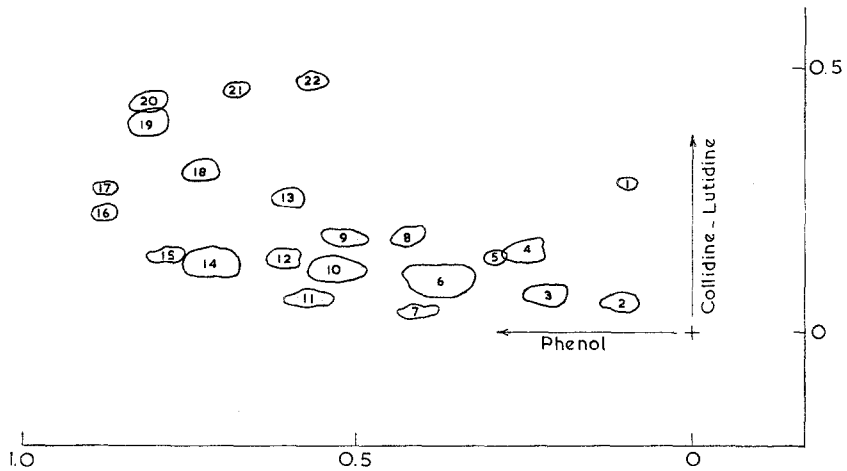

Fig. 1. Two-dimensional chromatogram, showing separation of the amino-acids in the protein and the non-protein fraction of the potato tuber.

1. Cystein + cysteine (after oxidation)
2. Aspartic acid
3. Glutamic acid
4. Serine
5. Glycine
6. Asparagine
7. Lysine
8. Threonine
9. Alanine
10. Glutamine
11. Arginine
12. Histidine
13. Methionine (after oxidation)
14. γ-Aminobutyric acid
15. Methionine sulphoxide
16. Proline
17. Pipecolic acid
18. Valine
19. Leucines
20. Phenylalanine
21. Tryptophane
22. Tyrosine

of 0.02 *M* ammonium molybdate and 0.1 ml of 30% H_2O_2 to 0.1 ml of the amino-acid solution in a watch glass. After drying, the residue is dissolved in water and transferred to the paper.

Ammonia and cyanide were not used in the cabinet during the phenol treatment.

Position of amino-acids on the chromatogram (see Fig. 1). The following amino-acids give distinct spots on the chromatogram: glycine, alanine, serine, cystine (after oxidation to cysteic acid), threonine, valine, methionine (after oxidation to methionine sulphone), the leucines, tyrosine, proline, aspartic acid, glutamic acid, arginine, lysine, γ-aminobutyric acid, asparagine and glutamine. An unknown spot was always present on the chromatogram of the soluble fractions. After most analyses had been performed this spot was found to be brought about by pipecolic acid. Leucine and isoleucine occupy the same position so that they are determined together. Glycine and asparagine more or less overlap so that their separate determination is difficult when both occur in considerable amounts. In the present investigation this point was not very important, since glycine occurs only as traces in the soluble non-protein fraction in which asparagine is an important constituent, while in the protein hydrolysate asparagine is absent. When glycine and asparagine occur in the same sample, hydrolysis of the asparagine to aspartic acid may be used to allow for the determination of both. In this case the sample has to be chromatographed before and after hydrolysis. The increase in aspartic acid upon hydrolysis gives the value for asparagine. To decide whether a certain spot on the chromatogram represents a certain amino acid, a second aliquot of the same sample, to which an additional amount of the expected amino acid has been added, is chromatographed. Phenylalanine, tryptophane and histidine cannot be determined by this chromatographic method. For their estimation see below.

Accuracy of the method. The amino-acids may be determined with an accuracy of 12% or less. Table III gives the recovery values for different amino-acids added to a potato extract. The amino-acids were added in amounts of 40 μg.

Phenylalanine.

Phenylalanine is estimated according to the method of Kapeller-Adler[6]). This is based on the oxidation and nitration of the amino-acid to 3,4-dinitrobenzoic acid which after reduction with NH_2OH gives a

TABLE III

Recovery of amino-acids added to a potato extract			
Amino-acid	Recovery (%)	Amino-acid	Recovery (%)
Alanine	96	Glutamic acid	106
Valine	98	Arginine.	105
Leucine	102	γ-aminobutyric acid. .	96
Tyrosine	94	Asparagine.	103
Aspartic acid	97	Glutamine	106

purple-blue colour with ammonia, presumably due to the formation of the ammonium salt of *o*-diaci-dihydro-dinitrobenzoic acid.

Reagents. (*a*) Nitration mixture: 20 g KNO_3 are dissolved in 100 ml concentrated H_2SO_4. (*b*) 30% $NH_2OH.HCl$. (*c*) 25% NH_4OH.

Method. An aliquot of an amino-acid solution is evaporated to dryness. 2 ml of (*a*) are added and the nitration is carried out by warming on the steam bath for 20 minutes. The residue is transferred with 9 ml of H_2O to a volumetric flask of 25 ml. The solution is cooled in an ice bath, and 2.5 ml of (*b*), also cooled in the ice bath, are added. After 1 minute the solution is diluted to the mark with ice-cooled ammonia (*c*). The mixture is allowed to stand for 45 minutes at room temperature and then the colour intensity is read at 560 mμ. For each sample a blank value obtained without the addition of $NH_2OH.HCl$ is measured.

Tyrosine and histidine give a yellow-brown colour which does not interfere at 560 mμ. The method is sensitive to 100 μg of phenylalanine.

Tryptophane.

Tryptophane is estimated according to the method of Brummer [2]) (see also Groot [3]). This is based on the formation of a red-violet colour with vanillin in a strongly acid solution. The method is used for protein hydrolysates only, as unknown substances in the potato extract interfere with the determination of this amino-acid.

Reagents. (*a*) 1/75 *M* solution of vanillin in H_2O. (*b*) 96% H_2SO_4 (*pro analysi*). (*c*) 1/450 *M* Na_2S. Prepared by dissolving an excess of Na_2S in H_2O; the solution is titrated with iodine and then diluted with water to give the desired concentration.

Method. 1 ml of amino-acid solution, 1 ml of (*c*) and 0.1 of (*a*) are added to a test tube and cooled in a NaCl-ice bath. Then 3 ml of (*b*) are slowly added; care is taken that the solution remains as cold as possible; it is shaken continuously during the addition of the H_2SO_4. The colour is allowed to develop at room temperature for 4½ h after which the flask is placed on a boiling water bath for 3–4 minutes. The colour intensity is measured at 605 mμ. The method is sensitive to 5 μg of tryptophane.

Histidine.

Histidine is determined by the diazo reaction of Pauly: formation by histidine of a yellow-red compound with freshly diazotized sulphanilic acid (Block and Bolling [1]). Since tyrosine gives the same reaction, this amino-acid has to be removed before histidine can be determined. Use is made of the fact that tyrosine may be oxidized by 0.1 *N* $KMnO_4$ in an acid solution, without histidine being affected. The amount of $KMnO_4$ required for complete removal of tyrosine from a protein-free potato extract was found to be 5 to 6 times higher than was needed for elimination of an equal amount of tyrosine in the absence of other substances. Upon neutralization of the $KMnO_4$-treated sample a yellow-brown colour, presumably due to MnO_2, is formed which interferes with the histidine determination. Therefore the neutralization should be stopped when a faint yellow colour of the solution is observed.

The amount of diazo reagent required for determination of histidine in potato extracts is generally considerably greater than that needed for histidine only. This is presumably due to the fact that in addition to tyrosine, potato extract contains an unknown compound which reacts with the diazo reagent. When this unknown compound is present in large amounts, it may interfere with the histidine determination. This may be prevented by using a smaller aliquot of the extract. To check the absence of this interference two different amounts of the extract are analysed. In addition the recovery of added histidine has to be checked.

Reagents. (*a*) 4.5 g of sulphanilic acid and 45 ml of 37% HCl in 500 ml H_2O. (*b*) 25 g $NaNO_2$ in 500 ml H_2O. (*c*) 5.5 g anhydrous Na_2CO_3 in 500 ml H_2O. (*d*) Solution of histidine-monohydrochloride 1 : 10000. The diazo reagent is prepared by adding 1.5 ml of (*a*) to 1.5 ml of (*b*) in a 50-ml volumetric flask kept in an ice bath. After 5 minutes cooling 6 ml of (*b*) are again added and after a further 5 minutes cooling the solution is diluted to 50 ml. The solution may be kept for 1 day when cooled in ice.

