Parameters for Carrot Quality

and the development of the Inner Quality concept

met Nederlandse samenvatting: Parameters voor wortelkwaliteit
en de ontwikkeling van het Innerlijke Kwaliteitsconcept

mit deutscher Zusammenfassung: Parametern für Möhrenqualität
und die Entwicklung des Begriffs der Inneren Qualität

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About the organic Food, Quality and Health association (FQH)

International Research Association for Organic Food Quality and Health – Association of research institutes and stakeholders, that aims to stimulate research in the field of organic food quality and health, to present publications, and to organise scientific conferences and exhibitions.
Website: www.organicfqhresearch.org

About the Louis Bolk Instituut

The Louis Bolk Instituut has been a pioneer in innovative scientific research on farming, nutrition and healthcare since1976. Broadening the basis of scientific research is the aim of the institute's work. Where conventional research methods do not suffice, new methods are looked for, such as: phenomenology, participatory research, pictomorphology and conscious-intuitive methods, Rudolf Steiners philosophy being a source of inspiration.

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Colofon

Louis Bolk Instituut, 2004, publication number GV V 05
organic Food, Quality and Health publication number FQH 04
ISBN 90-74021-34-4 Price € 15,-, without postage.
Also on www.louisbolk.nl
English editor: Anne MacDonald
German translation: Elke Bussler

Keywords: organic food quality, inner quality concept, carrot, fertilisation, nitrogen, light, morphology, saccharose, dry matter, nitrate, protein ratio, carotenes, sensoric properties, luminescence, biophotones, copper chloride crystallisations, biocrystallisation, pH, redox potential, electrical resistance, storage test.

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1.1 English summary

Parameters for carrot quality and development of the Inner Quality concept

Martin Northolt, Geert-Jan van der Burgt, Thiemo Buisman, Arne Vanden Bogaerde, Louis Bolk Instituut, 2004

Motivation

Consumers expect organic farming to produce healthy products that taste good. Indeed, many organically grown products have won the acclaim of the best chefs de cuisine, but organic certification does not *guarantee* good taste or a healthy product. In organic agriculture, as in conventional agriculture, more and more emphasis is laid on higher fertilisation levels, higher yields, earlier harvests and sometimes longer trade chains. All these factors can affect the quality experienced by consumers.
Food quality is more than the sum of exterior characteristics, distinct component substances and the absence of harmful contaminants. Consumers of organic products expect properties such as tastiness, ripeness, 'vitality' and 'coherence', which are not easy to define or measure. To assess this expectation, we need to develop a new quality concept: Inner Quality.

International Research Association for Organic Food Quality and Health (FQH)

The International Research Association for Organic Food Quality and Health (FQH) (www.organicfqhresearch.org) was established to promote research into the health effects of good quality organic food. This type of research requires a coherent concept of quality with empirical parameters and a research methodology for measuring health effects. The research presented in this report thus serves the long-term research objective of FQH.
In the first two FQH apple reports, the provisional concept of Vital Quality was introduced, and later further developed into Inner Quality. The present carrot study is a further step towards finding and defining relevant parameters for the Inner Quality of products. This must be done before the next question can be looked at, i.e. whether products with a high Inner Quality can indeed benefit health.

Partners and financers

The carrot experiment took place at the biodynamic Warmonderhof farm (NL). The participating institutions were Kwalis Qualitätsforschung Fulda GmbH (D), Meluna Bio-photon Research, Geldermalsen (NL), University of Kassel - Witzenhausen (D), Elektro-Chemisches Qualitätslabor (EQL) (D), Biodynamic Research Association, Denmark (BRAD) (DK) and Louis Bolk Instituut (LBI) (NL).
The project was financially supported by Stichting Triodos Fonds (NL), Rabobank (NL), Software AG Stiftung (D), Zukunftstiftung Landwirtschaft (D), and by all the participants mentioned.

Inner Quality: a coherent quality concept appropriate for living produce

In the second apple report, Inner Quality is defined as follows (with some words modified):
Inner Quality is an extension of the common quality criteria on exterior, presence of wanted and the absence of unwanted substances, and hygienic standards. Inner Quality refers to properties which result in a crop-specific product that is ripe, tasty and has sufficient storage quality. These properties develop during the growing season as a result of growth and differentiation processes and are dependent on the balanced relation between these processes. A similar concept may be developed in the future for products of animal origin.

The quality concept must meet various requirements. It must, of course, tie in with growers' practices, regardless of whether they are organic or conventional. It is, after all, the grower who nurtures the life processes in crops or livestock. The growth and differentiation processes can occur in varying proportions and degrees of mutual interaction (integration).
The quality concept should also establish a link between consumers' perception of product quality in the store and growers' perception of their crops. This will enable the grower to make adjustments in the course of the growing season, taking advantage of seasonal and soil variations, to realise optimum quality in the end product. Consumers' wishes are never uniform: their food preferences depend on individual preferences, and on their health and mood. There is a market for produce with different good qualities. So in the concept, there is no one uniform optimal quality for everyone. The quality concept presented in Table 1 therefore consists of three interlinked dimensions: crop management, life processes and properties of harvested products.

Methodology to develop the new quality concept

This carrot study is part of the FQH-related research programme of Louis Bolk Instituut to design and evaluate the new quality concept. The first apple study was primarily concerned with the design of the quality concept, and also examined growth and differentiation processes and investigated how these can be measured. In the second apple study, a course of validation for the concept was described and partly executed. Growers and researchers of other crops were approached to see whether they could recognize and subscribe to the concept and the two life processes and their integration (*face validity and content validity*). They agreed on the concept of growth and differentiation. Some examples of crops were published in a brochure on life processes in food crops. Difficulties arose with the concepts of integration and the term 'vitality', and work was done to improve them. The term 'vitality' was dropped because it caused too much confusion, and integration was defined as the proportion of growth and differentiation and the interaction or intermingling of the two.
A new path was taken in distinguishing the processes of growth and differentiation per organ (in the case of apple: fruit, blossom, shoot) and demonstrating the development during the growing season. Plant hormones were skipped as an aspect of growth processes.
The *consistency of the theoretical construction* was checked by comparing the quality concept with various hypotheses and results from literature. The concept agreed, e.g. with the growth-differentiation-balance hypothesis used in plant ecology research on resistance to pests and diseases.

Aims

To further understand and validate the Inner Quality concept, research was done on carrots, with the following goals:

1. To demonstrate the Inner Quality concept for carrots by showing the life processes growth, differentiation and integration of the two, by generally accepted and experimental parameters. The life processes are varied experimentally by different levels of nitrogen and light and different days of harvest (time axis). From literature and experience, it is known that nitrogen and light influence the intensity of growth and differentiation processes. A high nitrogen level stimulates growth and inhibits differentiation; light stimulates differentiation more than growth and, during the growth season, the intensity of differentiation increases. The integration factor involves the relation and interaction between the growth and the differentiation processes.
2. To select generally accepted parameters and to relate them with the experimental parameters for the different processes in carrots. Moreover, to select parameters which are easily measurable and cheap to use in practice.
3. To verify whether growth, differentiation and integration are relevant determinants of the inner quality of carrots by use of the experimental results.
4. To search for factors which clearly determine the integration of the growth and differentiation processes, in order to enable the growers to maximise the Inner Quality of the carrot root.

Table 3. The concept for Inner Quality for carrots, based on growth and differentiationprocesses and their integration
(a: validated in this study)

Inner Quality for carrots		
CROP MANAGEMENT FACTOR	PROCESSES	CARROT PARAMETERS
1. Growth		
• no limits in nutrients and water • warmth by ridges	• photosynthesis: primary metabolism • absorption of nitrogen and other nutrients • forming cells, tissues and organs • maintenance of basic metabolism	• high weight of leaves and root[a] • dark green, coarse leaves • growing leaves at harvest • high nitrate content[a] • little carrot taste • juicy and crunchy root • high emission of delayed luminescence[a]
2. Differentiation		
• light by large plant distance • reduced fertilisation	• refining, ordering • ripening: monosaccharides -> saccharose • secondary metabolisms	• fine leaves • discolouration of leaves at harvest • low leaves/root weight ratio • cylindrical root • stumpy root[a] • high saccharose content[a], sweetness • high dry matter content[a] • high carotenes content[a] • strong carrot taste • high emission ratio of delayed luminescence • pH[a]
Integration of 1 and 2		
• optimal proportion of growth and differentiation • appropriate varieties • disease-preventing soil • diversity of agro-ecosystem	• balanced relation of growth and differentiation, also depending on cultivar and time	• resistance to pests and disease • total sensoric appreciation • storability properties[a]

Design of the carrot experiment

A field experiment with carrots with completely randomised plots in three repetitions was designed. Three nitrogen levels were created at the levels: 0, 100 and 200 kg N per ha, in addition to the background soil nitrogen availability. Nitrogen was applied by means of blood meal pellets and feather meal pellets before and after sowing. Three levels of light were created using shadow nets, with 52, 85 and 100% light. The cultivar Rodelika was used, an open-pollinating cultivar from the biodynamic breeding programme of Dottenfelderhof. At intervals of two weeks, carrots were harvested and analysed for the fresh weight of leaves and root, morphology and dry matter of root. At three major harvests in August, September and

October, analyses also took place of saccharose, D-glucose and D-fructose, nitrate, total nitrogen, carotenes, sensory properties and the experimental parameters: pure/crude protein ratio, copper chloride crystallisation, delayed luminescence and electro-chemical parameters. During the whole growth period, pests and diseases were recorded.
Nitrogen levels differed significantly from day 41 after sowing. Shadow nets were set up at the beginning of July, creating the intended light differences in the second half of the season. Some morphological and chemical results showed significant unwanted differences between the field repetitions, which necessitated the use of average values measured in the three field repetitions, and thus influenced the statistical analysis of the study.
The warm, dry summer also reduced the effective differences in nitrogen levels applied.

Conclusions regarding the experiment and the parameters

- The morphological parameters generally responded best to light, then to time, and less to nitrogen. In general, results corresponded to expectations. The cortex/core ratio was an exception; an increase in time was expected, indicating according to literature a ripening process, but a decrease was measured. Most morphological parameters are easily observed by farmers. The absolute value will vary, depending on the cultivar used, the soil and the year (weather). This means that, given a cultivar and a soil (or a farm), morphological parameters can be used as indicators for growth, differentiation and integration, although it will be difficult to set fixed values as references to be used for a quality indication.
- Pests and diseases occurred infrequently and bore no relation to light or nitrogen level. This does not support the idea of pests and diseases as a parameter for well-integrated life processes. On the other hand, there are more factors than just light and nitrogen, influencing integration. If we *define* pests and diseases as an expression of imbalance, this carrot crop as a whole had poorly integrated growth and differentiation processes. The increase in Alternaria infection at the final harvest and the overall presence of Mildew for the last two months indicate, according to our definition, a poor integration.
- The different chemical contents of the roots responded to light and time, and only a little to nitrogen. In literature, different yield responses of carrots to nitrogen supply are described. Nitrate content and pure/crude protein ratio did not change conform to expectations as regards the time variable. We expect nitrate to decrease and the ratio to increase over time; they reacted in exactly the opposite way. The protein ratio also decreased in the pilot study.
- The highest total sensory appreciation of the carrots was realised without additional nitrogen and in full light, as was expected.
- The copper chloride crystallisation pictures produced by LBI and BRAD were unusual compared with the Triangle study of LBI, BRAD and University of Kassel – Witzenhausen and little effect of nitrogen, light and time were discernible. This was attributed to a non-optimal proportion of carrot juice and copper chloride. It appeared to be necessary to determine the optimal concentrations in advance before examining carrot samples.
- The delayed luminescence by Kwalis showed significant effects of all four parameters regarding the light levels, in line with expectations, but they were not significantly affected by nitrogen. Also as regards time, significant effects were found, mostly as expected. Only hyperbolicity ratio was expected to increase or have an optimum in the time series, but turned out to decrease. Also the Meluna luminescence measurements (carried out on the final harvest day only) had significant effects for three parameters in the light series, with initial emission white and total emission white increasing according to expectation, but slope white increasing contrary to expectations. Nitrogen had significant effects on initial emission white and total emission white (expected) and on slope white (not expected). The initial emission / slope white ratio was influenced by light and nitrogen and may indicate to integration.
- The electro-chemical parameters of Kassel had significant effects in the light and time series, but not in the nitrogen series. The parameter pH reacted in line with expectations. The redox potential and electrical resistance in the light series corresponded roughly to expectations, but those of the time series were contrary to expectations. Of the electro-chemical parameters of EQL (measured on the final harvest day only), pH (significant) and electrical resistance (tendency) increase in the light series as expected. With regard to nitrogen levels, significant effects are found for redox potential and electrical resistance with an optimum at medium nitrogen level, which was not expected.

- The storage test showed better storability at low nitrogen levels and full light, as was expected. Minimal loss of weight was found in the carrots from the expected optimal harvest day.
- Five parameters reacted in the opposite way to the variable time than was expected: cortex/core ratio (tendency), nitrate, pure/crude protein ratio (tendency), redox potential (tendency) and electrical resistance. Probably these parameters do not react to nitrogen when nitrogen occurs at a high level.

Conclusions regarding the quality concept

In Table 1a, the results of the study with respect to the quality concept are summarised.
The growth processes are measured by the fresh weight of leaves and root, and nitrate content and emission 30-50 white of delayed luminescence of root. They could not be demonstrated by differences in the greenness and coarseness of leaves or growing rate of leaves at harvest, probably because of the high base level of soil nitrogen. Also no correlation was found with carrot taste, juiciness and crunchiness. The carrot taste and core/cortex ratio of root could not be correlated with growth or differentiation processes.
The differentiation processes are measured by stumpiness, saccharose and carotenes content, dry matter and emission 30-50 ratio of delayed luminescence of root.
The integration of the two processes is, by definition, measured by total sensory appreciation, storability properties and resistance to pests and disease. The first two parameters showed differences in line with expectations; the latter parameter showed no difference between treatments, probably due to lack of discriminating the onset of the diseases. Slope white of delayed luminescence may be a differentiation parameter but reacted not accordingly, and needs further elaboration.
Integration is considered the balanced relation and interaction (or intermingling) of the growth and differentiation processes. The decrease in growth processes around the expected optimal harvest day was not found, probably because of the high level of nitrogen. The expected increase in differentiation processes was confirmed. The validation of integration needs to be further elaborated. Besides monitoring growth and differentiation parameters, integration parameters should also be measured. The measurement of integration by the parameters resistance to pests and disease, sensory properties and storability should be improved.

Recommendations for future research

- To further understand and communicate the Inner Quality concept, it is necessary to repeat the carrot study in two, preferably three countries to demonstrate the life processes and the corresponding parameters. Only a limited number of low-cost parameters should be used. These are: weight of leaves and root for growth processes; leaves/root weight ratio, stumpiness of root, saccharose, sweetness and dry matter for differentiation processes; prevalence of pests and disease, total sensory appreciation, weight loss and rot in a storage test for integration processes.
- In separate studies, delayed luminescence and copper chloride crystallisation should be investigated to validate parameters for life processes. Reference series of carrots can be obtained from the above-mentioned study.

1.2 Nederlandse samenvatting

Parameters voor peen kwaliteit en de ontwikkeling van het Innnerlijke Kwaliteitsconcept

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Motivatie

Consumenten verwachten dat de biologische landbouw gezonde, smaakvolle producten produceert. Inderdaad wordt door top-koks gezegd dat veel biologische producten een betere smaak hebben dan gangbare producten, maar het Eko-merk geeft geen garantie voor een goede smaak en een gezond product. In de biologische landbouw, evenals in de gangbare landbouw, wordt meer en meer nadruk gelegd op een zware bemesting, hoge opbrengsten, vroege oogsten en soms langere afzetketens. Al deze factoren kunnen de kwaliteit beïnvloeden, zoals die door de consument wordt ervaren.
Voedselkwaliteit is meer dan de som van uiterlijke kenmerken, enkele inhoudsstoffen en afwezigheid van schadelijke contaminanten. Consumenten van biologische producten verwachten eigenschappen zoals smaak, rijpheid, 'vitaliteit', 'coherentie' en gezondheidsbevordering welke niet gemakkelijk te definiëren en te meten zijn. Om deze eigenschappen te meten is er behoefte aan een nieuw kwaliteitsconcept: Innerlijke kwaliteit.

Internationale Research Association for Organic Food Quality and Health (FQH)

De internationale onderzoeksorganisatie Organic Food Quality & Health (www.organicfqhresearch.org) is opgericht om het onderzoek naar de gezondheidsaspecten van biologische producten te bevorderen. Dit onderzoek vereist een coherent kwaliteitsconcept met empirische parameters en een onderzoeksmethodologie voor het meten van gezondheidseffecten. Het onderzoek dat in dit rapport wordt gepresenteerd dient dus het lange termijn doel van FQH. In de eerste twee FQH appelrapporten werd het voorlopige concept van 'Vitale kwaliteit' geïntroduceerd, later werd dit verder ontwikkeld in 'Innerlijke kwaliteit'. Deze wortelstudie is een volgende stap voor het zoeken en definiëren van de relevante parameters voor de Innerlijke kwaliteit van producten. Dit moet worden gedaan voordat de volgende vraag kan worden onderzocht: zijn producten met een hoge Innerlijke kwaliteit gezonder voor de mens.

Partners en financiers

Het onderzoek met wortels vond plaats op de biodynamische boerderij Warmonderhof bij Dronten. De deelnemende instituten zijn Kwalis Qualitätsforschung Fulda GmbH (D), Meluna Biofotonen Onderzoek in Geldermalsen, Universiteit van Kassel - Witzenhauzen (D), Electro-Chemisches Qualitätslabor (EQL)(D), Biodynamic research Association Denmark (BRAD)(DK) en het Louis Bolk Instituut.
Het project werd financieel gedragen door de Stichting Triodos Fonds, Rabobank, Software AG Stiftung (D), Zukunftstiftung Landwirtschaft (D) en alle genoemde deelnemers.

Innerlijke Kwaliteit: een coherent kwaliteitsconcept voor levende producten

In het tweede appelrapport werd Innerlijke Kwaliteit als volgt gedefinieerd, met verandering van enkele woorden: Innerlijke kwaliteit is een uitbreiding van de gangbare kwaliteitscriteria voor uiterlijk en smaak, aanwezigheid van gewenste en afwezigheid van ongewenste stoffen, en hygiëne. Innerlijk kwaliteit verwijst naar de eigenschappen die resulteren in een gewasspecifiek product dat rijp en smaakvol is en voldoende bewaareigenschappen heeft.
Deze eigenschappen ontwikkelen gedurende het groeiseizoen als resultaat van groei- en differentiatieprocessen en zijn afhankelijk van een gebalanceerde relatie tussen deze processen. Een dergelijk concept kan in de toekomst ook worden ontwikkeld voor dierlijke producten.

Het kwaliteitsconcept moet voldoen aan verschillende eisen. Het moet aansluiten bij de ervaring van de telers, ongeacht of zij biologisch of gangbaar werken, omdat zij de levensprocessen in de gewassen en de dieren verzorgen. De groei- en differentiatieprocessen kunnen zich voordoen in verschillende intensiteit, verhouding en mate van onderlinge interactie (integratie).
Het kwaliteitsconcept zou ook een verbinding moeten zijn tussen de kwaliteitspercepties van de consument en de teler. Dit maakt het voor de teler mogelijk om voor en tijdens de groei van het gewas maatregelen te nemen, met gebruikmaking van variaties in seizoen en grondsoort, om de optimale kwaliteit van het eindproduct te realiseren. De wensen van de consumenten zijn niet uniform; zij hangen af van individuele verschillen, en van hun gezondheid en stemming. Er is een markt voor producten met verschillende kwaliteiten. Daarom is in het concept geen uniforme optimale kwaliteit voor iedereen. In het kwaliteitsconcept dat in tabel 1b wordt gepresenteerd zijn drie dimensies met elkaar verbonden: teeltmaatregelen, levensprocessen en eigenschappen van geoogste producten.

Methodologie voor het ontwikkelen van een nieuw kwaliteitsconcept

Dit wortelonderzoek is onderdeel van het aan FQH gerelateerde onderzoekprogramma dat het Louis Bolk Instituut uitvoert om een nieuw kwaliteitsconcept te ontwikkelen en te valideren. Het eerste appelonderzoek was gericht op het ontwikkelen van het kwaliteitsconcept , de groei- en differentiatieprocessen te demonstreren en de methoden daarvoor. In het tweede appelonderzoek werd de methode om het concept te valideren beschreven en gedeeltelijk uitgevoerd. Telers en onderzoekers werden benaderd om te zien of zij het concept en de twee levensprocessen en hun integratie kunnen herkennen en onderschrijven (*gezichtsvaliditeit en inhoudsvaliditeit*). Zij stemden in met het concept. Enkele voorbeelden werden gepubliceerd in een brochure over levensprocessen in gewassen. Omdat er problemen rezen met begrippen integratie en vitaliteit in het concept, werd gezocht naar verbetering en verduidelijking. Het begrip 'vitaliteit' kwam te vervallen omdat het in de literatuur in verschillende betekenissen wordt gebruikt. Het begrip integratie werd gedefinieerd als de verhouding van groei en differentiatie en de interactie daartussen.
Een nieuwe benadering werd gevonden door de groei- en differentiatieprocessen te beschrijven per orgaan (bij appel: vrucht, bloesem, loot) en de ontwikkeling daarvan gedurende het groeiseizoen aan te tonen. Plantenhormonen als een aspect van groeiprocessen kwamen te vervallen.
De *consistentie van de theoretische constructie* werd gecontroleerd door het kwaliteitsconcept met verschillende theorieën en resultaten in de literatuur te vergelijken. Het concept kwam daarmee goed overeen, bijvoorbeeld met de groei-differentiatie balans hypothese in het plantenecologisch onderzoek van de weerstand tegen ziekten en plagen.

Doelstellingen

Voor beter begrip en validatie van het Innerlijke kwaliteitsconcept werd het onderzoek met wortels uitgevoerd met de volgende doelstellingen:

1. Het demonstreren van het Innerlijke Kwaliteitsconcept door het aantonen van de levensprocessen groei, differentiatie en integratie van die twee, door algemeen geaccepteerde en experimentele parameters. De levensprocessen werden experimenteel gevarieerd door verschillende niveaus van stikstof en zonlicht en verschillende oogstdagen (tijd-as). Door literatuur en ervaring is bekend dat stikstof en zonlicht de intensiteit van groei- en differentiatieprocessen beïnvloeden. Een hoge stikstofgift stimuleert groei en remt differentiatie; zonlicht stimuleert differentiatie meer dan groei, en gedurende het groeiseizoen neemt de differentiatie toe. De integratie omvat de verhouding en interactie tussen de groei- en differentiatieprocessen.
2. Het selecteren van algemeen geaccepteerde parameters en ze relateren met de experimentele parameters voor de verschillende levensprocessen in wortel. Bovendien het selecteren van parameters die in de praktijk gemakkelijk en goedkoop zijn te gebruiken.
3. Het vaststellen dat groei, differentiatie en integratie relevante determinanten zijn voor de Innelijke kwaliteit van wortels.
4. Het zoeken van factoren die de integratie van groei en differentiatieprocessen duidelijk bepalen, om het de telers mogelijk te maken om de Innerlijke kwaliteit van wortels te verhogen.

Tabel 1b. Het Innerlijke Kwaliteit concept voor wortel, gebaseerd op groei, differentiatie en integratie (: gevalideerd in deze studie)*

Innerlijke Kwaliteit voor wortel		
TEELT MAATREGELEN	PROCESSEN	EIGENSCHAPPEN
om het groeiend gewas te beïnvloeden *voor communicatie met de teler*	in het gewas *voor communicatie met de teler*	van het gewas of geoogst product *voor communicatie met de teler, winkelier en consument*
1. Groei		
• geen beperking van nutriënten en water • Warmte door teelt op ruggen • Beperkte bemesting	• fotosynthese: primair metabolisme • opname van stikstof en andere nutriënten • vorming van cellen, weefsels en organen • onderhoud van basaal metabolisme	• hoog blad-en wortelgewicht* • donker groene en grove bladeren • groeiende bladeren bij de oogst • hoog nitraatgehalte • weinig wortelsmaak • sappige en knapperige wortel • hoge emissie bij luminescentie*
2. Differentiatie		
• licht door grote plantafstand • beperkte bemesting	• verfijning, ordening • rijping: monosaccharides -> saccharose • secondaire metabolismen	• fijne, symmetrische bladeren • lage loof/wortelgewicht ratio • stompe wortel* • hoog saccharosegehalte*, zoet • hoog drogestofgehalte* • hoog carotenengehalte* • sterke wortelsmaak • hoge emissie ratio bij luminescentie* • pH*
Integratie van 1 en 2		
• optimale verhouding van groei en differentiatie • passende rassen • ziektewerende bodem • diversiteit van het agro-ecosysteem	• gebalanseerde relatie van groei and differentiatie, ook afhankelijk van ras en tijd	• weerstand tegen ziekten en plagen • totale sensorische waardering • bewaareigenschappen*

Opzet van het wortelexperiment

Een experiment met wortelplanten op volledig gerandomiseerde behandelingsveldjes werd in drie herhalingen in het veld uitgelegd. Drie stikstofniveaus werden ingesteld door toevoegen van 0,100 en 200 kg N per hectare aan de bestaande beschikbare bodemstikstof. De stikstof werd toegevoegd door middel van bloedmeelpellets en verenmeelpellets respectivelijk voor en na zaaien. Drie zonlichtniveaus met 52, 85 en 100% licht werden ingesteld door schaduwnetten. Het zaadvaste ras Rodelika afkomstig van het biodynamische veredelingprogramma van Dottenfelderhof werd gebruikt. De wortelplanten werden met intervallen van 2 weken geoogst en onderzocht op nat-gewicht en morfologie van loof en wortel, en droge stofgehalte van de wortel. Bij drie grote oogsten in augustus, september en oktober werden ook bepaald: gehalten van saccharose, D-glucose en D-fructose (monosacchariden), nitraat, totaal stikstof en carotenen, sensorische eigenschappen, en de experimentele parameters zuiver/ruw eiwit ratio, koperchloride kristallisatie, luminescentie (biofotonen) en elektrochemische parameters. Gedurende het groeiseizoen werden ziekten en plagen geregistreerd.

De stikstofniveaus waren significant verschillend vanaf de 41e dag na het zaaien. De schaduwnetten werden begin juli geplaatst om in de tweede helft van het groeiseizoen de zonlichtverschillen te hebben. Bepaalde morfologische en chemische analyses vertoonden ongewenste verschillen in de veldherhalingen. Dit beïnvloedde de statistische analyse van de onderzoekresultaten.
De warme, droge zomer zorgde er ook voor dat de verschillen tussen de stikstofniveaus kleiner werden.

Conclusie over het experiment en de parameters

- De morfologisch parameters reageerden het sterkste op licht, daarna op tijd en minder op stikstof. De resultaten waren over het geheel genomen volgens de verwachtingen. De cortex/core ratio van de wortel was een uitzondering; een toename in de tijd werd verwacht dat volgens de literatuur een indicator voor rijping is, maar een afname werd gevonden. De meeste morfologische parameters kunnen gemakkelijk worden waargenomen door telers. De waarden kunnen variëren afhankelijk van ras, bodem en jaar (weer). Dit betekent dat voor een bepaald ras en bodem (bedrijf) morfologische parameters kunnen gebruikt worden als indicatoren voor groei, differentiatie en integratie, alhoewel het moeilijk is om waarden vast te stellen als referenties voor een kwaliteitsindicatie.
- Ziekten en plagen kwamen weinig voor en hadden geen relatie met het licht- en stikstofniveau. Dit ondersteunt niet de hypothese dat ziekten en plagen een parameter zijn voor goed geïntegreerde levensprocessen. Echter, er zijn meer factoren dan alleen licht en stikstof die integratie beïnvloeden. Als wij ziekten en plagen *definiëren* als een uitdrukking van onbalans, had dit wortelgewas als geheel matig geïntegreerde groei- en diferentiatieprocessen. De toename van de Alternaria infectie bij de laatste oogst en het algemeen voorkomen van Meeldauw gedurende de laatste twee maanden duiden volgens onze definitie op een matige integratie.
- De verschillende chemische inhoudsstoffen van de wortel reageerden op licht en tijd, maar weinig op stikstof. In de literatuur wordt voor wortels eveneens een beperkte invloed van stikstof op de opbrengst beschreven. Nitraatgehalte en zuiver/ruw eiwit-ratio veranderden niet volgens verwachting in de tijd. Wij verwachten een afname van nitraat en een toename van de ratio in de tijd; zij veranderden in tegenovergestelde richting. In de pilot wortelstudie nam de zuiver/ruw eiwit-ratio ook af.
- De hoogste totale sensorische waardering werd gerealiseerd zonder toevoeging van nitraat en in het volle zonlicht, zoals was verwacht.
- De koperchloride kristallisatiebeelden van LBI en BRAD waren ongewoon vergeleken met de Triangle studie van LBI, BRAD en Universiteit van Kassel - Witzenhausen en weinig effect van stikstof, licht en tijd werd gevonden. Dit werd toegeschreven aan een niet-optimale verhouding van wortelsap en koperchloride. Het blijkt dus noodzakelijk om de optimale verhouding vooraf vast te stellen.
- De luminescentiemetingen van Kwalis laten volgens verwachting significante effecten zien van alle vier parameters met betrekking tot de lichtniveaus, maar zij werden niet significant beïnvloed door stikstof. Ook ten aanzien van de tijd werden significante effecten gevonden, meestal volgens verwachting. Echter, de hyperboliciteit-ratio die in de tijd zou moeten toenemen of een optimum zou moeten hebben, bleek af te nemen. Ook de Meluna luminescentiemetingen, die alleen van de laatste oogst werden gedaan, hadden significante effecten bij drie parameters in de lichtseries, met volgens verwachting toenemende initiële-emissie-wit en totale-emissie-wit, maar de toenemende hellingshoek-wit was niet volgens verwachting. Stikstof had een significant effect op initiële-emissie-wit en totale-emissie-wit (verwacht) en hellingshoek-wit (niet verwacht). De initiële emissie/hellingshoek ratio die beïnvloed werd door licht en stikstof, zou een indicatie voor integratie kunnen zijn.
- De elektrochemische parameters van Kassel hadden significante effecten in de licht- en tijdseries, maar niet in de stikstofserie. Parameter pH reageerde volgens verwachting. De redox potentiaal en elektrische weerstand in de lichtserie reageerde ongeveer volgens verwachting, die in de tijdserie niet. Van de elektrochemische parameters van EQL, alleen van de laatste oogstdag gemeten, namen de pH (significant) en elektrische weerstand (trend) toe bij meer licht, zoals verwacht. Bij hoger stikstofniveau werden significante effecten gevonden bij redox potentiaal en elektrische weerstand met een optimum bij het middelste stikstofniveau, hetgeen niet was verwacht.
- De bewaarproef liet volgens verwachting een betere bewaarbaarheid zien bij laag stikstofniveau en volle zonlicht. Het gewichtsverlies was minimaal bij de wortels van de volgens verwachting optimale oogstdag.

