05 November 2015, Breda

3de docentendag CBBE
biobased products and refinery

CoE BBE

dr. ir. G.C.H. (Dorien) Derksen

Lectoraat Biobased Products

HZ University of Applied Sciences

avans hogeschool

Isolation of secondary metabolites

Natural Colours as a product

CoE BBE

Centre of Expertise BioBased Economy

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Company Rubia Natural Colours

Bio-Based Dyes and Pigments

cultivation, manufacturing, application, innovation and market

RUBIA 100% NATURAL COLOURS

Crop growing & processing

- new colours, new sources, new crops
- plant breeding and harvesting
- phytochemistry, knowledge on plant compounds and metabolism
- agrification, economic value of new sustainable crops
- cost efficient processing, sustainable, low in energy

RUBIA 100% NATURAL COLOURS

Rubia’s manufacturing capability

- breeding plants, up to 30 ha., licensed cultivars
- harvesting plants, 20 breeders
- plant pretreatment, wash, dry, grind
- extraction
- solid – liquid separation
- drying
- quality control, own laboratory
- packaging, up to 150 tpa finished product

a colourful future

Rubia NC product portfolio

- Rubia® Red: red & brown
- Rubia® Yellow: yellow & green
- Rubia® Blue and Rubia ® Black: blue, black and grey

different plant sources: madder, woad and weld

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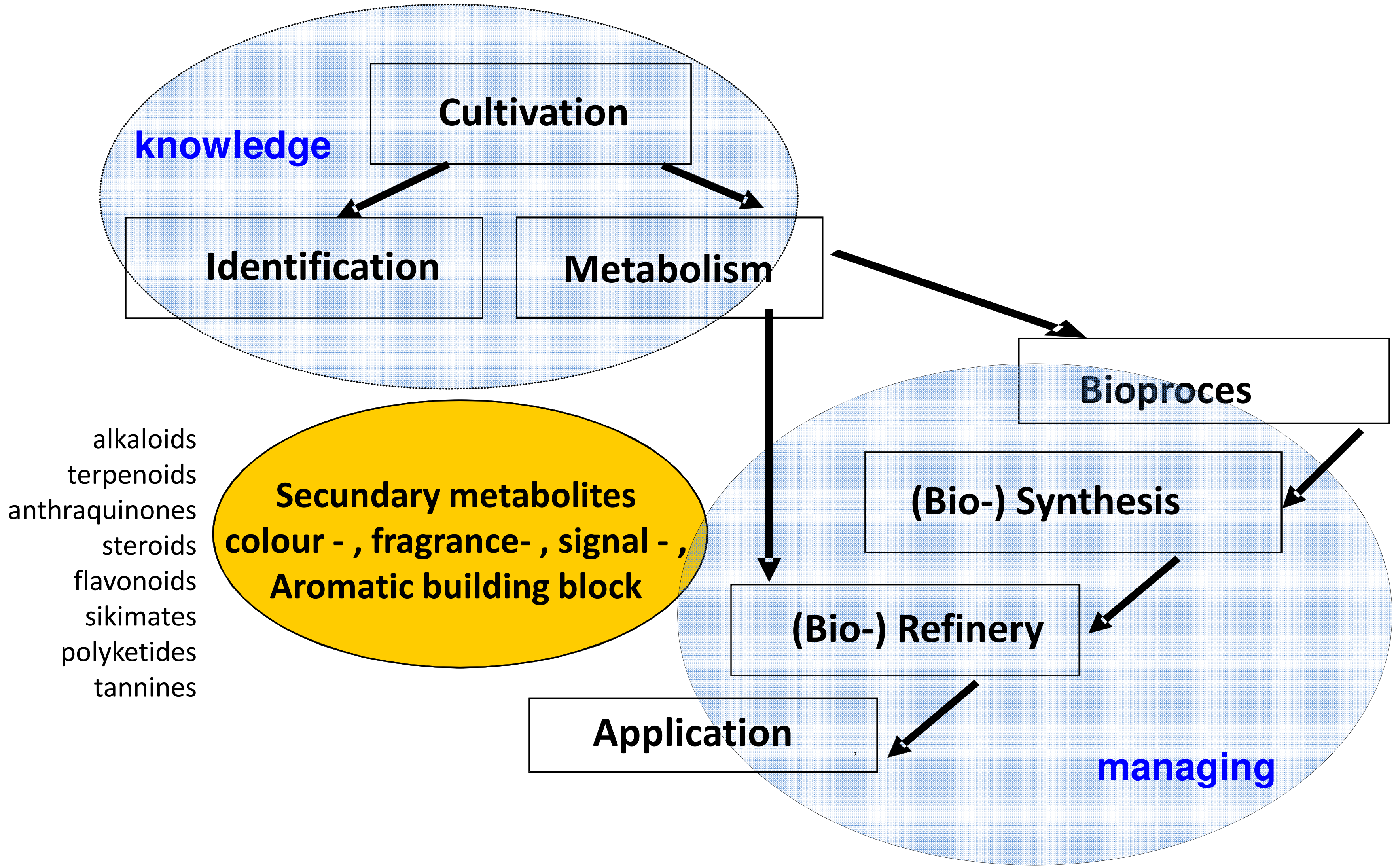
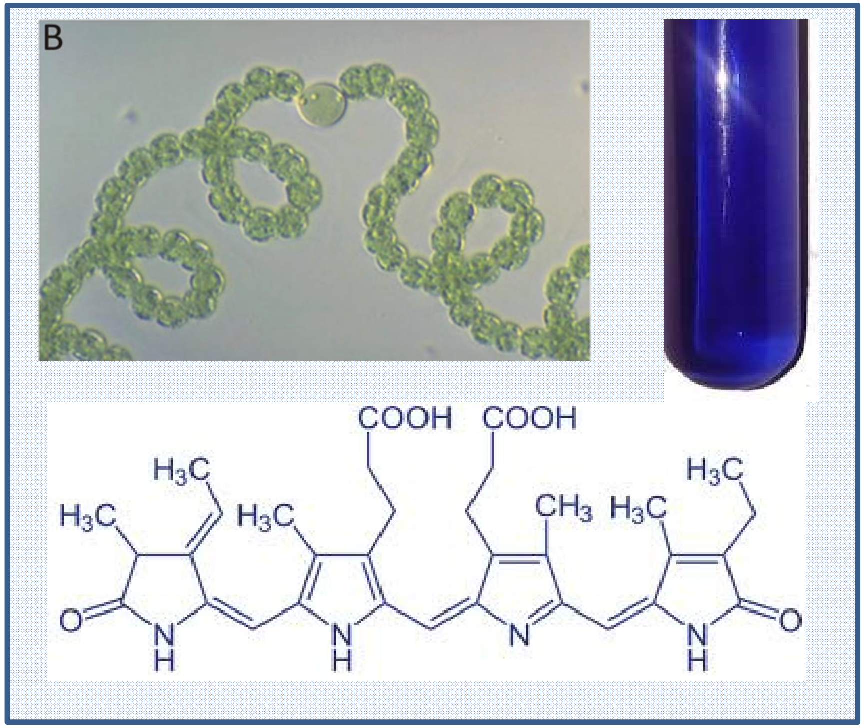
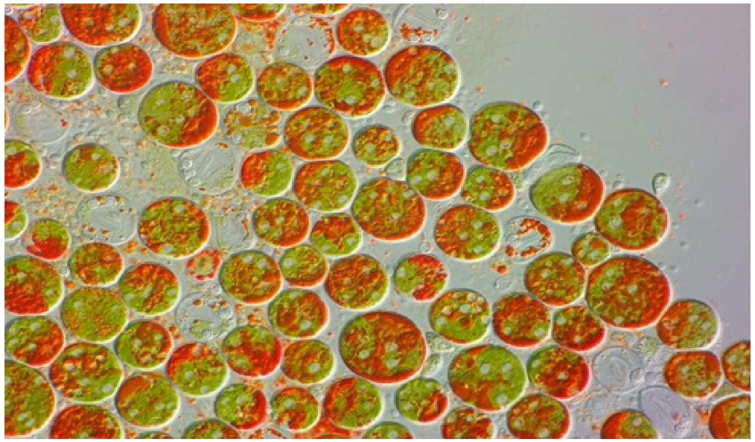
Biobased products