Method. To 15 ml of the protein-free amino-acid solution are added 1 ml of 30% H_2SO_4 and 3 ml of 0.1 *N* $KMnO_4$. The mixture is neutralized with 10% NaOH until it takes on a light yellow colour and then filtered through a dry filter paper. 0.5 and 1 ml of the filtrate are used for the histidine determination which is carried out as follows. 5 ml of (*c*) are pipetted into a 25-ml volumetric flask. To this solution sufficient H_2O is added so that the total volume of added water and aliquot of histidine solution is 2 ml. Subsequently 2 ml of the diazo reagent are added, immediately followed by the histidine-containing solution. When the yellow-orange colour is formed a second portion of 2 ml reagent is added. After 6 minutes incubation in a water bath at 15°C maximum colour intensity is reached. The latter is measured at 604 mμ. The method is sensitive to 10 μg of histidine.

RESULTS

a) Comparison of the amino-acid composition of the protein and the soluble non-protein fractions

Tables V to X contain the amino-acid analyses of the protein and the soluble non-protein fractions of potato tubers grown under widely differing nutritional conditions. In each case a comparison was made between tubers from plants highly deficient in one of the major nutrient elements and those from plants grown on the same field with an ample supply of this element.

In Tables V, VII, VIII and IX the amino-acid values for both protein and soluble non-protein fractions have been recorded; in Tables VI and X those of the non-protein fractions only are given. In the case of protein the figures have been calculated as g amino-acid per 100 g of hydrolysed protein (16 g N) and as

TABLE IV

Yield data and protein and soluble non-protein N-contents of the potato tubers used for the amino-acid analyses

Exp., year, variety	Manurial treatment *) (kg per ha)	Yield, q per ha (1 q = 100 kg)	mg N per 10 g dry weight	
			Protein fraction	Soluble non-protein fraction
1388, 1952, Noordeling	40 N	103	78.7	32.8
	160 N	183	107.4	66.6
1300, 1952, Noordeling	0 P_2O_5	56	113.6	79.7
	300 P_2O_5	249	102.1	73.1
1472, 1954, Noordeling	0 P_2O_5	44	85.5	84.4
	300 P_2O_5	290	62.5	44.8
1299, 1952, Noordeling	0 K_2O	108	106.4	106.9
	200 K_2O	290	93.4	86.4
1472, 1954, Noordeling	0 K_2O	83	86.3	92.9
	400 K_2O	290	65.6	46.7
1220, 1951, Voran	5 MgO	161	79.8	90.6
	160 MgO	383	63.6	56.3
1466, 1953, Noordeling	400 K, 150 N	299	99.8	69.7
,, ,, Voran	400 K, 150 N	455	54.7	54.6

*) N was supplied as ammonium nitrate limestone, P as superphosphate, K as potassium sulphate, and Mg as magnesium sulphate.

amino-acid nitrogen as a percentage of the sum of estimated amino-acid nitrogen (1st and 2nd column of each sample respectively). The values for the free amino-acids are similarly given as g amino-acid per 16 g animo-acid-N (3rd and 4th column of each sample) and as amino-acid nitrogen as a percentage of the total estimated amino-acid nitrogen (5th and 6th column). In the case of free amino-acids, values are given both including and excluding the amides.

When a comparison is made between the content of amino-acids occurring in the free state and that of amino-acids present in the protein, it will be seen that no correlation exists. In the soluble non-protein fraction the nitrogen of the amides constitutes more than 60% and sometimes more than 70% of the total amino-acid nitrogen. In the case of protein, at a maximum, 20% of the total amino-acid nitrogen could be combined as amides, assuming that all the ammonia in the hydrolysate came from these compounds.

When the amino-N of the amides is added to the amino-N of aspartic acid and glutamic acid respectively, it will be seen that

TABLE V

Composition of the protein and the soluble non-protein fractions of potato tubers grown at two nitrogen levels, stored for 4 months (Exp. 1388, 1952, *var.* Noordeling)

Amino compound	Low-N potatoes						High-N potatoes					
	Protein fraction *)		Soluble non-protein fraction *)				Protein fraction *)		Soluble non-protein fraction *)			
	g am.-acid per 100 g protein (16 g N)	N as % of sum of est. am.-acid-N (incl. NH_3)	g am.-acid per 16 g am.-acid-N		N as % of sum of est. am.-acid-N		g am.-acid per 100 g protein (16 g N)	N as % of sum of est. am.-acid-N (incl. NH_3)	g am.-acid per 16 g am.-acid-N		N as % of sum of est. am.-acid-N	
			incl. the amides	excl. the amides	incl. the amides	excl. the amides			incl. the amides	excl. the amides	incl. the amides	excl. the amides
Glycine	5.9	7.5	tr	tr	tr	tr	6.0	7.6	tr	tr	tr	tr
Alanine	5.1	5.4	1.0	2.3	1.0	2.3	5.0	5.4	0.6	2.2	0.6	2.2
Serine	6.8	6.2	1.0	2.3	0.8	2.0	7.2	6.6	0.6	2.2	0.5	1.8
Cystine + cysteine	1.1	0.7	0.7	1.6	0.4	0.9	1.3	0.8	0.3	1.1	0.2	0.6
Threonine	6.5	5.2	1.3	2.9	0.9	2.2	6.5	5.2	0.6	2.2	0.5	1.6
Valine	6.9	5.7	2.8	6.3	2.1	5.0	6.5	5.4	1.7	6.0	1.3	4.5
Methionine + methionine sulphoxide	2.8	1.7	1.9	4.3	1.1	2.6	3.1	2.0	1.0	3.3	0.6	2.0
Leucines	16.2	11.8	3.3	7.5	2.2	5.1	16.3	11.9	2.2	7.5	1.4	5.0
Phenylalanine	5.0	2.9	—	—	—	—	4.9	2.8	—	—	—	—
Tyrosine	4.2	2.2	2.1	4.8	1.0	2.4	4.6	2.4	1.3	4.5	0.6	2.2
Proline	3.8	3.2	tr	tr	tr	tr	3.8	3.2	tr	tr	tr	tr
Tryptophane	1.8	1.6	—	—	—	—	2.0	1.9	—	—	—	—
Aspartic acid	10.5	7.5	5.9	13.7	4.0	9.3	10.5	7.5	4.1	14.2	2.7	9.3
Glutamic acid	9.9	6.4	20.6	47.4	12.4	29.3	9.8	6.3	10.3	35.3	6.1	21.1
Arginine	5.8	12.7	5.4	12.4	11.0	25.9	5.5	12.1	4.8	16.5	9.6	33.2
Lysine	4.2	5.4	tr	tr	tr	tr	4.0	5.2	tr	tr	tr	tr
Histidine	2.5	4.6	1.0	2.3	1.7	4.0	2.6	4.8	0.6	1.9	0.9	3.2
γ-Aminobutyric acid	0.0	0.0	4.2	9.7	3.6	8.5	0.0	0.0	4.6	15.8	3.9	13.4
Asparagine	—	—	34.6	—	46.4	—	—	—	41.4	—	54.9	—
Glutamine	—	—	9.1	—	11.1	—	—	—	13.3	—	15.9	—
Ammonia	1.7	9.3	—	—	—	—	1.6	9.0	—	—	—	—

*) 91.9% and 76.2% of the Kjeldahl nitrogen of the protein and the soluble non-protein fractions of the low-N potatoes are accounted for by the amino-acid analyses of these fractions. For the high-N potatoes these values are 91.6% and 81.8%, respectively.

aspartic acid occurs even more abundantly than glutamic acid in the non-protein fraction.

The ratio asparagine-N : glutamine-N varies from 4.5 : 1 to 2 : 1. This ratio apparently is not only affected by the mineral nutrition of the potato plants but also by climatic conditions and variety.