- Vijf parameters reageerden in de tijd niet volgens verwachting: cortex/core ratio (trend), nitraat, zuiver/ruw eiwit ratio (trend), redox potentiaal (trend), en elektrische weerstand. Waarschijnlijk reageren deze parameters niet op stikstof bij een hoog stikstofniveau.

Conclusies over het kwaliteitsconcept

In tabel 1b worden de resultaten van het onderzoek met betrekking tot het kwaliteitsconcept samengevat. De groeiprocessen worden gemeten door het natgewicht van loof en wortel, nitraatgehalte en emissie 30-50 van de luminescentie van wortel. Zij konden niet worden gedemonstreerd door verschillen in groene kleur en grofheid van het loof of groei van het loof bij de oogst, waarschijnlijk door het hoge basisniveau van de bodemstikstof. Ook werd geen correlatie gevonden met de sappigheid en knapperigheid van de wortel. De wortelsmaak en cortex/core ratio van wortel konden niet gecorreleerd worden met groei- en differentiatieprocessen.
De differentiatieprocessen worden gemeten door stompheid, saccharose- en caroteengehalte, droge stofgehalte en emissie 30-50 ratio van de luminescentie van wortel. De hellingshoek van de luminescentie reageerde niet volgens verwachting als differentiatieparameter, en dient verder te worden onderzocht.
De integratie van de twee processen wordt, per definitie, gemeten door totale sensorische appreciatie, bewaarbaarheid en weerstand tegen ziekten en plagen. De eerste twee parameters laten verschillen zien volgens verwachting; de laatste parameter werd niet beïnvloed door de verschillende behandelingen, waarschijnlijk omdat het begin van de schimmelinfecties niet nauwkeurig werd vastgesteld. Integratie wordt beschouwd als de gebalanceerde relatie en interaktie van de groei- en differentiatieprocessen. De afname in groeiprocessen ten tijde van de verwachte optimale oogstdag werd niet gevonden, waarschijnlijk vanwege het hoge bodemstikstofniveau. De toename van de differentiatieprocessen werd wel gevonden.
De validatie van integratie behoeft verder onderzoek. Behalve het meten van groei- en differentiatieparameters moeten ook integratieparameters gemeten worden. Het meten van integratie door de parameters weerstand tegen ziekten en plagen, sensorische eigenshappen en bewaarbaarheid moet worden verbeterd.

Aanbevelingen voor verder onderzoek

- Voor verder begrip en communicatie van het Innelijke kwaliteitsconcept, is het nodig om de wortelstudie te herhalen in twee, bij voorkeur drie landen waarbij de levensprocessen en de daarbij behorende parameters worden gedemonstreerd. Slechts een beperkt aantal goedkope parameters worden gebruikt. Deze zijn: nat-gewicht van loof en wortel voor groeiprocessen; loof/wortelgewicht ratio, stompheid van wortel, saccharose, zoetheid en droge stofgehalte voor differentiatieprocessen; voorkomen van ziekten en plagen, totale sensorische waardering, gewichtsverlies en rot tijdens een bewaarproef voor integratie.
- In afzonderlijke studies moeten luminescentie en koperchloride kristallisatie (voedingskristallisatie) worden onderzocht om parameters voor de levensprocessen te valideren. Daarvoor kunnen referentie-series van wortels uit bovenstaande studie worden verkregen.

1.3 Deutsche Zusammenfassung

Parameter für die Qualität von Möhren

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Louis Bolk Institut, 2004.

Begründung

Konsumenten erwarten, dass in der ökologischen Landwirtschaft gesunde und wohlschmeckende Produkte erzeugt werden. Viele Produkte aus ökologischem Anbau werden in der Tat von Spitzenköchen gerühmt, doch stellt ein Öko-Zertifikat noch keine *Garantie* für ein wohlschmeckendes und gesundes Produkt dar. Ebenso wie in der herkömmlichen Landwirtschaft wird auch in der ökologisch Landwirtschaft in zunehmendem Maße Wert gelegt auf höhere Düngermengen, höhere Erträge, frühere Ernten und bisweilen längere Handelsketten. All diese Faktoren können sich auf das Qualitätserleben des Verbrauchers auswirken.
Die Qualität eines Nahrungsmittels ist mehr als die Summe äußerlicher Merkmale, ein paar charakteristische Inhaltsstoffe und das Nichtvorhandensein schädlicher Verunreinigungen. Konsumenten ökologischer Produkte verlangen Eigenschaften wie Schmackhaftigkeit, Reife, "Vitalität" und "Zusammenhang", die nicht einfach zu definieren oder zu messen sind. Um diese Erwartungen zu erfüllen, müssen wir einen neuen Qualitätsbegriff entwickeln: Innere Qualität.

Internationale Forschungsvereinigung für ökologische Lebensmittelqualität und Gesundheit (FQH)

Die *International Research Association for Organic Food Quality and Health* (FQH) (www.organicfqhresearch.org) wurde gegründet, um die Forschung über die Gesundheitseffekte hochwertiger ökologischer Nahrungsmittel zu fördern. Dazu ist ein kohärenter Qualitätsbegriff mit nachvollziehbaren Parametern und Forschungsmethoden zur Beurteilung von Gesundheitseffekten erforderlich. Das langfristige Forschungsziel der FQH bildet den Rahmen der hier vorgelegten Untersuchung.
In den ersten beiden FQH-Untersuchungen an Äpfeln wurde zunächst der vorläufige Begriff der Vitalen Qualität eingeführt, der später zur Inneren Qualität weiterentwickelt wurde. Die vorliegende Untersuchung an Möhren stellt einen weiteren Schritt zum Finden und Definieren relevanter Parameter für die Innere Qualität von Produkten dar. Erst wenn Qualität umschrieben und messbar ist, kann die nächste Frage untersucht werden, nämlich ob Produkte mit einer hohen Inneren Qualität tatsächlich der Gesundheit zugute kommen.

Partner und Geldgeber

Der Möhrenversuch wurde auf dem biologisch-dynamischen Hof Warmonderhof (NL) durchgeführt. Die folgenden Institute waren daran beteiligt: Kwalis Qualitätsforschung Fulda GmbH (D), Meluna Biofotonen-Onderzoek, Geldermalsen (NL), Universität Kassel - Witzenhausen (D), Elektro-Chemisches Qualitätslabor (EQL) (D), Biodynamic Research Association Denmark (BRAD) (DK) und Louis Bolk Institut (LBI) (NL).
Das Projekt wurde finanziell ermöglicht von Stichting Triodos Fonds (NL), Rabobank (NL), Software AG Stiftung (D), Zukunftsstiftung Landwirtschaft (D) sowie von allen oben genannten Teilnehmern.

Innere Qualität: ein kohärenter Qualitätsbegriff für lebende Produkte

Im Bericht über den zweiten Versuch an Äpfeln wurde Innere Qualität, mit Abwandlung einiger Worte, folgendermaßen definiert:
Innere Qualität ist eine Erweiterung der üblichen Qualitätskriterien, die sich auf Aussehen, Vorhandensein erwünschter und Nichtvorhandensein unerwünschter Bestandteile sowie hygienische Normen beziehen.

Innere Qualität hat mit Eigenschaften zu tun, die ein arttypisches Produkt ergeben, das reif, wohlschmeckend und lagerfähig ist. Diese Eigenschaften entwickeln sich während der Wachstumsperiode als Ergebnis von Wachstums- und Differenzierungsprozessen. Sie sind abhängig von einem ausgewogenen Verhältnis zwischen diesen Prozessen. Ein ähnlicher Begriff könnte in der Zukunft für Produkte tierischer Herkunft entwickelt werden.
Der Qualitätsbegriff sollte unterschiedliche Anforderungen erfüllen. So muss er natürlich an die Arbeitsweise des Landwirts anknüpfen, unabhängig davon, ob dieser nun ökologisch oder konventionell arbeitet. Schließlich ist es der Landwirt, der die Lebensprozesse in Pflanze oder Tier unterstützt. Die Wachstums- und Differenzierungsprozesse können in unterschiedlichem Verhältnis zueinander stehen und einander in unterschiedlichem Maße beeinflussen (Integration).
Der Qualitätsbegriff sollte auch eine Verbindung zwischen den verschiedenen Gesichtspunkten herstellen, unter denen Verbraucher die Qualität eines Produktes im Laden beurteilen, und die Landwirte ihre Gewächse. Dies macht es möglich, dass der Landwirt im Laufe der Wachstumsperiode lenkend eingreifen kann, um unter Ausnutzung der saison- und bodenartbedingten Verschiedenheiten die Qualität des Endproduktes zu optimieren. Verbraucher haben keine einheitlichen Wünsche; sie wählen ihre Nahrungsmittel je nach individueller Vorliebe, Gesundheits- und Gemütszustand. Es besteht ein Markt für unterschiedliche gute Qualitäten. Eine einzige, einheitliche, für jedermann optimale Qualität gibt es daher nach diesem Konzept nicht. Der in Tabelle 1 dargestellte Qualitätsbegriff besteht denn auch aus drei miteinander verbundenen Aspekten: Kulturmaßnahmen, Lebensprozessen und den Eigenschaften der geernteten Produkte.

Methodologie zur Entwicklung des neuen Qualitätsbegriffs

Diese Untersuchung an Möhren ist Teil des Forschungsprogramms des Louis Bolk Instituts, das, im Rahmen der Arbeit innerhalb der FQH-Vereinigung, die Entwicklung und Prüfung des neuen Qualitätsbegriffs zum Ziel hat. In der ersten Apfeluntersuchung lag der Schwerpunkt auf der Entwicklung des Qualitätsbegriffs, auf Wachstums- und Differenzierungsprozessen sowie der Art und Weise diese zu beurteilen. In der zweiten Untersuchung an Äpfeln wurde ein Verfahren zur Validierung dieses Begriffs umrissen und wurde ein Teil der Arbeiten dazu ausgeführt. Anbauer und Untersucher anderer Kulturen wurden angesprochen, um zu sehen, ob der Begriff und die beiden Lebensprozesse und ihre Integration für sie erkennbar und akzeptabel sind (*Inhaltsvalidität*). Für die Begriffe Wachstum und Differenzierung war dies der Fall. Eine Reihe von Beispielen wurde in einer Broschüre über Lebensprozesse in Nahrungspflanzen veröffentlicht (Bloksma und Huber, 2002). Die Begriffe "Integration" und "Vitalität" erwiesen sich als problematisch und wurden weiter ausgearbeitet. Auf den Begriff der "Vitalität" wurde schließlich verzichtet, weil er zu viel Anlass zu Verwirrung gab, und "Integration" wurde definiert als das Verhältnis zwischen Wachstum und Differenzierung nebst ihrer wechselseitigen Beeinflussung oder Vermischung.
Ein neuer Weg wurde eingeschlagen mit der Unterscheidung der Wachstums- und Differenzierungsprozesse nach Organen (im Fall von Äpfeln: Frucht, Blume, Zweig) und der Darstellung ihrer Entwicklung in Beziehung zum Jahreslauf. Auf die Einbeziehung der Pflanzenhormone als Aspekt der Wachstumsprozesse wurde verzichtet.
Die *Konsistenz des theoretischen Konstrukts* wurde durch den Vergleich des Qualitätsbegriffs mit einer Reihe von Hypothesen und Erkenntnissen aus der Literatur geprüft. Der Begriff stimmt zum Beispiel gut überein mit der Growth-Differentiation-Balance-Hypothese aus der pflanzenökologischen Forschung über den Widerstand gegen Krankheiten und Schädlinge.

Tabelle 1c. Der Begriff der Inneren Qualität bei der Möhre, auf der Grundlage von Wachstum, Differenzierung und Integration (?: in dieser Untersuchung validiert)*

Innere Qualität bei Möhren		
KULTURMAßNAHMEN Zum Korrigieren des Produkts während der Wachstumsperiode *mit dem Erzeuger zu besprechen*	PROZESSE Während des Anbaus *mit dem Erzeuger zu besprechen*	EIGENSCHAFTEN des Feldbestandes oder des Endprodukts *mit dem Erzeuger, dem Verbraucher und dem Händler zu besprechen*
1. Wachstum		
• Keine Begrenzung durch Nährstoff- oder Wassermangel • Wärme durch Dämme • Verminderte Düngung	• Photosynthese: Primärstoffwechsel • Aufnahme von Stickstoff und übrigen Nährstoffen • Bildung von Zellen, Geweben und Organen • Aufrechterhaltung des Grundstoffwechsels	• hohes Gewicht von Kraut und Wurzel* • Kraut dunkelgrün und rau • wachsende Blätter zum Zeitpunkt der Ernte • hoher Nitratgehalt • schwacher Möhrengeschmack • saftige und knackige Wurzel • hohe Emission in der Lumineszenz-Anregungs-Spektroskopie*
2. Differenzierung		
• Licht durch weiten Pflanzabstand • Verminderte Düngung	• Verfeinerung, Ordnung • Reifung: Monosaccharide -> Saccharose • Sekundärstoffwechsel	• zierliche, symmetrische Blätter • niedriges Verhältnis Kraut-/ Wurzelgewicht? • abgestumpfte Wurzel* • hoher Saccharosegehalt*, Süße • hoher Gehalt an Trockensubstanz* • hoher Karotingehalt* • ausgeprägter Möhrengeschmack • hohe Emission Ratio in der Lumineszenz-Anregungs-Spektroskopie* • pH*
Integration von 1 und 2		
• optimales Verhältnis zwischen Wachstum und Differenzierung • passende Sorten • krankheitshemmender Boden • Vielfalt des Agro-ökosystems	• ausgewogenes Verhältnis zwischen Wachstum und Differenzierung, für die jeweilige Sorte zum jeweiligen Zeitpunkt	• Widerstand gegen Schädlinge und Krankheiten • sensorische Gesamtbewertung • Lagereigenschaften*

Ziele

Um zu einem besseren Verständnis und zur Validierung des Begriffs der Inneren Qualität zu gelangen, wurde eine Untersuchung an Möhren durchgeführt. Diese hatte die folgenden Ziele:

1. Darstellung des Begriffs der Inneren Qualität im Fall von Möhren durch Aufzeigen der Lebensprozesse Wachstum, Differenzierung und Integration dieser beiden anhand allgemein anerkannter wie auch

experimenteller Parameter. Die Lebensprozesse wurden durch Unterschiede in Stickstoffgehalt, Sonnenlicht und Ernteterminen (Zeitachse) im Versuch variiert. Aus der Literatur und aus der Erfahrung ist bekannt, dass Stickstoff und Sonnenlicht die Intensität der Wachstums- und Differenzierungsprozesse beeinflussen. Ein hoher Stickstoffgehalt fördert das Wachstum und hemmt die Differenzierung; Sonnenlicht fördert die Differenzierung mehr als das Wachstum, und die Intensität der Differenzierung während der Wachstumsperiode nimmt zu. Der Faktor Integration bezieht sich auf das Verhältnis und die Wechselwirkung zwischen den Wachstums- und Differenzierungsprozessen.

2. Auswahl allgemein anerkannter Parameter und sie relatieren mit den experimentellen Parameter für die verschiedenen Prozesse in der Möhre. Außerdem die Auswahl von Parametern, die einfach zu beurteilen und preiswert im praktischen Gebrauch sind.
3. Prüfung anhand der Versuchsergebnisse, ob Wachstum, Differenzierung und Integration relevante Determinanten für die Innere Qualität von Möhren sind.
4. Ausfindig machen von Faktoren, anhand derer sich die Integration der Wachstums- und Differenzierungsprozesse deutlich feststellen lässt. Dies soll es den Erzeugern ermöglichen, die Innere Qualität der Möhre zu maximieren.

Anordnung des Möhrenversuchs

Ein Feldversuch mit Möhren auf vollständig randomisierten Parzellen in drei Wiederholungen wurde angelegt. Drei N-Düngungsstufen wurden verglichen: 0, 100 und 200 kg N/ha, zusätzlich zu dem sowieso im Boden verfügbaren Stickstoff. Der Stickstoff wurde in Form von zu Pellets gepresstem Blutmehl und Federmehl vor und nach der Aussaat zugeführt. Mit Hilfe von Schattennetzen wurden drei Lichtstufen mit 52, 85 und 100 % Licht eingerichtet. Die benutzte Sorte war Rodelika, eine samenfeste Sorte aus dem biologisch-dynamischen Züchtungsprogramm des Dottenfelderhofs. In Abständen von zwei Wochen wurden Möhren geerntet und wurden das Frischgewicht von Kraut und Wurzel sowie die Trockenmasse der Wurzel festgestellt und die Morphologie beurteilt. Bei drei größeren Ernten im August, September und Oktober wurde auch der Gehalt an Saccharose, D-Glukose und D-Fruktose, Nitrat, Gesamtstickstoff und Karotinen analysiert und wurden die sensorischen Eigenschaften sowie die experimentellen Parameter beurteilt: Verhältnis zwischen Reinprotein und Rohprotein, Kupferchloridkristallisation, Lumineszenz-Anregungs-Spektroskopie und elektrochemische Parameter. Während der gesamten Wachstumsperiode wurden Schädlinge und Krankheiten aufgezeichnet.

Die Stickstoffgehalte wiesen ab Tag 41 nach der Aussaat signifikante Unterschiede auf. Die Schattennetze wurden Anfang Juli angebracht, so dass die beabsichtigten Lichtunterschiede in der zweiten Hälfte der Saison eingeführt wurden. Einige der morphologischen und chemischen Resultate zeigten unerwünschte, signifikante Unterschiede zwischen den Feldwiederholungen. Darum mussten jeweils die Durchschnittswerte der drei Feldwiederholungen verwendet werden, mit den entsprechenden Folgen für die statistische Analyse des Versuchs.

Zudem traten die tatsächlichen Unterschiede zwischen den N-Düngungsstufen durch den warmen und trockenen Sommer weniger deutlich zutage.

Schlussfolgerungen bezüglich des Versuchs und der Parameter

- Die morphologischen Parameter reagierten im Allgemeinen am besten auf Licht, an zweiter Stelle auf den Erntetermin und am wenigsten auf Stickstoff. Im Allgemeinen entsprachen die Ergebnisse den Erwartungen, wobei das Verhältnis der Rinde zum Zentralzylinder eine Ausnahme bildete. Man erwartet, dass dieses mit der Zeit zunimmt, was nach der Literatur auf einen Reifungsprozess hindeutet; gemessen wurde jedoch eine Abnahme. Die meisten morphologischen Parameter sind von Bauern leicht zu beobachten. Der absolute Wert wird je nach der benutzten Sorte, dem Boden und dem Jahr (Wetter) variieren. Dies bedeutet, dass mit einer bestimmten Sorte auf einem bestimmten Boden (oder einem bestimmten Hof) morphologische Parameter als Indikatoren für Wachstum, Differenzierung und Integration verwendet werden können. Dagegen wird es schwierig sein, feste Bezugswerte anzugeben, die für eine Qualitätsandeutung angewendet werden könnten.
- Schädlinge und Krankheiten kamen in geringem Ausmaß vor; sie standen nicht im Zusammenhang mit Licht oder Stickstoffgehalt. Dies spricht nicht für die Vorstellung von Schädlingen und Krankheiten als Parametern für gut integrierte Lebensprozesse. Andererseits wird die Integration von mehr Faktoren

beeinflusst als nur von Licht und Stickstoff. Wenn wir Schädlinge und Krankheiten als Ausdruck eines gestörten Gleichgewichts *definieren*, waren die Wachstums- und Differenzierungsprozesse dieses Möhrenbestandes als Ganzem nicht gut integriert. Die zum letzten Erntetermin festgestellte Zunahme des Alternaria-Befalls und das Vorkommen von Mehltau auf dem gesamten Feld während der letzten zwei Monate deuten nach unserer Definition auf eine schwache Integration hin.

- Die Gehalte an den verschiedenen chemischen Bestandteilen der Wurzeln reagierten auf die Faktoren Licht und Zeit, doch nur wenig auf Stickstoff. In der Literatur wird ein begrenzter Ertragseffekt einer zusätzlichen Stickstoffversorgung auf Möhren beschrieben. Der Nitratgehalt und das Verhältnis zwischen Reinprotein und Rohprotein bestätigten nicht die Erwartungen bezüglich der Zeitvariable. Wir erwarten, dass der Nitratgehalt mit der Zeit abnimmt und der Anteil des Reinproteins zunimmt; sie verhielten sich jedoch in entgegengesetzter Weise. Der Anteil des Reinproteins ging in der Pilotstudie ebenfalls zurück.
- Die höchste sensorische Gesamtbewertung der Möhren wurde, wie erwartet, ohne zusätzlichen Stickstoff und bei vollem Licht erzielt.
- Die vom LBI und der BRAD erzeugten Kupferchloridkristallisationsbilder zeigten im Vergleich zu der *Triangle Study* von LBI, BRAD und Universität Kassel – Witzenhausen ungewöhnliche Erscheinungen, während nur geringe Wirkungen von Stickstoff, Licht und Erntetermin unterschieden werden konnten. Dies wurde dem Umstand zugeschrieben, dass das Verhältnis von Möhrensaft und Kupferchlorid nicht optimal war. Es erwies sich als notwendig, vor der Untersuchung der Möhrenproben erst die optimalen Konzentrationen zu bestimmen.
- Die von Kwalis durchgeführten Versuche zur Lumineszenz-Anregungs-Spektroskopie zeigten entsprechend den Erwartungen signifikante Effekte für alle vier Parameter hinsichtlich der Lichtstufen; von Stickstoff ließ sich dagegen kein signifikanter Einfluss feststellen. Auch in Bezug auf Zeit waren signifikante Wirkungen zu beobachten, die den Erwartungen größtenteils entsprachen. Nur der Kurvenverlaufsquotient (Ausdruck für das Maß der Hyperbolizität), bei dem mit einer Zunahme oder einem Optimum in der Zeitserie gerechnet wurde, nahm wider Erwarten ab. Die Lumineszenzmessungen von Meluna (die ausschließlich für den letzten Erntetermin durchgeführt wurden) ergaben ebenfalls für drei Parameter signifikante Wirkungen in der Lichtserie, wobei die Anfangslumineszenz sowie die Gesamtemission nach Anregung mit Weißlicht erwartungsgemäß zunahmen, ebenso wie - wider Erwarten - die Abklingkurve der Emission nach Anregung mit weißem Licht. In Bezug auf Stickstoff war, wie erwartet, eine signifikante Wirkung auf die Anfangslumineszenz sowie die Gesamtemission nach Anregung mit Weißlicht zu beobachten, wie auch, was nicht zu erwarten war, auf die Abklingkurve. Das Verhältnis zwischen Anfangslumineszenz und Abklingkurve zeigte eine Wirkung von Licht und Stickstoff und könnte einen Hinweis auf Integration darstellen.
- Die in Kassel untersuchten elektrochemischen Parameter zeitigten signifikante Effekte in der Licht- und in der Zeitserie, nicht jedoch in der Stickstoffserie. Der pH-Parameter verhielt sich erwartungsgemäß. Das Redoxpotential und der elektrische Widerstand entsprachen in der Lichtserie im Großen und Ganzen den Erwartungen, während in der Zeitserie das Gegenteil der Fall war. Von den vom EQL (nur für den letzten Erntetermin) erfassten elektrochemischen Parametern nahmen in der Lichtserie pH (signifikant) und elektrischer Widerstand (tendenziell) wie erwartet zu. Was die Stickstoffstufen betrifft, waren für das Redoxpotential und den elektrischen Widerstand signifikante Wirkungen mit einem Optimum auf der mittleren Stickstoffstufe zu verzeichnen, was den Erwartungen nicht entsprach.
- Der Lagerfähigkeitstest ergab wie erwartet eine bessere Lagerfähigkeit bei niedrigem Stickstoffgehalt und vollem Licht. Der geringste Gewichtsverlust wurde bei den Möhren beobachtet, die zu dem erwartungsgemäß optimalen Zeitpunkt geerntet wurden.
- Fünf Parameter widersprachen bezüglich der Zeitvariable den Erwartungen: Verhältnis der Rinde zum Zentralzylinder (Tendenz), Nitrat, Verhältnis zwischen Reinprotein und Rohprotein (Tendenz), Redoxpotential (Tendenz) und elektrischer Widerstand. Wahrscheinlich reagieren diese Parameter nicht auf Stickstoff, wenn dieser in reichlichem Maße vorhanden ist.

Schlussfolgerungen bezüglich des Qualitätsbegriffs

In Tabelle1c sind die Ergebnisse der Untersuchung bezüglich des Qualitätsbegriffs zusammengefasst. Messgrößen für die Wachstumsprozesse sind das Frischgewicht des Krautes und der Wurzel sowie der Nitratgehalt und die Emission 30-50 Weißlicht der Wurzel in der Lumineszenz-Anregungs-Spektroskopie. Dabei konnten keine Unterschiede im Grün und in der Rauheit der Blätter oder der Wachstumsrate der Blätter zum Zeitpunkt der Ernte nachgewiesen werden, wahrscheinlich wegen des hohen Ausgangsgehalts an Stickstoff im Boden. Auch ein Zusammenhang mit Geschmack, Saftigkeit und Knackigkeit der Möhre ließ sich nicht feststellen. Der Möhrengeschmack und das Verhältnis Zentralzylinder/Rinde der Wurzel konnten nicht mit Wachstums- oder Differenzierungsprozessen in Zusammenhang gebracht werden.
Die Differenzierungsprozesse werden anhand der abgestumpften Form, der Gehalte an Saccharose und Karotinen, der Trockenmasse sowie der Emission 30-50 Ratio der Wurzel in der Lumineszenz-Anregungs-Spektroskopie beurteilt.
Die Integration beider Prozesse wird per definitionem anhand der sensorischen Gesamtbewertung, der Lagerungseigenschaften und des Widerstandes gegen Schädlinge und Krankheiten beurteilt. Die ersten beiden Parameter wiesen Unterschiede auf, die den Erwartungen entsprachen; der letzte zeigte keine Unterschiede zwischen den Behandlungen, wahrscheinlich weil der Ausbruchsort der Krankheiten nicht klar erkannt wurde. Die Abklingkurve der Emission nach Anregung mit Weißlicht in der Lumineszenz-Anregungs-Spektroskopie könnte als Integrationsparameter betrachtet werden, aber das hat sich hier nicht bestätigt Eine weitere Ausarbeitung ist erforderlich.
Integration wird als ausgewogenes Verhältnis und Wechselwirkung (oder Vermischung) von Wachstums- und Differenzierungsprozessen betrachtet. Eine Abnahme der Wachstumsprozesse um den erwartungsgemäß optimalen Erntetermin war nicht zu beobachten, wahrscheinlich wegen des hohen Stickstoffgehalts. Die erwartete Zunahme der Differenzierungsprozesse hat sich bestätigt. Zur Validierung des Integrationsbegriffs ist eine weitere Ausarbeitung erforderlich. Außer der Beobachtung der Wachstums- und Differenzierungsparameter sollten auch Integrationsparameter beurteilt werden. Die Beurteilung der Integration anhand der Parameter Widerstand gegen Schädlinge und Krankheiten, sensorische Eigenschaften und Lagerfähigkeit sollte verbessert werden.

Empfehlungen für weitere Untersuchungen

- Zu einem vertieften Verständnis und einer besseren Verständigung über den Begriff der Inneren Qualität ist es notwendig, die Möhrenuntersuchung in zwei oder vorzugsweise drei Ländern zu wiederholen, um die Lebensprozesse und die dazugehörigen Parameter zu veranschaulichen. Dabei sollte nur eine begrenzte Anzahl Parameter verwendet werden, die mit geringem Aufwand zu messen sind. Dies sind die folgenden: Gewicht von Kraut und Wurzel für die Wachstumsprozesse; Kraut/Wurzel-Gewichtsverhältnis, abgestumpfte Wurzel, Saccharose, Süße und Trockenmasse für die Differenzierungsprozesse; Vorkommen von Schädlingen und Krankheiten, sensorische Gesamtbewertung, Gewichtsverlust und Fäulnis in einem Lagerungstest für die Integrationsprozesse.
- In getrennten Versuchen sollten die verzögerte Lumineszenz und die Kupferchloridkristallisation untersucht werden, um die Parameter für die Lebensprozesse zu validieren. Vergleichsserien von Möhren können aus der oben genannten Untersuchung gewonnen werden.