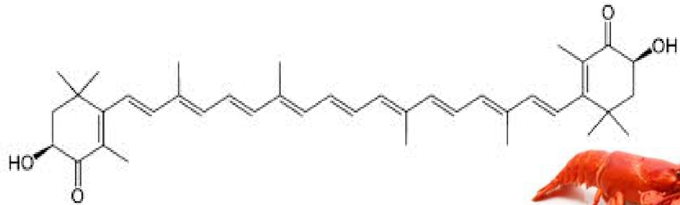


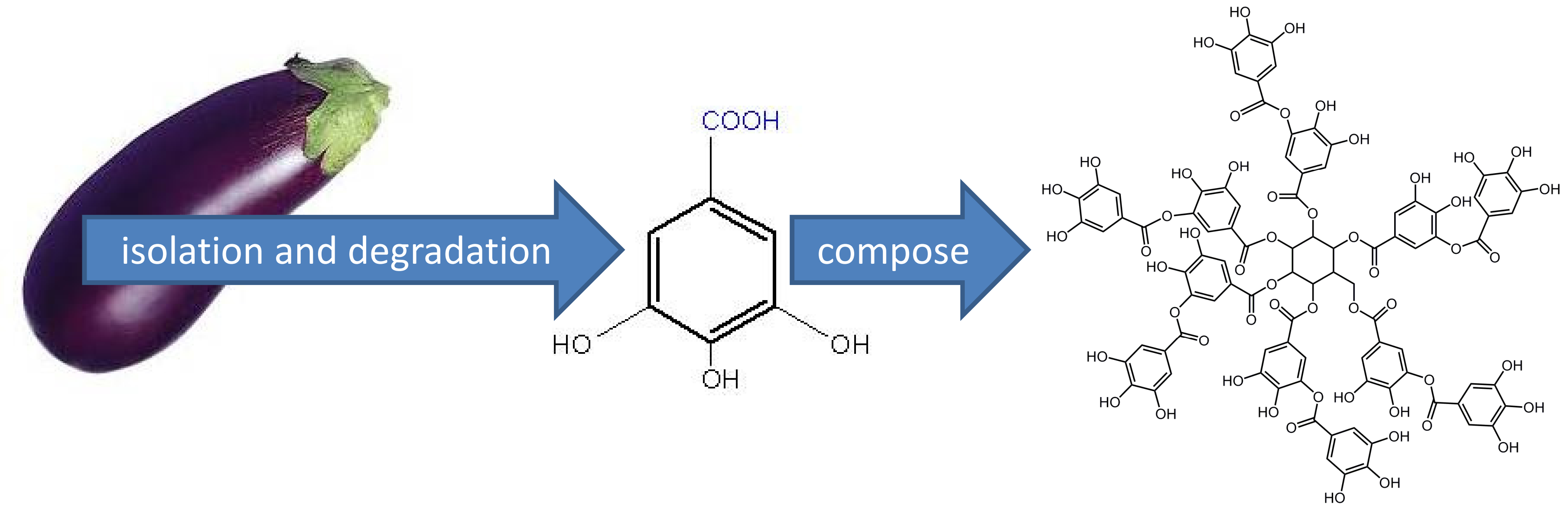
Secondary metabolites from micro organism