TABLE VI

Composition of the soluble non-protein fraction of potato tubers grown at two phosphorus levels, stored for 6 months (Exp. 1300, 1952, *var.* Noordeling)

Amino compound	Low-P potatoes				High-P potatoes			
	Soluble non-protein fraction				Soluble non-protein fraction			
	g am.-acid per 16 g am.-acid-N *)		N as % of sum of est. am.-acid-N *)		g am.-acid per 16 g am.-acid-N *)		N as % of sum of est. am.-acid-N *)	
	incl. the amides	excl. the amides	incl. the amides	excl. the amides	incl. the amides	excl. the amides	incl. the amides	excl. the amides
Glycine	tr	tr	tr	tr	tr	tr	tr	tr
Alanine	0.6	2.1	0.6	2.1	0.6	2.0	0.6	2.0
Serine	1.3	4.8	1.1	4.1	1.1	3.7	0.9	3.1
Cystine + cysteine	0.7	2.5	0.4	1.4	0.6	2.0	0.3	1.1
Threonine	1.0	3.8	0.8	2.8	1.0	3.6	0.8	2.7
Valine	1.5	5.4	1.1	4.1	2.2	7.4	1.6	5.5
Methionine + methionine sulphoxide	0.6	2.1	0.3	1.3	0.5	1.8	0.3	1.0
Leucines	1.7	6.1	1.1	4.1	1.6	5.3	1.0	3.6
Phenylalanine	1.5	5.6	0.8	3.0	1.6	5.3	0.8	2.8
Tyrosine	1.7	6.2	0.8	3.0	2.2	7.5	1.1	3.6
Proline	0.8	2.8	0.6	2.2	1.0	3.4	0.8	2.6
Tryptophane	—	—	—	—	—	—	—	—
Aspartic acid	4.0	14.8	2.6	9.7	4.4	15.0	2.9	9.9
Glutamic acid	9.4	34.7	5.4	20.1	7.0	24.1	4.1	13.9
Arginine	4.0	14.8	7.2	26.4	3.5	11.9	6.2	21.3
Lysine	tr	tr	tr	tr	tr	tr	tr	tr
Histidine	1.6	5.7	2.6	9.7	1.9	6.6	3.3	11.2
γ-Aminobutyric acid	2.0	7.3	1.7	6.2	5.4	18.4	4.6	15.6
Asparagine	44.9	—	59.6	—	38.8	—	51.6	—
Glutamine	11.1	—	13.3	—	16.0	—	19.2	—
Ammonia	—	—	—	—	—	—	—	—

*) 80.9% of the Kjeldahl nitrogen of the soluble non-protein fraction of the low-P potatoes is accounted for by the amino-acid analyses of this fraction. For the high-P potatoes this value is 82.0%.

When the amides are excluded, glutamic acid, arginine, and γ-aminobutyric acid account for more than 50% of the amino-acid nitrogen of the soluble non-protein fraction. On a weight basis glutamic acid is generally by far the most important amino-acid of this fraction followed by γ-aminobutyric acid in the second and arginine in the third place. On a nitrogen basis arginine is generally the most important amino-acid, owing to its high N-content.

In contrast to the non-protein fraction, γ-aminobutyric acid

TABLE VII

Composition of the protein and the soluble non-protein fractions of potato tubers grown at two phosphorus levels, stored for 1 month (Exp. 1472, 1954, *var.* Noordeling)

Amino compound	Low-P potatoes						High-P potatoes					
	Protein fraction *)		Soluble non-protein fraction *)				Protein fraction *)		Soluble non-protein fraction *)			
	g am.-acid per 100 g protein (16 g N)	N as % of sum of est. am.-acid-N (incl. NH_3)	g am.-acid per 16 g am.-acid-N		N as % of sum of est. am.-acid-N		g am.-acid per 100 g protein (16 g N)	N as % of sum of est. am.-acid-N (incl. NH_3)	g am.-acid per 16 g am.-acid-N		N as % of sum of est. am.-acid-N	
			incl. the amides	excl. the amides	incl. the amides	excl. the amides			incl. the amides	excl. the amides	incl. the amides	excl. the amides
Glycine	5.4	7.6	0.5	1.9	0.6	2.3	5.8	8.0	1.0	2.8	1.1	3.4
Alanine	4.1	4.8	0.6	2.5	0.6	2.4	4.3	5.0	0.7	2.1	0.7	2.1
Serine	5.8	5.8	0.8	3.1	0.7	2.6	6.0	5.9	1.3	3.9	1.1	3.4
Cystine + cysteine	0.6	0.4	0.2	0.7	0.1	0.4	0.7	0.5	0.3	0.7	0.1	0.4
Threonine	6.0	5.3	0.8	3.3	0.6	2.4	5.8	5.1	1.0	3.0	0.8	2.3
Valine	7.3	6.7	2.2	8.8	1.7	6.6	7.6	6.7	2.8	8.3	2.1	6.2
Methionine + methionine sulphoxide	1.9	1.3	1.2	4.8	0.7	2.8	2.0	1.4	2.0	5.7	1.2	3.5
Leucines	17.0	13.8	1.7	6.7	1.3	4.5	17.9	14.1	2.1	6.2	1.4	4.1
Phenylalanine	5.0	3.3	1.9	7.6	1.0	4.0	5.3	3.3	1.8	5.3	1.0	2.8
Tyrosine	4.5	2.7	1.2	4.6	0.6	2.2	4.6	2.6	2.7	7.8	1.3	3.7
Proline	3.6	3.3	tr	tr	tr	tr	3.6	3.2	tr	tr	tr	tr
Tryptophane	1.8	1.8	—	—	—	—	1.9	1.9	—	—	—	—
Aspartic acid	8.0	6.4	2.2	8.6	1.4	5.6	8.1	6.3	2.7	7.8	1.8	5.1
Glutamic acid	9.5	6.9	7.6	30.3	4.5	18.0	10.0	7.0	10.9	31.8	6.5	19.0
Arginine	3.2	7.8	3.4	13.4	6.8	27.0	3.4	8.0	2.9	8.6	5.9	17.1
Lysine	3.7	5.3	tr	tr	tr	tr	3.9	5.5	tr	tr	tr	tr
Histidine	3.1	6.4	0.8	3.2	1.4	5.4	3.0	6.0	1.3	3.8	2.2	6.4
γ-Aminobutyric acid	0.0	0.0	4.1	16.4	3.5	14.0	0.0	0.0	8.6	25.0	7.3	21.2
Asparagine	—	—	37.3	—	52.4	—	—	—	32.3	—	43.1	—
Glutamine	—	—	18.6	—	22.3	—	—	—	18.7	—	22.4	—
Ammonia	1.7	10.3	—	—	—	—	1.6	9.5	—	—	—	—

*) 82.5% and 70.0% of the Kjeldahl nitrogen of the protein and the soluble non-protein fractions of the low-P potatoes are accounted for by the amino-acid analyses of these fractions. For the high-P potatoes these values are 84.8% and 71.5%, respectively.

does not occur in the protein. Arginine accounts for approximately 10 per cent of the protein-N, glutamic acid for about 6 per cent. A number of amino-acids, however, occur much more abundantly in the protein than in the free state. These are glycine, alanine, serine, threonine, the leucines, proline and lysine.

b) Effect of the mineral nutrition on the amino-acid composition of the protein.

Although the mineral nutrition of the potato plants in this investigation had a pronounced effect on the yield and on the protein content of the tubers (see Table IV) no effect on the amino-

TABLE VIII

Composition of the protein and the soluble non-protein fractions of potato tubers grown at two potassium levels, stored for 3 months (Exp. 1299, 1952, *var.* Noordeling)