2 Introduction

2.1 Background

In search of a quality concept for organic food production: 'Inner Quality'

Consumers expect organic production to provide a healthy product with a good taste. Sometimes this is the case, sometimes it is not. Organic production does not guarantee a good taste or a healthy product. Besides, there is still disagreement about what tastes good, which quality is healthy, and how to achieve this in the cultivation of plants. In the conventional vision, product quality is mainly based on external, nutrient and sensory properties and is strongly directed by traders and fashions. In organic food shops, consumers expect properties such as tastiness, ripeness, 'vitality' and 'coherence', which are not easy to define or measure. In the past, some experimental parameters were proposed to estimate 'vitality' and 'coherence'. The organic production chain demands scientifically validated food quality concepts and parameters.

This study is carried out under the international research association for Organic Food, Quality and Health (FQH). The association aims to develop new concepts for food quality that dovetail with the principles of organic production, but which are also suitable for innovations in conventional production. It also is a platform for researchers, working on this quality theme, directed towards cooperation and gears research programmes one to another.

Three studies with apples and carrots have been realised by the Louis Bolk Instituut. In the first apple study (Apple-1) (Bloksma et al., 2001), the concept of Vital Quality was developed, based on the life processes of growth and differentiation and their integration. In the second apple study (Apple-2), the concept was further developed and renamed the Inner Quality concept (Bloksma et al., 2004). The previous pilot project on carrots has been reported (Northolt et al., 2002) and extended in the present carrot study. Table 2 gives the variable factors used in the studies and the presumed effects on the life processes.

Table 2. Overview of crops and variable factors used in the quality concept studies and the presumed effect on the intensities of life processes ***(arrows indicate the positive or negative effect, small or large)***

Crop	harvest	Factor	Growth	Differentiation	Integration
Apple-1	2000	Bearing (yield)	↓↓		
		Light	↑	↑↑	↑
		Ripening (harvest dates)	↑	↑↑	
		Post-harvest ageing	↓↓		↓
Apple-2	2002 (2001-2003)	Nutrients	↑↑	↓	
		Compost & fertiliser			↑?
		BD-preparations			↑?
Carrot	2002 (pilot) 2003	Nutrients	↑↑	↓	
		Light	↑	↑↑	↑
		Ripening (harvest dates)	↑	↑↑	

The apple and carrot studies are used to develop the quality concept as well as to acquire references for the experimental parameters with product samples from crops grown under controlled conditions. The concept of life processes can also be a basis for quality management in other crops (Bloksma and Huber, 2002). In the meantime, FQH partners studying experimental parameters worked on validating their individual measurement methods. The field experiments conducted in this study provide them with reference data.

Working sequence in the experiments

First, in the first apple study, a preliminary quality concept was used based on commonly accepted physiological and morphological knowledge and described in terms of the life processes: growth, differentiation and integration. Appropriate parameters were selected to assess the intensities of life processes in apples. In field experiments, management factors influencing the life processes were varied, and the resulting crop properties were measured. Besides using generally accepted or golden standard parameters, experimental parameters such as delayed luminescence and copper chloride crystallisation pictures (Meier-Ploeger and Vogtmann, 1988) were also determined, to see if they correlate with the generally accepted parameters for the same process. Next, a preliminary quality concept based on the results of the experiments was elaborated.

Conclusions from the apple experiments

On the basis of the two apple experiments, sets of growth and differentiation parameters were distinguished. The parameters of tree growth are: fruit-bearing, twig growth, leaf size and colour, nitrogen content in buds, nitrogen and magnesium content in leaves, and scab infestation. The parameters of fruit growth are: fruit size and weight, firmness, acidity, nitrogen content, amino acid content, protein content, taste, growth score on copper chloride crystallisation (CC) pictures, initial emission of delayed luminescence, and susceptibility to fruit rot.
Parameters of tree differentiation are: autumn colours and bud formation. Parameters of fruit differentiation are fruit colour, shape of fruit, starch conversion, differentiation score of CC pictures, and hyperbolic decline of delayed luminescence.

For most plant processes, both growth and differentiation are important. Many parameters can therefore express both growth and differentiation, depending on which is the limiting factor. For example, bud formation in apple trees is a parameter of growth when nutrients are the limiting factor. If light exposure is the limiting factor, bud formation acts as a differentiation parameter in a light exposure series. Another example of parameters with interaction between growth and differentiation is the content of secondary metabolites in the plant, such as phenols, vitamins, aromas and the red pigments of fruit. The formation of the primary metabolites (assimilates) is a growth process; the subsequent conversion to secondary metabolites is a differentiation process. Whether a correlation with either growth or differentiation is found, depends on when the measurements are done and what the limiting factors are.

Parameters which could potentially give an indication of the degree of integration are: resistance to pests and disease, total sensoric appreciation, ratio of pure/crude proteins, integration score of CC images, and species-specific spectral range in delayed luminescence. Generally accepted methods exist for the first two. The last three have only recently been validated for carrots and wheat (Kahl et al., 2005). The results obtained do not yet allow clear interpretation as regards the assessment of quality. To this end, the quality concept must provide a context for these assessments, and still more reference series, as used in the two apple studies, must be established.

CC pictures and delayed luminescence dovetail well with the quality concept since both techniques can be evaluated on all three aspects: growth, differentiation and integration. The apple studies produced successful growth series, and for both parameters, the growth aspect correlated with many generally accepted growth parameters. However, the electro-chemical measurements showed too much variation and did not correlate with the growth and differentiation parameters.

Life processes in carrots in literature

The carrot belongs to the family of the Apiaceae. The wild carrot is an indigenous species in Europe and Asia. In the book of herbs of Dodonaeus (1554), three types of carrots were distinguished: the wild carrot, the yellow carrot and the red carrot. All types have the Latin name *Daucus carota L.* var. *sativa*. Parsnip, parsley, fennel and celeriac also belong to this family. The yellow carrot has been used to develop different varieties with different root forms.

Various parameters have been associated with growth, differentiation and integration. Plant mass and size are parameters for growth. Fine, symmetrical forms of leaves are parameters for differentiation according the definition. This also holds true for discolouration of leaves. An increase in maturity of carrots is reflected in changes in the root from conical to cylindrical, from pointed to stumpy, from an increase in the cortex/core (phloem/xylem) ratio and a decrease in the leaves/root weight ratio (Wistinghausen, 1979; Schulz and Köpke, 1992). Maturity is considered to be similar to integration (Bloksma et al., 2004 a), however the said parameters might also be related to differentiation. Pests and disease are related to integration (Bloksma et al., 2004 b). Carrots are rich in sugar but do not contain starch. The total sugar content which increases in the second half of the growing period (Bokhorst, 1985) may be related to differentiation or integration. The pure/crude protein ratio increases during maturation (Wistinghausen, 1975) and may indicate integration. Carotenes are secondary metabolites and may indicate differentiation. The taste of carrots indicates a balance between vegetative growth and maturation (Bokhorst in: Visser and De Vries, 1979) and can therefore be related to integration. Balzer-Graf (2001) relates CC pictures to carrot typicity and therefore to integration. Changes such as loss of weight and rot during storage may also be related to integration.

Conclusions from the carrot pilot study

For the pilot study, the hybrid Yukon was used (Northolt et al., 2003). The study included a series of different levels of nitrogen and another series of different levels of light. Since it did not include repetitions, the results are preliminary in nature.
The parameters were evaluated according to the assumed succession of the growth and differentiation phases and the assumption that nitrogen mainly affects growth, and light affects both growth and differentiation. In this way, some parameters could be related to growth: aspects of leaf morphology, leaf and root weight, content of root: nitrate; free amino acids; total soluble solids (Brix); density of CC pictures by LBI. Some other parameters could be related to differentiation: aspects of leaf morphology; leaves/root weight ratio; emission 30-50 ratio of delayed luminescence; filling of plate, side needle angle and head branches of CC pictures by LBI; perradiation, specificity, coordination and integration of CC pictures by BRAD. Two parameters, pests and disease, and Bovis value, could be related to both growth and differentiation. Only one parameter is thought to be related only to integration: coherence of CC pictures by LBI, whereas Bovis value could be related to growth and differentiation.

Aims of the present study

1. To demonstrate the Inner Quality Concept for carrots by showing the life processes growth, differentiation and integration by generally accepted and experimental parameters. The life processes are experimentally varied by different levels of nitrogen and light and by different days of harvest (time axis). From literature and experience, we know that they influence the intensity of the growth and differentiation processes. High nitrogen levels stimulate growth and inhibit differentiation; light stimulates differentiation more than growth, and during the growth season, the intensity of differentiation or ripening increases. The integration factor involves the balance and interaction between the growth and differentiation processes.
2. To select generally accepted parameters and to relate them with the experimental parameters for the different processes in carrots. Moreover, to select parameters that are easily measurable and cheap to use in practice.
3. To verify whether growth, differentiation and integration are relevant determinants of the inner quality of carrots by use of the experimental results.
4. To search for factors which determine the integration factor of the growth and differentiation processes more clearly, in order to enable growers to maximise the inner quality of the carrot root during the cultivation process.

Outline of the study

Carrots were grown at different levels of light and soil nitrogen. Plants were harvested at intervals over time before and after the advised harvest day (day 136). Leaves and roots were examined for a large number of morphological and chemical parameters. Moreover, the roots were investigated to determine sensory

properties and a number of experimental parameters, such as copper chloride pictures, delayed luminescence, electro-chemical properties and protein ratio. Furthermore, the roots were used in a storage test. The parameters were determined in seven specialised laboratories. Some experimental parameters were measured in two laboratories to compare method and results.

Reading this report

After this introduction (Chapter 2), you will find the way of validating the quality concept (Chapter 3) and the current carrot experiment (Chapters 4, 5, 6 and 7), including background, method and results of each parameter (Chapter 7). In Chapters 8 and 9, the two strands are brought together: the carrot results are discussed in the context of the Inner Quality concept. Of course, the work is not finished, and Chapter 10 looks at future prospects and asks further questions.

We wrote the report for several target groups:

Target group	Read chapter:
People who want a general idea about the development of the Inner Quality concept with carrots as an example	1: An extended summary and translations in Dutch and German 10: Conclusions
People more interested in the Inner Quality concept	3, 5, 9
People mainly interested in carrot growing	2.1, 4, 5, 6, 7
People interested in measuring quality	7, 9, 10

2.2 Cooperation

Participants:

- Louis Bolk Instituut (NL), Dr M. Northolt, Ir. G. van der Burgt, M. Huber M.D., Ing. P. Doesburg, Ir. M. Zanen and trainees T. Buisman and A. Vanden Bogaerde: coordination, field observations, harvests, copper chloride crystallisations, morphological measurements, statistics, quality concept, report;
- Warmonderhof farm (NL), T. Verdonschot: preparation of the test field;
- Kwalis Qualitätsforschung Fulda GmbH (D), Dr J. Strube and Dr P. Stolz: delayed luminescence, amino acid analysis, protein ratio and quality concept;
- Meluna Bio-photon Research (NL), Drs. R. and E. van Wijk: delayed luminescence and quality concept;
- Electro-chemisches Qualität Labor (D), Dipl.Ing.agr. H. Heilmann: electro-chemical measurements and quality concept;
- Biodynamic Research Association Denmark (DK), Dr J-O. Andersen: copper chloride crystallisations, quality concept and member of FQH scientific advisory committee;
- University of Kassel - Witzenhausen (D), Dr J. Kahl, M. Desche: electro-chemical measurements.

Further collaboration with:

- Silliker Laboratories (NL);
- CSO Centrum voor smaakonderzoek (NL), Dr B. Cramwinckel;
- Forschungsinstitut für biologischen Landbau (FiBL)(CH), Dr M. Koller;
- Universität Gesamthochschule Kassel (D), Dipl.-Ing. Agr. M. Fleck.

Funding:

This project is financed by:

- Louis Bolk Instituut (NL);
- Software AG Stiftung (D);
- Rabobank (NL);
- Stichting Triodos Fonds (NL);
- Zukunftsstiftung Landwirtschaft (D);
- Kwalis Qualitätsforschung Fulda GmbH (D);
- Meluna Bio-photon Research (NL);
- Biodynamic Research Association, Denmark (DK).

Our thanks to them all!

3 Development of the Inner Quality concept

3.1 Introduction

In the first apple report (Bloksma et al., 2001), the Vital Quality concept based on organic principles was presented as a concept in nutritional quality. This concept was based on the life processes of growth and differentiation and their integration (i.e. balance and interaction). The concept had two interest groups: growers on processes and consumers on product properties. These two dimensions are connected by life processes which provide consistency in the concept.

To recap, the aim of this quality concept was to design a concept for food quality with three purposes:
The first purpose is to develop a concept that links product properties to management measurements by growers. Growers would recognise the quality at the growing stage and take appropriate and timely measures. They manage the life processes to optimise the quality. Life processes can be defined in conventional plant physiological terms, as a link to generally accepted science.
The second purpose is to support the inner nutritional quality towards which (organic) agriculture strives: a tasty, healthy product, described in positive terms. Thus not only in negative terms, as modern food safety standards emphasise (no residues, no microbes, etc.). Also, to verify the assumption by organic agricultural communities that healthy food needs to be mature and to be coherent. Coherence is defined as a high degree of organisation in the system.
The third purpose is to relate quality to human health concepts used by holistic health workers and dieticians. For them, growth, differentiation and integration are meaningful terms.

A balanced relationship of life processes for optimal food quality is not a new idea. We have already encountered this mode of thought in the biodynamic tradition (e.g. Schuphan, 1961; Engquist, 1963; Klett, 1968; Pettersson, 1970; Koepf, Pettersson and Schaumann, 1976; Bisterbosch, 1994; Kunz, 1999; Bauer, 1999) and in scientific plant physiology (e.g. Herms and Mattson, 1992; Lerdau et al., 1994; Galston, 1994). What is new is using these life processes as a framework for a coherent quality concept including various quality properties. In the long term, they can also be linked to human and animal health. Another new facet of this concept is that it does not include a single optimum quality. There is some freedom in choosing a more growth-related or a more differentiation-related quality. For example, some consumers like green, firm, juicy apples (emphasis on growth) and others prefer blushed, sweet, aromatic apples (emphasis on differentiation).

3.2 Validation process

The steps required to validate a new concept are given by Streiner and Norman (2001) and completed for the apple and carrot studies by Bloksma et al. (2004) (p. 27). This validation process is important to avoid going around in circles when it comes to new properties and new parameters, especially at the integration level. It provides a route for logical reasoning.

Since 2001, the concept has been discussed in communities of practical workers (*face validation* by farmers, dieticians and consumers) and in communities of scientific experts and compared with literature (*content validation*), such as international conferences on organic fruit growing (Bloksma, 2002 b), the Round Table Conference in May 2003 at LBI, Driebergen as part of the second apple study. This led to some new terms and new positioning of old properties in the newly designed concept for Inner Quality for apples in Bloksma et al. (2004 b)(p. 3).

In this report, a similar concept is developed for carrots in Table 1 of the summary. The experimental parameters are compared with the generally accepted parameters for *convergent validity*. For integration, where *construct validity* is of major importance, further literature research and work on aspects such as self-regulation and coherence are necessary.

In the meantime, the various partner laboratories have worked on the *predictive validity* of their parameters. Recently, predictive validations of copper chloride crystallisation pictures, delayed luminescence, pure/crude protein ratio, but not yet successfully of electro-chemical parameters, were presented at the symposium New approaches in food quality analysis, Berlin, 13 - 14 November, 2003 (Bloksma et al., 2004 a).

3.3 Improvement of the quality concept since 2002

Since the first apple study (Bloksma et al., 2001) the concept is tested in the second apple study (Bloksma et al., 2004) and the first carrot project (Northolt et al., 2003), and described for other products (Bloksma and Huber, 2002). Growers of flowers, fruits and seed crops use cultural measures to influence growth and differentiation as two interacting life processes. They recognise the conflicting demands of growth (quantity) and differentiation (ripening) which particularly concerns the quality in the crops. Growers of apple take measures to regulate fruit and twig growth, see Bloksma et al. (2004) (p. 3). Potato, carrot and leaves vegetable growers mainly focus on growth processes and only some of them recognise the benefits of differentiation. Some lettuce growers strive to grow 'mature lettuce' instead of 'baby lettuce'.

As the previous description of the concept lacked a time dimension, Bloksma et al. (2004) introduced the development of the crop in the season as a time axis and worked out the balance between growth and differentiation in each developmental stage based on literature on apple physiology.

Growth is not equally distributed in the various organs of plant. In carrot, leaves grow vigorously in July whereas the root grows thicker in August and September. This means that growth and differentiation must be considered per organ and during time.

The integration of the life processes of growth and differentiation has two aspects: a balance aspect and an interaction aspect.
The balance aspect is commonly recognised. An understanding of balance can be obtained by looking at examples of out-of-balance situations. Plants grown in shadow are tall and weak and lack differentiation by light; a lettuce crop growing fast with much fertiliser lacks taste and is susceptible to diseases; emergency flowering in a dry location lacks vigour (too little growth); aphids suck growing substances (amino acids) and this results in dry, mummified fruits (too little growth). The growth-differentiation-balance hypothesis (GDBH) in plant defence strategies (Herms and Mattson, 1992; Lerdau et al., 1994) is summarized by Bloksma et al.(2004)(p.29), and is very much in line with the Inner Quality concept. In the GDBH hypothesis growth is used for primary metabolism and differentiation for secondary metabolism.
The second aspect, the presumed interaction or intermingling of growth and differentiation, is still unknown in plant physiology. It is recognised and desired by biodynamic growers of food crops and users of biodynamic field preparations, who express this as 'plant-typicity'. The balance aspect of integration is expressed in a moderate resistance to stress and diseases and for fruits in aromatic and crispy. The interaction aspect of integration can be described as the maturation or maximal natural development which is used for the crop as a whole, including the vegetative growth, flowering and fruiting. Plant physiologists admit that there is a lack of understanding of the physiological basis for plant resistance and self-regulation. This needs further elaboration.

Secondary metabolism or ripening is a clear differentiation process. Secondary metabolites, e.g. instance phenols, vitamins, and colour pigments, are produced when enough primary metabolites are available from the preceding growth process. Both processes should be in balance to obtain a high amount of secondary

metabolites. Therefore a considerable amount of natural plant resistance chemicals (e.g. phenols), colour pigments and flavour is the result of integration.

Refining and ordering the growing plant cells produces fine forms, leaf serration, colours, etc. and is a formative process recognised by growers and plant physiologists (F. Buwalda, pers. com.). In organic agriculture communities this process is called differentiation. Some plant physiologists use differentiation only to describe the formation of new organs or phases in the plant development. But every cell and organ has stages with emphasis on growth or on differentiation. As an example out of the apple cultivation: the formation of flower buds starts a new phase. Depending on the crop and organ we use 'ordered', 'differentiated' or 'refined' to describe the result of differentiation.

The term Vital Quality was the former name of the concept emphasising the relationship with life and human health. However, it appeared that this term leads to confusion because of the many different ways in which the word vitality is already used. For some people vitality is the result of growth (strong growing green mass) and for others it is the result of integration (resistance, self regulation, staying healthy). Cell biologists recognise the term cell vitality for a cell which grows well. Seed growers generally use the term seed vitality for seeds that germinate easily. Balzer-Graf (2001) uses vitality in the integrated meaning in her picture-forming methods. We prefer the term Inner Quality as it emphasises the inner properties such as taste and health rather than just the external properties.

Research on the Inner Quality concept aims to develop a quality scale based on growth, differentiation and integration. We are working to link life processes and product properties to human health. The first ideas are mentioned in the booklet about life processes (Bloksma and Huber, 2002). In this context we are looking for research models to test products with different levels of Inner Quality in animals and human beings. This approach is not further described in this report.

4 Design of the carrot experiment

In order to demonstrate the life processes growth, differentiation and integration in relation to carrot plants, a test field with homogeneous and relatively low levels of nitrogen content was planted on a farm with a great deal of experience in carrot growing. The carrot plants were grown on plots at different levels of light and available nitrogen to vary their life processes. Periodically, the carrot plants were harvested to determine a number of parameters of the leaves and roots that could reflect the intensity of the life processes. Some parameters were determined in two laboratories to compare the results.

4.1 Choice of test field

The test field was different to the one used for the pilot carrot study of which the clayish loam soil contained too much available nitrogen. It was located near Dronten in the Flevopolder in central Holland on the biodynamic, mixed farm Warmonderhof. The soil is clay, moderate in organic matter (2.3%) and well-drained. Due to the fact that the groundwater level varies between 0.9 and 1.3 m below surface level and to the soil properties, capillary rising of soil water will occur during dry periods, thus preventing soil drought and stress to crop growth.

The Warmonderhof farm started at this location in 1993 and soil conditions are likely to be stabilised after conversion to biodynamic farming. The farm as a whole has a restricted manure application policy. The 2002 pre-crop was onion, and no manure was applied after the onion crop. Because of this, moderate autonomous nitrogen availability can be expected.

For the last ten years, the whole field, in which the test field was situated, had always been used for one crop (no split field), and no deviant spots or in-field differences were experienced by the farmer. In 2003, the whole field, including the test field, was used to cultivate carrots. Soil cultivation and crop treatments, except for the nitrogen application and construction of shadow nets, were done by the farmer, at the same time and in a similar way to the surrounding carrot field.

4.2 Layout of the test field and design of experiment

Varying factors

To investigate the life processes, three factors were introduced in the experiment:

1. Nitrogen: three levels of available nitrogen were created by the addition of 0, 100 and 200 kg N per ha, with code N1, N2 and N3;
2. Light: three levels of light were created using shadow nets: 52, 85 and 100% light, with code L1, L2 and L3;
3. Harvest time: to follow the life processes over time, seven harvest days were planned. On four of these harvest days, chemical analysis was done only (minor harvests), and on the other three, chemical analysis was done together with determination of different experimental parameters (major harvests). Morphological parameters were measured on all harvest days.

Field design

The design of the experiment was as follows:

- Four repetitions, of which numbers 2, 3 and 4 were used for sampling. The first could be used in case of trouble with one of the other replicates, but was not used except for morphological observations;
- Within each repetition: nine randomised experimental plots (3 nitrogen levels, 3 daylight levels);
- All plots were located on one line, running approx. east-west, 180 m long, beginning 20 m from the field border. The plots were located within a larger carrot field. The location of the test field on a long line was chosen because: firstly, the experimental field could easily be treated as the rest of the field: all 'normal' crop practices were done in the same way and at the same time as the surrounding carrot crop; and secondly, the influence of the shadow nets on the microclimatic conditions of the plots is less when compared with clustered plots;
- Every plot was 5 m long and consisted of six ridges 75 cm apart, giving a width of 4.5 m. In each plot, only 3 m length and 3 m width (four ridges) were used for sampling, excluding the outer ridges and the first and last m from data collection.

For an overview, see Figure 1.

Field and crop conditions

The carrots were sown on 25 April (day 0) in perfect soil and weather conditions. The aim was 60-80 plants per m of ridge, but due to an underestimation of seeds per m, the number of plants was higher. The open-pollinated cultivar Rodelika, bred by the biodynamic breeding programme of Dottenfelderhof, Germany, was used. The seeds were obtained from Bingenheimer Saatgut AG, Germany. An open-pollinated cultivar was chosen because such a cultivar could react more to varying environmental conditions than a hybrid cultivar. A disadvantage is the variability among the individual plants.

Germination was very uneven in time and space, resulting in a strong variation in number of plants per m (minimum 47, maximum 130) and in differences in growth stage. This heterogeneity did not even out during the season. This will have influenced the results of the experiment.

Soil nitrogen levels

In order to vary the available soil nitrogen, animal meal fertiliser was applied in three doses. Blood meal pellets were applied on 17 April, one week before sowing, and feather meal pellets were applied on 15 May and 5 June. The pellets were introduced in the topsoil by the different soil tillages after sowing. Five times during the growing period, the level of plant-available nitrogen (NO3-N) was measured (see Section 6.1). For that, from each plot, 20 soil samples were taken (0-30 cm), mixed, dried and ground. Analysis was done by means of the RQ-flex method.

Light levels

In order to vary the level of light, shadow nets were constructed at the start of July, after complete settlement of the crop, approx. 1 m above soil level. Because of the east-west orientation of the plots and the use of only the central part of 3 m out of 5 m length, the plots used for sampling had the light conditions mentioned above, except at the very start and end of the day. Measurement of light levels under the shadow nets was carried out with a lux meter twice under clear skies, and the results were compared with measurements not underneath the nets.

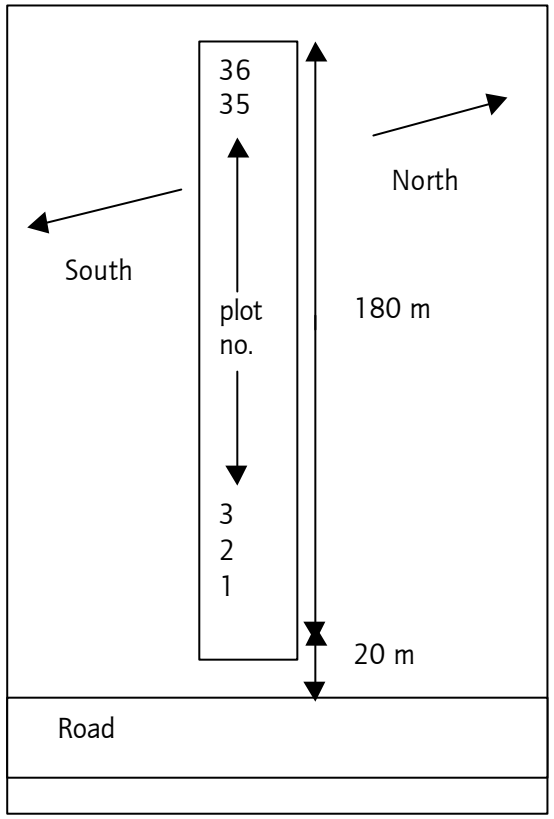
Figure 1. Design of the test field

A: Repetition number (1-4), plot number (1-36), code for level of light (L1-L3) and level of nitrogen (N1-N3).
B: Test field situation within the larger field.
C: Carrot ridges, plot and subplot for sampling.

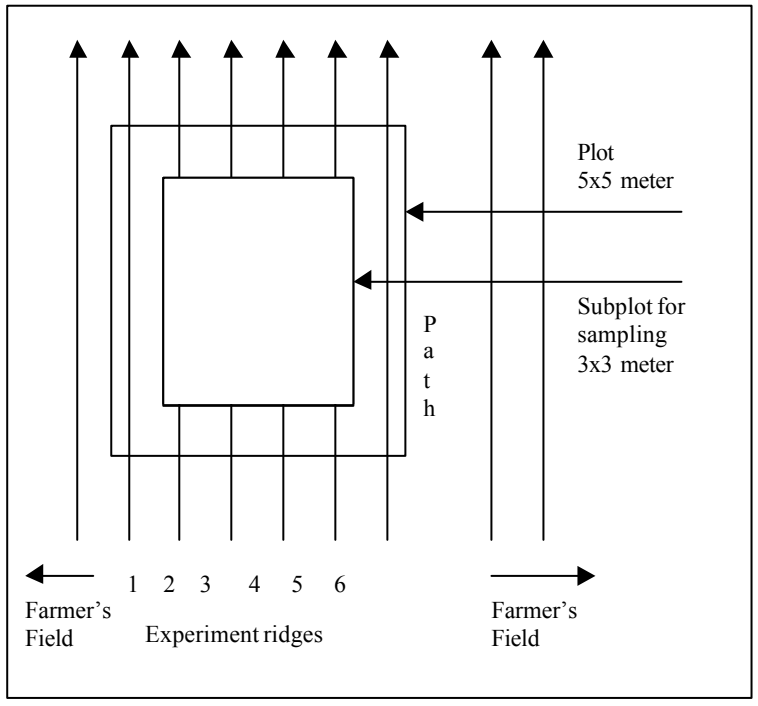
A

Repetition 4:	
36	L2N1
35	L1N1
34	L1N2
33	L3N1
32	L3N2
31	L1N3
30	L3N3
29	L2N2
28	L2N3
Repetition 3:	
27	L1N1
26	L1N3
25	L3N1
24	L3N3
23	L2N1
22	L1N2
21	L2N3
20	L2N2
19	L3N2
Repetition 2:	
18	L1N2
17	L1N1
16	L3N3
15	L3N1
14	L2N2
13	L2N1
12	L1N3
11	L2N3
10	L3N2
Repetition 1:	
9	L1N1
8	L3N2
7	L3N3
6	L1N3
5	L2N3
4	L1N2
3	L3N1
2	L2N2
1	L2N1

B



C



Harvest time

Seven times, 60 cm of carrot plants were harvested from each plot for morphological measurements and simple chemical analysis of 25 plants (minor harvest).
Three times, besides the said 60 cm, another 200 cm of carrot plants were harvested and used for morphological measurements, simple chemical analyses and determination of the experimental parameters (major harvest).