i.p. yeasts and algae





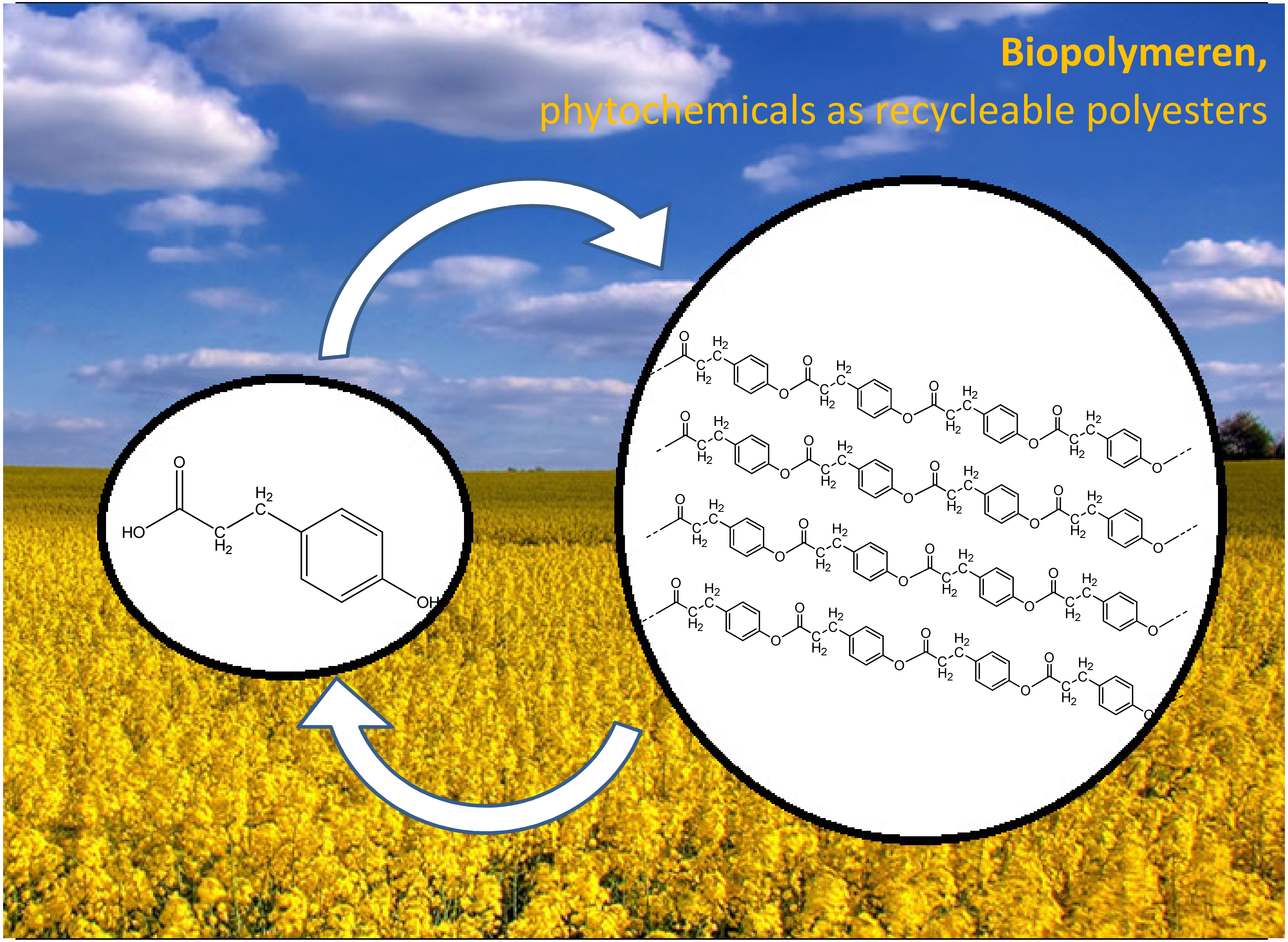




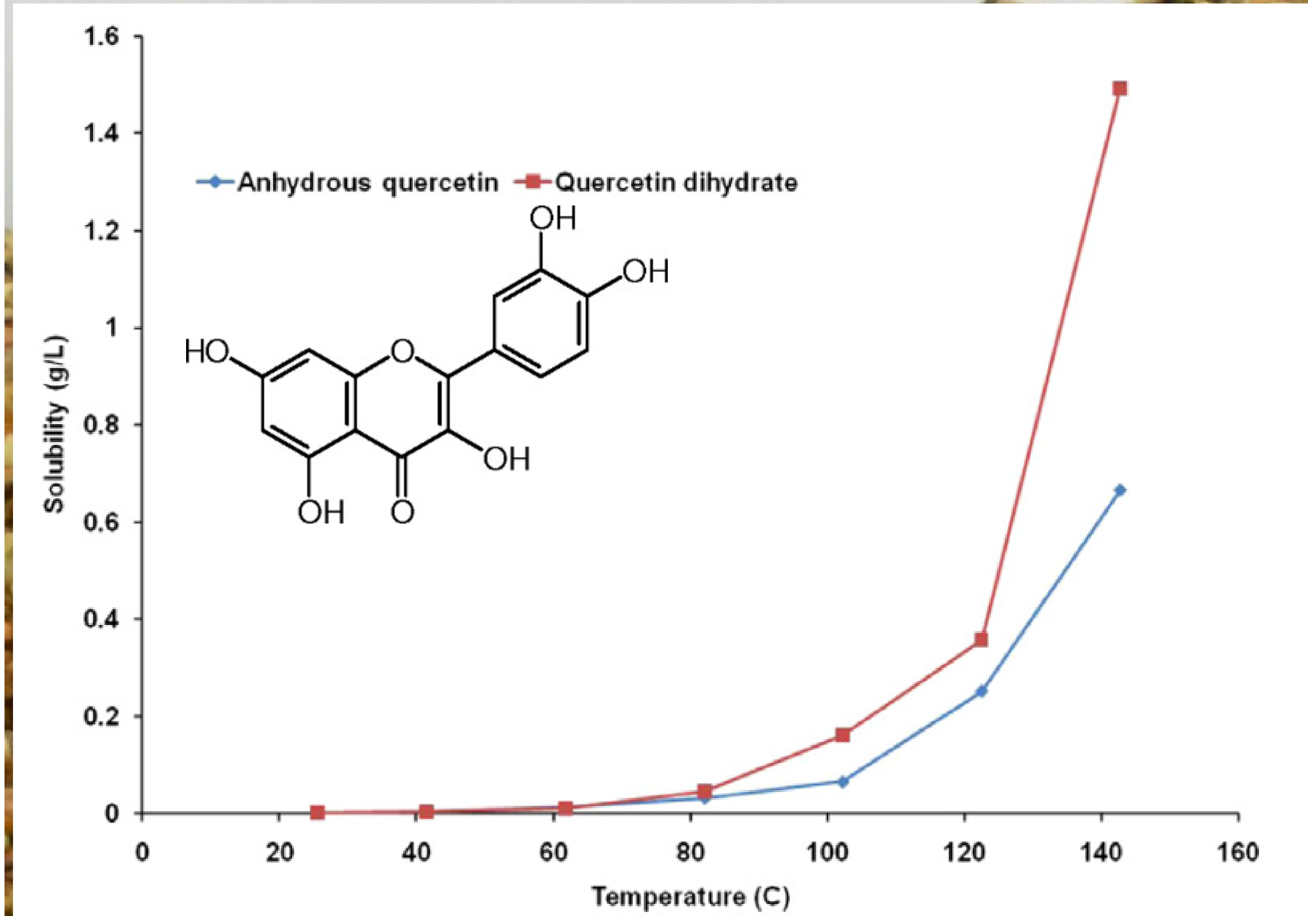
Natural dyes and pigments

- Tannins, polyphenols
  - Building block
  - Encapsulation
  - Light stability

Other colours


Biopolymeren,
phytochemicals as recycleable polyesters

Quercetine: meerwaarde uit reststroom uien



- Sub-kritisch water (160 °C)
- Geen organische oplosmiddel

- Uienpellen 17000 ton/jaar in Zuidwest Nederland
- Biogas productie
- Digestaat als meststof

Antioxidant
Kleurstof papier, textiel en levensmiddelen

Isolation of secondary metabolites

Natural Colours as a product

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Applied Research on Natural Colours in practical

- HBO, ROC (KLAC), GCC, MKB (company)
- Chemistry, Chemical Technology, Biobased TeCH, Other

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Possibilities

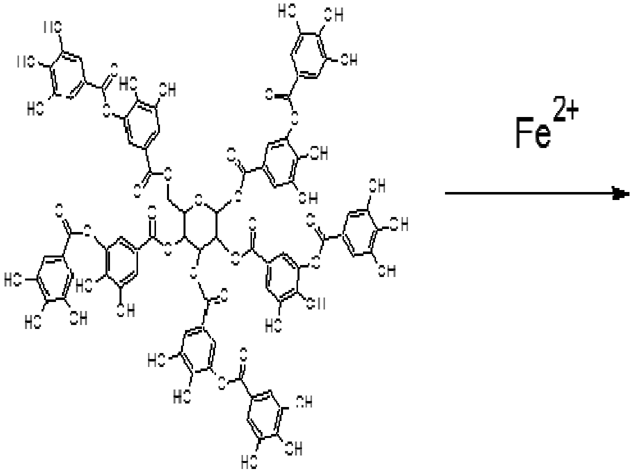
Present Setting

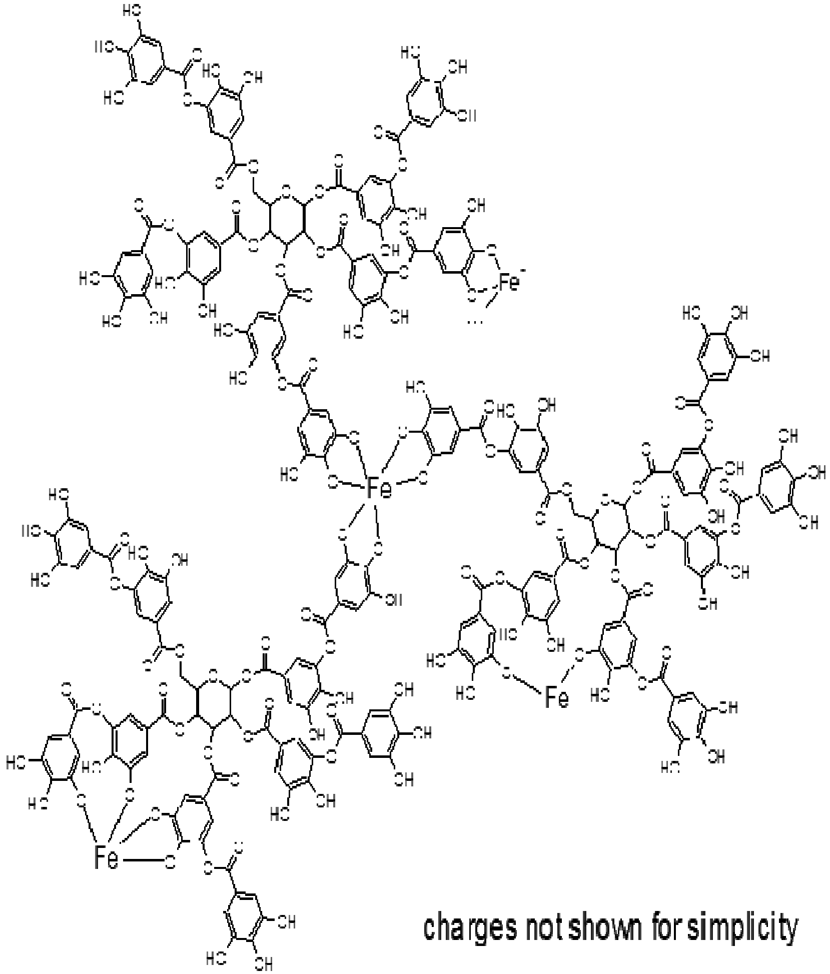
1. Welke onderzoeksprojecten →strategic innovation.
2. Emerged → Funnelling
   – Example Rubia; Dyeing of textile,
     Improvement in mass balance total process,

"FIND the NICE to know instead of NEED to know"