Amino compound	Low-K potatoes						High-K potatoes					
	Protein fraction *)		Soluble non-protein fraction *)				Protein fraction *)		Soluble non-protein fraction *)			
	g am.-acid per 100 g protein (16 g N)	N as % of sum of est. am.-acid-N (incl. NH_3)	g am.-acid per 16 g am.-acid-N		N as % of sum of est. am.-acid-N		g am.-acid per 100 g protein (16 g N)	N as % of sum of est. am.-acid-N (incl. NH_3)	g am.-acid per 16 g am.-acid-N		N as % of sum of est. am.-acid-N	
			incl. the amides	excl. the amides	incl. the amides	excl. the amides			incl. the amides	excl. the amides	incl. the amides	excl. the amides
Glycine	5.3	7.0	tr	tr	tr	tr	5.4	6.9	tr	tr	tr	tr
Alanine	5.9	6.6	0.6	1.7	0.5	1.7	6.0	6.6	0.6	1.8	0.6	1.7
Serine	6.6	6.2	1.3	4.1	1.1	3.4	6.0	5.6	1.5	4.6	1.3	3.8
Cystine + cysteine	1.0	0.6	0.4	1.3	0.3	1.0	0.7	0.4	0.5	1.6	0.3	0.8
Threonine	6.7	5.6	1.2	3.7	0.9	2.7	6.5	5.3	1.3	3.8	1.0	2.8
Valine	5.9	5.0	3.0	9.2	2.3	6.9	6.2	5.1	2.6	7.6	1.9	5.7
Methionine + methionine sulphoxide	2.5	1.7	1.5	4.5	0.9	2.7	2.3	1.4	1.7	4.9	1.0	2.9
Leucines	15.3	11.6	1.9	5.9	1.3	3.9	15.2	11.3	1.7	5.1	1.1	3.4
Phenylalanine	5.5	3.3	2.3	6.9	1.2	3.7	5.4	3.2	1.4	4.2	0.8	2.2
Tyrosine	5.2	2.8	5.2	15.9	2.5	7.7	5.0	2.7	3.3	9.7	1.6	4.7
Proline	4.3	3.7	tr	tr	tr	tr	5.2	4.4	tr	tr	tr	tr
Tryptophane	1.9	1.8	—	—	—	—	1.8	1.7	—	—	—	—
Aspartic acid	10.3	7.7	2.8	8.4	1.8	5.6	10.5	7.7	3.9	11.4	2.5	7.5
Glutamic acid	9.4	6.4	5.8	17.8	3.5	10.6	9.9	6.6	8.6	25.3	5.1	15.1
Arginine	4.2	9.5	4.6	14.0	9.2	28.0	4.4	9.8	5.2	15.3	10.4	30.7
Lysine	4.6	6.2	0.7	2.2	0.9	2.7	5.0	6.7	0.8	2.4	1.0	2.9
Histidine	2.6	5.0	0.6	1.8	1.0	3.1	2.4	4.6	0.6	1.9	1.1	3.1
γ-Aminobutyric acid	0.0	0.0	6.4	19.5	5.4	16.6	0.0	0.0	5.2	15.5	4.5	13.1
Asparagine	—	—	37.9	—	50.3	—	—	—	38.6	—	51.2	—
Glutamine	—	—	14.1	—	16.8	—	—	—	12.3	—	14.8	—
Ammonia	1.6	9.3	—	—	—	—	1.8	10.0	—	—	—	—

*) 88.2% and 87.8% of the Kjeldahl nitrogen of the protein and the soluble non-protein fractions of the low-K potatoes are accounted for by the amino-acid analyses of these fractions. For the high-K potatoes these values are 89.9% and 89.1%, respectively.

acid composition of this protein was observed (see Tables V, VII, VIII and IX). This was found to be true in all three cases, nitrogen, potassium and phosphorus. Since the composition of the soluble non-protein fraction varied considerably under the influence of the mineral nutrition, this result once more demonstrates the lack of correlation between the amino-acid composition of the protein and that of the soluble non-protein fractions.

When a comparison is made between the proteins of tubers grown on the same field with different manurial treatments and those of tubers grown on different fields and in different years, it will be seen that the agreement in amino-acid composition in the former case is more complete than in the latter. Although it is possible that small differences in protein composition of potato tubers may occur under the influence of varying climatic conditions, it is also possible that small differences in the analytical technique have been responsible for the observed differences.

c) *Effect of the mineral nutrition on the amino-acid composition of the soluble non-protein fraction.*

The amino-acid composition of the non-protein fraction was found to be much less constant than that of the protein. Tables V, VII, VIII and IX contain the results for samples in which both protein and non-protein fractions had been analysed. In Table VI and X the results for a few further samples have been recorded in which non-protein nitrogen only was investigated.

N i t r o g e n. Tubers from nitrogen-deficient potato plants were considerably poorer in both protein and soluble non-protein nitrogen than those from plants dressed amply with ammonium nitrate. The non-protein nitrogen content was more strongly affected by the nitrogen dressing than the protein content, the respective values for protein and non-protein fractions being 78.7 (N-deficient) and 107.4 (high-N), 32.8 and 66.6 mg N per 10 g of dry matter, respectively.

More than 70% of the estimated nitrogen of this fraction was accounted for by amides in the case of fully manured plants. In the tubers of nitrogen-deficient plants this value was 57.5%. The relative content of asparagine was increased by 18 per cent following the nitrogen manuring, that of glutamine by 43 per cent.

When the amides were excluded, arginine contributed 33 per cent

TABLE IX

Composition of the protein and the soluble non-protein fractions of potato tubers grown at two potassium levels, stored for 2 months (Exp. 1472, 1954, *var.* Noordeling)

Amino compound	Low-K potatoes						High-K potatoes					
	Protein fraction *)		Soluble non-protein fraction *)				Protein fraction *)		Soluble non-protein fraction *)			
	g am.-acid per 100 g protein (16 g N)	N as % of sum of est. am.-acid-N (incl. NH_3)	g am.-acid per 16 g am.-acid-N		N as % of sum of est. am.-acid-N		g am.-acid per 100 g protein (16 g N)	N as % of sum of est. am.-acid-N (incl. NH_3)	g am.-acid per 16 g am.-acid-N		N as % of sum of est. am.-acid-N	
			incl. the amides	excl. the amides	incl. the amides	excl. the amides			incl. the amides	excl. the amides	incl. the amides	excl. the amides
Glycine	4.8	6.8	tr	tr	tr	tr	5.8	8.0	tr	tr	tr	tr
Alanine	4.4	5.2	0.8	3.2	0.8	3.1	4.3	5.0	0.9	2.7	0.9	2.6
Serine	5.8	5.8	1.6	6.4	1.3	5.3	6.0	5.9	1.3	3.8	1.1	3.2
Cystine + cysteine	1.0	0.6	0.2	0.7	0.1	0.4	0.7	0.5	0.2	0.7	0.1	0.4
Threonine	5.1	4.5	0.6	2.6	0.5	1.9	5.8	5.1	1.1	3.3	0.8	2.4
Valine	6.8	6.2	3.1	12.8	2.3	9.6	7.6	6.7	3.1	9.3	2.3	6.9
Methionine + methionine sulphoxide	2.2	1.8	1.2	4.9	0.7	2.9	2.0	1.4	2.0	5.9	1.2	3.5
Leucines	17.5	14.2	1.6	6.5	1.1	4.4	17.9	14.1	2.7	8.0	1.8	5.4
Phenylalanine	5.5	3.5	1.2	5.0	0.7	2.7	5.3	3.3	1.7	5.0	0.9	2.7
Tyrosine	3.4	1.9	3.0	12.2	1.5	5.9	4.6	2.6	2.3	6.8	1.1	3.3
Proline	3.9	3.6	tr	tr	tr	tr	3.6	3.2	tr	tr	tr	tr
Tryptophane	1.9	1.9	—	—	—	—	1.9	1.9	—	—	—	—
Aspartic acid	8.7	6.9	2.9	11.9	1.9	7.9	8.1	6.3	3.0	9.0	2.0	5.9
Glutamic acid	9.6	6.9	5.2	21.2	3.0	12.3	10.0	7.0	11.2	33.6	6.5	19.4
Arginine	3.8	9.3	2.8	11.6	5.1	20.9	3.4	8.0	3.1	9.3	5.6	16.6
Lysine	3.7	5.3	tr	tr	tr	tr	3.9	5.5	tr	tr	tr	tr
Histidine	2.6	5.3	0.7	2.9	1.2	4.9	3.0	6.0	1.2	3.7	2.1	6.3
γ-Aminobutyric acid	0.0	0.0	5.2	21.1	4.4	18.0	0.0	0.0	8.4	25.1	7.1	21.3
Asparagine	—	—	37.7	—	50.1	—	—	—	32.3	—	42.9	—
Glutamine	—	—	21.3	—	25.5	—	—	—	19.7	—	23.7	—
Ammonia	1.6	10.1	—	—	—	—	1.6	9.4	—	—	—	—

*) 82.4% and 73.3% of the Kjeldahl nitrogen of the protein and the soluble non-protein fractions of the low-K potatoes are accounted for by the amino-acid analyses of these fractions. For the high-K potatoes these values are 84.8% and 69.7%, respectively.

of the total estimated nitrogen of this fraction in fully manured plants as against 25.9 per cent in N-deficient plants. Since the nitrogen of arginine can be easily made available in living plants, this amino-acid may function in a similar way to the amides as a temporary storage product for excess nitrogen.