Number	Date	Days after sowing	Harvest
1	2 Jul.	68	minor
2	16 Jul.	82	minor
3	30 Jul.	96	minor
4	11 Aug.	108	major
5	27 Aug.	124	minor
6	8 Sep.	136	major
7	6 Oct.	164	major

The second, major harvest was based on the length of the optimal growing season as indicated by the seed producer: 120-140 days. This harvest is supposed to be the optimal moment, and the first and third major harvests were done 4 weeks earlier and 4 weeks later, respectively. The minor harvest, planned for 150 days after sowing, was omitted to reduce costs.

Preparatory activities for morphological study and chemical analysis

On all harvest dates, 60 cm per plot of each replicate were carefully harvested. The carrot plants were transported to the institute, where they were washed. Abnormal carrots were registered for deformation of the roots and for pests and disease, and these carrots were not used for further analyses. The remaining plants were laid out in order: smallest - largest root and counted, Then 28 plants of medium-sized roots per plot were selected for measurement, leaving out the plants with the smallest and largest roots.
The 28 carrot plants were photographed in a row from small to large root (see Figure 5), and a second photograph was taken of the plant with the smallest, medium and largest root of the 28 plants. The medium carrot plant was used to make a line-up of leaves (only for field repetition 2). This line-up is a sequence of leaves stuck on a sheet of paper with the oldest leaf on the left and the youngest leaf on the right (see Figure 6). The other 25 plants per plot were used for several measurements: length of root, length of longest leaf, fresh-weight of leaves, fresh-weight of root, number of leaves, and ranking of leaf number. These measurements were done on two days: the day of harvest and, after storage in plastic bags at 6°C, the day after. After these measurements, the central □ of each of the 25 carrots was cut out and used for analysis of dry matter, saccharose, D-glucose, D-fructose, nitrate, total nitrogen and total carotenes.

Preparatory activities for other analyses

At the major harvests, another 200 cm per plot of each repetition were harvested. The leaves of the carrots were removed immediately and the roots were not washed. Of these, the smallest and largest carrots were removed, resulting in approx. 160 carrots to be distributed among the cooperating laboratories for determination of the sensory properties, copper chloride crystallisation, delayed luminescence, electro-chemical parameters and free amino acids and crude protein (Kjeldahl).

4.3 Statistical analyses

Significant effects of nitrogen, light, harvest time and field repetitions on the parameters

At first, the mixed Anova model was used with morphological parameters to discover significant effects between different levels of light and nitrogen, harvest day and repetition in the field. Light, nitrogen and harvest day are fixed effects and repetition is a random effect. The model showed a significant effect of repetition with most morphological parameters, especially root weight and length.

The effect of repetition can be the result of variations in soil in the test field. However, the appearance and available nitrogen in the soil and previous crops showed no indication of heterogeneity of structure or nutrients. Another possibility can be the way the 25 carrots were selected, thus influencing the per-plot-variation, but it is very unlikely that this per-plot-variation, based on these 25 plants, does not represent the variation of the whole plot. Thus we do not have a satisfactory explanation of the repetition effect.

This complication forced us to treat data, not as repetitions, but simply as extra data per condition. This operation reduces the strength of the statistical output and the conclusions based on it. The new Anova model consisted only of the fixed factors light, nitrogen and harvest day for the overall tests and only of the fixed factors light and nitrogen for the per-day tests.

For the final model in each parameter, i.e. after removing the non-significant (interaction) factors, the assumption of normality of the residues was considered. These were all satisfactory. To determine which categories of significant factor vary significantly, a Bonferroni or Tukey correction for multiple testing was applied, respectively for a few and a lot of pairs to be tested. All Anova tests were performed in SPSS. Error bars as presented in the figures in the annexes are 1.96 times the standard error based on the experimental data.

Correlations

A correlation matrix was made for the morphological parameters only.

5 Overview: Hypotheses and results of the different parameters

In Table 3, an overview of the effects of the varying factors on the different measured parameters and the expected values based on literature on carrots and plant physiology is given. The effects are extracted from Chapter 7, where each parameter is described in terms of background, method, results and relation to growth, differentiation and integration.

Key to table 3:

Arrows:
Examining all the results of the individual parameters, we could group them as follows:
The varying factors nitrogen, light and time could have different effects on the parameter. The parameters could react to an increase in nitrogen, light or time as follows: an increase in parameter value (arrow up), a maximum (arrow up-down), a decrease (arrow down), a minimum (arrow down-up), no effect found (no symbol in the table).

Symbols:
X significant (with 95% certainty from statistical analysis);
O trend, but not significant due to a great deal of variation;
a until day 108;
b on day 136;
c on day 164;
lN at low nitrogen levels;
hN at high nitrogen levels;
lL at low light levels;
mL at medium light levels;
hL at high light levels.

Examples: Xc, O: significant on day164 and a trend on all other days;
O hNb: trend only at high nitrogen levels and on day 136.

Grey cell indicates the **expectation**, based on literature on carrot and plant physiology.
Open cell: no significant effect, no trend.

Table 2. Effects of nitrogen, sunlight and time on the carrot parameters

Key: Arrows indicate increase or decrease of p-arameter. Grey cell indicates the expectation. X: significant,
a: until day 108, b: on day 136, c: on day 164,
LN: at low nitrogen, hN: at high nitrogen, lL: at low light, mL: at medium light, hL: at high light

Parameters	Increasing nitrogen				Increasing light				Increase of time			
	↗	↘	↘	↗	↗	↘	↘	↗	↗	↘	↘	↗
weight leaves	X a					X b				X		
fine leaves					O a							
discolouration leaves												
growing leaves at harvests												
weight root					X				X			
leaves/root weight ratio							X				X	
cylindricity root	O lLc		O hLc		X							
stumpness root					X				X			
orangeness root	O				O							
cortex/core ratio root											O	
Pests and diseases									X c			
monosaccharides											X	
saccharose					X				X			
monosac./saccharose ratio							X c Ob				X	
dry matter					X				X a			
nitrate	X						X		X			
crude protein	O hL								O			
pure/crude protein ratio											O	
carotenes					X				X			
total sensoric appreciation			X ab		X				O			

Table 2 (continued)

Parameters	Increasing nitrogen				Increasing light				Increase of time			
	↗	↘	↘	↗	↗	↘	↘	↗	↗	↘	↘	↗
Luminescence Kwalis												
emission 30-50 white	O c				X						X	
emission 30-50 ratio					X					X		
hyperbolicity white	O c				X					X		
hyperbolicity ratio					X						X	
Luminescence Meluna												
initial emission white					X lNc							
total emission white					X lNc							
total emission ratio							O lNc					
slope white		Xc			Xc							
initial emis./slope white			O c				O c					
pH					X				X			
redox potential (Eh')	X hLb		X lLb		X hNb		O lNb		O bc			
electrical resistance (R)			O		X						X	
storage test: rot	O											
storage test: weight loss							X ab					X

6 Evaluation of test field

6.1 Soil and nitrogen supply

With a 10-year history of biodynamic farming, the soil can be considered converted to organic farming. With a soil organic matter content of 2.3% and a restricted manure application, the nitrogen availability without manure application will be moderate on this farm. The result of modelling this situation by means of the NDICEA model (Koopmans and Bokhorst, 2000 a, 2000 b; Burgt, van der, 2004) is shown in Figure 2 for the N1 treatment (no additional nitrogen). The modelled nitrogen level in the topsoil corresponds well with measurements during crop growth (black squares in Figure 2).

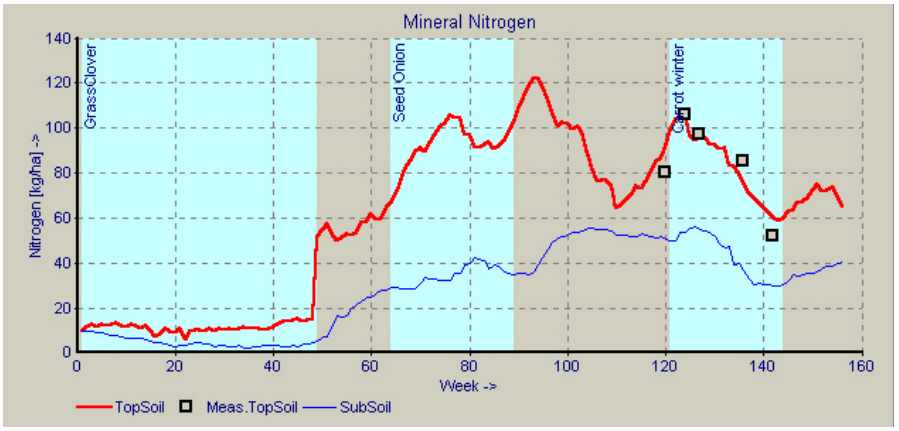


Figure 2. Modelling of available nitrogen

Light bars: crop growth period.
Thick line: NO3-N in topsoil 0-30 cm, N in kg/ha.
Thin line: NO3-N in subsoil 30-70 cm, N in kg/ha.

The difference in nitrogen availability between the N-treatments does not occur directly after application of blood meal and feather meal pellets which started eight days before sowing. This may be caused by drought (see Section 6.2). The pellets are only incorporated in the topsoil, which frequently dried out during this hot and partly rainless season. A significant difference in the levels of available nitrogen (nitrate, 0-30 cm depth) started on day 41 (Annex 6.1).

The yield on the adjacent carrot field was very high: about 90 tons per ha. This illustrates that the drought had no or only a very limited influence on crop growth. Nevertheless, the drought may have had a big influence on this experiment. The NDICEA modelling indicates limited water availability in the topsoil (data not shown), which means that the crop has taken a relatively large amount of water out of deeper soil layers. Since this water contains nitrogen, the nitrogen uptake of the crop may have been mainly from deeper soil layers. This means that nitrogen levels in the topsoil may have had less influence on the crop than foreseen, compared to a "normal" year.

6.2 Light and weather

The weather conditions in 2003 were rather extreme. The weather in May was normal. At the end of May, a warm, dry period started. In June, 255 hours of sunshine were recorded, with 192 being the 30-year average for the Netherlands (KNMI weather data), and only half the average rainfall. July again was warm and dry, with virtually no rain between 4 and 23 July. August was very warm, very dry and had a great deal of sunshine. During the first three weeks of August, almost no rain was recorded in the Netherlands. By the end of August, 22 mm rainfall was recorded, compared with 62 mm as a 30-year average. In September, the temperature had dropped, but it was again a sunny and rather dry month. During the week before and on the day of the final harvest on 6 October, there were several rain showers. This relatively wet period, shortly before the harvest, is the reason for some of the roots splitting, 1-2 days after the harvest.

Light interception by the shadow nets was measured on two days (9 July, 15.00 h., and 14 July, 11.00 h.) using a lux meter under open sky conditions. The light was reduced to 52% and 85% (average values) for the L1 and L2 plots. There were only very small differences amongst measurements within the shadowed plots (measurements on 12 July and 27 August), indicating that the shadow nets were homogeneous.

Due to the lack of rainfall and high temperatures, irrigation was applied twice: at the end of July and in the middle of August, 20 mm water each time. No drought stress was observed on the crop. This does not mean that there has not been any influence on the crop. During the gathering of soil samples it was observed that ridges without shadow nets were drier and tougher than ridges under shadow nets.

6.3 Intended and realised variables in the test field

Three factors were adjusted: nitrogen, light and (harvest) time, in three replicates.

Nitrogen

Although the nitrogen applications have realised a substantial difference in available nitrogen in the ridge, the effect on the crop was less than expected. This may be caused by relatively high water and nitrogen uptake from of deeper soil layers because of a drought in the topsoil.
The differences in the soil nitrogen measurements do not indicate any significant light influence (Annex 6.1). Annex 6.1 also shows that the field replicates were not significantly different in available nitrogen.

Light

In the carrot pilot project, mould infection occurred in those plots where the shadow nets were placed directly after the seedlings came up. Because of this, the nets in this experiment were placed after complete settlement of the seedlings in early July. The nets were homogeneous, and light quantity on the subplot used for analysis (3 x 3 m) was as expected. So, in the second part of the growing season, the variable light was as intended. The generally accepted standard for growth 'dry matter production' corresponds with the light levels (see Annex 7.4).

Time

The optimum harvest time for the cultivar Rodelika, as indicated by the seed firm, is 120 - 140 days. The main harvests were realised on day 136, and four weeks earlier and later. Small harvests were done at intervals of two weeks. Only the final small harvest, planned in between the second and third large harvests, was skipped for financial reasons.
In summary, the spread of harvests over time was good.

Nitrogen and time

The time series is supposed to create a gradual increase in differentiation processes (ripening). This is an autonomous physiological process, but is influenced by the environment. In this case, the soil was still rich in nitrogen at the end of the growing period (about 90 kg N ha^{-1} in 0 - 70 cm), which will stimulate continuing growth or reduce differentiation.

7 Methods and results of analyses of the carrot plants

The results of the different parameters are described below. The results of the parameters with replicates are statistically processed and the significant factors and tendencies are shown in tables and figures. Parameters without replicates, i.e. copper chloride crystallisation by BRAD, crude protein, protein ratio, and rot forming during storage, are only shown in figures if a trend appears.

7.1 Morphology of root and leaves

Introduction

The carrot plant has many morphological properties that may be related to the life processes of growth, differentiation and integration. Some of them are generally accepted: fresh weight and length and number of leaves are related to growth; the same is true of fresh weight and length of root. Other morphological properties may relate to differentiation and integration. Since morphological properties can be easily recognised by growers, it is very inviting to select morphological parameters that can be used to manage the life processes. Therefore a large number of morphological properties are measured in this study to discover parameters that are related to differentiation and integration. They are described by other researchers and are reviewed below.

The carrot is a biannual plant. In the first year, it develops leaves and a root until the end of the growth season in autumn. When the leaves have reached their maximum length, the root has also reached its maximum length. Then the leaves start to spread out and the root thickens. At the end of the growth period, the leaves start to age and turn yellow and brown, and some of them die off and the root uproots itself slightly (Visser and de Vries, 1979). In the second year, the plant uses the root as reserve organ and forms new leaves and flowers.

Reid and English (2000) state that the density of plants greatly influences growth. When the density is high, the leaf stalks become long and thin due to competition for light. The *mass and size of the leaves* increase when a lot of fertiliser is used, whereas a lot of light results in small leaves. Koeijer (2002) reports differences between carrot varieties with regard to *fine, symmetrical morphology* and *discolouration of leaves* (brown, yellow, red). Klett (1968) reports that carrot leaves exposed to a lot of light, show a tendency to xeromorphy; they are relatively small and strong, and the surface is hairy. Leaves exposed to a little light are hydromorph: they are broad, large and round and do not have hairs. The cuticle is reinforced, which results in a high turgor. Such leaves, when chopped off, wither quickly. Causes of variations in the *length and weight of the root* are germination time, differences in light exposure, soil differences and carrot variety. Open-pollinated varieties are genetically variable, whereas hybrids are genetically similar. The shape of the root of the open-pollinated variety Rodelika used for this study is slightly conical and similar to the shape of the carrot variety Flakkeese (see Figure 3). Koeijer (2002) mentions that an *orange, shiny and stumpy carrot root* indicates differentiation.
Rosenfeld (1998) found that *cylindricity* is connected with some chemical variables and might be used as a criterion for fully developed roots, together with root weight. Bleasdale and Thompson (1963) developed a method to calculate cylindricity. Furthermore, a method is reported for quantitatively studying the variability in size and shape of carrot roots of which 15 measurements were taken (Umiel et al.,1972). Since both methods are labour intensive, in this study, cylindricity is determined using a scale between two extreme forms (see Figure 6).

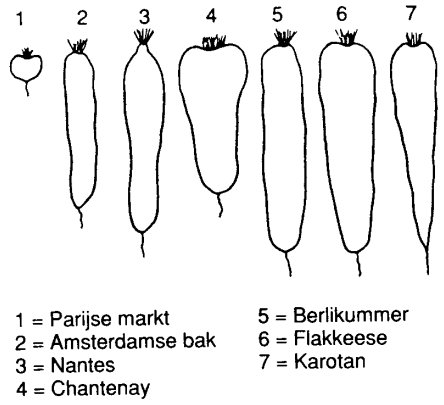


Figure 3. Root forms of different carrot varieties

The root increases in size by secondary growth of the vascular cambium. This produces xylem to the interior (core) and phloem to the exterior (cortex) (Esau, 1940). The ratio between cortex and core is an indication of differentiation of carrots, because the fraction of the core decreases during ripening in favour of the cortex (Schulz and Köpke, 1992; Koeijer, 2002). When the growth of the carrot plant is rapid, the cortex/core ratio of the root decreases (Bokhorst, pers. com.). The colour of the xylem core is paler than the phloem cortex due to a lower carotene content. The amount of carotene and colour of the root are determined genetically and by circumstances in the field. The colour of the root is more orange/reddish in dry, warm summers than in wet, cold summers. Generally, the carotene concentration correlates with an orange/red colour and increases with light, fertiliser and age. Low temperatures or a high water intake produces pale roots.

The hypothesis is that a wide availability of nitrogen in the soil stimulates growth processes and inhibits differentiation processes, and that light stimulates differentiation processes and also stimulates growth processes. Table 4 shows the hypothetical correlation between the chosen parameters and the growth and differentiation processes. In the table, a distinction is made between: 1) generally accepted parameters, 2) parameters which logically result from growth and differentiation processes, and 3) parameters which do not logically result from growth and differentiation processes, but may result from these considering the effects of nitrogen and light.

Table 4. Parameters that may relate to growth (G) and differentiation (D). Acceptability: 1 = generally accepted; 2 = results logically from G and D processes; 3 = does not result logically.
Arrows indicate positive or negative change.

Number	Parameter	Acceptance of parameter	Hypothetical correlation between the parameter and 'growth'	Hypothetical correlation between the parameter and 'differentiation'	Comments	Selected for this study
1	Fresh weight of leaves	1	Weight ↗ = G ↗	_	Extremely high temperatures can cause a decrease in fresh weight of leaves during time	+
2	Length of leaves	1	Length ↗ = G ↗	_	-	+
3	Number of leaves	1	No. of leaves ↗ = G ↗	_	During harvest leaves can break off	+
4	Coarse vs. fine morphology of leaves	2	Coarse ↗ = G ↗	Fine ↗ = D ↗	Statistical analysis not possible because of absence of repetition of measurements	+
5	Symmetrical vs. chaotic leaves	2	Chaotic ↗ = G ↗	Symmetrical ↗ = D ↗	Statistical analysis not possible because of absence of repetition of measurements	+
6	Discolouration of leaves	2	-	Discolouring ↗ = D ↗	Statistical analysis not possible because measurements include a general impression of the different plots	+
7	Fresh weight of the root	1	Weight ↗ = G ↗	-	Excessive water intake after drought can be a cause of a strong increase in this parameter	+
8	Length of the root	1	Length ↗ = G ↗	-	-	+
9	Leaves/root weight ratio	2		Ratio ↘ = D ↗		+
10	Conical vs. Cylindrical root	2	Conical ↗ = G ↗	Cylindrical ↗ = D ↗	Shape of a carrot is mainly dependent on the variety	+
11	Pointed vs. stumpy root	2	-	Stump ↗ = D ↗	-	+
12	Orangeness of the root	2		Intensive orange ↗ = D ↗	Analysis of the colour of the root is difficult because of differences in colour due to differences in moisture of the root	+
13	Cortex/core ratio of the cross-section of the root	3	-	Cortex/core ratio ↗ = D ↗		+
14	Growing leaves at the end of the growth period	1	Still growing ↗= G ↗	-	Statistical analysis is not possible because measurements of one single plant	+
15	Shiny vs. dull surface of the root	3	-	-	-	+
16	Smooth vs. ribbed surface of the root	3	-	-	-	+

17	Die-off of leaves	1	Die-off ↗ = G ↘	-	Statistical analysis is not possible because measurements include measurements of one single plant	+
18	Shape of the leaf blade	3	-	-		+
19	Deformed and cracked roots	2	-	Cracked roots ↗ = D ↘	Carrot with a high level of integration may reduce deformed and cracked roots	+
20	Spreading out of leaves	3		-	Statistical analysis is not possible because measurements of one single plant	+
21	Greenness of leaves	3	Dark green ↗ = G ↗	-	-	+
22	Differentiated pattern of cross-section of carrot root	3	-	Differen-tiated ↗ = D ↗	-	+
23	Horizontal vs. vertical leaves	3	-	-	Overlap with shape of the leaf blade	-
24	Density of leaves per m^2	3	Density ↗ = G ↗	-	Overlap with fresh weight of leaves	-
25	Density of a single leaf	3	Density ↗ = G ↗	-	Overlap with fresh weight of leaves	-

Method

Preparatory activities

28 carrot plants per plot were photographed in a row from small to large, one photo per plot (see Figure 4). A second photo per plot was taken of the smallest carrot plant, the middle carrot plant and the largest carrot plant. Of these three plants, the middle carrot plant is used to make a line-up of leaves (see below), and the other two are not used any more. The remaining 25 plants were used for several measurements. These measurements took place on the day of harvest and, after storage in plastic bags at 6°C, the day after.

Figure 4. Example of a row of 28 carrot plants

Almost all observations were done in repetition no. 2, 3 and 4; a few (14, 17 and 20 in Table 2) in repetition 1. In repetition 1, one single marked plant per plot was observed and measured after removal of the adjacent plants (on day 124 after sowing). Since the cotyledons were not then there, the sequence of leaves from old to new could not be traced back. Therefore, the number of stages in a leaf is determined as it is related to the sequential number of the leaf. For instance, the second leaf has two stages (the leaf apex was not counted). This method is reliable until the 8th leaf. Further, one photo per plot of repetition 1 at five stages is taken.

The line-up of leaves, which was made of repetition 2 only, is the sequence of leaves of one plant stuck on paper. The oldest leaf is on the left, the newest leaf on the right (Figure 5). The line-ups of leaves are then compared.

Figure 5. Example of a line-up of leaves on harvest day 124

The parameters were determined as follows:

1. Fresh weight of leaves: of 25 plants after separation from the roots.
2. Length of leaves: the length of the longest leaf of each of the 25 carrot plants.
3. Number of leaves of each of the 25 carrot plants.
4. Coarse vs. fine morphology: examination of the line-ups of leaves.
5. Symmetrical vs. chaotic leaves: examination of the line-ups of leaves.
6. Discolouration of leaves: changes in colour in the leaves at the end of growth.
7. Fresh weight of the root: of each root of 25 plants.
8. Length of the root: of each root of 25 carrot plants; it is defined as the distance from the base of the leaf stems to the rounded termination at the root tip or to a root thickness of 2 mm.
9. Leaves/root weight ratio is derived from parameters 1 and 7.
10. Conical vs. cylindrical roots: photos of 28 carrot plants were viewed on a computer-screen and the roots were scored on a scale of 1 for conical to 10 for cylindrical (see Figure 6). This was only done for the last three harvest days: 124, 136 and 164.
11. Pointed vs. stump roots: photos of 28 carrot plants were viewed on a computer screen and the roots are scored on a scale of 1 for pointed to 10 for stumpy (see Figure 7).
12. Colour of the root: visual examination on the day after the date of harvest.
13. Cortex/core ratio of the cross-section of the root: 25 carrot roots were cut at 1/3 length from the upper end and the diameter of the carrot root and the diameter of the core is expressed as cortex/core ratio.

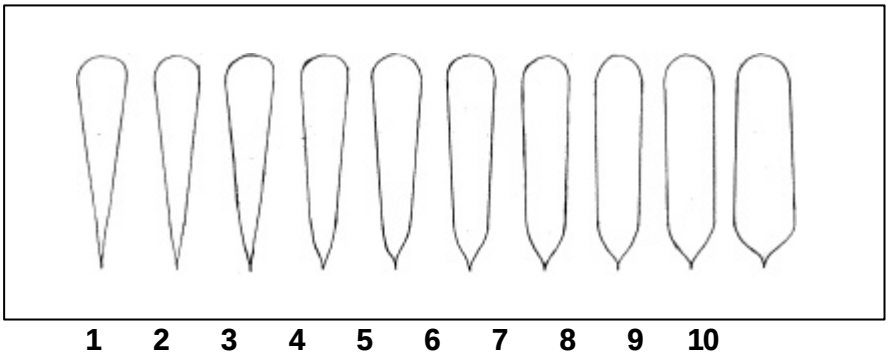


Figure 6. Scale for the parameter conical vs. cylindrical roots

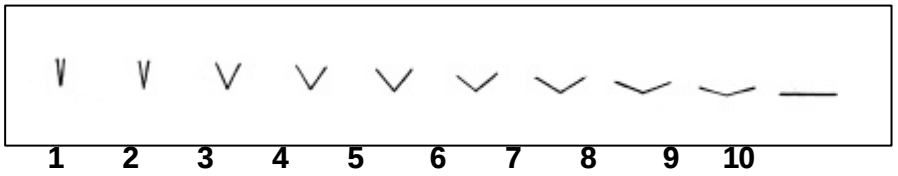


Figure 7. Scale for the parameter pointed vs. stumpy roots

14. Growing leaves at the end of the growth period: marked plants of repetition 1 are monitored five times.
15. Shiny vs. dull surface of the carrot root: photos of the 28 carrot plants were viewed on a computer-screen.
16. Smooth vs. ribbed surface of the carrot root: photos of 28 carrot plants were viewed on a computer screen.
17. Die-off of leaves: marked plants of repetition 1 are monitored five times.
18. Shape and size of the leaf blade: examination of the line-ups of leaves.
19. Deformed and cracked roots: counting after harvest, roots with these deviations were not included in the 28 plants used for the further examination.
20. Spreading out of leaves: one plant per plot in repetition 1 was used and adjacent plants were removed. Because the position of leaves is influenced by neighbouring plants, this parameter is excluded from this study.
21. Greenness of leaves: some examinations with a colour index were carried out, but the measurements are too variable due to differences in colour within a single leaf, the variable background, and the change in colour of leaves due to differences in moisture. Consequently, this parameter is excluded from this study.
22. Differentiated pattern of the cross-section of the carrot root: not included due to lack of time.

Results

Of 19 morphological parameters studied, data on the following 6 parameters, for 7 harvest days, were statistically processed: fresh weight of leaves, length of leaves, fresh weight of the root, length of the root, conical vs. cylindrical roots and pointed vs. stumpy roots.

1. Fresh weight of leaves
In Annex 7.1.1, it is shown that the weight of leaves increased until day 124 and decreased thereafter. The sharp decrease on day 136 may be due to an error in the selection procedure, see also 7 below, or the drought (see Section 6.2). Nitrogen level had a positive effect on weight on day 108 only, with a significant difference between levels 0 and 200 N.
Light had an effect on weight on day 136 only, with an optimum at 85% light. The difference between 52% and 85% light is significant.

2. Length of leaves
Annex 7.1.1 shows that the length of leaves reached at all levels of light is at a maximum on day 124. Earlier on, light tended to have a decreasing effect whereas after day 124, both 52% and 100% light correlated with shorter leaves. Nitrogen together with light had a significant effect on day 96 and day 124, but no clear trend can be described.

3. Number of leaves
The method to determine the parameter is not used consistently over time. Moreover, the leaves broke off easily during harvesting and handling. Therefore this parameter is discarded.

4. Coarse vs. fine morphology of leaves
On harvest day 82, the leaves were still weak and very fine. It took about a month from then on to develop strong, broad leaves. On days 96 and 108, the line-ups of leaves showed that more light causes more refinement of the leaves. However, on days 136 and 164, the line-ups of leaves showed no differences related to light.

5. Symmetrical vs. chaotic leaves
On days 108 and 124, the line-ups of leaves showed more chaotic leaves at 52% light compared with 85% and 100% light. However, on day 164, the leaves at all three levels of light were rather chaotic, possibly due to poor weather conditions during harvest and to senescence of the leaves.

6. Discolouration of leaves
The expected yellow and orange colours did not develop in autumn. No differences between the different treatments were found.

7. Fresh weight of the root
Annex 7.1.2 shows a continuous increase in fresh weight of the root over time, except on day 136. This cannot be explained by a change in growth conditions and must be caused by a systematic measurement error. The fresh weight of the root is positively correlated with light. The nitrogen level had no effect on the weight.

8. Length of the root
Annex 7.1.2 shows that the length of root generally increased over time, but not on day 136. As was said previously, this must be caused by a systematic measurement error. Light had a positive effect on the root length. On day 164, the roots at 85% and 100% light were significantly longer than at 52% light.

9. Leaves/root weight ratio
Annex 7.1.2 shows a rapid decrease in the leaves/root weight ratio until day 124, and was most rapid at 85% and 100% light. After day 136, decrease in the ratio was less.

10. Conical vs. cylindrical roots
Annex 7.1.3 shows an increase in cylindricity with more light. On day 164, the effect of light on cylindrical roots was most pronounced at low nitrogen levels and disappeared at higher nitrogen levels.

11. Pointed vs. stumpy roots
Annex 7.1.3 shows the change in pointed to stumpy over time. More light correlated with increasing stumpiness of the root, where the difference between 85% and 100% was negligible.

12. Colour of the root
The colour of the roots changed after day 124, depending on both light and nitrogen. Roots grown at 100% light and high nitrogen levels were more orange than those grown at 52% light and low nitrogen levels.

13. Cortex/core ratio of the root
Annex 7.1.4 shows that the cortex/core ratio had a tendency to decrease over time, except for some treatments on day 136. This can be caused by drought. Due to the high standard deviation, differences between treatments were not statistically significant. No effect of light and nitrogen is found. The Annex also shows a weak correlation between the width of the root and the cortex/core ratio, where thicker carrot roots had a relatively smaller cortex and a larger core.