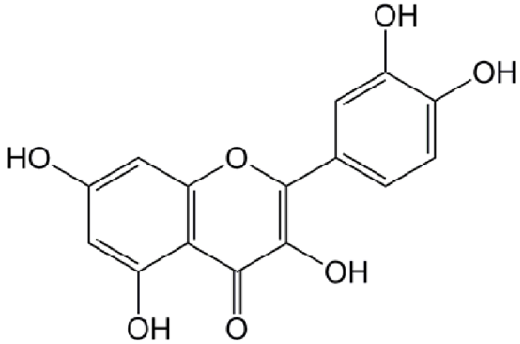
– Example Rubia; Deeper black
  Application of knowledge on colour stability
– Example RIGO, Van Houtum; Pigment application paint, paper

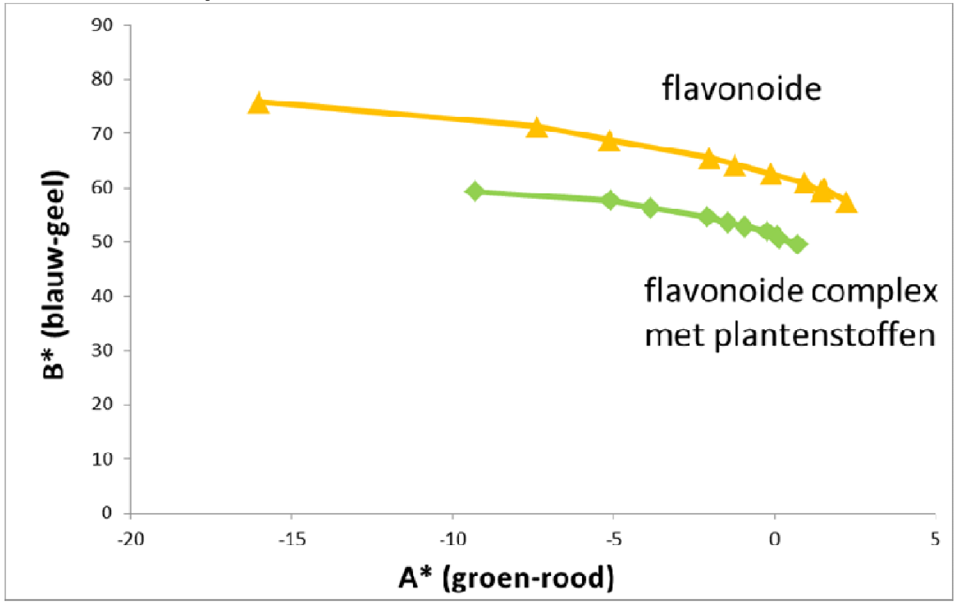
2. Complexation of tannic acids

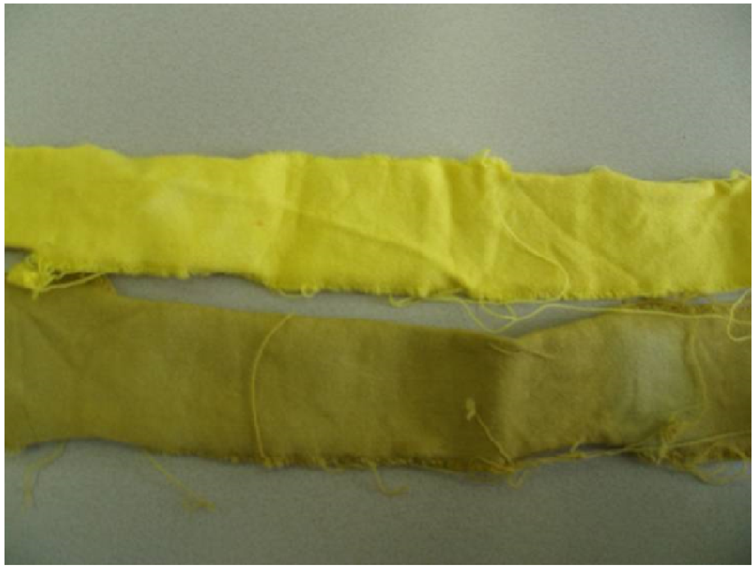




4. Light stability of natural dyes







6. Connect and support

Living colours

SIA-RAAK-MKB

Give interior a natural and bio based ambiance

Lectoraat Innovatie Bouwproces en Techniek

A rendezvous between

chemistry and art

Lectoraat Biobased Products

AKV|St.Joost

Possibilities

Present Setting

1. Laboratories AVANS →
   - organic-, analytical and biobased chemistry
   - Chemical technology
2. Green Chemistry Campus, GCC →
   - Light stability research
   - Molecular work

Future Adding

3. ROC → KLAC (kleurstoffen applicatie centrum)
   - MBO, HBO students work together with masters and with
   - Small medium enterprises
   - Application work