TABLE X

Composition of the soluble non-protein fraction of potato tubers grown at two magnesium levels, stored for 6 months (Exp. 1220, 1951, *var.* Voran)

Amino compound	Low-Mg potatoes				High-Mg potatoes			
	Soluble non-protein fraction				Soluble non-protein fraction			
	g am.-acid per 16 g am.-acid-N *)		N as % of sum of est. am.-acid-N *)		g am.-acid per 16 g am.-acid-N *)		N as % of sum of est. am.-acid-N *)	
	incl. the amides	excl. the amides	incl. the amides	excl. the amides	incl. the amides	excl. the amides	incl. the amides	excl. the amides
Glycine	tr	tr	tr	tr	tr	tr	tr	tr
Alanine	0.6	2.1	0.6	2.0	1.1	3.2	1.1	3.1
Serine	1.1	3.4	0.9	2.9	1.3	3.9	1.1	3.2
Cystine + cysteine	tr	tr	tr	tr	tr	tr	tr	tr
Threonine	1.7	5.6	1.3	4.1	1.3	3.7	0.9	2.7
Valine	3.7	11.9	2.8	8.9	4.1	12.0	3.2	9.0
Methionine + methionine sulphoxide	tr	tr	tr	tr	tr	tr	tr	tr
Leucines	2.2	7.2	1.5	4.8	2.0	5.7	1.3	3.8
Phenylalanine	2.2	7.0	1.1	3.7	1.4	4.0	0.7	2.1
Tyrosine	3.3	10.6	1.6	5.1	4.6	13.5	2.2	6.5
Proline	1.7	5.4	1.3	4.1	1.5	4.3	1.1	3.3
Tryptophane	—	—	—	—	—	—	—	—
Aspartic acid	4.2	13.7	2.8	9.1	5.7	16.6	3.8	10.9
Glutamic acid	5.7	18.3	3.4	10.9	7.7	22.5	4.6	13.4
Arginine	2.5	8.2	5.1	16.4	2.4	7.0	4.8	14.1
Lysine	1.1	3.4	0.9	2.8	1.0	2.8	0.8	2.2
Histidine	1.8	5.7	3.0	9.7	1.0	2.9	1.7	4.9
γ-Aminobutyric acid	5.7	18.3	4.8	15.5	8.4	24.3	7.1	20.7
Asparagine	42.5	—	56.3	—	38.7	—	51.5	—
Glutamine	10.6	—	12.8	—	11.8	—	14.2	—
Ammonia	—	—	—	—	—	—	—	—

*) 86.1% of the Kjeldahl nitrogen of the soluble non-protein fraction of the low-Mg potatoes is accounted for by the amino-acid analyses of this fraction. For the high-Mg potatoes this value is 94.6%.

Likewise, γ-aminobutyric acid occurred more abundantly in normal than in the nitrogen-deficient potatoes.

Although the relative contents of most other amino-acids were somewhat higher in nitrogen-deficient tubers, this was particularly true of glutamic acid.

P h o s p h o r u s. P-deficient potatoes were found to be higher in protein and particularly higher in soluble non-protein nitrogen than those with an ample P-supply (see Table IV). In agreement with the nitrogen experiment (Table V) a higher proportion of the

TABLE XI

Effect of the nitrogen nutrition of potato plants on the amino-acid content of the tubers (calculated as mg amino-acid per 10 g of dry matter, *cf* Table V)

Amino compound	Low-N potatoes		High-N potatoes	
	Protein fraction	Soluble non-protein fraction	Protein fraction	Soluble non-protein fraction
Glycine	29.0	tr	40.3	tr
Alanine	25.1	1.6	33.6	2.2
Serine	33.5	1.6	48.3	2.1
Cystine + cysteine	5.4	1.1	8.7	1.1
Threonine	32.0	2.0	43.6	2.2
Valine	33.9	4.4	43.6	5.9
Methionine + methionine sulphoxide	13.8	3.0	20.8	3.3
Leucines	79.7	5.2	109.4	7.4
Phenylalanine	24.6	—	32.9	—
Tyrosine	20.7	3.3	30.9	4.4
Proline	18.7	tr	25.5	tr
Tryptophane	8.9	—	13.4	—
Aspartic acid	51.7	9.5	70.5	14.0
Glutamic acid	48.7	32.9	65.8	35.1
Arginine	28.5	8.6	36.9	16.3
Lysine	20.7	tr	26.9	tr
Histidine	12.3	1.6	17.5	1.9
γ-Aminobutyric acid	0.0	6.7	0.0	15.6
Asparagine	—	55.3	—	141.3
Glutamine	—	14.6	—	45.4
Ammonia	8.4	—	10.7	—

nitrogen of the latter fraction occurred as amides. In the second sample no difference in relative amide content was found.

In both cases asparagine occurred much more abundantly in the soluble fraction of the P-deficient tubers than in that of tubers supplied amply with phosphorus. In one experiment glutamine contributed to the soluble nitrogen of P-deficient and normal tubers in equal amounts; in the second its contribution was lower in the case of phosphorus deficiency.

Of the amino-acids, tyrosine was found to occur in somewhat higher concentration in the soluble non-protein fraction of P-manured tubers than in that of P-deficient ones, an observation which is in agreement which earlier estimations [8]). Phenylalanine which may be considered to be the precursor of tyrosine was lower in the normal tubers.

Glutamic acid was found more abundantly in the non-protein fraction of phosphorus-deficient than in that of normal tubers. This

TABLE XII

Effect of the phosphorus nutrition of potato plants on the amino-acid content of the tubers (calculated as mg amino-acid per 10 g of dry matter, *cf* Table VII)

Amino compound	Low-P potatoes		High-P potatoes	
	Protein fraction	Soluble non-protein fraction	Protein fraction	Soluble non-protein fraction
Glycine	28.9	1.8	22.6	1.9
Alanine	21.9	2.3	16.8	1.4
Serine	31.0	2.9	23.4	2.6
Cystine + cysteine	3.2	0.7	2.7	0.5
Threonine	32.1	3.1	22.6	2.0
Valine	39.0	8.2	29.7	5.5
Methionine + methionine sulphoxide	10.2	4.5	7.8	3.8
Leucines	90.8	6.3	69.9	4.1
Phenylalanine	26.7	7.1	20.7	3.5
Tyrosine	24.0	4.3	18.0	5.2
Proline	19.2	tr	14.1	tr
Tryptophane	9.6	—	7.4	—
Aspartic acid	42.8	8.0	31.6	5.2
Glutamic acid	50.8	28.2	39.1	21.3
Arginine	17.1	12.5	13.3	5.7
Lysine	19.7	tr	15.2	tr
Histidine	16.6	3.0	11.7	2.5
γ-Aminobutyric acid	0.0	15.3	0.0	16.7
Asparagine	—	137.8	—	62.9
Glutamine	—	68.7	—	36.3
Ammonia	9.1	—	6.3	—

was true in the sample in which glutamine was lower in the P-deficient tubers. This might indicate that in potatoes the formation of glutamine from glutamic acid proceeds less readily in the case of an inadequate supply of phosphorus. In the second sample, in which no differences in glutamine content were observed, the glutamic acid content was slightly lower in phosphorus-deficient tubers.

Arginine contributed considerably more to the nitrogen of the soluble non-protein fraction in phosphorus-deficient tubers than in normal ones. Similar results have been observed in those cases in which the nitrogen supply of the plants was high. Arginine is apparently used by potato plants in a similar way to amides as a storage compound for excess nitrogen.

γ-Aminobutyric acid occurred in much lower amounts in P-deficient potatoes than in tubers from plants amply supplied with phosphorus; this was true of both samples. Since γ-aminobutyric

acid was found to be more abundant in plants amply supplied with nitrogen (see Table V) and since phosphorus-deficient tubers behaved like those amply supplied with nitrogen as far as their amide and arginine contents were concerned, it may be concluded that the formation of γ-aminobutyric acid was specifically inhibited in P-deficient potatoes.