14. Growing leaves at the end of the growth period
The results of this parameter did not show clear effects of fertiliser or light.

15. Shiny vs. dull surface of the root
No differences were found in shininess or dullness of the surface of roots.

16. Smooth vs. ribbed surface of the carrot root
The results are only indicative, because the photos were examined only briefly. Roots grown at 100% light and high nitrogen levels seemed to show a smoother surface, whereas roots grown at 52% light and low nitrogen levels seemed to show a more ribbed surface.

17. Die-off of leaves
No differences related to the different treatments were found.

18. Shape and size of the leaf blade
It is observed that the shape of the leaf blades of one plant changed from round to more triangular over time. Comparing the line-ups of plants of the different treatments, it appeared that the shape of the leaves (from the 7th leaf onwards) became more triangular at increasing light levels.

19. Deformed and cracked roots
Annex 7.2 shows an overall significant effect of light and time, i.e. more deformed roots at 52% light and significantly more at 100% light, combined with a tendency towards more deformed roots over time.
A few carrots got a longitudinal fracture during harvesting or washing. After day 164, during transport and/or storage before examination in the laboratories, more carrots got fractures. The fractured carrots were discounted.

Correlations
The correlation between the six parameters is shown in Annex 7.1.5. Only a few parameters have a high correlation with each other: fresh weight and length of root; fresh weight of root and pointed vs. stumpy root; and fresh weight and length of leaves. A somewhat lower correlation is found between fresh weight of leaves and length of root, and pointed vs. stumpy and conical vs. cylindrical root. The low correlation between fresh weight of leaves and fresh weight of the root is due to the decrease in fresh weight of leaves on days 136 and 164.

Discussion
It is expected that weight and length of leaves, and weight and length of the root are generally accepted parameters for growth. The weight and length of root were not affected by increasing nitrogen, confirming the results of other investigators who found higher leaf weight but not higher root weight after nitrogen addition (Wistinghausen, 1975; M. Koller, pers. com.). Although the weight and length of root increased only with more light, they should still be considered as parameters for growth.

The morphology of leaves showed a tendency to change with more light: they tended to become finer, more symmetrical and more triangular. This may indicate differentiation.
The leaves/root weight ratio decreased regularly over time without the fluctuations seen by the leaves and root weight. It may be an indicator of differentiation, because of the effect of both time and light.
Over time, and correlated with increasing fresh weight, it is expected that the root becomes more stumpy and cylindrical. In this study, the stumpiness correlated with both light and time, and therefore this parameter may indicate differentiation. The cylindricity was affected by both light and nitrogen, where increasing nitrogen had a tendency to inhibit the cylindricity, but only on the final day of harvest and under high light conditions. However, this may be an indication of a differentiation parameter. The absence of the expected increase in time can probably be assigned to the lack of data for the first three days of harvest.
The orangeness of the root appeared stronger at both highest levels of light and nitrogen. However, the carotene content which is responsible for colour, was not affected by nitrogen level. Consequently, the meaning of the colour is unclear. The cortex/core ratio of the root decreased over time. The ratio is correlated with the root size and not with the level of light. Therefore, in this case, the ratio is not considered to be an indicator of differentiation. The smoothness of roots increased with increasing nitrogen and light and therefore might be growth parameters (tendency only).
In conclusion, weight and length presented as growth parameters, whereas leaf morphology (fine, symmetrical and triangular forms), leaf/root weight ratio, stumpiness and cylindricity (tendency) of root presented as differentiation parameters.

7.2 Pests and diseases

Introduction
In general, the plant as it appears in the field is the result of environmental conditions during the growing season. However, a change in an environmental condition does not necessarily mean a change in the plant or crop. This depends on its ability to resist, or react to environmental change. In the same way, the presence of pests or diseases in the plant's environment do not necessarily mean that the plant will be affected. The resistance to, or the reaction to attack by insects or micro-organisms is a reaction of the plant as a whole, and is considered to be an expression of the highest level of organisation of the plant. In the Inner Quality concept, this is known as the integration level. Pests and diseases are two of the few parameters in this concept that can be directly connected to this level.

Method
Before each harvest, a short field survey was carried out. If any pest or disease was observed on the leaves, the percentage of affected plants per plot was estimated. The washed carrot roots from every plot (see Chapter 4) were visually examined and damage due to a particular pest or disease was estimated.

Results
Only late in the season (day 136), was mildew found on the leaves of most plants, as was the case in the adjacent carrot field with another cultivar. No differences could be observed between the treatments, and the presence of the disease was comparable to the adjacent carrot field. The disease manifested itself a little more towards the final harvest (day 164), but no records were made.

During the season, some roots were affected by carrot fly (Psila rosae), but the attack rate was low. From the first harvest day up to day 136, the percentage of affected roots increased from 0% to 2% (not significant). No nitrogen or light effect was observed. On day 164, the percentage of affected roots suddenly became significantly higher (from 2% to 10%), caused by Alternaria spp. No nitrogen or light effect was observed.

Discussion

Although some time effects were observed, treatments by light and nitrogen did not create differences in susceptibility to the observed pest and diseases. The high percentage of Alternaria-affected roots shortly before harvest has been frequently observed in the Netherlands in recent years and is a serious problem, but remarkably the (small) nitrogen and (larger) light differences had no effect on susceptibility.

7.3 Monosaccharides and saccharose in roots

Introduction

Monosaccharides are produced by photosynthesis in leaves and are used for growth and maintenance. The surplus is stored in the root to be used the following year of this biannual plant. At the start, mainly fructose and glucose are stored, later on an increasing amount of saccharose (Wistinghausen, 1975). The sugar content of the root usually increases up until 60 – 90 days after sowing and may stabilise thereafter at a level of 6 – 12% of dry matter (Schoneveld and Zwanepol, 1991). However, Bokhorst (1985) found an increase in sugar content at a late stage and attributed this to ripening. High temperatures and drought have a positive effect on sugar concentration. Sugar is an energy and carbon source for growth. High nitrate uptake mobilises sugar to be used as an energy source for nitrogen assimilation, thus having a negative effect on sugar concentration (Schoneveld and Zwanepol, 1991). This explains the negative correlation of saccharose and nitrate content of carrot roots found by Fleck et al (2001). He considers a low monosaccharide/disaccharide ratio as an indicator of ripeness.

Method

On harvest days 108, 136 and 164, samples of 25 roots from each plot were used for analyses. The determination of D-glucose, D-fructose and saccharose content was carried out by Silliker Laboratories in Ede, NL, using the method of Boehringer Mannheim 716.260.

Results

In Annex 7.3, the glucose and fructose contents are combined in the monosaccharide content. The figures show that the monosaccharide content decreased strongly until day 136, whereas the saccharose content increased all the time, but less between days 136 and 164. The saccharose content increased at higher levels of light. No effect of nitrogen level on monosaccharide or saccharose content was found. The monosaccharide/saccharose ratio decreased until day 136. It decreased at higher levels of light.

Discussion

Monosaccharide and saccharose contents show an opposing pattern confirming the results of other investigators (Fleck et al., 2001; Wistinghausen, 1975). The increasing saccharose content and as a result the decreasing monosaccharide/ saccharose ratio are the results of a differentiation process. Both parameters are stimulated at higher levels of light and over time and are therefore considered to be differentiation parameters.
The absence of a nitrogen effect cannot be explained.

7.4 Dry matter, nitrate and total nitrogen in roots

Introduction

In many products, dry matter of fresh weight is known to increase during the ripening process. It is also frequently reported that a high nitrogen availability during growth leads to a lower dry matter content. Organically grown products often have a higher dry matter content than conventional products due to a lower supply of fertiliser (Wistinghausen, 1979).
Nitrate is actively absorbed by plants from the soil. The main part of nitrate is transported to the leaves, stored in the cells or reduced to ammonium. The other part is reduced in the root and converted to amino acids and amides and transported to the leaves. The total nitrogen content of products which include nitrate, amino acids, amides and proteins, can vary considerably, depending i.a. on nitrogen availability during growth. The nitrate content increases when light is limited. Since nitrogen availability is one of the varying factors in this study, the total nitrogen content is a measure of the absorption of nitrogen.

Method

Samples of 25 roots per plot were used for analysis. The analysis was done by Silliker Laboratories in Ede, NL. Dry matter of fresh weight was determined for the seven harvest days after drying at 70°C. The nitrate content was determined according NEN-EN 12014-7 for the three main harvests. The total nitrogen content was determined using the Dumas method described in AOAC 998.06 for the three main harvests.

Results

The results are shown in Annex 7.4. The dry matter increased over time from ca. 10% on day 68 to ca.14 – 16% on day 136, depending on light levels, and decreased slightly on day 164 at 52% and 100% light. Dry matter increased with light levels, while the nitrogen level had no effect. Nitrate content increased until day 136 and was affected by both nitrogen and light levels. It increased at higher nitrogen levels and decreased at higher light levels.
Over time, total nitrogen content increased from day 108 to day 136, and then it decreased. Light only had a significant effect on day 164: total nitrogen content decreased when light levels increased from 52% to 85%, while a further increase to 100% had no significant effect.

Discussion

The differences in dry matter content over time and due to light levels are in line with expectations. A dry matter content of 14-16% is high for carrots, but normal for this Rodelika cultivar. The decrease in dry matter on the final harvest day may be due to the weather. Heavy rainfall occurred shortly before the final harvest, which may have caused a temporary increase in water uptake. We cannot explain the absence of a nitrogen effect on dry matter.
The nitrate content is higher when light is limited. This corresponds to experience and the nitrogen metabolism in plants. The positive correlation between nitrogen level and nitrate content is also in line with expectations, assuming an excess of available soil nitrogen. The EU regulation for baby food limits the nitrate content of carrots to 250 mg/kg, and after 1 November 2004 to 200 mg/kg. This limit can be met with sufficient light and less than 100 kg N/ha. The total nitrogen content increased from day 108 to 136 and afterwards it decreased sharply. This indicates a remarkable change in nitrogen metabolism during the final period. Before day 136, the root grows and the nitrogen content (per kg fresh weight) also increases, indicating nitrogen absorption higher than dry matter production. After day 136, the root still grows, but the net nitrogen absorption seems to be very low, and nitrogen level has no effect. This change indicates a sharp decrease in assimilation of nitrates in nitrogen compounds, except for free amino acids and protein (see Section 7.5). This means that after day 136, the synthesis of amino acids and protein is preferred to synthesis of other nitrogen compounds.
Dry matter, nitrate and total nitrogen content are products of primary metabolism, and thus indicators of growth. The sharp change in assimilation of nitrogen compounds may point to intensification of the differentiation process.

7.5 Protein and free amino acids in roots

Introduction

Wistinghausen (1975) assessed the quality of agricultural crops by measuring pure protein in relation to crude protein (Kjeldahl-N) which includes free amino acids. He found that the pure/crude protein ratio increased at the end of the growth season. In this study the protein ratio is determined with the following consideration: the plant takes up nitrogen as nitrate and uses it to make amino acids and then proteins, mainly present in carrot as enzymes. This process will continue when all growth factors are optimal (supply of water, light intensity, temperature, major and minor nutrients). If growth stagnates, protein synthesis is inhibited and free amino acids may accumulate, for instance when nitrate supply is high. This results in a decreasing protein ratio. Synthesis of amino acids and protein requires much energy and is therefore highly dependent on the supply of sugars. The proportion of protein and free amino acids expresses the physiological state of the plant and is also called 'physiological amino acid status'. High pure protein content indicates that mass building is nearly finished and that sugar supply and nitrogen uptake are in balance. High free amino acids content compared to pure protein content indicates unbalanced growth which leads to susceptibility to pests and diseases and low storage quality (Trophobiose theory of Chaboussou (1987)).

Method

At harvest days 108, 136 and 164 samples of 24 roots from each plot were used for the analyses. The analysis is performed by Kwalis.
For the determination of free amino acids in the roots, the laboratory used the method of VDLUFA, Anlehnung Bd. VI method C 30.3 which included blending, denaturation and separation of protein by trichloracetic acid, identification and quantification by use of an animo acid analysator with ninhydrin derivatisation and UV detection.
Crude protein (i.e. amino acids, peptides and protein) was determined by use of the Kjeldahl method, described in VDLUFA, vol. III, method 4.1.1.
The protein ratio was calculated as: (crude protein minus free amino acids)/crude protein.

Results

In Annex 7.5 it is shown that crude protein tends to increase during the season. There were no clear trends demonstrating the influence of light or additional nitrogen. Only at 100% light additional nitrogen gave consequently a higher crude protein content.
The pure/crude protein ratio decreased during the season which was not expected. There were no clear trends demonstrating the influence of light or additional nitrogen on protein ratio.

Discussion

The increase of crude protein and the decrease of pure/crude protein ratio during time resulting from an increase of free amino acids confirm the results of the pilot carrot study, but it is different from the increase of the protein ratio reported by Wistinghausen (1975). The decrease of protein ratio during time indicates a limited conversion of amino acids into protein, also at high level of light. This suggests that the level of light and synthesis of sugar is not the limiting factor.
During the period of the harvests the average root weight increases from roughly 25 to 70 gram whilst the dry matter percentage of the roots increases only slightly. Growth defined as 'increase in dry matter' is still going on, but the total nitrogen content decreases sharply (see §7.4). So the pure protein build-up in this period is much lower than dry matter production, and also low compared to nitrogen uptake. This means that the synthesis of nitrogen compounds other than free amino acids and protein was relatively small. According to literature this indicates lack of differentiation or integration.

7.6 Carotenes in roots

Introduction
Alpha- and beta-carotenes constitute the main part (90-95%) of the carotenoids which give carrots their orange colour. They are an important source of vitamin A and play a role as antioxidants. They are not soluble in water and their concentration increases during the growth period until 90-120 days after sowing and remains roughly constant thereafter. Carotene synthesis is stimulated by dry, warm weather with an optimal temperature of 17°C, and by fertiliser (Schuphan, 1976; Schoneveld and Zwanepol,1991).

Method
On harvest days 108, 136 and 164, samples of 24 roots from each plot were used to determine carotenes. The analysis was done by Silliker Laboratories using method AOAC 45.104.95. This method comprises a column for separation of carotenes.

Results
In Annex 7.6, the increase in carotenes over time and the effect of light is shown. The effect of light increased over time, but on harvest day 164, 100% light did not produce more carotenes than with 85% light. Addition of nitrogen to the soil had no effect on the carotene content.

Discussion
Results from literature give no clear arguments for carotenes as a growth or differentiation parameter. The increase in carotenes during crop growth in this experiment is in line with expectations. Increasing light also caused an increase in carotenes. Light influences both growth and differentiation, but together with the time effect, we tend to classify carotenes as a differentiation-related parameter. Since carotenes are carrot-typical, further research should clarify this.

7.7 Sensory properties of roots

Introduction
The taste and flavour of carrots can be directly experienced on the tongue and through nose of the consumer. Taste and flavour are known to be dependent on the type of cultivar, and for a certain cultivar, they are influenced by soil conditions, weather and agricultural practices such as fertilisation. Visser and de Vries (1979) reported that with a moderate supply of fertiliser, vegetative growth and ripening are in balance and optimal taste develops.

Method
On harvest days 108, 136 and 164, samples of two roots from each plot were used for determination. The roots from all plots were similar with respect to weight and shape. The sensory properties were investigated by the CSO Centrum Voor Smaakonderzoek in Wageningen. All 54 carrots were tested by a team of four experienced testers and a score was given for eight parameters and total appreciation.

Results
In Annex 7.7, the sensory properties are shown. The total appreciation is summarised in the figures. On days 108 and 136, the total appreciation of the roots from the plots with 52% and 85% light was negatively affected by the addition of nitrogen, while the roots from plots with 100% light were very much appreciated, no matter what the level of nitrogen. On day 164, the said effect of nitrogen level was not found; on the contrary, the negative effect of 52% light was neutralised by nitrogen level.
The attributes sweetness, acidity/astringency, soapiness and crunchiness showed interaction with light and nitrogen level on all harvest days. Yet, some optimal conditions were found: the sweetness is highest at

100% light without nitrogen level, the acidity/astringency and the soapiness were lowest at 100% light without nitrogen level (not shown).
The bitterness was lowest at 100% light without nitrogen level. On harvest day 108, the carrot taste was maximal at100% light and without nitrogen level, but on harvest days 136 and 164, the effects of light and nitrogen were unclear. On harvest day 108, the juiciness decreased with more light and nitrogen level; however, on harvest day 164 (after heavy rainfall), it increased with more light. On harvest day 108, the toughness increased with more light and nitrogen level; however, on harvest day 164, it decreased with more nitrogen addition.

Discussion

Different parameters show interaction between the level of light and nitrogen addition level, which means that the effect of one factor depends on the level of the other. Nevertheless, it can be concluded that the combination of high intensity of light and no additional nitrogen provides the best conditions for growing carrot roots with good sensory properties. Because the taste panel used no fixed standards, the results of the three harvests are not comparable and therefore no trend over time is described.
Total appreciation and carrot flavour are typical of the product, and can be considered as the result of both growth and differentiation, and therefore they may be an indicator of integration.

7.8 Copper chloride crystallisation of roots

Introduction

The copper chloride crystallisation (CC) method has been developed from the viewpoint that living organisms do not just exist as substances, but have structuring and organising forces. These forces control the form and function of the organism. The CC method was developed around 1930 by Pfeiffer in Switzerland and since then, has gradually further developed into a method by which experts can distinguish different effects of features (Engquist, 1963), e.g. farming systems, fertilisation practice, varieties and processing of crop and food samples. The CC method is mostly applied with juice from a living organism. The juice is mixed with a solution of copper chloride, put on a glass dish and incubated in a climate chamber to evaporate water and to crystallise the solids. The pattern of the crystals is then examined. It is thought that the type of crystal patterns reveals the character of the structuring forces, here called life processes, in the growing product. Three institutions: LBI, BRAD in Denmark and the University of Kassel in Germany have standardised the CC method in a combined study (Triangle project) using carrot and wheat (Kahl et al., 2005). LBI and BRAD used the standardised method in this carrot study with standardised carrot extract concentrations, without determining the mixing ratio as was done in the Triangle project (Andersen et al., 2003).

Methods

On harvest days 108,136 and 164, nine samples of 24 roots (8 out of each replicate) representing the nine treatments were refrigerated for one day and shipped in isolated boxes within 24 hours to BRAD. BRAD examined the samples of harvest day 108 over three days, whereby each sample was used twice to make extract replicates. The samples of harvest days 136 and 164 were each examined over two days using only one extract. On the same days, LBI received nine samples in duplicate which were refrigerated for one and two days respectively before examination.

The visual evaluation of the pictures was done on the basis of the morphological parameters developed in the Triangle project. However, it appeared in both laboratories that the pictures deviated considerably from the pictures obtained with carrots in the Triangle project. The pictures were judged to be of very poor quality. Probably the mixing ratios of the extracts were not optimal. BRAD used pictures of carrots and wheat of the Triangle project as a reference, whereas LBI used extreme pictures from the samples as a reference. Because of the difficult judgements, great caution must be exercised when comparing these results with those of other evaluations.

BRAD and LBI chose six respectively eight of the 14 morphological parameters to characterise the pictures. BRAD used: integration, substance spirals, dense radial formation, beweglichkeit, regularity of ramifications, and zonal dissolution. LBI used: centre-coordination, durchstrahlung, beweglichkeit, clear stems formed, fullness with side needles, substance spirals and lemniscate forms. The definitions of the parameters are described by Kretschmer (2003). Moreover, LBI scored for the 'group parameters' growth (based on durchstrahling, fullness with side needles, substance spirals, lemniscate forms and quernadeln), differentiation (based on centre coordination, beweglichkeit, clear stems formed and quernadeln) and integration (based on foremost centre-coordination). These three parameters were scored with different reference images per harvest day. As a result, no pronouncements can be made for the time factor. BRAD scored only four samples (consisting of the extremes of the treatments) of harvest day 164 for the 'group parameters' growth (based on certain morphology of stems and presence of dense radial formations and substance spirals), differentiation (based on certain morphology of stems and needles, and mainly absence of dense radial formations and substance spirals) and integration (based on certain morphology of stems and needles, and total absence of dense radial formations and substance spirals.

Results and discussion

Researchers into crystallisation in both BRAD and LBI came up against unexpected results. In the currently ongoing Triangle project, to validate the crystallisation method, a method with a mixing ratio matrix was worked out to determine the optimal mixing ratio of carrot juice, as well as of copper chloride that produces the most informative crystallisation pictures. As a result, an optimal mixing ratio was determined and many pictures of different carrot samples were produced up until the present carrot study.
This apparently optimal mixing ratio was used for the samples of harvest day 108. To the researchers' surprise, unusual looking pictures appeared. A dilemma arose, as changing the mixing ratio for harvest day 136 would disturb the time effect series. So a decision was made to continue with this mixing ratio, in the expectation that a constant and logical change in appearance would appear. This unfortunately did not occur.
The pictures remained unusual on harvest days 136 and 164 and were considered unsuitable for reliable judgement. Therefore the results are not shown. The lesson to be learned from this experience is that, whenever possible, a mixing ratio matrix should be made with the samples under investigation, to determine the optimal mixing ratio. This should ideally be done shortly before the examination takes place.

7.9 Delayed luminescence of roots

7.9.1 Introduction

Plants in darkness emit a permanent and extremely low radiation of light (Ruth and Popp, 1976; Popp et al., 1979). Emission becomes stronger when preceded by illumination (light-induced delayed luminescence or delayed luminescence). The intensity of the delayed luminescence decreases and, after some time, is identical to the low permanent emission. The transition time depends on the sample material and varies from a few seconds to several days. Delayed luminescence is correlated with biological processes, such as germination capacity of seeds (Saeki et al., 1990). Popp and Li (1993) showed that delayed luminescence of highly interconnected or coherent biological systems decays according to a hyperbolic law.

Different methods of excitation (white light and different colours) and different measurements of delayed emission (initial emission, total emission over a period of time, shape of curvature of emission) can be used. In this study, the cooperating laboratories, Kwalis and Meluna, used somewhat different measurements. The initial emission (intensity at t=1-2 s) was measured by Meluna. The total emission was measured somewhat differently by Kwalis and Meluna (see Subsections 7.9.1 and 7.9.2). The shape of the decay curve was measured by hyperbolicity by Kwalis and Meluna. The steepness of the decay curve was measured by slope by Meluna. Both laboratories used white light and different colours for excitation. Kwalis used colour filters for red, green, yellow and blue light excitation. Meluna used red and blue light only. Both laboratories measured the broadness of spectral ranges by expressing the emissions in relation to blue.

The broadness of spectral ranges of various products correlated with the growth and ripening conditions (Strube, 1998; Strube and Stolz, 1999, 2000, 2001, 2002). Excitation with different colours showed a broad spectral range with maximum emission after excitation with red, green and yellow light for the ripest apples and the apples most exposed to light (Bloksma et al., 2001). After maximum ripeness, the emission decreases due to loss in structure. Dormant seeds show a small spectral range, i.e. the emission is restricted mainly to blue, which is interpreted as minimum vegetative activity (Strube and Stolz, 2002). Both hyperbolicity and slope of the decay curve are related to the capacity to hold the emitted energy. The slope value seems to be more related to this capacity, while the hyperbolicity (expressed as ChiE or ChiE/ChiH) means the accuracy of fit to the hyperbolic decay curve. Values closer to zero indicate for both parameters a better organised (interconnected or coherent) biological system.

From the apple studies, it was concluded that initial emission and total emission are related to growth, whereas the broadness of spectral range might be related to differentiation. The hyperbolicity might be related to differentiation and integration, and hyperbolicity ratio (hyperbolicity red/blue) might be related to integration (Bloksma et al., 2004 b).

7.9.2 Delayed luminescence by Kwalis

Method

Ten carrots per plot harvested on day 108, 136 and 164 were washed on arrival and used for measurements. Two opposite sides of each carrot were used for recording luminescence and the values were averaged. For excitation, white, yellow, red and blue light was used. The emission after white excitation was measured 30–50 seconds after excitation and mentioned here as emission 30-50 white (in previous publications R40). Emission 30-50 ratio yellow/blue (previously R40Y/B) was calculated after yellow and blue excitation for 30–50 seconds after excitation. Emission 30-50 ratio yellow/blue indicates the broadness of the spectral range. Hyperbolicity white (previously ChiE/H50W) of the curvature was measured for 1–25 s after white light excitation and the hyperbolicity ratio red/blue (previously ChiE/H50red/bl) after red and blue light excitation.

Results

The results are summarised in Annex 7.8.1. In the overall model, that is over the three harvest days, the four parameters are positively correlated with light. However, only two parameters are significantly correlated with light on one particular day: emission 30-50 white on day 108 and hyperbolicity ratio red/blue on day 136. Nitrogen had an effect, although not a significant one, on emission 30-50 white and hyperbolicity white on day 164 only: they increased with more nitrogen (not shown).
The three parameters: hyperbolicity white, hyperbolicity ratio red/blue and emission 30-50 ratio yellow/blue reached highest values on day 136 and similar or slightly lower values on day 108.
The emission 30-50 white was highest on day 108 and decreased over time.

Discussion

Emission 30-50 white decreases over time and is indicative of the transition from growth to differentiation. The higher emission 30-50 white values at more light (significant) and not at more nitrogen indicates that the transition from growth to differentiation is slowed down under light conditions. It agrees with the continuous growth under light conditions and with the correlation of fresh weight of root with light, whereas nitrogen has no effect on weight (Section 7.1, Results, 7th item).
The strong decrease in the emission 30-50 ratio, as well as the hyperbolicity ratio (red/blue) on day 164 indicates that the differentiation is characterised by these parameters. The spectral shift that is prominent in differentiation is observed earlier in low light conditions.

7.9.3 Delayed luminescence by Meluna

Method

Only carrots from harvest day164 were measured. Per plot, two carrots were used. After washing, immediately before measurement, the middle part of 4 cm long was cut longitudinally through the centre, resulting in a flat inner side and convex outside. The flat inner side was used for recording luminescence. For excitation, white, blue and red lights were used. The delayed luminescence was characterised by the initial emission (at 1-2 s), total emission for 0.1-120 s and slope of the curve. Initial emission, slope and hyperbolicity (previously ChiE/H) were measured using white light for excitation, whereas total emission was measured using white, blue and red lights.

Results

The results are shown in Annex 7.8.2. The figure of initial emission white is very similar to the figure of total emission white; apparently these parameters were influenced in the same way by nitrogen and light. At low nitrogen, they increased with more light, but at high nitrogen, an opposite trend appeared. At low light, they increased with more nitrogen, but not at higher levels of light. The slope white increased with more light and reached its maximum with intermediate nitrogen. The total emission ratio red/blue was hardly affected by nitrogen or light, except an increased value at low nitrogen and low light levels. The hyperbolicity (not shown) was optimal for all measurements, and was not affected by nitrogen or light. The initial emission / slope white ratio (not shown) decreased with more nitrogen and more light.

Discussion

On harvest day 164, the initial emission white and total emission white showed almost the same, large differences with regard to light and nitrogen. They indicate for growth. The slope is also positively correlated with light intensity.
The higher slope value at increasing light levels means that carrots with more light released photons faster, which indicates a less organised system and so for less differentiation. The growth parameters indicate that, with increasing light, the condition of growth is better maintained on harvest day 164. This 'growth' effect may be caused by the wet weather shortly before the harvest.
The initial emission / slope white ratio is related to growth and differentiation, and thus for integration.

Comparing the results of day 164 of Meluna and Kwalis, it appears that the initial and total emission on the one hand, and emission 30-50 white on the other hand react similarly to nitrogen and light. Hyperbolicity by Meluna is optimal for all carrots and showed no differences, and therefore slope values are used for distinction. In contrast, Kwalis detected effects on hyperbolicity. Apparently, the two laboratories had different parameters for differentiation.

7.10 Electro-chemical parameters of roots

Introduction

Life processes in plants and animals can be described as chains of electro-chemical or redox reactions. Haas (1998) developed a bio-electrical theory to derive an electrical energy value for food from measurements of pH, redox potential and electrical resistance (Hoffmann, in: Meier-Ploeger and Vogtmann, 1988). He suggested that food with high reducing power, later expressed as low P-value, promotes health. The P-value is calculated from the three stated parameters which are affected by the growth conditions of the product.
The *pH-value* is the best known electro-chemical parameter to measure proton concentration or acidity. The pH is measured by potentiometry using suitable electrodes. The measured mV value is logarithmically transformed to the pH-value. The potentiometric equilibrium is at pH 7 and 0 mV. A difference in pH of 1 (at 25°C) equals 59 mV. In plants, proton activity has energy aspects.
The *redox potential Eh* (mV) reflects the gradient of electrons which life processes utilise for their cellular work (Kollath, 1978). The redox potential represents the equilibrium between oxidising and reducing

substances. When redox potential is low, plant cells have more energy for their activity. Traditionally, the flow of electrons is considered to be the main form of respiratory energy transport in an organism with oxygen as the terminal electron acceptor.
Electrical resistance R (Ohm) gives an indication of the dissipation of electrolytes in plant cells. High values of electrical resistance indicate that electrolytes are more integrated in membranes and cell organelles. Low values indicate free-moving electrolytes, which might be a sign of deterioration in plant cells and tissues.

In the first apple study, pH and redox potential had a weak correlation with growth and differentiation respectively (Bloksma et al., 2001). In the second apple study, the electro-chemical parameters showed no effects (Bloksma et al., 2004 b). Because the calculated P-value has a higher variation than the measured parameters, this report only gives the results of the measured parameters.