Isolation of secondary metabolites

Natural Colours as a product

Case study

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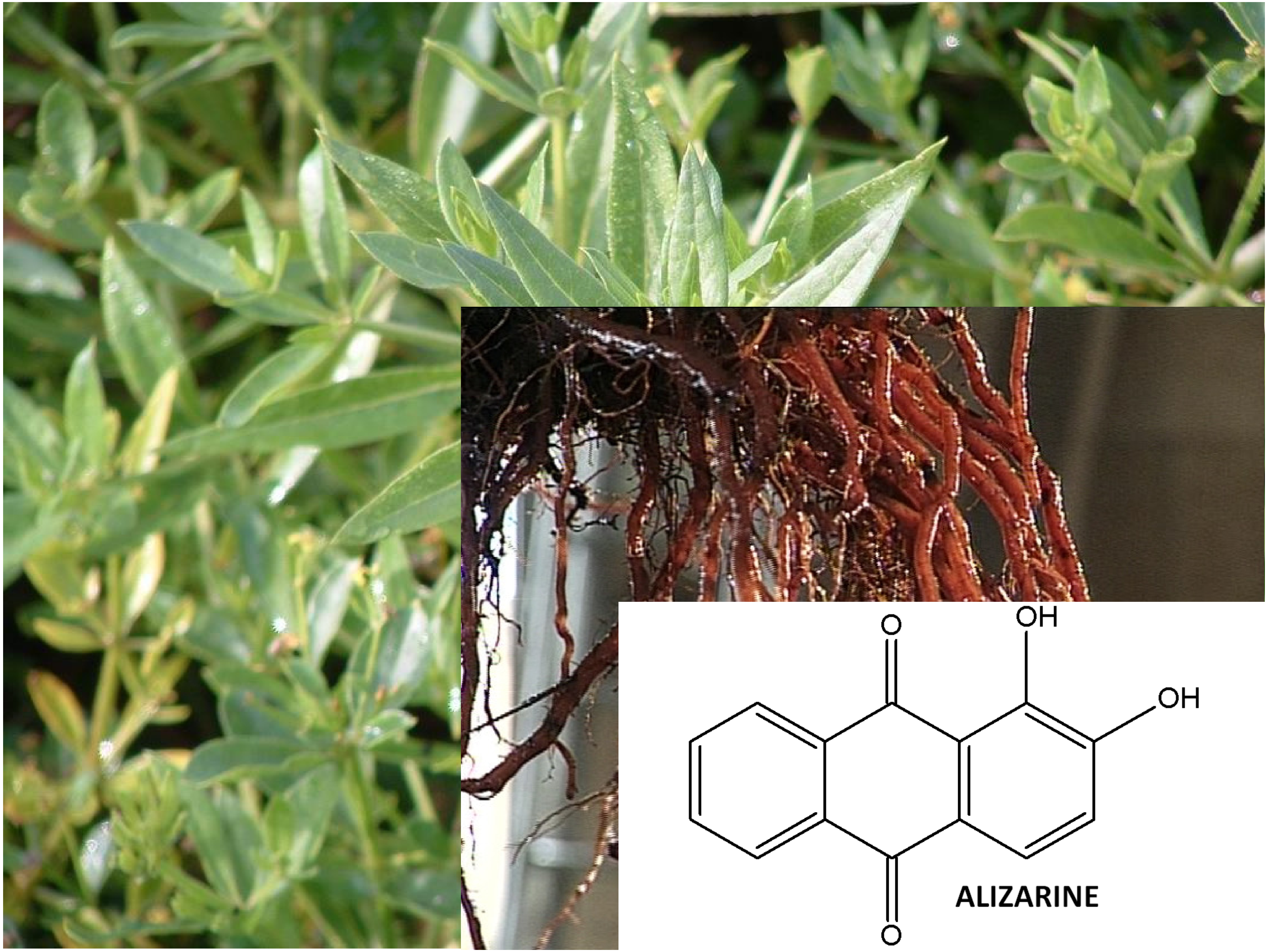
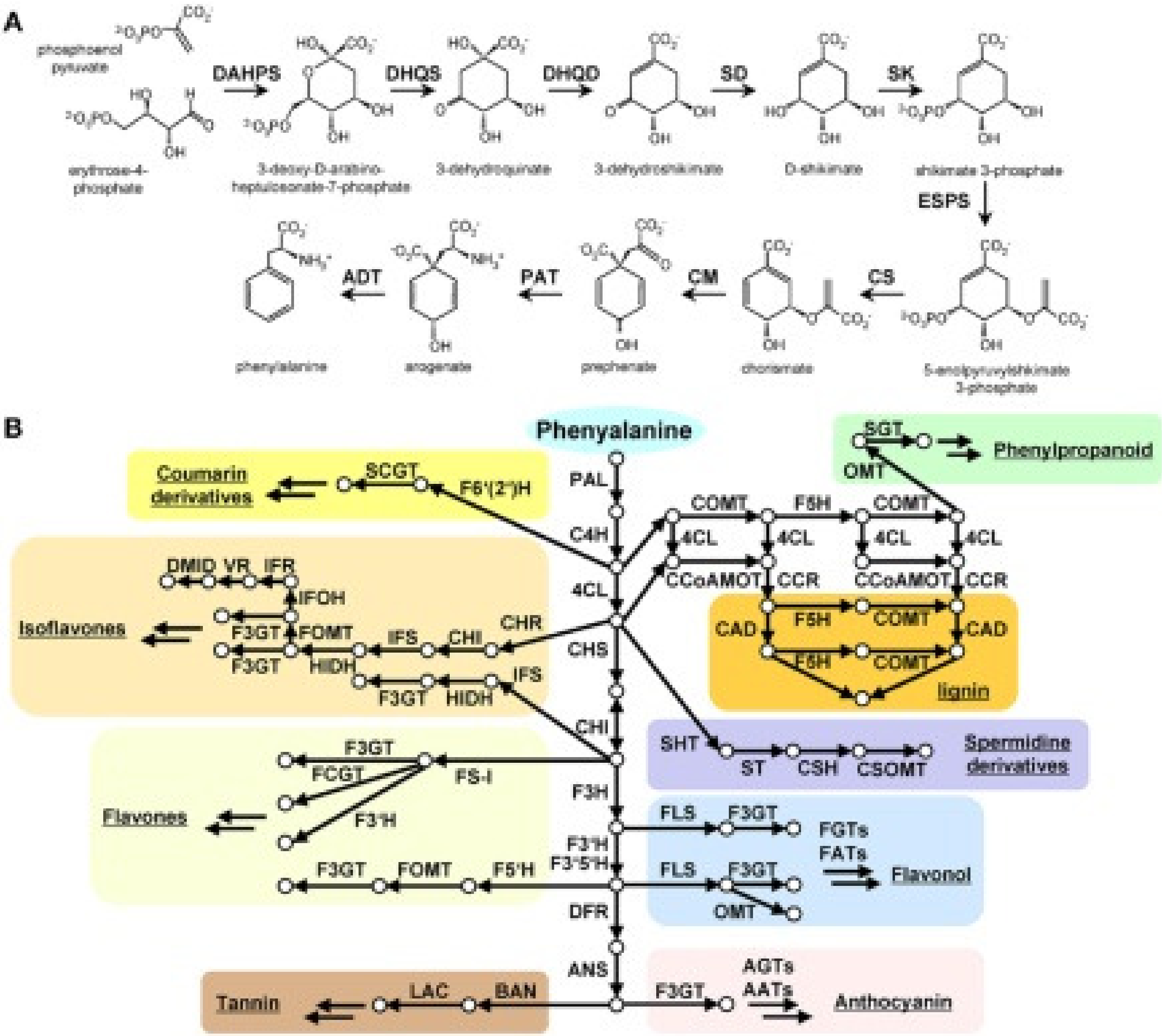