P o t a s s i u m. Potassium-deficient plants are generally considerably higher in soluble non-protein nitrogen than are those amply supplied with potassium. The protein may or may not be higher in the K-deficient plants (see Table IV).

In agreement with the nitrogen and phosphorus experiments, the amides were found to contribute more to the amino-acid-nitrogen in K-deficient tubers than in normal ones. In one case their contribution was 75.6 per cent in the K-deficient sample and 66.6 per cent in the K-manured sample; in the second sample no differences in the relative amide contents were found.

TABLE XIII

Effect of the potassium nutrition of potato plants on the amino-acid content of the tubers (calculated as mg amino-acid per 10 g of dry matter, *cf* Table VIII)

Amino compound	Low-K potatoes		High-K potatoes	
	Protein fraction	Soluble non-protein fraction	Protein fraction	Soluble non-protein fraction
Glycine	35.2	tr	31.5	tr
Alanine	39.2	3.3	35.0	2.7
Serine	43.9	7.9	35.0	6.9
Cystine + cysteine	6.7	2.6	4.1	2.4
Threonine	44.5	7.2	38.0	5.8
Valine	39.2	17.9	36.2	11.5
Methionine + methionine sulphoxide	16.6	8.9	13.4	7.4
Leucines	101.8	11.5	88.8	7.7
Phenylalanine	36.6	13.5	31.5	6.4
Tyrosine	34.6	30.9	29.2	14.6
Proline	28.6	tr	30.4	tr
Tryptophane	12.6	—	10.5	—
Aspartic acid	68.5	16.4	61.3	17.2
Glutamic acid	62.5	34.7	57.8	38.2
Arginine	27.9	27.2	25.6	23.1
Lysine	30.6	4.3	29.2	3.7
Histidine	17.3	3.5	14.0	2.8
γ-Aminobutyric acid	0.0	38.0	0.0	23.4
Asparagine	—	224.8	—	172.0
Glutamine	—	83.5	—	55.0
Ammonia	10.6	—	10.5	—

When the amides were excluded, valine was found to contribute in somewhat greater amounts to the non-protein nitrogen in K-deficient tubers than in those with an ample K-supply. Tyrosine also occurred more abundantly in the K-deficient potatoes. The latter observation is in agreement with earlier investigations in which it was found that the content of tyrosine as well as that of *o*-diphenols is considerably higher in the case of an inadequate supply of potassium.

Glutamic acid occurred in a considerably smaller concentration in the non-protein fraction of K-deficient tubers than in that of plants amply supplied with potassium.

M a g n e s i u m. Tubers from magnesium-deficient plants were also found to be higher in protein and in soluble non-protein nitrogen than those from normal plants. Asparagine contributed slightly more to the soluble non-protein nitrogen in the magnesium-deficient samples. Alanine, glutamic acid and γ-aminobutyric acid occurred less abundantly in Mg-deficient tubers, threonin, the leucines, phenylalanine, arginine and histidine occurred in somewhat greater amounts.

Effect of mineral nutrition on the nutritional value of potato tubers.

From a nutritional point of view the amounts of amino-acids per unit of dry matter, contained in the protein and occurring in the free state, are important data; therefore, in Tables XI to XIV these values have been calculated per 10 g of dry matter. It will be seen that, for a number of amino-acids, the amount occurring in the protein is more than 10 times higher than that occurring in the free state. In the case of serine, cystine, threonine, valine, methionine, tyrosine, aspartic acid, glutamic acid and arginine, however, the free amino-acids contribute considerably more to the total amount of amino-acids per unit of dry matter. In the case of tyrosine, glutamic acid and arginine the free amino-acids may constitute 30–50 per cent of the total amount.

When a comparison is made between samples of varying manurial treatment, it will be seen that large differences occur. Tubers from plants amply supplied with nitrogen contained from 30 to 40 per cent higher amounts of practically every amino-acid. The content of γ-aminobutyric acid, asparagine and glutamine was more than doubled. In the case of phosphorus and potassium, the tubers from

TABLE XIV

Effect of the potassium nutrition of potato plants on the amino-acid content of the tubers (calculated as mg amino-acid per 10 g of dry matter, *cf* Table IX)

Amino compound	Low-K potatoes		High-K potatoes	
	Protein fraction	Soluble non-protein fraction	Protein fraction	Soluble non-protein fraction
Glycine	25.9	tr	23.8	tr
Alanine	23.7	3.3	17.6	1.8
Serine	31.3	6.6	24.6	2.6
Cystine + cysteine	5.4	0.7	2.9	0.5
Threonine	27.5	2.7	23.8	2.2
Valine	36.7	13.2	31.2	6.3
Methionine + methionine sulphoxide	11.9	5.0	8.2	4.0
Leucines	94.3	6.7	73.4	5.4
Phenylalanine	29.7	5.2	21.7	3.4
Tyrosine	18.3	12.6	18.9	4.6
Proline	21.0	tr	14.8	tr
Tryptophane	10.3	—	7.8	—
Aspartic acid	46.9	12.3	33.2	6.1
Glutamic acid	51.7	21.9	41.0	22.7
Arginine	20.5	12.0	13.9	6.3
Lysine	19.9	tr	16.0	tr
Histidine	14.0	3.0	12.3	2.5
γ-Aminobutyric acid	0.0	21.8	0.0	17.0
Asparagine	—	159.0	—	65.3
Glutamine	—	89.7	—	39.9
Ammonia	8.6	—	6.6	—

TABLE XV

Content of protein and soluble non-protein nitrogen of different potato varieties

Variety	N as % of dry matter		Ratio, protein-N: soluble non-protein-N
	Protein	Soluble non-protein	
Noordeling	1.02	0.85	1.20
Voran	0.89	0.97	0.92
Rode Star	0.77	0.65	1.18
Gloria	0.80	0.65	1.23
Libertas	0.68	0.54	1.26
Eigenheimer	0.84	0.76	1.11
Bintje	0.65	0.82	0.79
Bevelander	0.86	0.85	1.01
Eersteling	0.85	1.12	0.76
Industrie	0.75	0.64	1.17
Wilpo	0.80	0.69	1.16
Record	0.85	0.64	1.33

deficient plants had considerably higher contents of most amino-acids.

Amino-acid content of different potato varieties

It is a well-known fact that, independently of nutritional conditions, different potato varieties may have a widely diverging nitrogen and protein content. Table XV gives the results of an experiment with 12 potato varieties grown on the same field under similar conditions. It will be seen that not only the contents of

TABLE XVI

Composition of the soluble non-protein fractions of the potato varieties Noordeling and Voran, stored for 4 months (Exp. 1466, 1953)

Amino compound	Noordeling				Voran			
	Soluble non-protein fraction				Soluble non-protein fraction			
	g am.-acid per 16 g am.-acid-N *)		N as % of sum of est. am.-acid-N *)		g am.-acid per 16 g am.-acid-N *)		N as % of sum of est. am.-acid-N *)	
	incl. the amides	excl. the amides	incl. the amides	excl. the amides	incl. the amides	excl. the amides	incl. the amides	excl. the amides
Glycine	tr	tr	tr	tr	tr	tr	tr	tr
Alanine	0.7	2.6	0.7	2.6	1.2	3.5	1.2	3.5
Serine	0.9	3.2	0.7	2.6	1.0	2.9	0.9	2.4
Cystine + cysteine	0.3	0.9	0.1	0.5	0.2	0.7	0.1	0.4
Threonine	0.5	2.0	0.4	1.5	0.5	1.5	0.4	1.1
Valine	2.3	8.3	1.7	6.2	3.0	8.5	2.2	6.3
Methionine + methionine sulphoxide	tr	tr	tr	tr	tr	tr	tr	tr
Leucines	1.8	6.7	1.2	4.5	2.2	6.4	1.5	4.3
Phenylalanine	1.0	3.6	0.5	1.9	1.3	3.6	0.7	1.9
Tyrosine	1.9	6.8	0.9	3.3	2.5	7.1	1.2	3.4
Proline	tr	tr	tr	tr	tr	tr	tr	tr
Tryptophane	—	—	—	—	—	—	—	—
Aspartic acid	4.1	14.8	2.7	9.8	6.9	19.6	4.6	12.9
Glutamic acid	9.2	33.2	5.4	19.7	10.6	30.2	6.3	18.0
Arginine	3.3	12.0	6.7	24.2	4.1	11.8	8.3	23.7
Lysine	tr	tr	tr	tr	tr	tr	tr	tr
Histidine	1.5	5.3	2.5	8.9	1.4	4.0	2.4	6.8
γ-Aminobutyric acid	4.7	17.2	4.0	14.6	6.4	18.1	5.4	15.4
Asparagine	41.8	—	55.4	—	40.1	—	53.1	—
Glutamine	14.1	—	16.9	—	9.8	—	11.8	—
Ammonia	—	—	—	—	—	—	—	—

*) 72.6% of the Kjeldahl nitrogen of the soluble non-protein fraction of the variety Noordeling is accounted for by the amino-acid analyses of this fraction. For the variety Voran this value is 74.5%.