In this carrot study, the University of Kassel measured samples for harvest day 108, 136 and 164, while the Electro-chemisches Qualität Labor at Kirchberg/Jagst (D) measured samples for harvest day 164 only.

Methods

Measurements by University of Kassel:
16 carrots per plot were mixed to a sample size of 48 carrots per treatment on harvest days 108 and 164. On harvest day 136, 24 carrots per plot were used. The mixed sample was washed and divided in 4 subsamples with similar size distribution. After removal of the upper and lower quarters, juice was made with a Greenstar machine and adjusted to 25°C. The pH, specific conductivity, and redox potential in relation to the standard hydrogen electrode (Eh', mV) were measured using EQC apparatus. The electrical resistance (R, Ohm) was calculated from the specific conductivity (R = 1/mS x 1000). Because of unreliable redox measurements on harvest day 108, the gold and Pt-electrodes of Hamilton were replaced by a special Pt electrode of EQC for future harvest days.

Measurements by Electro-chemisches Qualität Labor:
17 carrots per plot were mixed to a sample size of 51 carrots. However, the mixed samples of plot N1L2, N2L1, N2L3 and N3L2 contained 25 – 33 carrots. The mixed samples were washed and divided into 5 subsamples. Juice was made using a Braun 4290 240 W machine and measurements were carried out using WTW Inolab apparatus with gold and calomel electrodes.

Results

Results of University of Kassel - Witzenhausen (DE):
The results in Annex 7.10.1 show that the pH increases over time and is positively related to light levels on harvest day 164 only. However, the increase was minimal (from ± 6.3 to 6.4). On days 136 and 164, the redox potential Eh' was significantly affected by light and nitrogen-interaction, but no clear trends can be seen, except for a positive effect of light at a high nitrogen level on day 136. The electrical resistance, R, decreased from day 108 to 136, and stayed at the same level thereafter. On days 108 and 136, R tends to increase at higher levels of light.

Results of Electro-chemisches Qualität Labor (EQL):
The results in Annex 7.10.2 are limited to harvest day 164 only. They show that the pH increased with more light, but the effect of light depended on the level of nitrogen. The redox potential Eh' and the electrical resistance R have an optimum at an intermediate nitrogen level.

Comparison UK and EQL:
Some similarities and differences between the results of UK and EQL are found. The pH values of both laboratories increase with the light levels, but the differences found by EQL are larger. For the Eh' and R, UK recorded a clearer light than nitrogen effect, while EQL only recorded a nitrogen effect. For the Eh', UK found no trend in the N-effect on day 164, while EQL recorded a nitrogen optimum at middle N. The same can be seen for the resistance, R.
The values of pH, Eh' and R of the two laboratories were at the same level.

Discussion
It can be concluded that the pH is positively related to light, increases over time, and can therefore be considered as a differentiation parameter. Resistance, R, also shows clear features of a differentiation parameter: increase with more light and a tendency to decrease with more nitrogen. The decrease in time (between first and third harvest days) is not in line with expectations for a differentiation parameter. Eh' does not show clear features for either laboratory.

7.11 Storage of roots

Introduction
From a physiological point of view, storage of carrots is a natural process. Storage in soil is the transition from the vegetative phase in the first year to the generative phase in the second year. At low temperature and high humidity, carrots can be stored for months with little loss of taste, moisture and nutritious compounds such as sugars, carotenes, vitamin C and proteins. Some changes take place (e.g. the conversion of a part of saccharose into fructose and glucose). This conversion starts at higher temperatures as preparation for the second growth period.
With respect to the Inner Quality concept, a good storability indicates a balance between growth and differentiation processes or high integration. Therefore, two storage tests were carried out. One storage test was a challenge at intermediate temperature and low humidity; the other test, at low temperature and high humidity, was more similar to storage in practice, and only performed on the final harvest day.

Method
Storage test at intermediate temperature and low humidity: On harvest days 108, 136 and 164, 75 carrots of each treatment were packed in a storage net. The nine nets were stored in a cellar at 16-19°C and approximately 50-70% RH. The nets were weighed before, during and after storage. At the end, the rotten carrots were counted. The nets for harvest day 108 were weighed after 4 and 9 weeks; the nets for harvest day 136 after 5 and 12 weeks; and the nets for harvest day 164 after 8 weeks.
Storage test at low temperature and high humidity: On harvest day 164, 50 carrots of each treatment were packed in a perforated plastic bag and stored at 6°C and approximately 98% RH. After 3 months, the bags were weighed and the rotten carrots were counted.

Results
The incidence of rot during the storage test at intermediate temperature and low humidity was as follows. The carrots for harvest day 108 were hardly affected by wet (bacterial) rot or dry (fungal) rot. The carrots for harvest day 136 showed 1-15% dry rot without correlation with light and nitrogen levels. The carrots for harvest day 164 showed 2-20% dry rot without correlation with light and nitrogen levels. These results are not presented.
The weight losses during the said storage test are shown in Annex 7.10. Generally, the weight loss is less at higher levels of light, whereas it has no correlation with the level of nitrogen. Comparing the weight loss of the different harvests, it is found that it was at its lowest with harvest day 136 and at its highest with harvest day 164. Light reduced the weight loss up until day 136; after this, the effect of light was less clear. The storage test at low temperature and high humidity showed that 0-6% of the carrots had wet rot, with the lowest incidence at low levels of nitrogen.

Discussion
The weight loss and occurrence of rot during storage is a property of the whole organism, and therefore an indicator of integration. The integration was optimal at the highest level of light and on day 136 and decreased thereafter.

8. Discussion of the varying factors

In this chapter, the effects of the varying factors on the different parameters are compared, and also compared with expectations based on literature on carrot and plant physiology. For this purpose, Table 3 in Chapter 5 and the results of the individual parameters in Chapter 7 are used.

8.1 Disturbing effects

Although the test field was situated apparently in a homogeneous soil and physical situation, and previously used homogeneously, most of the statistically processed morphological parameters showed significant differences among the repetitions in the test field; however, nitrogen availability did not (Annex 6). Observed irregularities in plant density and germination speed have caused inhomogeneous plots and are the source of a relatively high in-plot variation. Nevertheless, it cannot be explained how this has led to the significant differences among the repetitions.
This complication forced us to treat the data not as repetitions but simply as extra data per condition. This operation reduces the strength of the statistical output and the conclusions based on it.

8.2 Nitrogen level

Since we consider the lowest nitrogen level as optimal, we expected increasing or decreasing response curves only, and not optimum or minimum response curves. As mentioned in Section 6.1, the differences in available nitrogen level may have been smaller than foreseen. This reduces the possibilities of finding significant nitrogen effects.

The nitrogen level had no effect on the weight and length of the root, which confirms the results of Wistinghausen (1975) and M. Koller (pers. com.). Some other parameters were affected by nitrogen addition. The weight and length of leaves increased with more nitrogen, as did the nitrate content, crude protein content and the orangeness of the root, although the latter could not be confirmed by higher carotene content. These effects are in line with expectations.
The form of the roots became less cylindrical with increasing nitrogen levels in full light on the final harvest day but, at reduced light, it became more cylindrical. This confirms only partly the expectation of a carrot with a cylindrical shape when ripe.
Total sensoric appreciation, sweetness, carrot taste and juiciness decreased at higher nitrogen levels, while acidity/astringency, bitterness and soapiness increased. This confirms the practical experience that a high dose of fertiliser has an adverse influence on the taste of carrots. The negative effect of nitrogen addition on juiciness measured on harvest day 124 only was not expected.
The copper chloride crystallisation pictures produced by LBI and BRAD were not optimal or typical of carrots, due to a change in the method applied. This is probably why neither laboratory could demonstrate the expected differences caused by different levels of nitrogen.
The slope of luminescence by Meluna showed a maximum value at the middle nitrogen level, again unexpected. The hyperbolicity white of Kwalis had a tendency to increase with more nitrogen, which was expected.
The electro-chemical parameters of Kassel were not affected by nitrogen level.
The redox potential and electrical resistance by EQL tend, unexpectedly, to maximise at the middle nitrogen level.
The storage tests showed only a tendency to more rot at higher nitrogen levels, as expected.

Overall, the response to nitrogen has been smaller than to light and time. The general nitrogen level in the ripening period of the crop was high, also in the low-nitrogen plots, and may have had an effect on the results of the time series.

8.3 Light level

The different levels of light were realised as planned. Compared to the pilot study, the shadow nets were placed after full settlement of the crop, and therefore have had effect only in the second half of the growing season. This diminished the possibilities of finding characteristic differences in leaf shape and line-up of leaves because a fully developed leaf at the start of the growth period without shadow will not change its shape after the erection of shadow nets.
Since the light levels were 52%, 85% and 100%, differences can be expected to be smaller, between 85 and 100%, then between 52 and 85%. This was for some parameters indeed the case.

Most non-experimental parameters were affected by different levels of light as was expected. The weight and length of leaves were maximal at middle light level. The weight and length of root, saccharose (and sweetness), carotene (and orangeness) content, dry matter, total sensory appreciation (thanks to fewer off-flavours) and carrot taste increased with more light.
Other parameters also increased with more light: fine, symmetrical, triangular leaves (trend), cylindricity and stumpiness of roots, juiciness (only on the final harvest day).
Carrots grown at high light levels had less weight loss during storage.
Some experimental parameters were positively affected by more light, as expected: total emission white and slope white of luminescence by Meluna, pH and electrical resistance by Kassel, and pH by EQL.
Redox potential by Kassel had a tendency to decrease against expectation and redox potential by EQL was not affected.

Overall, the response to light has shown significant and expected effects.

8.4 Harvest time

After the warm, dry summer period, the late summer was fairly normal, and the harvest dates were evenly distributed over time. The above ground conditions were normal during the crop's ripening period. Overall decreasing light and temperature coincide with ripening processes in nature (Bockemühl, 1982). The soil itself displayed the characteristic of ongoing growth stimulation and more or less inhibition of ripening processes as described by Lammerts van Bueren (1990) and Visser and De Vries (1979). This might indicate that the variable time does not coincide fully with increased stimulation of differentiation processes. Together with the given optimum harvest moment (120-140 days), suggesting the possibility of finding an optimum in some of our measured parameters, this makes interpretation of the time-related processes complex.
In summary: the created time series may not represent clearly defined differentiation series.

Most non-experimental parameters changed over time as was expected. Weight and length of leaves, and cortex/core ratio decreased at the end of the season. Weight and length of root, stumpiness, dry matter, content of saccharose, crude protein (trend), and carotenes (and orangeness) increased over time, whereas the monosaccharides/saccharose ratio decreased over time. The increase in nitrate concentration can be explained by less light later in the season and an abundance of nitrogen, even in the plots without additional nitrogen.
The weight loss during storage was minimal for harvest day 124, the supposed optimal harvest day.

Not expected was the decrease (trend) of pure/crude protein ratio, when compared with the results of other investigators. This may be the resulting effect of ongoing nitrogen uptake from the nitrogen-rich soil, and limited nitrogen assimilation into pure protein due to weaker light conditions.
The sensory parameters of the various harvests cannot be easily compared due to lack of a standard. Total appreciation of the final harvest tended to be the best.
One experimental parameter increased during time: pH by Kassel. Other experimental parameters decreased over time: hyperbolicity and emission 30-50 white by Kwalis, and electrical resistance by Kassel. Hyperbolicity and emission 30-50 ratio by Kwalis was at a maximum on harvest day 124.

8.5 Comparison with the carrot pilot study in 2001

Nitrogen, light and time influenced the parameters of the pilot study in 2001 (with Yukon carrot and clay loam soil) partly in a similar fashion and partly differently to this study.
Leaf/root weight ratio, root weight, root shape (cylindricity), and sweetness were affected in the same way. These parameters are probably independent of carrot variety and soil type and could be used universally for carrot studies.
Nitrate, total nitrogen, crude protein, pure/crude protein ratio and dry matter reacted in the same way for the most part. The difference that was noticed might be due to the carrot variety and soil type or other circumstances in the field. This has to be made clear before these parameters can be used universally for carrots.
Other parameters (saccharose, carotenes, emission 30-50 ratio and hyperbolicity ratio of delayed luminescence, electrical-chemical parameters and total sensory appreciation) reacted very differently.

9 Discussion of parameters, life processes and the Inner Quality concept

9.1 Overview of results

In this chapter, the parameters are evaluated using following considerations (as shown in Table 2, Section 2.1):

- Growth parameters are positively affected by increasing soil nitrogen and light;
- Differentiation parameters are negatively affected by soil nitrogen and positively affected by light, and increase over time (or decrease when related inversely);
- Integration parameters can be affected by soil nitrogen and light depending on the proportion of the two, and can reach a maximum value (or minimum when related inversely).

We compare them with the following generally accepted parameters, based on observations obtained from many agricultural crops:

- Growth processes: fresh weight of leaves; fresh weight of root;
- Differentiation processes: fine leaves (finer leaves are more differentiated, by definition); dry matter of root (dry matter is higher when growth is limited and differentiation is sufficient);
- Integration: pests and diseases; total sensory appreciation; rot during storage; and weight loss during storage, because they are all related to the whole organism.

To enable recommendations for use of parameters in future, we included cost and strength (usefulness, validation, linked to consumers' quality) in the evaluation process.

The evaluation of the parameters is presented in Table 5 and further discussed in Chapter 9.2.

Key to Table 5:

Black cells:	generally accepted parameter;
Grey cells:	expectation according to the Inner Quality concept, literature and experience;
x:	the result shows a significant correlation with generally accepted parameter;
o:	the result shows a tendency towards a correlation with generally accepted parameter.
Question marks are explained in Section 9.2.	

Table 5 Relationships between parameters and lifeprocesses

Parameters	growth	differentation	integration	cost (+: low, -: high)	validated method	recommendation
Morphology						
weight leaves	**x**			+	+	+
length leaves	o			+	+	-
fine leaves		**o**		-	-	+?
symmetrical leaves		o		-	-	-
triangleness leaves		o		-	-	-
weight root	**x**			+	+	+
length root	o			+	-	-
leaves/root weight ratio		x?		+	+	-
cylindricity root		o		+	-	-
stumpness root		x		+	+	+
orangeness root		o		+	-	-
cortex/core ratio root				-	+	-
shininess root				+	-	-
smoothness root	o			+	-	-
deformed roots				+	-	-
Pests and deseases				+	+	+
Chemical content roots						
monosaccharides				+	+	-
saccharose		x		+	+	+
monosacch./saccharose ratio		o		+	+	-
dry matter		**x**		+	+	+
nitrate	x?			+	+	?
total nitrogen				+	+	-
crude protein				+	+	-
pure/crude protein ratio				-	+	-
carotenes		**x**		-	+	-
Sensoric properties						
total sensoric appreciation			**o**	-	+?	+?
sweetness		o		-	+?	+?
acid-astringentness		o		-	+?	+?
bitterness		o		-	+?	+?
carrot taste		o		-	+ ?	+?
soapiness		o		-	+ ?	+?
juiciness				-	+?	+?
crunchiness				-	+?	+?
toughness				-	+?	+?

Table 5 (continued)

Parameters	growth	differentation	integration	cost (+: low, -: high)	validated method	recommendation
Luminescence Kwalis						
emission 30-50 white	x			-	+	+
emission 30-50 ratio		x		-	+	+
hyperbolicity white	o			-	+	-
hyperbolicity ratio		x ?		-	+	?
Luminescence Meluna						
initial emission white	x			-	+	+
total emission white	x			-	+	+
total emission ratio				-	+	-
hyperbolicity white				-	+	-
slope white	x			-	+	-
initial emission/slope white			o	-	+	?
Electro-chem.Kassel						
pH		x		+	+	+
redox potential				-	+	-
electrical resistance		x ?		-	+	-
Electro-chem. EQL						
pH		x		+	+	+
redox potential				-	+	-
electrical resistance				-	+	-
Storage test						
rot			**o**	+	-	?
weight loss			**x**	+	+	+

9.2 Comments on specific parameters

Morphology

The morphological parameters have the great advantage that they can be directly observed and easily measured. Root weight (generally accepted growth parameter) and shape of the root (cylindricity or filling and stumpiness of root) are parameters used by farmers, although only partly quantified. Leaves/root weight ratio and stumpiness of root react to nitrogen and light as differentiation parameters. Cylindricity is probably related to stumpiness, but it is not validated in this study.
The change in leaf shape (symmetry and round vs. triangular form) and fineness could not be validated as a differentiation parameter, because of the weak sensitivity to light. Leaf colour and spread of the leaves (not quantified in this study) are used by farmers as ripening characteristics and need further investigation to be regarded as differentiation parameters, but they could also be specific to the type of cultivar.
We recommend use of root and leaf weight as growth parameters. Further study is necessary to validate leaves/root weight ratio as a differentiation parameter, together with study of leaf properties (firmness, colour, discolouration and decay of leaves) which may also indicate differentiation.
The cortex/core ratio in this study could not be related to the proportion of growth and differentiation as is suggested in another study. This needs more research.

Pests and diseases

Per definition, this is a parameter indicating integration, but further investigation is necessary to demonstrate the relation between proportion of growth and differentiation. For this purpose, plants could be deliberately challenged by infection.

Chemical content of roots

In the conversions from nitrate to protein and from monosaccharides to saccharose, the ratio between precursor and product is of interest. We need more in-depth plant physiological knowledge to be able to definitely include these parameters with the differentiation parameters. We also need more research to overcome the opposing results of our two carrot studies compared to literature.
Dry matter is an easy and clear parameter. It is relatively easy to make (per cultivar) development curves as reference for the judgement of a certain crop. Together with fresh root weight, two curves can be made: dry matter production per day, and dry matter content. The first could be used as a growth parameter, the second as a differentiation parameter which increases over time according the definition.
Saccharose content indicates differentiation. Monosaccharide/saccharose ratio indicates differentiation in the final harvest only and is therefore a less suitable parameter.
Carotenes, typical components of carrots, are secondary metabolites and thus a differentiation parameter which increases with more light and over time in this study. However, according to literature, fertiliser does increase the carotene content of the root which is not expected of a differentiation parameter. Only if we understand this better, does it make sense to use this parameter to assess the Inner Quality.

Sensory properties

Taste is an important parameter in marketing. A trained taste panel can produce reliable results. The parameter could win interest if a method could be developed to consistently compare harvests. Total appreciation is per definition an integration parameter and is composed of single taste components which could be related to either growth or differentiation. Sweetness is correlated with sugar content and thus related to differentiation. Carrot taste reacts to nitrogen and light as a differentiation parameter. Acidity/astringency, bitterness and soapiness react as growth parameters, but this is questionable for the latter two. Juiciness and toughness do not react in line with expectations; we recommend their incorporation in future research.

Copper Chloride Crystallisations

The unique selling point of the crystallisation method is the creation of pictures (crystallisation plates) which might express different life processes of the organism. Although the crystallisation method has a

long history, validation work with carrot and wheat has been recently undertaken by three laboratories (Kahl et al., 2005). In their validation study, including a concentration matrix to optimise the information expressed in the pictures, different characteristics in the pictures were distinguished which could be reproduced. However, in the current carrot study, the concentration matrix was not carried out and the pictures did not express the characteristics usually produced by the carrot. Therefore the results are excluded.
We recommend that the participating laboratories include the concentration matrix in the method protocol to avoid such problems in future. Moreover, the investigation of carrots grown under different fertiliser and light conditions should be repeated to demonstrate the relationship of the different characteristics of the pictures with growth, differentiation and integration.

Delayed luminescence

The concept of an organism emitting light according to its inner degree of energetic organisation is attractive. The results can be mathematically understood and elaborated. The relation between the single parameters on the one hand, and growth and differentiation on the other, is generally known. This makes them attractive for further research, especially if a database of a certain product is built up. In this report, we experienced difficulties in comparing the methodology and used parameters of both laboratories. Of the Kwalis parameters, the emission 30-50 white indicates growth and emission 30-50 ratio indicates differentiation, but the hyperbolicity ratio did not show a differentiation pattern. The Meluna parameters initial emission white and total emission white showed also a growth pattern. The slope white of Meluna showed a growth pattern, but was expected to be a differentiation parameter. The initial emission / slope white ratio might be an indicator for integration because it relates growth and differentiation parameters. It can be validated by use of generally accepted parameters.
The delayed luminescence method could gain in strength by harmonising the laboratory protocols and by validation.

Electro-chemical parameters

These parameters are easier to understand by mainstream scientists than delayed luminescence and copper chloride crystallisation. The pH and possibly electrical resistance could be related to differentiation, but redox potential was unexplainably influenced by nitrogen and light and could therefore not be related to any life process; thus it seems to be of no use for the Inner Quality concept.
It cannot be ruled out that further investigations will eventually shed more light on this matter.
The Dp-value (not shown in this study) can be calculated from the three mentioned parameters and may be related to differentiation or integration. The calculation of Dp-value is only worthwhile if the standard deviation of the three parameters is small compared with the measured differences, which was not the case. For a better understanding, we need a better insight into the physiological background of these parameters.

Storage tests

The storability is a direct expression of the product's resistance to stress. Because it reveals something on the level of integration and is cheap, it should form part of every quality test. The test should be standardised and validated. Contrary to the others parameters, this is not a measurement at one moment; it needs time.

9.3 Reflections on the Inner Quality concept

In this study, as was the case in the Apple-2 report (Bloksma et al., 2004 b), several parameters have obtained more credibility with regard to the life processes growth and differentiation. Some (experimental) parameters correlate well with the generally accepted parameters.
With respect to integration, both Apple-2 report and this study show promising parameters, although the integration aspect still needs further clarification.

The definition of integration as a balanced relation between growth and differentiation depending on cultivar and time (Table 1) suggests a continuous balanced relation between the two life processes from germination to harvest, and not an increase to a maximum or optimum as is the case in a ripening process. So we suggest, for further development of the Inner Quality concept, that a clear distinction be made between integration and ripening:
Integration is a continuous balanced relation between the growth and differentiation processes from germination to harvest; the proportion of the intensity of growth and differentiation processes can vary over time.
Growth processes include primary physiological processes such as photosynthesis, uptake of nutrients, tissue and organ formation and maintenance.
Differentiation processes are mostly plant-specific: refining and ordering plant forms, and include processes that are known as ripening such as storage of saccharose and production of flavours and coloured pigments.
The integration is well balanced over time when growth processes decrease and differentiation processes intensify leading to a closure of a life phase in a species-specific product.
A high Inner Quality can be reached by growing a plant with a high level of integration and by harvesting the product when the required ripeness is reached.

Firstly, the parameters used in this study can be ordered according to their relation to the physiological history at the moment of measurement.

1. Independent and varying. These parameters can go up and down, independent of the previous period. Nitrate belongs to this group, and so possibly may amino acids and monosaccharides.
2. Dependent and varying. These parameters result from the previous period, but can increase or decrease. They express the state of being at the time of measurement together with previous influences. Many experimental parameters belong to this group, e.g. leaf weight.
3. Dependent and cumulative. These parameters are irreversibly built up over time, such as root weight, dry matter, stumpiness of roots.

Secondly, we have to be very clear about the difference between processes and properties resulting from the processes. Strictly speaking, root weight is not a growth parameter, whereas increase in root weight per time unit is. The stumpiness of the roots is the result of growth (there are carbohydrates to be stored) and differentiation (the shape of the root changes). So the differentiation process is, in this case, the change in root shape, and the resulting measured root shape can be used as a parameter for a ripe product: sufficient differentiation in relation to growth. Whether it is possible to make use of more ratio parameters to exclude a growth component in a presumed differentiation parameter should be investigated.

Thirdly, we suggest further quantification. If we define (as has been contributed to in this report) a few parameters for growth only and strictly, and some for differentiation only and strictly, we can construct a quantitative, mathematical model for the presence and intensity of growth and differentiation processes over time. This forms the basis of a quantitative construction of hypothetical growth and differentiation curves as presented in the Apple-2 report. This can be validated in field trials and correlated with pests and diseases during cultivation (one of the few direct integration parameters) and parameters of the resulting carrots, such as taste and storage quality, and experimental parameters for integration. After validation, a quantitative growth/differentiation model is available. This model can serve as a reference model, and abbreviations during crop growth can be noted.

10 Conclusions and recommendations

10.1 Life processes

The life processes of the carrot plant were demonstrated by various parameters in this study.
Growth processes, including photosynthesis, absorption of nitrogen and other nutrients and formation of cells, tissues and organs, are measured by the parameters weight of leaves and roots and emission 30-50 white of delayed luminescence. The nitrate content may indicate growth but has to be investigated further.
Differentiation processes, including refining, ordering, ripening and secondary metabolisms, are measured by the parameters root stumpiness, saccharose, sweetness, dry matter and emission 30-50 ratio of delayed luminescence. Some other parameters may indicate differentiation processes but have to be further investigated: leaves/root weight ratio, monosaccharides/saccharose ratio, carotenes, initial and total emission white, hyperbolicity ratio and slope white of delayed luminescence.
Integration of growth and differentiation is measured by resistance to pests and disease, total appreciation and storage test. Some other parameters may indicate integration but need further investigation: carrot taste, slope white of delayed luminescence.
Copper chloride crystallisation did not produce clear pictures due to failure of the method applied and should be further investigated since, in the Apple-1 and -2 studies (Bloksma et al., 2001, 2004 b), it was one of the indicators of the life processes.
Electro-chemical parameters, except possibly pH, did not indicate growth processes in this study or in the Apple-1 and -2 studies.

10.2 Selection and validation of parameters

Parameters are useful if they are validated for the life processes and measure a broad range. It should be borne in mind that growth and differentiation can change over time and can be different for the different crop varieties.
Weight of leaves and root are validated to measure growth and are easy and cheap to determine.
Stumpiness of root, dry matter, saccharose content and sensory properties are validated to measure differentiation and are cheap and easy to measure. Sweetness is difficult to measure quantitatively, but can be used for a quick examination. The cheap parameters monosaccharide/saccharose ratio and carotenes and the expensive parameters emission 30-50 ratio of delayed luminescence can indicate differentiation processes which are not measured by the above-mentioned parameters and therefore should be investigated further.
Total sensory appreciation, resistance to pests and disease, weight loss and rot in a storage test are the parameters of choice to measure integration. The attack rate of pests and disease and rot and weight loss during storage did not show any major differences in this study, but they are promising and should be further investigated.

10.3 Relevance of life processes for Inner Quality

During the different stages of development of the carrot plant, the proportion of growth processes and differentiation processes can be different in order to maintain a high integration level. A high level of integration is expressed by absence of pests and disease before harvest and good storability after harvest. It is possible to draw figures of parameters expressing the intensity of the growth and differentiation

processes over time and to point out what proportion of intensities are desired during the development stages to obtain a high integration level. This proportion of intensities should be determined in further investigations.
It is defined that a carrot root has a high Inner Quality, if the integration level at all stages of development and storage is high. The hypothesis is that carrots with a high integration level are beneficial for consumers' health. This should be confirmed by research and is beyond the scope of this study.
Growers can measure parameters of growth and differentiation processes during the growth season and relate these parameters to integration parameters to understand the balanced relation of growth and differentiation over time. For instance, if certain intensities of growth and differentiation lead to an attack of insects or an infection by fungi, then the relation is not optimal. The same is true for storability.

10.4 Factors determining the integration of growth and differentiation

Carrot growers can steer the crop towards a high Inner Quality by taking the right measures at the right time. These measures include choice of soil type and soil fertility, watering and timing of harvest.
Growth is improved by soil with a good soil structure and without limits in nutrients - especially nitrogen - and water. Also warmth, in combination with sufficient available water, stimulates growth. In late spring, this can be realised more easily on clayish soils than on sandy soils. Later in the season, growth processes are supposed to become less prominent, in favour of differentiation processes, leading to a ripe product. In the Inner Quality concept, dry warmth and limited nitrogen availability support the shift from the growth-dominated phase to the differentiation-dominated phase. For carrots harvested in the cool, moist autumn, the requirements for optimal ripening processes are not met. In combination with a clayish soil, which was the case in this study, this can result in ongoing growth processes. New leaves are being formed, there is no discolouration of leaves to yellow and red, and nitrogen uptake continues, but there is not enough light to convert it into proteins.
Light is an important factor for differentiation, but cannot be managed by the farmer, except plant density: a low plant density results in more light.
Time may help in reaching sufficient differentiation by a hot, sunny autumn. Growth processes should then be reduced by absence of nitrogen.
In conclusion, high Inner Quality of carrots on clayish soils can be obtained with limited nutrients (no additional fertiliser or manure), possibly a little less plants per ha, and late harvest. This might result in a lower yield with high Inner Quality. On sandy soils (not in this study), the concept of steering towards a high Inner Quality is the same, but the measures will be different. The autonomous soil processes on sandy soils might favour differentiation processes, while the growth processes tend to be weak.

10.5 Recommendations

- For further understanding and communication of the Inner Quality concept, it is necessary to repeat the carrot study in two, preferably three countries to demonstrate the life processes and the corresponding parameters. Only a limited number of parameters, validated in this study, and of low cost, should be used. These are: weight of leaves and root for growth processes; leaves/root weight ratio, stumpiness of root, saccharose, sweetness and dry matter for differentiation processes; prevalence of pests and disease, total sensory appreciation, weight loss and rot in a storage test for integration processes.
  The difference between integration and ripening must be clarified and communicated to growers and the trade:

- Integration is considered as a optimal (changing) relation and interaction of growth and differentiation;
- Ripening, being a part of the differentiation processes, occurs during the last phase of a plant development process in which growth processes decrease and stop, and differentiation processes become dominant and, in the end, also stop;
- High inner quality is obtained when a product has a optimum relation and interaction of growth and differentiation throughout its development and is harvested when the desired ripeness is reached.