O
OH
OH
O
ALIZARINE

RUBIA 100% NATURAL COLOURS

Applicatie

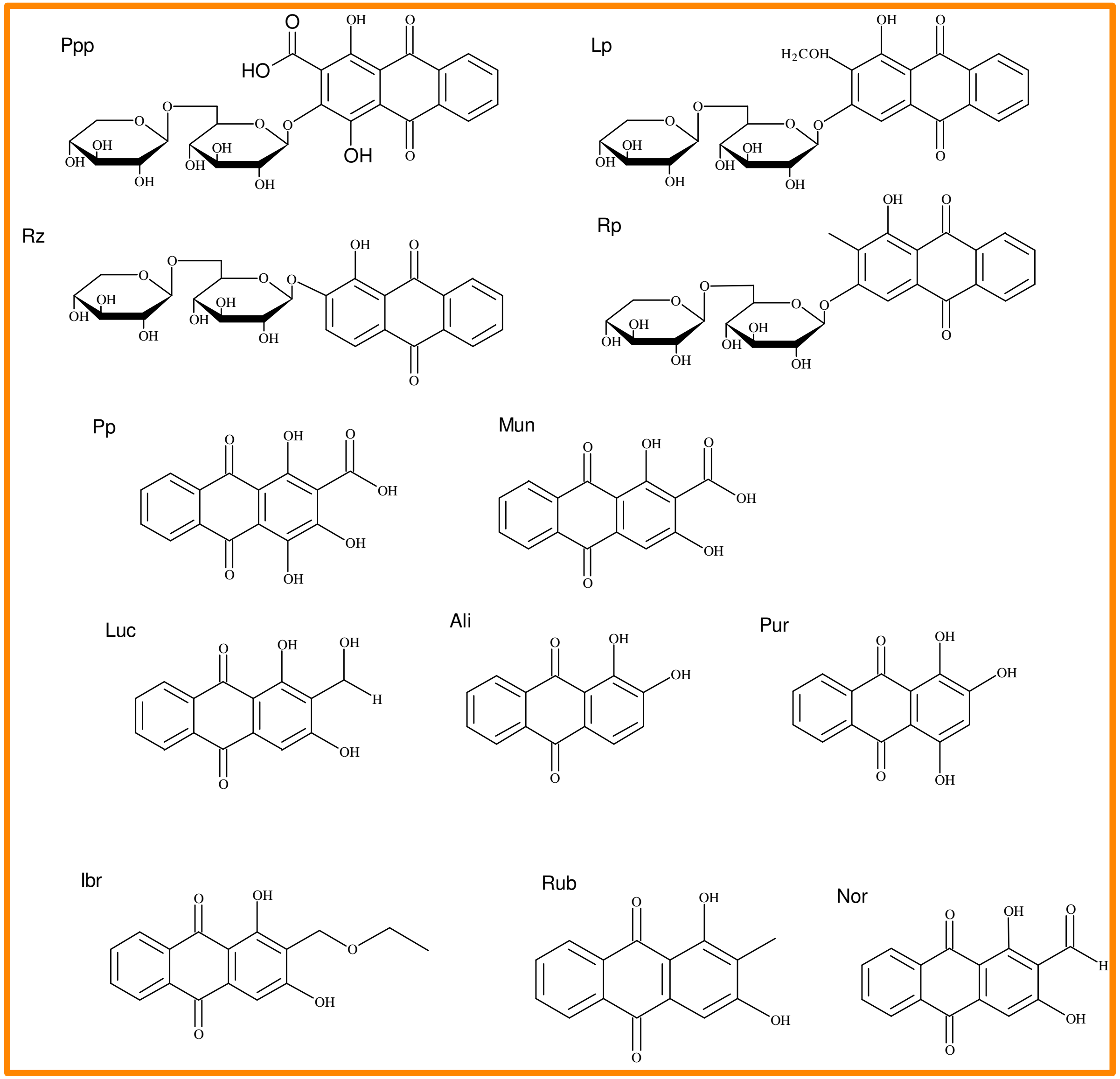
a colourful
future

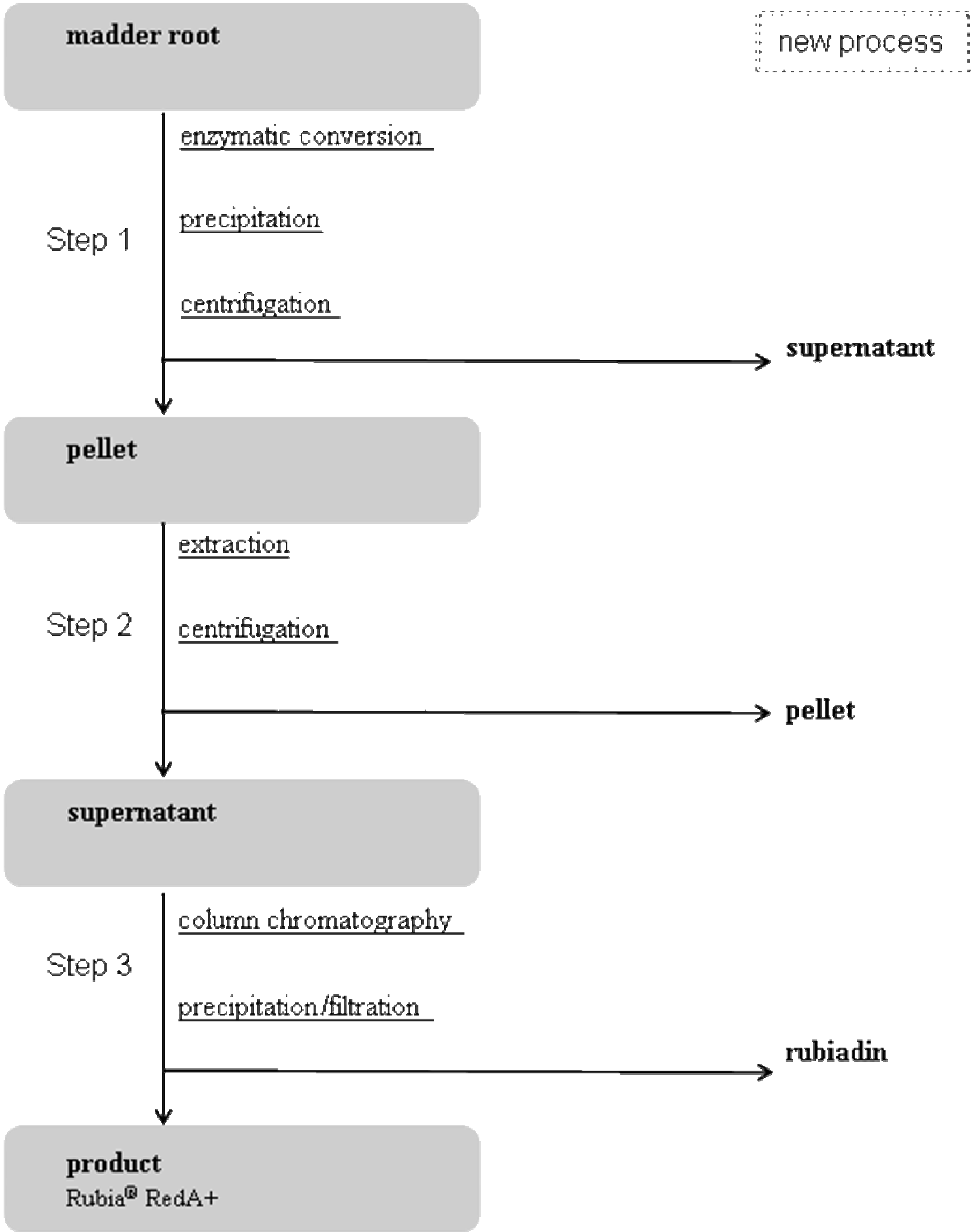

A
phosphoenol pyruvate
erythrose-4-phosphate
DAHPS
3-deoxy-D-arabino-heptulosonate-7-phosphate
DHQS
3-dehydroquinate
DHQD
3-dehydroshikimate
SD
D-shikimate
SK
shikimate 3-phosphate
ESPS
5-enolpyruvylshikimate 3-phosphate
CS
chorismate
CM
prephenate
PAT
arogenate
ADT
phenylalanine
B
Phenyalanine
PAL
C4H
4CL
CHS
CHI
F3H
F3'H
F3'5'H
DFR
ANS
Coumarin derivatives
SCGT
F6'(2'')H
Isoflavones
DMID VR IFR
IFOH
F3GT FOMT
F3GT HIDH
IFS CHI CHR
F3GT HIDH IFS
Flavones
F3GT
FCGT
FS-I
F3'H
F3GT FOMT F5'H
Tannin
LAC BAN
SGT
OMT
Phenylpropanoid
COMT F5H COMT
4CL 4CL 4CL 4CL
CCoAMOT CCR CCoAMOT CCR
CAD F5H COMT CAD
F5H COMT
lignin
SHT
ST CSH CSOMT
Spermidine derivatives
FLS F3GT
FGTs
FATs
FLS F3GT
OMT
Flavonol
F3GT
AGTs
AATs
Anthocyanin