TABLE XVII

Comparison between the amino-acid composition of the protein of tubers from crosses *) between different *Solanum* species × *Solanum tuberosum* and the average values for eight different samples from *Solanum tuberosum* (*var.* Noordeling)

Amino compound	Grams of amino-acid per 16 g of N					
	WT 41-7	AT	DTT 9-8	51-155-13	51-42-2	Noordeling
Glycine	3.9	3.9	3.7	4.0	3.5	5.5
Alanine	4.1	5.2	4.2	4.0	4.4	5.0
Serine	4.3	4.0	4.2	4.6	4.0	6.2
Cystine + cysteine	0.8	1.2	0.8	0.8	0.7	0.9
Threonine	4.3	4.2	4.0	3.3	3.7	6.2
Valine	7.9	7.8	8.9	8.4	8.3	6.7
Methionine + methionine sulphoxide	1.8	1.9	1.8	1.8	1.7	2.4
Leucines	18.2	17.4	20.2	18.8	19.1	16.5
Phenylalanine	—	—	—	—	—	5.2
Tyrosine	4.6	5.7	—	5.8	6.1	4.5
Proline	5.1	4.0	4.2	5.5	4.0	4.0
Tryptophane	—	—	—	—	—	1.9
Aspartic acid	10.7	12.0	11.5	11.5	11.6	9.5
Glutamic acid	10.3	10.7	10.6	8.8	8.6	9.7
Arginine	3.9	3.6	3.8	3.6	4.0	4.3
Lysine	3.4	3.3	3.9	3.7	3.2	4.2
Histidine	—	—	—	—	—	2.7
γ-Aminobutyric acid	0.0	0.0	0.0	0.0	0.0	0.0
Asparagine	—	—	—	—	—	—
Glutamine	—	—	—	—	—	—
Ammonia	1.5	1.5	1.6	1.4	1.5	1.6

*) Samples received from Dr. H. J. Toxopeus, Institute of Agricultural Plant Breeding Wageningen. WT 41—7 = *S. andigenum* × *S. tuberosum;* AT = *S. andigenum* (resistant to the potato root eelworm) × *S. tuberosum;* DTT—9—8 = (*S. demissum* × *S. tuberosum*) × *S. tuberosum*; 51—41—2 = (*S. andigenum* × *S. demissum*) × *S. tuberosum*; 51—153—13 = (*S. demissum* × *S. rybinii*) repeatedly backcrossed with *S. tuberosum*. All varieties of *S. andigenum* used are very different from each other.

protein and soluble non-protein nitrogen vary considerably but also the ratio, protein nitrogen: soluble non-protein nitrogen. The latter is of considerable interest from a nutritional point of view as the soluble non-protein fraction consists of more than 50 per cent of amides which have a low nutritional value. The variety Noordeling which was employed in most of the above-mentioned experiments had a ratio, protein nitrogen : soluble non-protein nitrogen, of 1.20 as compared with 0.92 in Voran.

In a subsequent experiment, in which tubers of the variety Noordeling contained 102.7 mg protein-N and 69.7 mg soluble non-protein-N and those of the variety Voran 54.7 mg protein-N and

54.6 mg soluble non-protein-N per 10 g of dry matter, the non-protein fraction was analysed for amino-acids. Table XVI contains the results of this investigation. It will be seen that the difference between the two varieties in amino-acid composition of this fraction was relatively small. Asparagine and particularly glutamine contributed considerably more to the nitrogen of this fraction in the Noordeling than in the Voran varieties. This is in agreement with the nutrition experiments described above in which a high concentration of soluble non-protein nitrogen was correlated with a high proportion of the nitrogen of this fraction occurring in the amide form.

No data concerning the protein content of the two varieties are available. Protein analyses were made on a number of crosses between different *Solanum* species × *Solanum tuberosum*. For comparison the average values for 8 analyses of the variety Noordeling are given (see Table XVII). It will be seen that small differences occur in the contents of glycine, serine, threonine, valine, the leucines and aspartic acid.

DISCUSSION

The purpose of the present study was to investigate the effect of the mineral nutrition of potato plants on the amino-acid composition of the protein and the soluble non-protein fractions of the tubers. Such information is not only important from a plant-physiological point of view, but is also of interest in the study of human and animal nutrition. In many countries the potato is one of the main foodstuffs and according to Groot [3]) 10–25% of the total amount of protein consumed in human nutrition in the Netherlands consists of potatoes.

The separation between the protein and the soluble non-protein fractions was made by heat coagulation after the addition of a few drops of acetic acid and a small amount of sodium chloride. The amount of precipitated nitrogen compound was found to agree well with the amount precipitated by trichloroacetic acid. The latter is generally considered to represent the protein fraction.

The protein as used in the present investigation was of a satisfactory purity (approximately 90%). In contrast to the alcohol-insoluble fraction, analysed by Thompson and Steward, [14]) it did not contain the peptides.

From the investigations of Kiesel *et al.* [7]), Groot *et al.* [4]), and Hofstee [5]) it has become clear that the protein of the potato tuber consists of at least two different fractions, viz. one with globulin characters (insoluble in water after dialysis), the so-called tuberin, and a second which behaves like an albumin (soluble in water after dialysis), the so-called tuberinin. These fractions occur in a ratio of 7 : 3; both are coagulated by heat so that it may be concluded that the protein analysed in the present investigation consists of a mixture of tuberin and tuberinin. Kiesel *et al.* have analysed both fractions for a number of amino acids; they found no differences between them.

Although, in the past, a number of analyses have been made of the amino-acid composition of potato protein, it was not before the development of the quantitative paper-chromatographic method by Thompson and Steward [12]) [13]) that the complete composition of potato protein was given [14]). It must be stressed, however, that these authors did not hydrolyse the isolated protein

TABLE XVIII

Comparison of the amino-acid composition of potato-tuber protein as obtained by various authors (g amino-acid per 16 g protein-N)

Amino-acid	Sjollema and Rinkes [9]) (tuberin)	Kiesel *et al.* [7]) tube-rin	Kiesel *et al.* [7]) tube-rinin	Groot [3])	Slack [10])	Thompson and Steward [14])	Present paper (average of 8 samples)
Glycine	—	—	—	—	—	5.6	5.5 ± 0.16
Alanine	4.9	—	—	—	—	6.2	5.0 ± 0.29
Serine	—	—	—	—	—	3.7	6.2 ± 0.30
Cystine + cysteine	4.4	—	—	0.9	2.1	—	0.9 ± 0.11
Threonine	—	—	—	6.9	5.9	3.2	6.2 ± 0.21
Valine	1.1	—	—	7.6	6.1	7.7	6.7 ± 0.18
Methionine + meth. sulphoxide	—	—	—	2.6	2.3	1.5	2.4 ± 0.19
Leucines	12.2	—	—	14.6	17.5	17.6	16.5 ± 0.39
Phenylalanine	3.9	—	—	5.9	6.6	12.7	5.2 ± 0.10
Tyrosine	4.3	3.6	3.7	—	—	3.9	4.5 ± 0.22
Proline	3.0	3.9	4.2	—	—	3.5	4.0 ± 0.21
Tryptophane	—	—	—	2.3	1.6	—	1.9 ± 0.04
Aspartic acid	—	—	—	—	—	12.0	9.5 ± 0.45
Glutamic acid	4.6	—	—	—	—	14.6	9.7 ± 0.09
Arginine	4.2	6.6	6.4	4.8	6.0	6.0	4.3 ± 0.37
Lysine	3.3	—	—	3.6	7.7	8.0	4.2 ± 0.18
Histidine	2.3	2.0	2.2	2.2	2.2	—	2.7 ± 0.11
Ammonia	1.8	—	—	—	—	—	1.6 ± 0.03

but the residue which was left after thorough extraction of ground potato tissue with hot alcohol. This treatment presumably left the peptides in the insoluble fraction. A comparison of the results obtained by various authors is given in Table XVIII.