  We suggest that exclusive growth and exclusive differentiation parameters be further quantified to construct growth and differentiation curves (see the hypothetical curves in the Apple-2 report). They can be correlated with the integration parameters pests and disease, total sensory appreciation and storability to build up a reference life process model.

- in separate studies, delayed luminescence and copper chloride crystallisation should be investigated to validate parameters for the life processes. Reference series of carrots can be obtained from the above-mentioned study.

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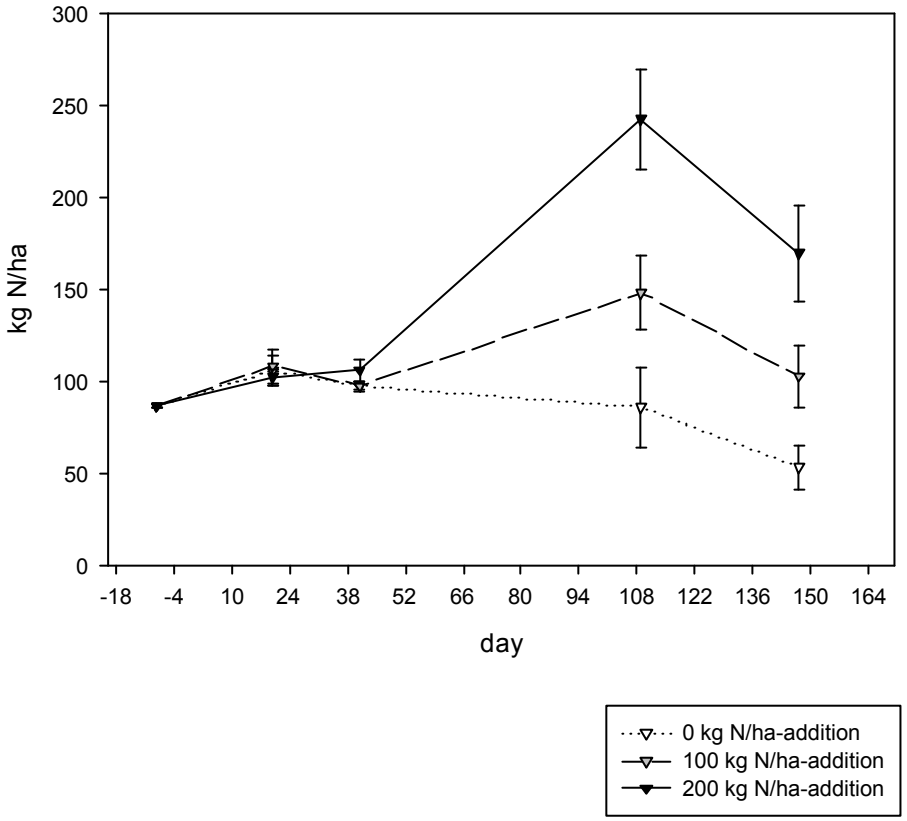
Annexes

Annex 6.1 Measured available nitrogen (NO3-N) in topsoil (0-30 cm).

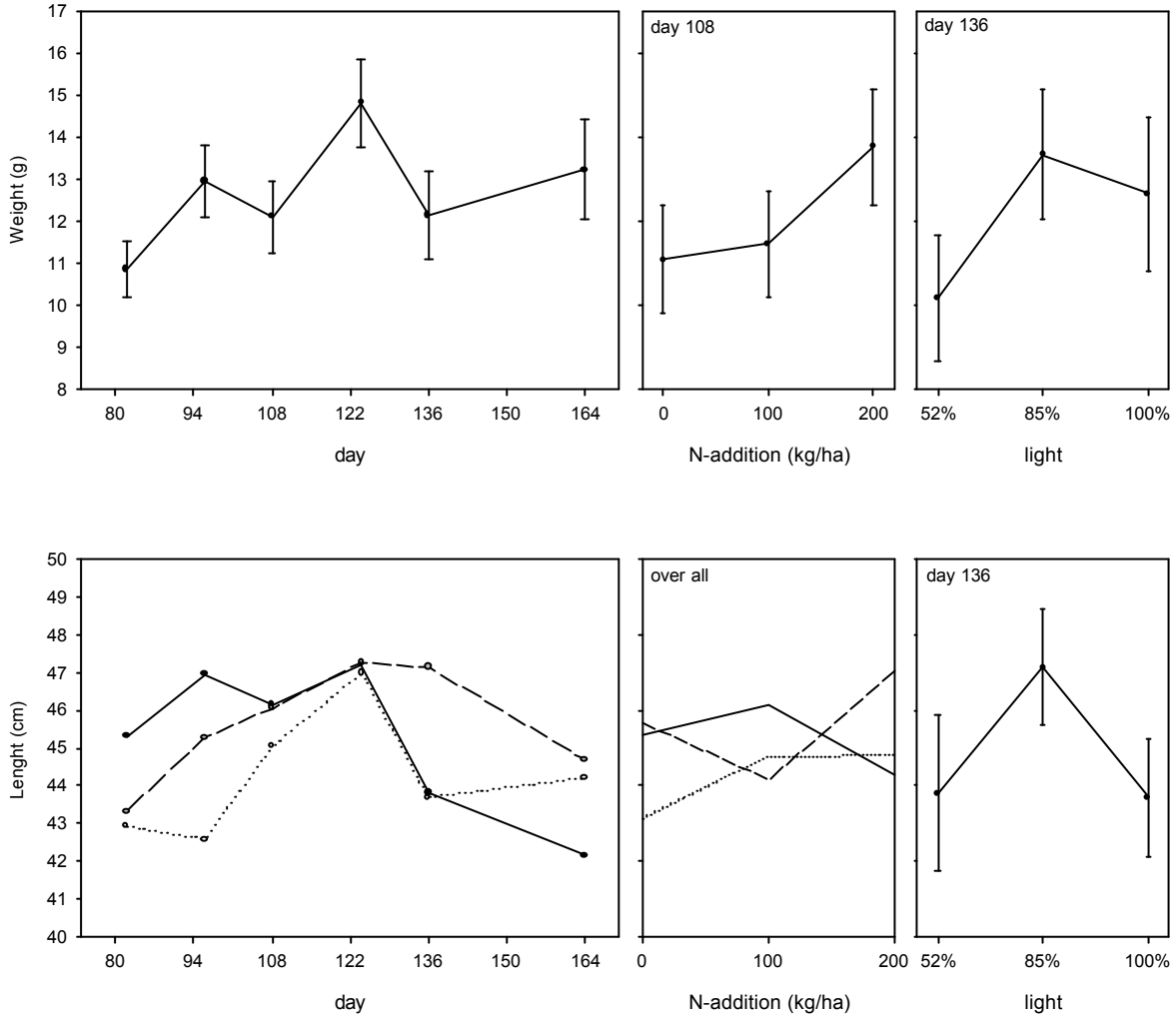


Results ANOVA

Harvest	Parameter	Signif. factor(s) in model	Category(ies)
Overall	Nsoil	N***	all
Day 20	Nsoil	\	
Day 41	Nsoil	N**	1&3
Day 109	Nsoil	N***, H**	N: all; H: 2&4, 3&4
Day 147	Nsoil	N***	all

(*,** and *** are resp. p <0,05,<0,01 and <0,001)

Annex 7.1.1 Fresh weight of leaves and length of leaves
(Key: ── 52% light, – – 85% light and 100% light)

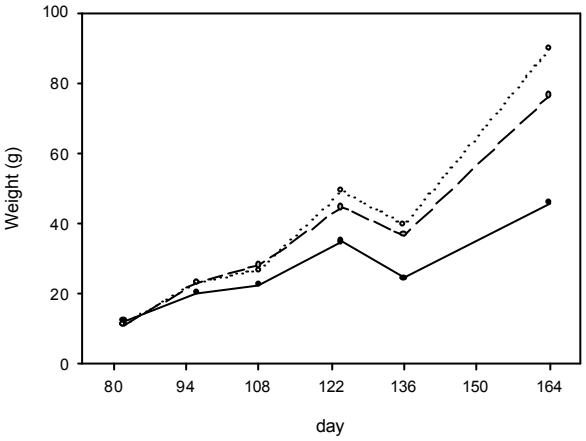


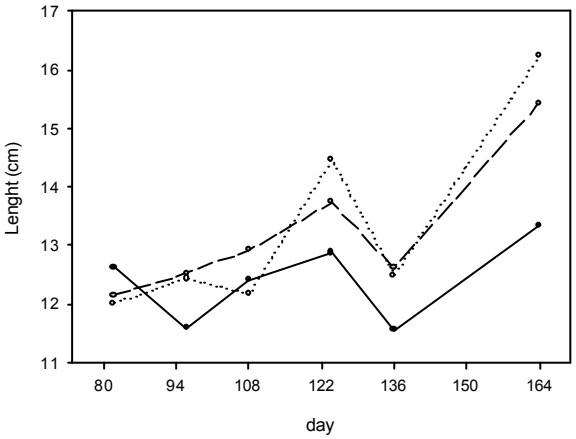
Results ANOVA

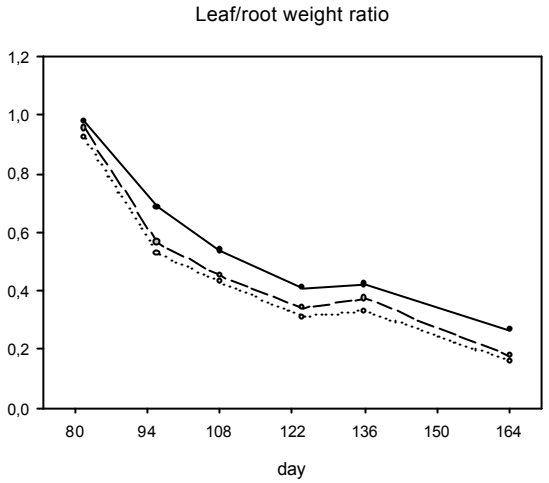
Harvest	Parameter	Signif. factor(s) in model	Category(ies)
Overall	Weight leaf Lenght leaf	day*** Lxday** & LxN***	82&96&124&164, 108&124, 136&164
Day 96	Lenght leaf	LxN**	
Day 108	Weight leaf	N*	1&3
Day 124	Lenght leaf	LxN*	
Day 136	Weight leaf Lenght leaf	L* L*	1&2 1&2, 2&3

(*,** and *** are resp. p <0,05,<0,01 and <0,001)

Annex 7.1.2 Fresh weight of roots, length of roots and ratio fresh weight of leaves to fresh weight of roots
(Key: ── 52% light, – – 85% light and 100% light)





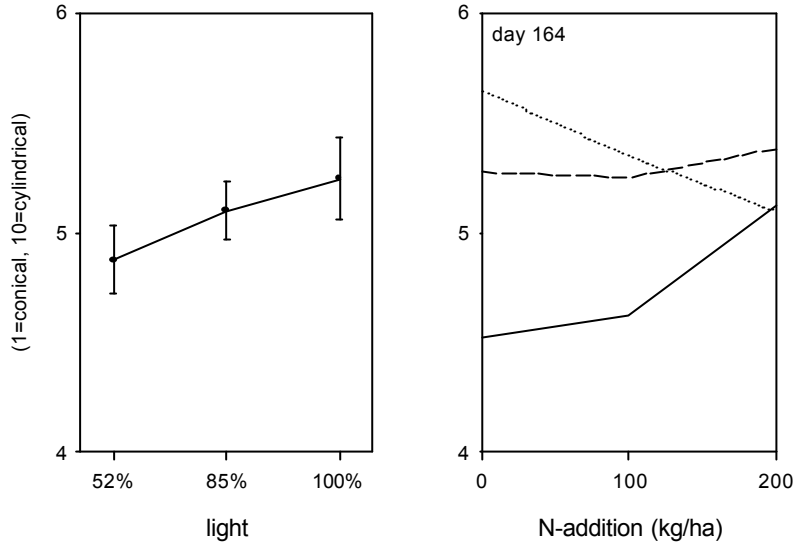


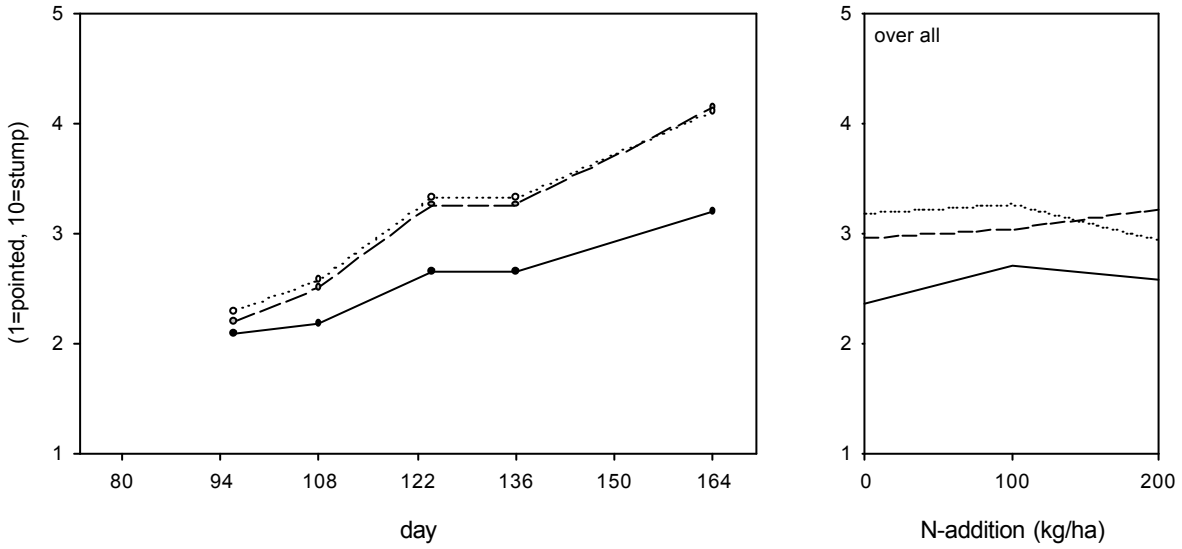
Results ANOVA

Harvest	Parameter	Signif. factor(s) in model	Category(ies)
Overall	Weight root	Lxday***	
	Length root	Lxday**	
	Leaf/root weight ratio	L***, day***	L: all; day: all except 124&136
Day 96	Leaf/root weight ratio	L***	1&2,1&3
Day 108	Weight root	L*	1&2
	Leaf/root weight ratio	L***	1&2,1&3
Day 124	Weight root	L**	1&2,1&3
	Leaf/root weight ratio	L***	1&2,1&3
Day 136	Weight root	L**	1&2,1&3
	Leaf/root weight ratio	L**	1&3
Day 164	Weight root	L***	1&2,1&3
	Lenght root	L***	1&2,1&3
	Leaf/root weight ratio	L***	1&2,1&3

(*,** and *** are resp. p <0,05,<0,01 and <0,001)

Annex 7.1.3 Conical vs. cylindrical roots and pointed vs. stumpy roots
(Key: ── 52% light, – – 85% light and 100% light)



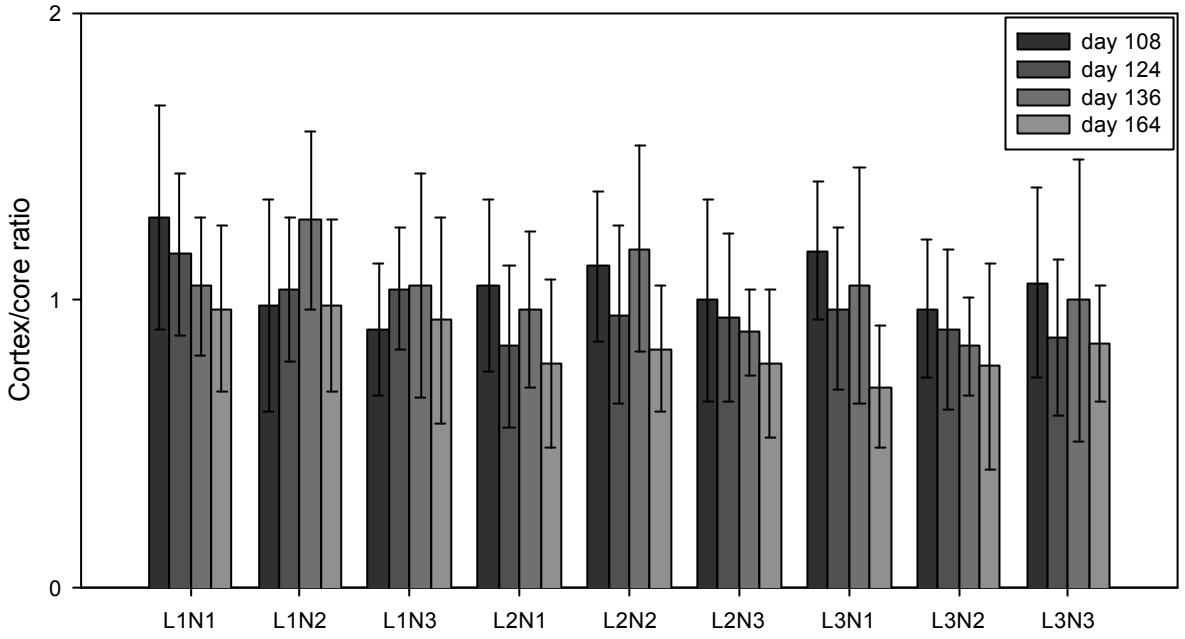


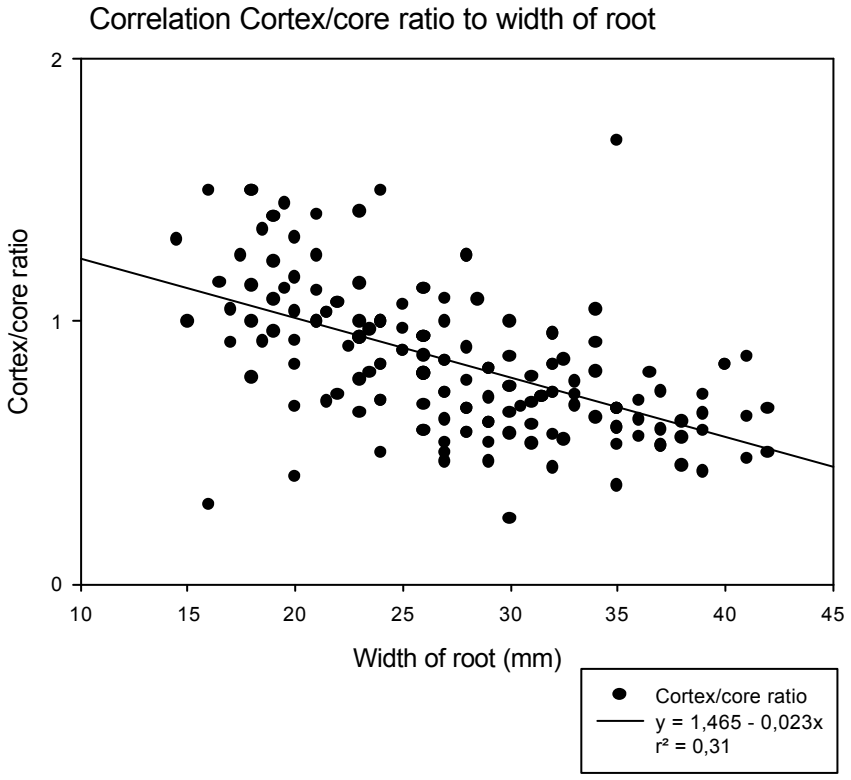
Results ANOVA

Harvest	Parameter	Signif. factor(s) in model	Category(ies)
Overall	Cylindrical Stumpiness	L*** Lxday* & LxN**	1&3
Day 108	Stumpiness	L***	1&2,1&3
Day 124	Stumpiness	L**	1&2,1&3
Day 136	Stumpiness	L**	1&2,1&3
Day 164	Cylindrical Stumpiness	LxN** L***	L1N1, L1N2 vs L3N1 1&2,1&3

(*,** and *** are resp. p <0,05,<0,01 and <0,001)

Annex 7.1.4 Cortex/core ratio (mean and standard deviation) of the 9 treatments on 4 harvest days (Key: L1, L2 and L3 are resp. 52%, 85% and 100% light; N1, N2 and N3 are resp. 0, 100 and 200 kg N ha^{-1}.) and correlation cortex/core ratio to width of root



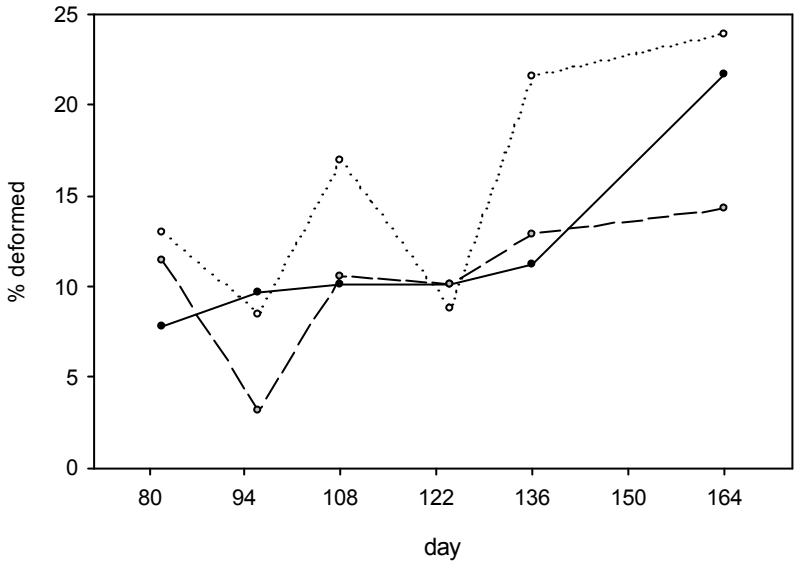


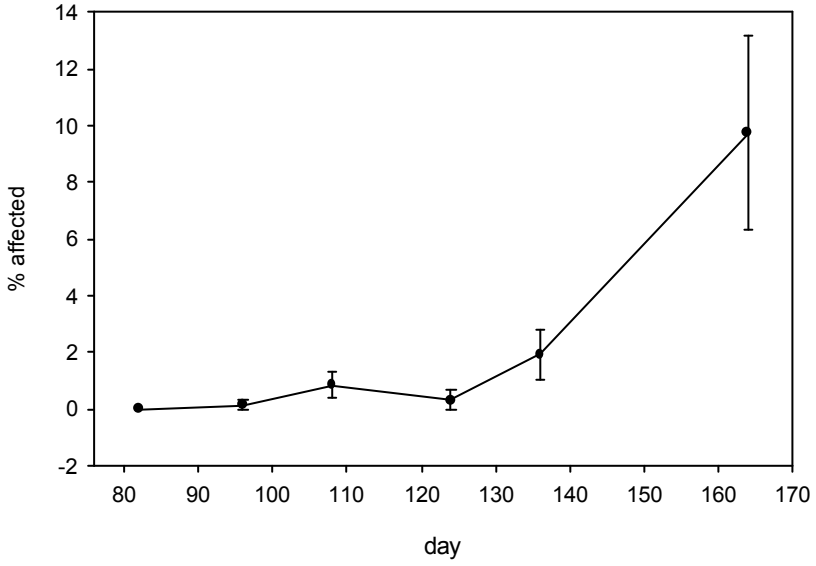
Annex 7.1.5 Correlation table of the six morphological parameters which were statistically processed

		Length of roots	Fresh weight of roots	Length of leaves	Fresh weight of leaves	Pointed vs. stumpy roots	Conical vs. cylindrical roots
Length of roots	Corr. Coefficient	1	,72(**)	,36(**)	,60(**)	,48(**)	,24(*)
	Sig. (2-tailed)		,000	,000	,000	,000	,033
	N		162	162	162	135	81
Fresh weight of roots	Corr. Coefficient	-	1	,25(**)	,54(**)	,77(**)	,29(**)
	Sig. (2-tailed)			,001	,000	,000	,009
	N			162	162	135	81
Length of leaves	Corr. Coefficient	-	-	1	,69(**)	-,07	-,04
	Sig. (2-tailed)				,000	,395	,706
	N				162	135	81
Fresh weight of leaves	Corr. Coefficient	-	-	-	1	,17	,03
	Sig. (2-tailed)					,054	,775
	N					135	81
Pointed vs. stumpy roots	Corr. Coefficient	-	-	-	-	1	,60(**)
	Sig. (2-tailed)						,000
	N						81
Conical vs. cylindrical roots	Corr. Coefficient	-	-	-	-	-	1
	Sig. (2-tailed)						
	N						

** Correlation is significant at the 0.01 level (2-tailed).
* Correlation is significant at the 0.05 level (2-tailed).

Annex 7.2 Percentage of deformed and affected roots
(Key: ── 52% light, – – 85% light and 100% light)



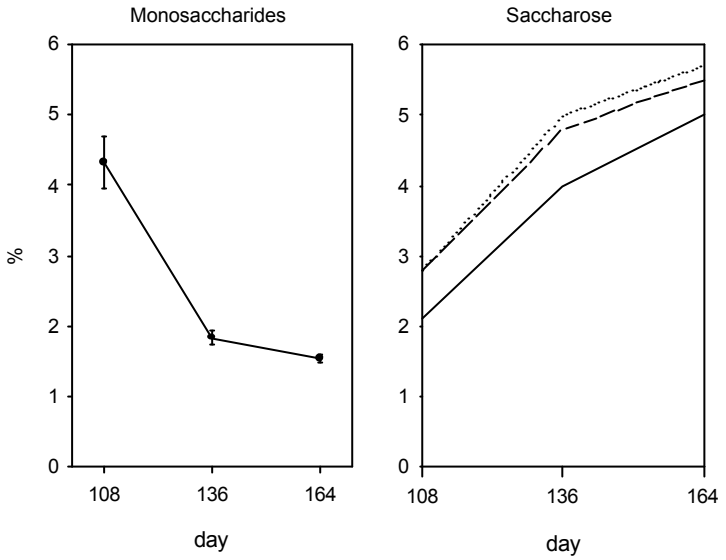


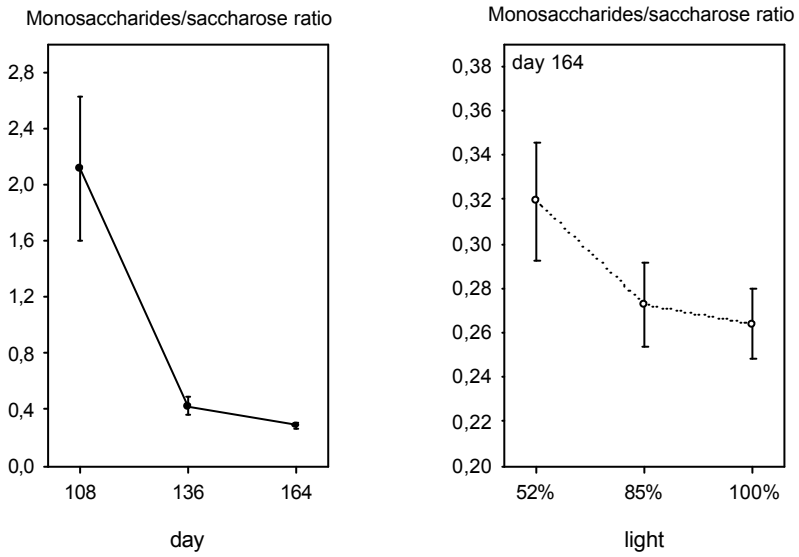
Results ANOVA

Harvest	Parameter	Signif. factor(s) in model	Category(ies)
Overall	Deformed Affected	L*, day*** day***	L: 2&3; day: 82&164, 96&136&164, 124&164 day 164 from rest
Day 124	Deformed	\ tendency LxN	

(*,** and *** are resp. p <0,05,<0,01 and <0,001)

Annex 7.3 Monosaccharides and saccharose content and monosaccharides/saccharose ratio. (Key: ── 52% light, – – 85% light and 100% light)