Research

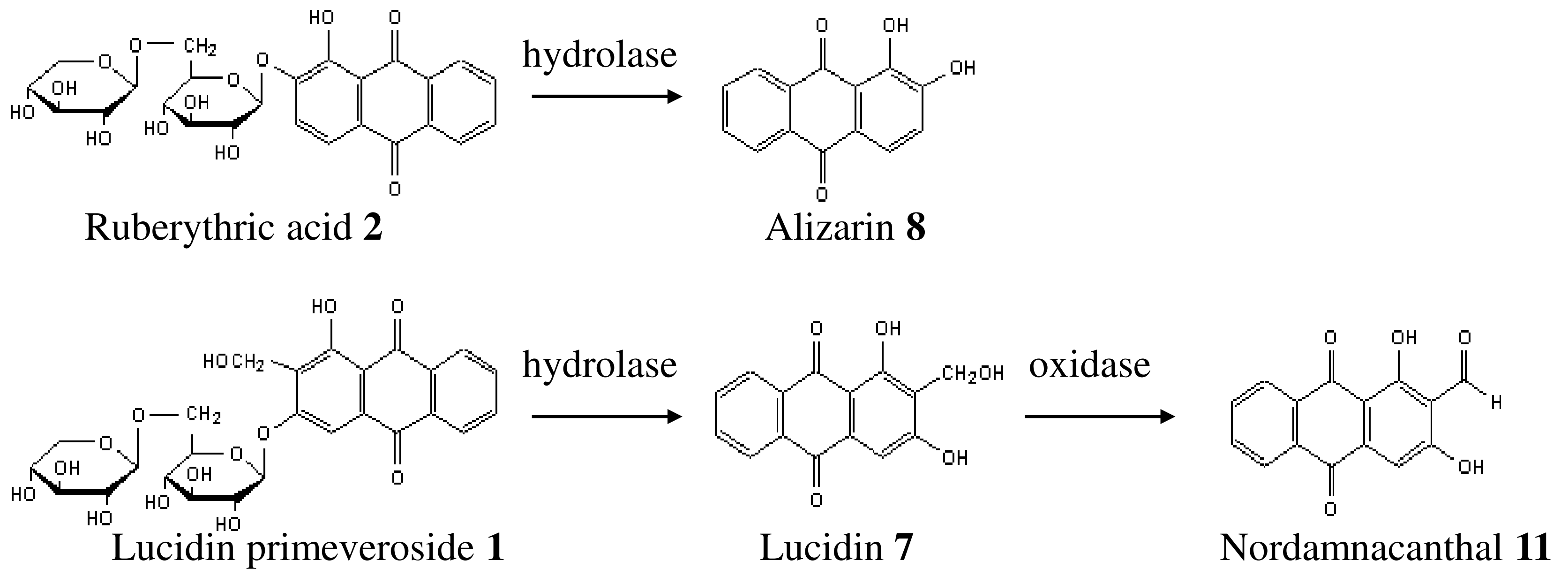
Compounds

1. Water soluble
   sucrose, salts (40%)
2. Fibres, 40%
3. Anthraquinones
   Glycosides
   COOH
   Aglycones
   (5-15%)

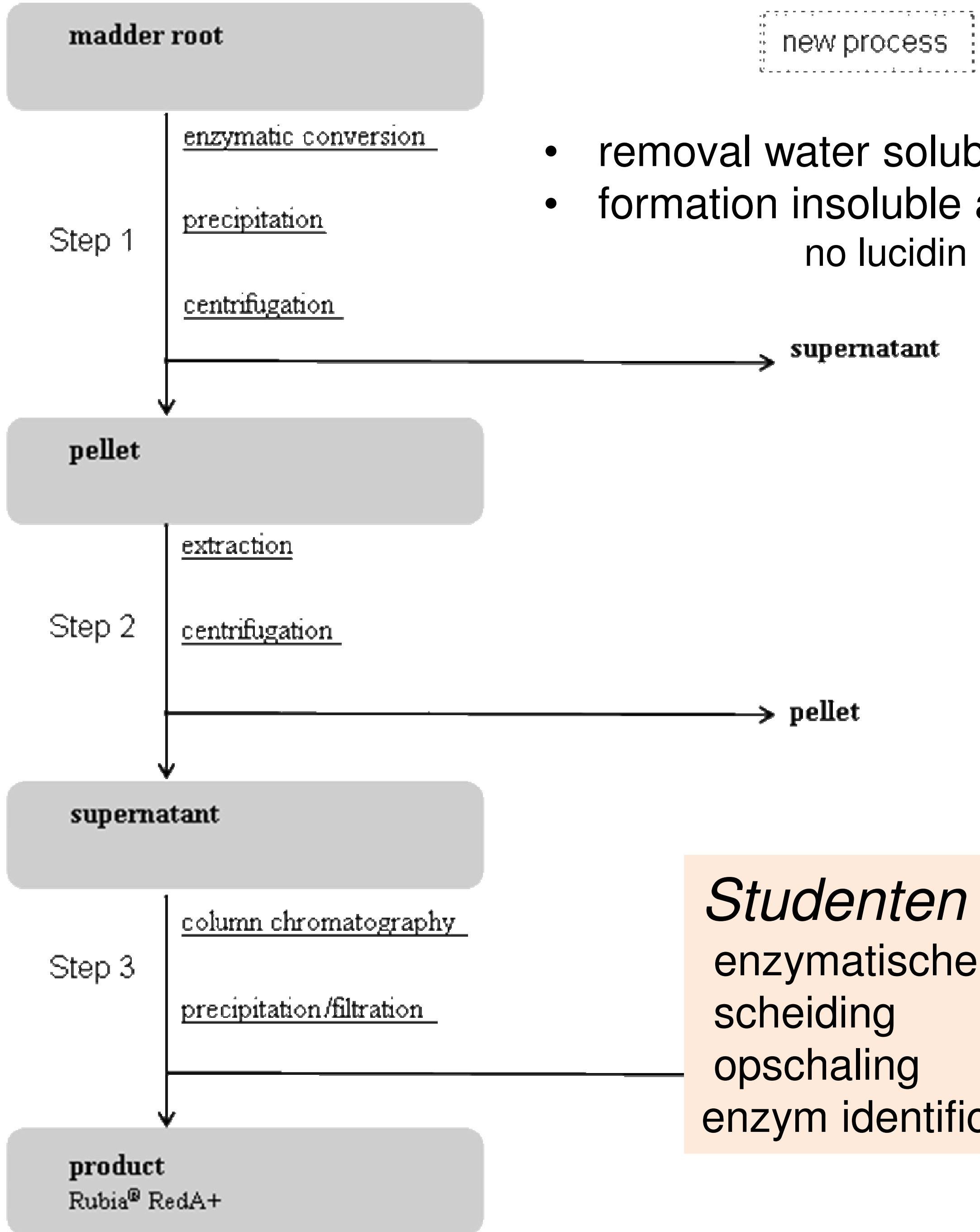



madder root
new process
enzymatic conversion
precipitation
Step 1
centrifugation
supernatant
pellet
extraction
Step 2
centrifugation
pellet
supernatant
column chromatography
Step 3
precipitation/filtration
rubiadin
product
Rubia® RedA+

step 1, enzymatic conversion



G.C.H. Derksen, M. Naayer, T.A. van Beek, A. Capelle, I.K. Haaksman, H.A. van Doren, Æ. De Groot, Chemical and enzymatic hydrolysis of anthraquinone glycosides from madder roots, Phytochem. Anal. 14 (2003) 137-144



- removal water soluble compounds (30%)
- formation insoluble aglycones

no lucidin

Studenten CT en chemie

enzymatische omzetting, T t K O_2

scheiding

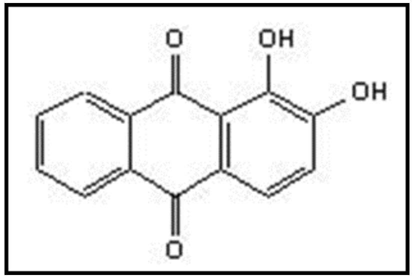
opschaling

enzym identification

step 2, extraction

Isolation anthraquinones from plant fibres, by extraction
→ No organic solvent and low in energy