It will be seen that, for a number of amino-acids, the values found by Thompson and Steward [14]) agree satisfactorily with those found in the present investigation. For alanine, phenylalanine, aspartic acid, glutamic acid, arginine and lysine, higher values were found by Thompson and Steward [14]); for serine, threonine, and methionine, lower values were found by these authors. In the case of threonine, methionine, and phenylalanine the values obtained in the present study are in agreement with those of the other authors mentioned. The value for phenylalanine of Thompson and Steward [14]) which was determined chromatographically is presumably too high, due to contamination with some leucine which produces, weight for weight, much more colour than phenylalanine. Phenylalanine should be estimated by a separate method. The values for arginine and lysine which are considerably lower in the present study then in the work of Thompson and Steward [14]) are similar to those of Groot [3]) and Sjollema and Rinkes [9]), while the much higher values of Thompson and Steward [14]) agree with those of Slack [10]).

It must be stressed that Groot [3]) analysed the same variety as used in the present investigation, viz. Noordeling. Although it is possible that the protein of different varieties of potato and that of potatoes grown under widely differing climatic conditions is different in amino-acid composition, the evidence obtained so far for this hypothesis is very poor. Steward and Thompson [11]) give the values for two varieties to show that varietal differences in amino-acid composition of tuber protein occur. It is highly probable, however, that the differences recorded by these authors depend on an erroneous calculation. In the protein of their variety Sebago 14 per cent amide-N was found, in the case of the variety Katadin no value for amide-N is given. On the assumption that the amide content of the latter variety would also have amounted to 14 per cent, all values for this variety have to be multiplied by 0.86 to make them comparable to those for the variety Sebago. When this calculation is carried out almost exactly the same amino-

acid composition of the tuber protein is found in both varieties.

The data of Table XVII of the present paper give some indication that the amino-acid composition of the protein from crosses between different *Solanum* species × *Solanum tuberosum* is slightly different from the average value found in Noordeling. The differences are small, however, and since the tubers of the crosses and those of Noordeling were grown in different years, and on different sites, it is highly questionable whether these data may be used as evidence for varietal differences in the protein composition of potato tubers.

The main conclusion of the present investigation is that the amino-acid composition of potato protein is independent of the mineral nutrition of the plants. This was found to be true of nitrogen, phosphorus and potassium supply. Although the protein content of tubers from plants with pronounced symptoms of N-, P-, or K-deficiency may be quite different from that of plants with an ample supply of these nutrient elements, the amino-acid composition of the protein of these tubers is not affected by the mineral nutrition.

The composition of the soluble non-protein fraction is much less constant than that of the protein fraction. It is affected by the mineral nutrition, variety, and presumably by other factors like climatic conditions, age, *etc.*

Tubers from plants with a liberal nitrogen supply, which generally have a considerably higher content of soluble non-protein nitrogen, were found to contain a greater proportion of this fraction in the form of amides than those from plants with a moderate or a poor nitrogen supply (70.8 and 57.5% respectively in the samples tested); the glutamine content was increased even more than that of asparagine. Apparently both amides may be used as storage products for nitrogen. This conclusion is not in agreement with previous statements in the literature according to which asparagine should have the main function in the storage of ammonia.

In the case of phosphorus deficiency and potassium deficiency the potato tubers were higher in protein and particularly in soluble non-protein nitrogen. The contribution of the amides to the latter fraction was considerably greater in phosphorus- and potassium-deficient tubers than in those with a normal supply of these nutrients. This rise in relative amide-N was completely accounted for by asparagine in the case of P-deficiency. In one experiment

the relative glutamine content was even considerably lower in P-deficient tubers. In the case of K-deficiency both asparagine and glutamine contributed to the rise in relative amide content.

Of the free amino-acids, the relative contents of tyrosine, glutamic acid, arginine and γ-aminobutyric acid were found to be affected by the mineral nutrition.

Tyrosine occurred much more abundantly in K-deficient potatoes than in those with a normal K-supply. This is in agreement with earlier investigations [8]) in which the high tyrosine content of K-deficient potatoes was found to be related to the high liability of such tubers to enzymatic blackening. In phosphorus-deficient tubers the tyrosine content was lower than in normal tubers.

Glutamic acid occurred more abundantly in the non-protein fraction of nitrogen-deficient potatoes than in that of normal tubers. Potassium-deficient potatoes which are always rich in non-protein nitrogen behaved in a similar way: their relative glutamine content was considerably lower than that of tubers with an ample K-supply. Phosphorus-deficient tubers, however, although also considerably higher in soluble non-protein nitrogen than tubers with a normal P-supply were as high as or higher in relative glutamic-acid content than the latter.

Arginine was found in greater amount in the non-protein fraction of potatoes with an ample nitrogen supply than in nitrogen-deficient tubers; P-deficient and K-deficient potatoes behaved in a similar way. This amino acid, like the amides, is apparently associated with ammonium storage.

γ-Aminobutyric acid contributed considerably more to the soluble non-protein nitrogen in tubers from plants dressed amply with nitrogen than in nitrogen-deficient tubers. This was true to some extent also in the K-deficient potatoes as compared with the K-manured ones, but the phosphorus-deficient tubers behaved quite differently; their relative γ-aminobutyric acid content was quite low when compared with normal plants.

From a nutritional point of view it is important to know that the free amino-acids in some cases may contribute considerably to the total amount of amino-acids contained in the potato. In the case of amino-acids essential to human nutrition (threonine, valine, methionine, leucine and iso-leucine, phenylalanine, tryptophane and lysine) this is true of valine, methionine, phenylalanine and

possibly tryptophane. Lysine, which, from a nutritional point of view, may occur in somewhat too low a concentration in potato protein, is only present in trace amounts in the soluble non-protein fraction. The fact that mineral nutrition may have a pronounced effect on the protein content, and as a result of this on the amino-acid content of potatoes is also important for the human and animal nutrition specialist. Tubers from plants supplied amply with nitrogen may contain 30 per cent more of the essential amino-acids than those from N-deficient plants. Tubers from P-deficient and K-deficient plants behave in general like those from plants manured with ample nitrogen. It must be stressed that this effect of mineral nutrition on amino-acid content works mainly through the protein content of the tuber and only to a minor extent through the soluble non-protein fraction. The latter fraction consists of 50–60 per cent of amides which have only a minor value in human nutrition; increased nitrogen supply may raise this percentage to 70–75 per cent.

SUMMARY

The paper-chromatographic method of Thompson and Steward [12] [13] has been used for the estimation of the amino-acid composition of the protein and of the free amino-acids in potato tubers grown under carefully controlled manurial conditions. A comparison has been made between tubers from plants deficient in nitrogen, phosphorus and potassium, respectively, and those from plants grown on the same field with an optimum supply of these elements.

Although the mineral nutrition of the potato plant may have a pronounced effect on the protein content of the tubers, the amino-acid composition of the protein was found to be independent of the supply of nitrogen, phosphorus and potassium.

The amino-acid composition of the soluble non-protein fraction was found to vary considerably under the influence of a different mineral nutrition. Conditions which increase the nitrogen content of the potato (ample N, deficiency in P or K) increase the proportion of the amides in the soluble non-protein fraction. Both asparagine and glutamine were found to be associated with nitrogen storage.

Arginine and in some cases, γ-aminobutyric acid, behaved like the amides; in P-deficient tubers, however, the relative content of γ-aminobutyric acid was much lower than in normal tubers, although the total non-protein-N was high.

The tyrosine content of P-deficient tubers was lower, that of K-deficient tubers much higher than normal. Glutamic acid contributed much less

abundantly to the soluble non-protein fraction when the nitrogen supply of the plants was high.

From the point of view of human and animal nutrition it is important to know that in some cases the free amino-acids may contribute considerably to the total amino-acids of the potato tubers, furthermore, that the mineral nutrition may have a pronounced effect on the amino-acid content of the potato, mainly through its effect on the protein content.

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