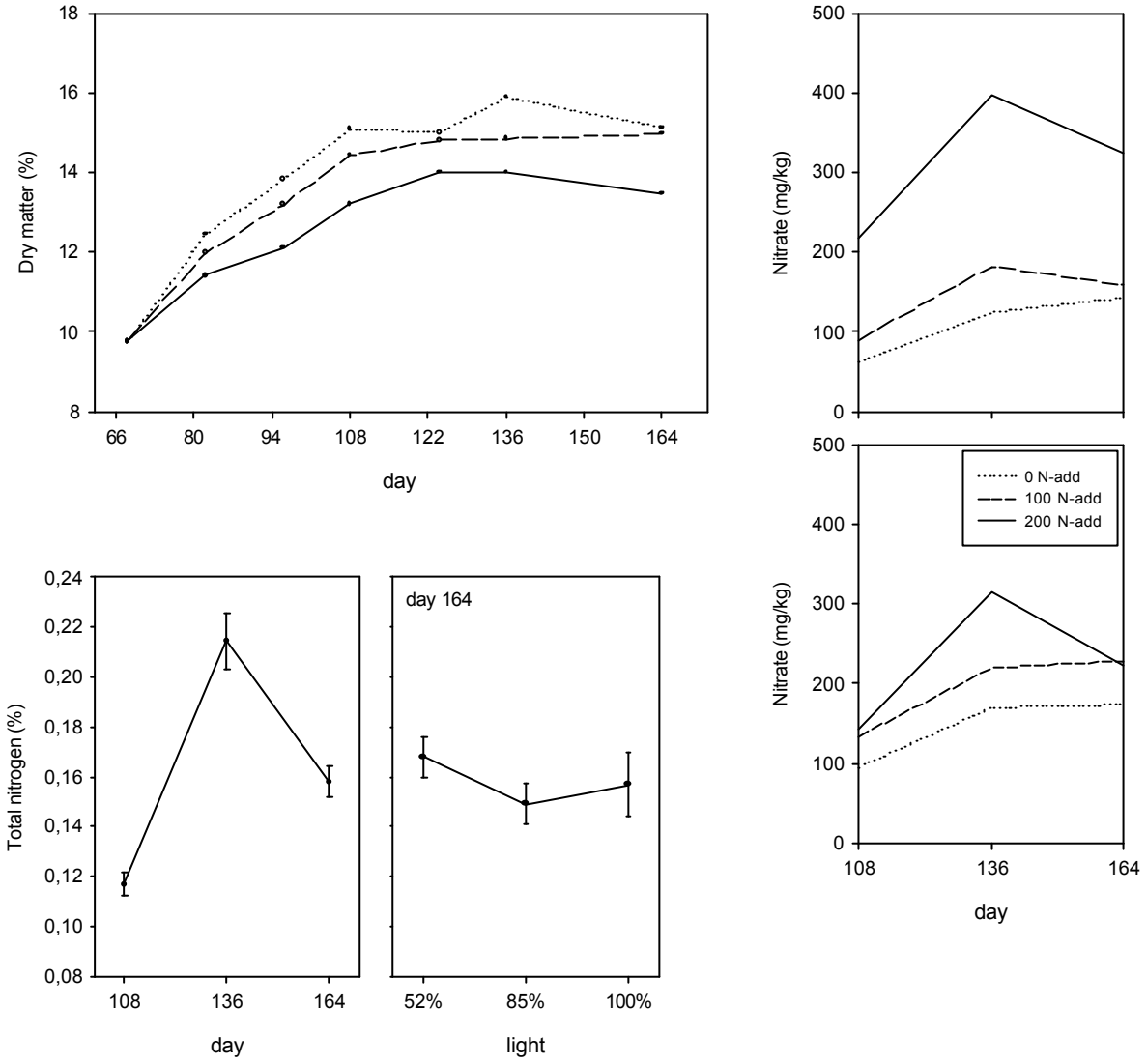


Results ANOVA

Harvest	Parameter	Signif. factor(s) in model	Category(ies)
Overall	Monosaccharides	day***	1&2,1&3
	Saccharose	L***, day***	L: 1&2,1&3; day: all
	Monosaccharides/saccharose	day***	1&2,1&3
Day 136	Saccharose	L**	1&3
	Monosaccharides/saccharose	\ tendency L (p=0,085)	
Day 164	Saccharose	L***	1&2,1&3
	Monosaccharides/saccharose	L**	1&2,1&3

(*,** and *** are resp. p <0,05,<0,01 and <0,001)

Annex 7.4 Dry matter, nitrate and total nitrogen
(Key: ── 52% light, – – 85% light and ····· 100% light)

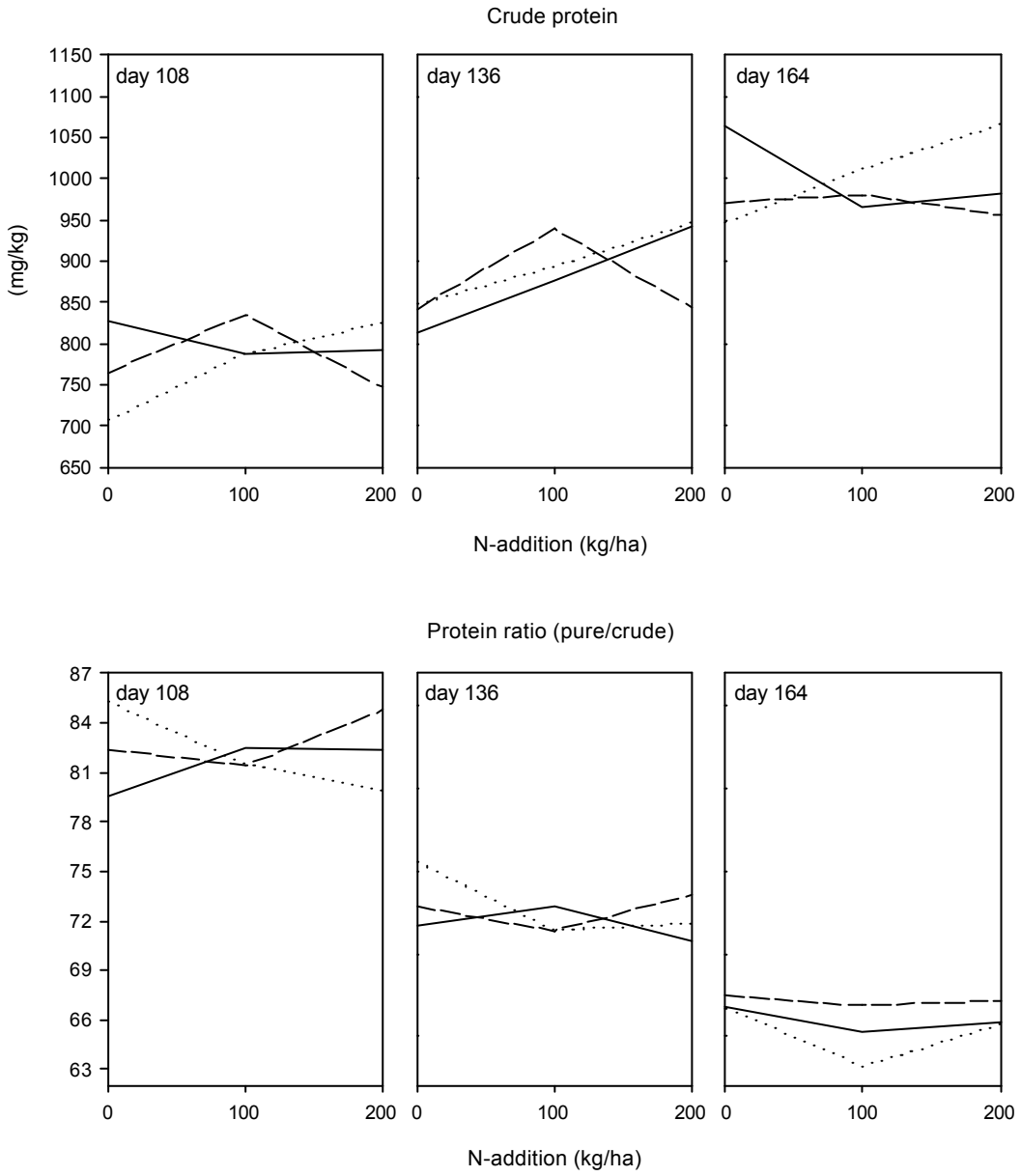


Results ANOVA

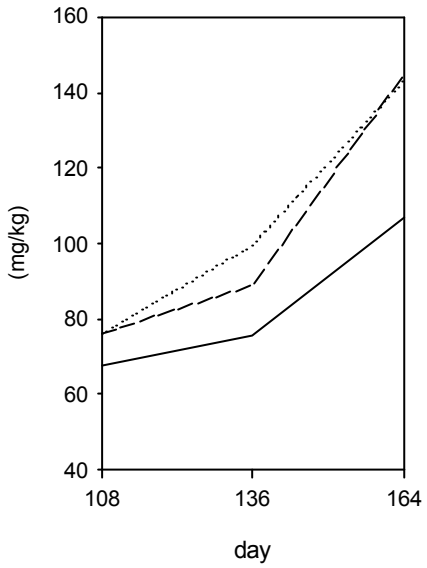
Harvest	Parameter	Signif. factor(s) in model	Category(ies)
Overall	Dry matter	L***, day***	L: all; day: 82 and 96 from rest
	Nitrate	L***, N** and day***	L: 1&2,1&3; N: 1&3; day: 1&2,1&3
	Total nitrogen	day***	all
Day 82	Dry matter	L***, tendency N (p=0.057)	all
Day 96	Dry matter	L***	all
Day 108	Dry matter	L***; tendency LxN (p=0.075)	1&2,1&3
	Nitrate	L***; tendency N (p=0.073)	1&2,1&3
Day 124	Dry matter	L**	1&2,1&3
Day 136	Dry matter	L***	1&3, tendency 2&3
	Nitrate	L***, N*	L: 1&2,1&3; N: 1&3
Day 164	Dry matter	L***	1&2,1&3
	Nitrate	L***	1&2,1&3
	Total nitrogen	L*	1&2

(*,** and *** are resp. p <0,05,<0,01 and <0,001)

Annex 7.5 Crude protein and protein ratio
(Key: ── 52% light, – – 85% light and 100% light)



Annex 7.6 Carotenes content
(Key: ── 52% light, – – 85% light and 100% light)

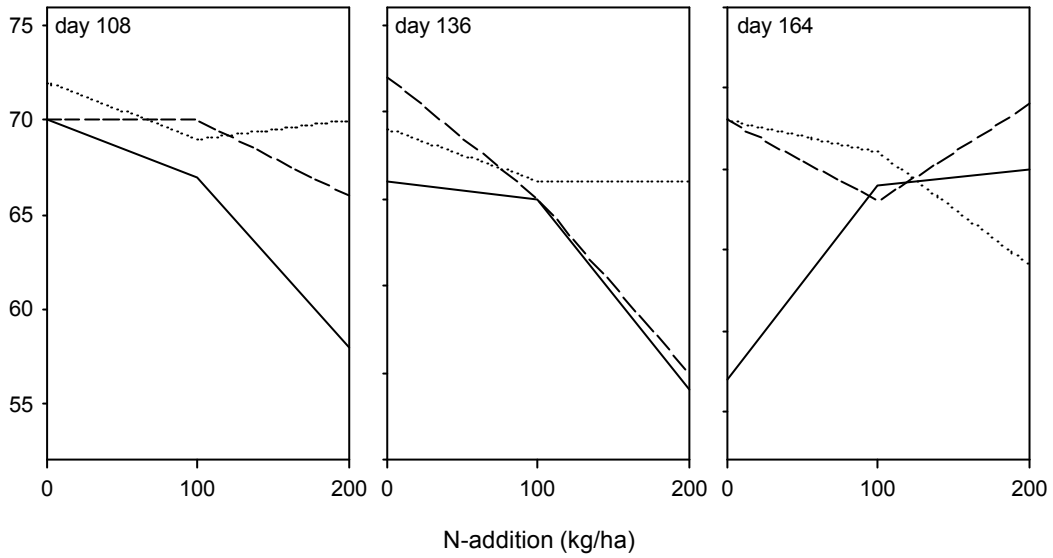


Results ANOVA

Harvest	Parameter	Signif. factor(s) in model	Category(ies)
Overall	Carotenes	Lxday***	
Day 108	Carotenes	L*	tendency 1&2,1&3
Day 136	Carotenes	L***	all
Day 164	Carotenes	L***	1&2,1&3

(*,** and *** are resp. p <0,05,<0,01 and <0,001)

Annex 7.7 Sensory properties. Figures show total appreciation
(Key: ── 52% light, – – 85% light and 100% light)

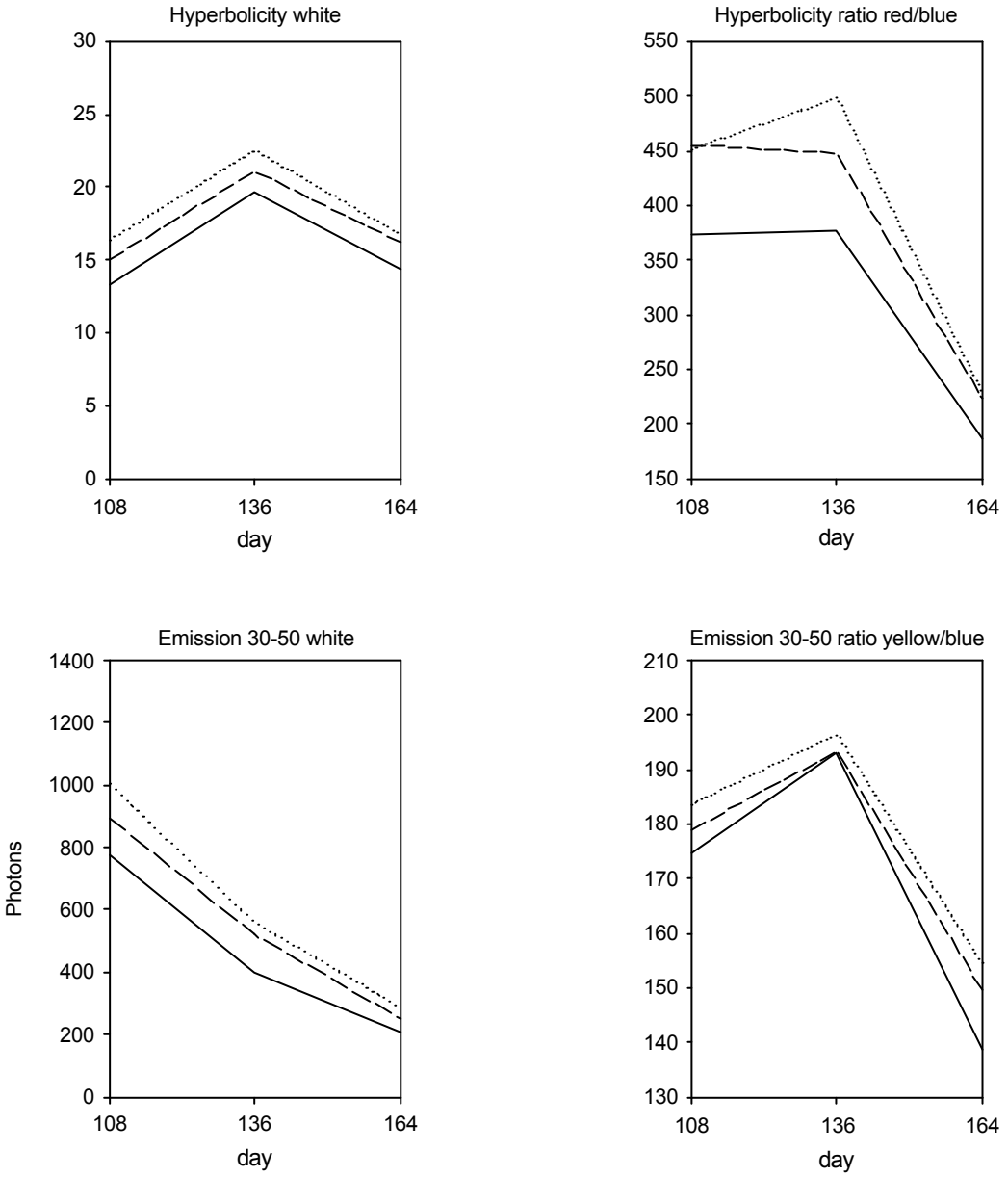


Results ANOVA

Harvest	Parameter	Signif. factor(s) in model
Day 108	sweet	LxN**
	acid/astringent	LxN***
	bitter	LxN**
	carrot taste	L**, N*
	soapy	LxN*
	juicy	L*, N**
	crunchy	LxN*
	tough	L**, N**
	total appreciation	**L**, N****
Day 136	sweet	LxN**
	acid/astringent	L*, N***
	bitter	LxN*
	carrot taste	LxN*
	soapy	LxN*
	juicy	LxN**
	crunchy	LxN***
	tough	LxN***
	total appreciation	**L*, N***
Day 164	sweet	LxN**
	acid/astringent	LxN***
	bitter	\
	carrot taste	LxN***
	soapy	LxN**
	juicy	L*
	crunchy	LxN**
	tough	N**
	total appreciation	**LxN*****

(*,** and *** are resp. p <0,05,<0,01 and <0,001)

Annex 7.8.1 Hyperbolicity and emission 30-50 of luminescence by Kwalis
(Key: ── 52% light, – – 85% light and 100% light)

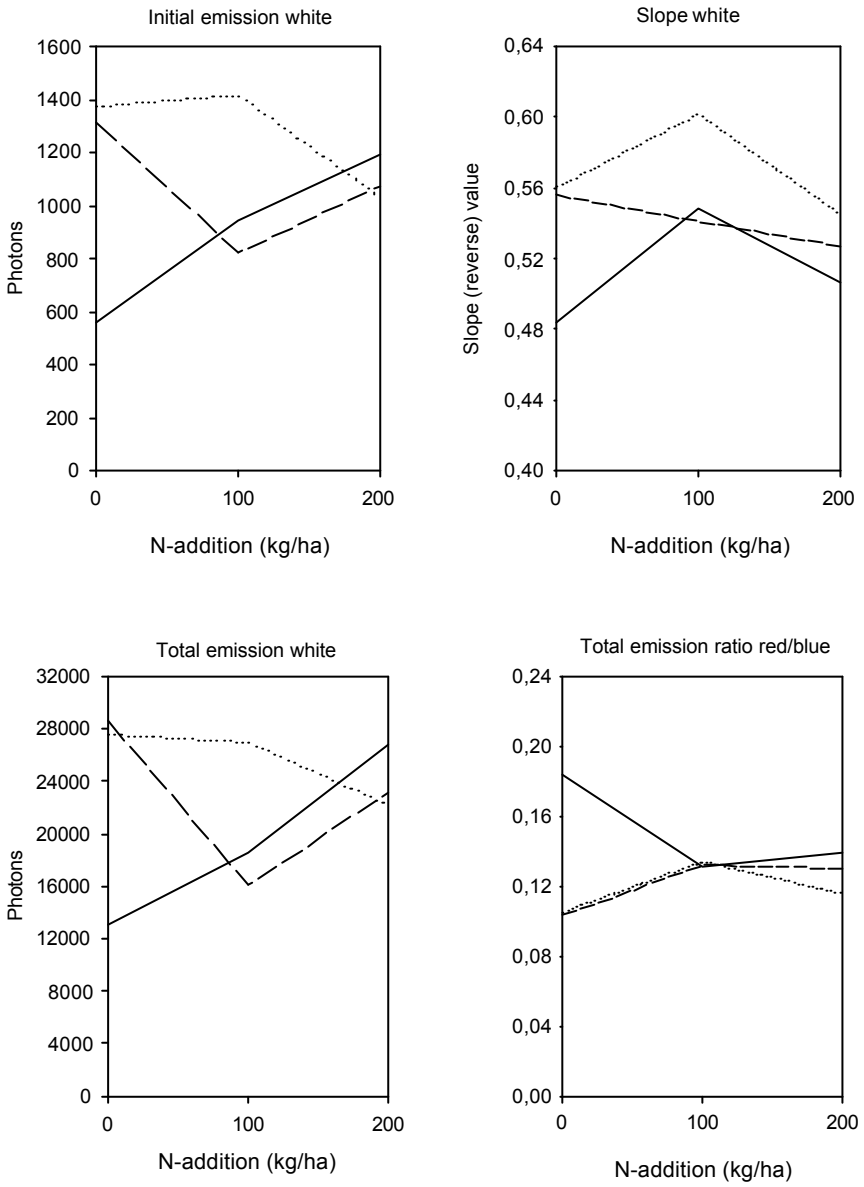


Results ANOVA

Harvest	Parameter	Signif. factor(s) in model	Category(ies)
Overall	Hyperbolicity white	L*, day***	L: 1&3; day: 1&2,2&3
	Hyperbolicity ratio red/blue	L*, day***	L: 1&3; day: 1&3,2&3
	Emission 30-50 white	L***, day***	L: 1&2,1&3; day: all
	Emission 30-50 ratio yellow/blue	L**, day***	L: 1&3; day: all
Day 108	Emission 30-50 white	L**	1&3
	Emission 30-50 ratio yellow/blue	\ strong tendency L (p=0.074)	
Day 136	Hyperbolicity ratio red/blue	L**	1&3
	Emission 30-50 white	\ tendency L (p=0.1)	
Day 164	Hyperbolicity white	\ strong tendency N (p=0.076)	
	Emission 30-50 white	\ strong tendency L (0.068) & N (p=0.082)	
	Emission 30-50 ratio yellow/blue	\ strong tendency L (p=0.068)	

(*,** and *** are resp. p <0,05,<0,01 and <0,001)

Annex 7.8.2 Initial emission, slope value and total emission of luminescence by Meluna on day 164. (Key: ── 52% light, – – 85% light and 100% light)

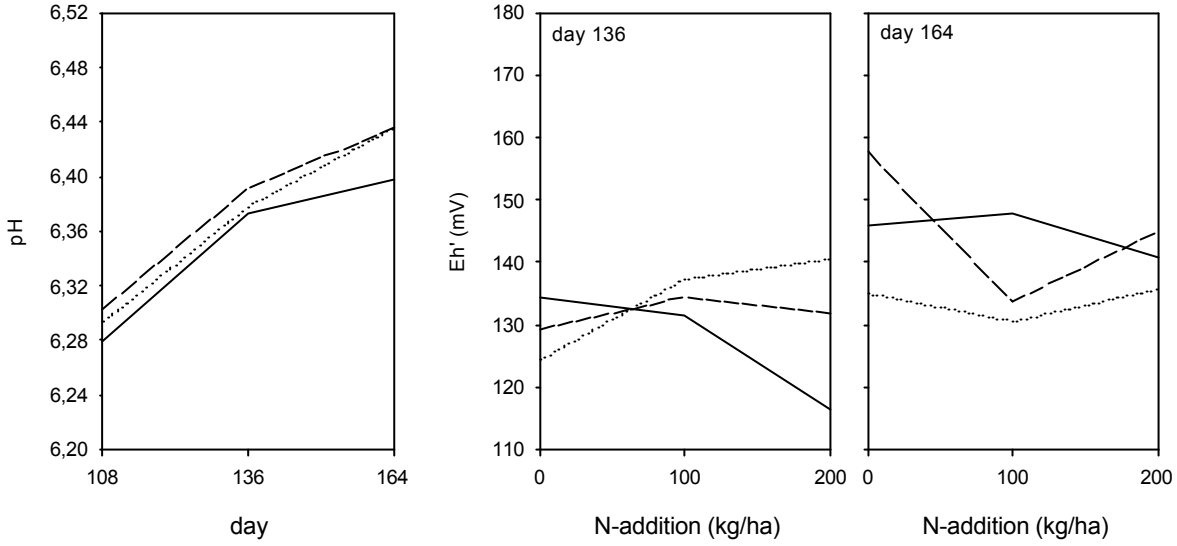


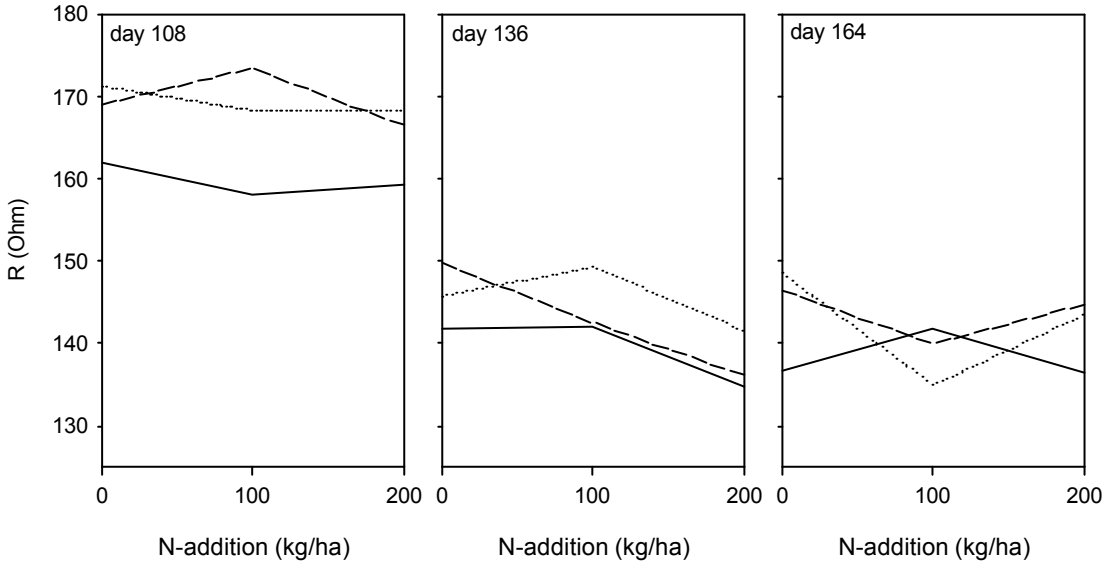
Results ANOVA

Harvest	Parameter	Signif. factor(s) in model	Category(ies)
Day 164	Initial emission white	LxN***	L: 1&3 (tend.1&2 and 2&3); N: 2&3,1&2
	Slope white	L***, N**	
	Total emission white	LxN***	
	Total emission ratio red/blue	LxN**	

(*,** and *** are resp. p <0,05,<0,01 and <0,001)

Annex 7.10.1 Electro-chemical parameters pH, Eh´ and R by the University of Kassel
(Key: ── 52% light, – – 85% light and 100% light)



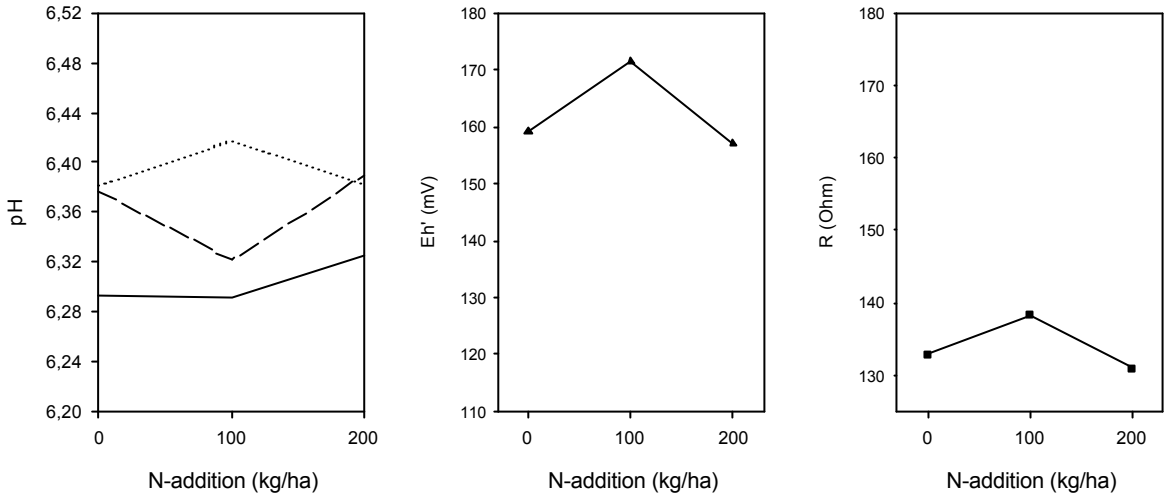


Results ANOVA

Harvest	Parameter	Signif. factor(s) in model	Category(ies)
Overall	pH	L***, day***	L: 1&2,1&3; day: all
	Eh'	LxNxday**	
	R	L***, N**, day***	L: 1&2,1&3; N: 1&3; day: 1&2,1&3
Day 108	Eh'	no measurements	
	R	L**	1&2,1&3
Day 136	Eh'	LxN***	L1N3 vs L3N3 & L3N2
	R	L**, N***	L: 1&3; N: 1&3,2&3
Day 164	pH	L**	1&2, 1&3
	Eh'	LxN*	L3N2 vs L2N1
	R	tendency LxN (p=0.071)	

(*,** and *** are resp. p <0,05,<0,01 and <0,001)

Annex 7.10.2 Electro-chemical parameters pH, Eh´ and R on harvest day 164 by the Electro-chemisches Qualität Labor.
(Key: ── 52% light, – – 85% light and 100% light)

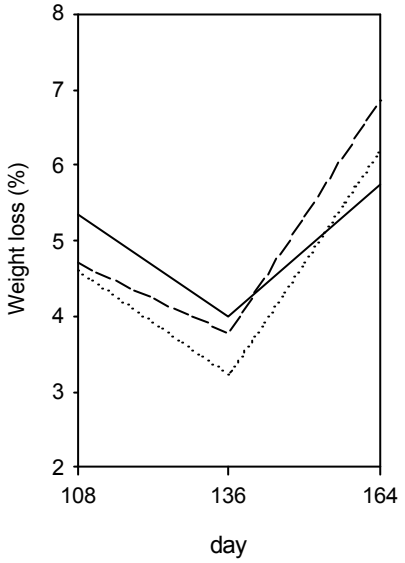


Results ANOVA

Harvest	Parameter	Signif. factor(s) in model	Category(ies)
Day 164	pH	LxN**	L1&3 for N1; L1&2&3 for N2
	Eh'	N*	tendency 2&3
	R	N**, tendency L (p=0.067)	2&3

(*,** and *** are resp. p <0,05,<0,01 and <0,001)

Annex 7.11 Weight loss per week of roots from different days of harvest during storage at 16-19°C and ca. 70% R.H.
(Key: ── 52% light, – – 85% light and 100% light)



Results ANOVA

Harvest	Parameter	Signif. factor(s) in model	Category(ies)
Overall	Weight loss (%)	Lxday*	
Day 108	Weight loss (%)	\ tendency L (and LxN and N)	
Day 136	Weight loss (%)	\ tendency L (p=0,076)	

(*,** and *** are resp. p <0,05,<0,01 and <0,001)