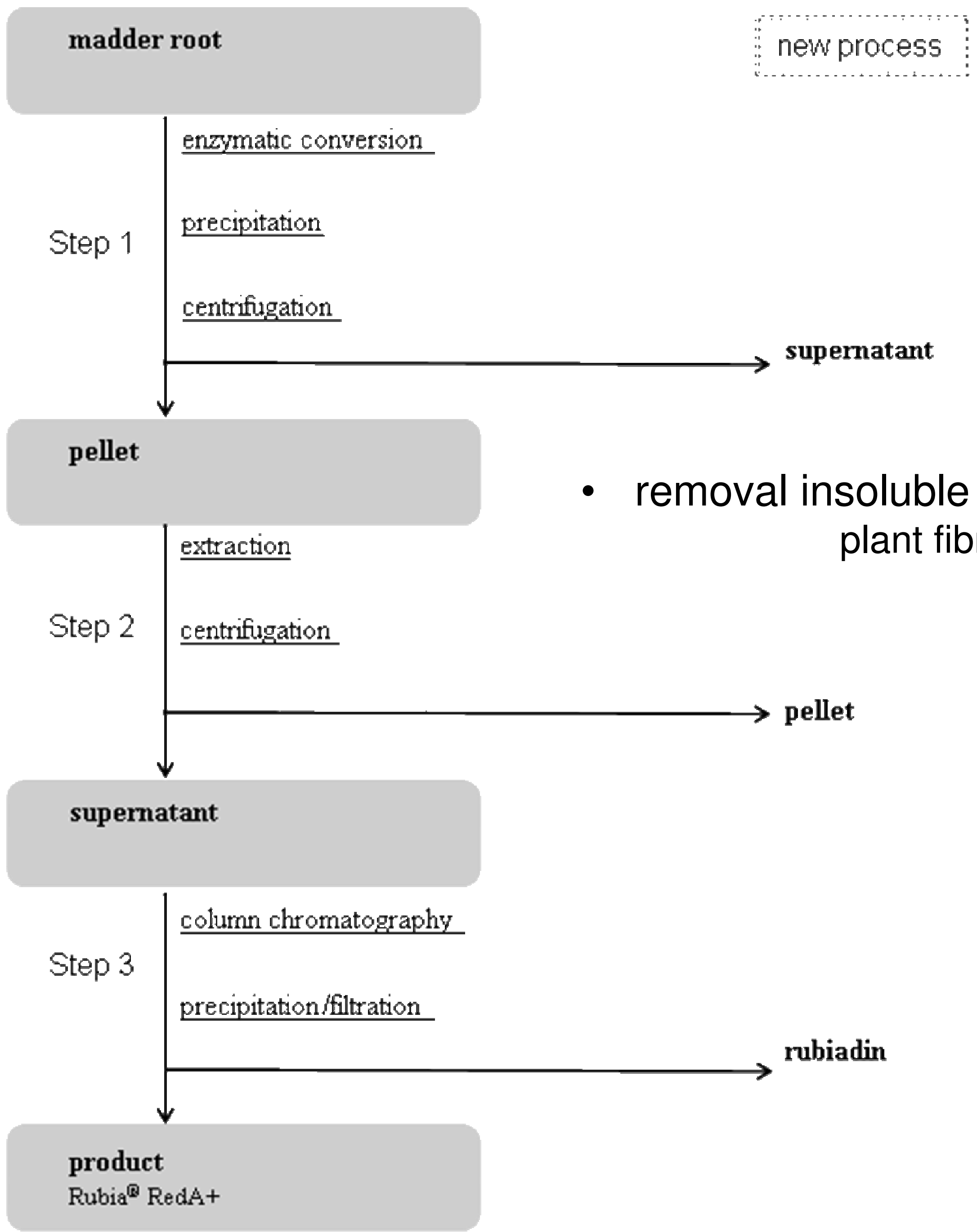
Several possibilities tested
Alkaline extraction (pH=10)
→ drawback yield not always as high as expected



			reflux I	I		rt			NaOH	II	III
grondstof	gedr. pulp	g	5	5						5	5
	ali startprod	%	1.4	1						1.4	1.3
uitvoering	RatioRL	x	9	1						20	20
resultaat	m. oplosbaar	g	0.37	0.						1.07	0.86
	m. onoplosbaar	g	4.6	4						4.4	3.7
	% m. oplosbaar	%	7.5	9						19.4	18.9
	[alizarine] sup.	mg/g	0.95	0.						.60	0.50
	% opl. Ali	%	56	8						83	76
	% ali eindproduct	%	9.5	10						5.1	5.5
			Buchner EtOH-water 1:1 v/v RRL 1:10	Watten EtOH-wate RRL 1:20					beetje HCl voor de analyse	n , 0.04 M art = 11.5 nd = 9.5	Watten NaOH, 0.04 M pH start = 12.4 pH eind = 9.8 ds in oven

Students CT and chemistry

extraction
yield
mass balances
energy balance



- removal insoluble compounds
  plant fibres (40%)

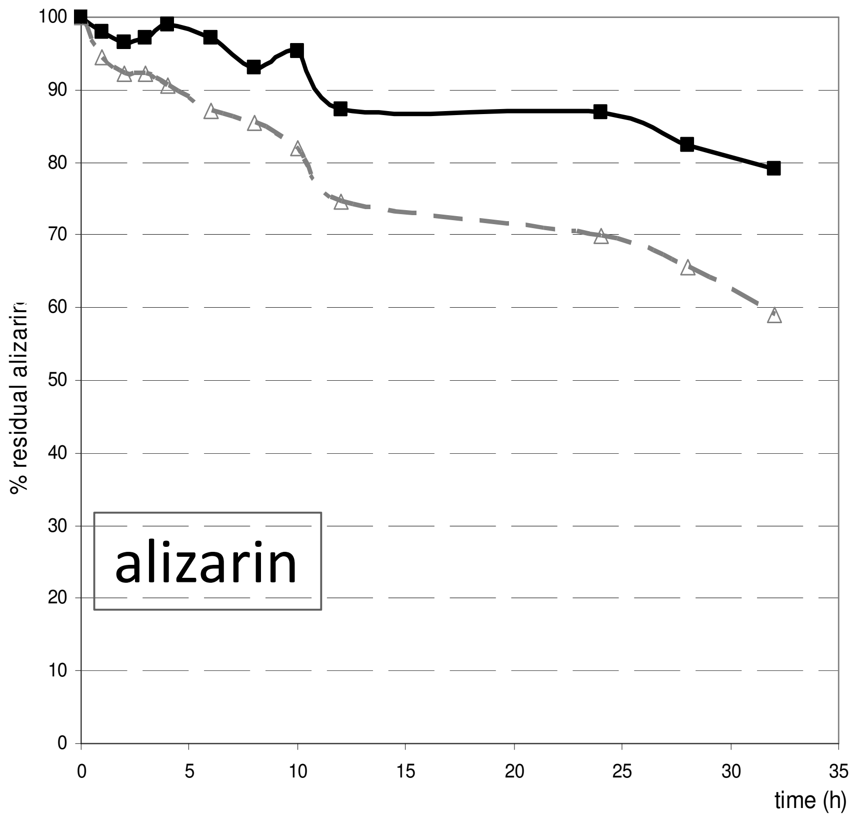
Step 3: Affinity chromatography
Selection resin, Static tests, Corresponding isotherm profile

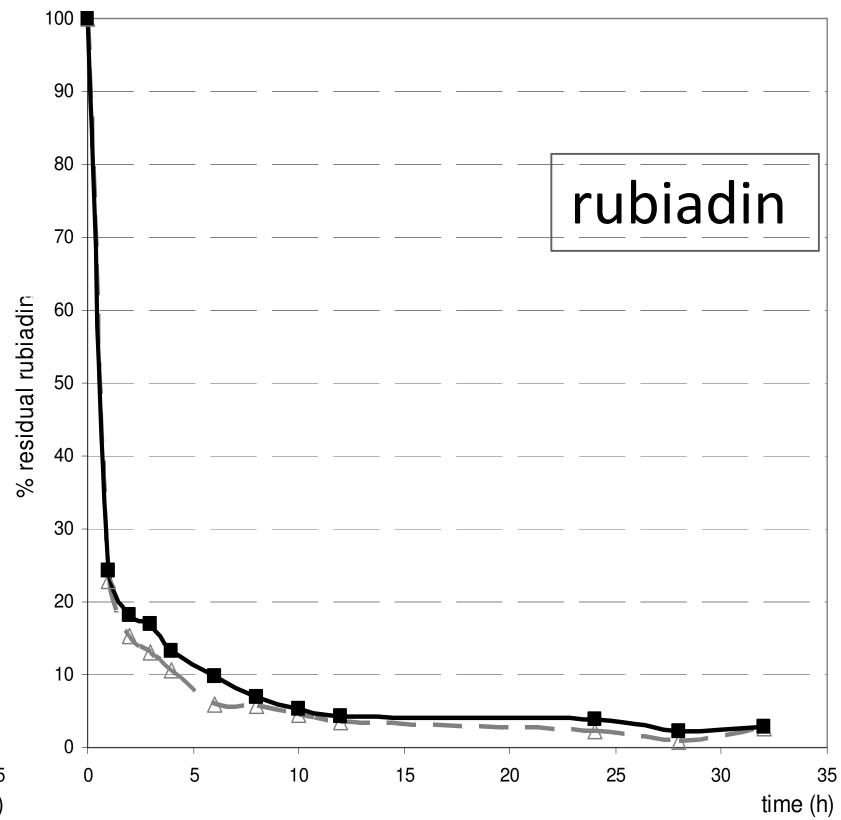
15 different resins investigated here described:

resin 1

resin 2

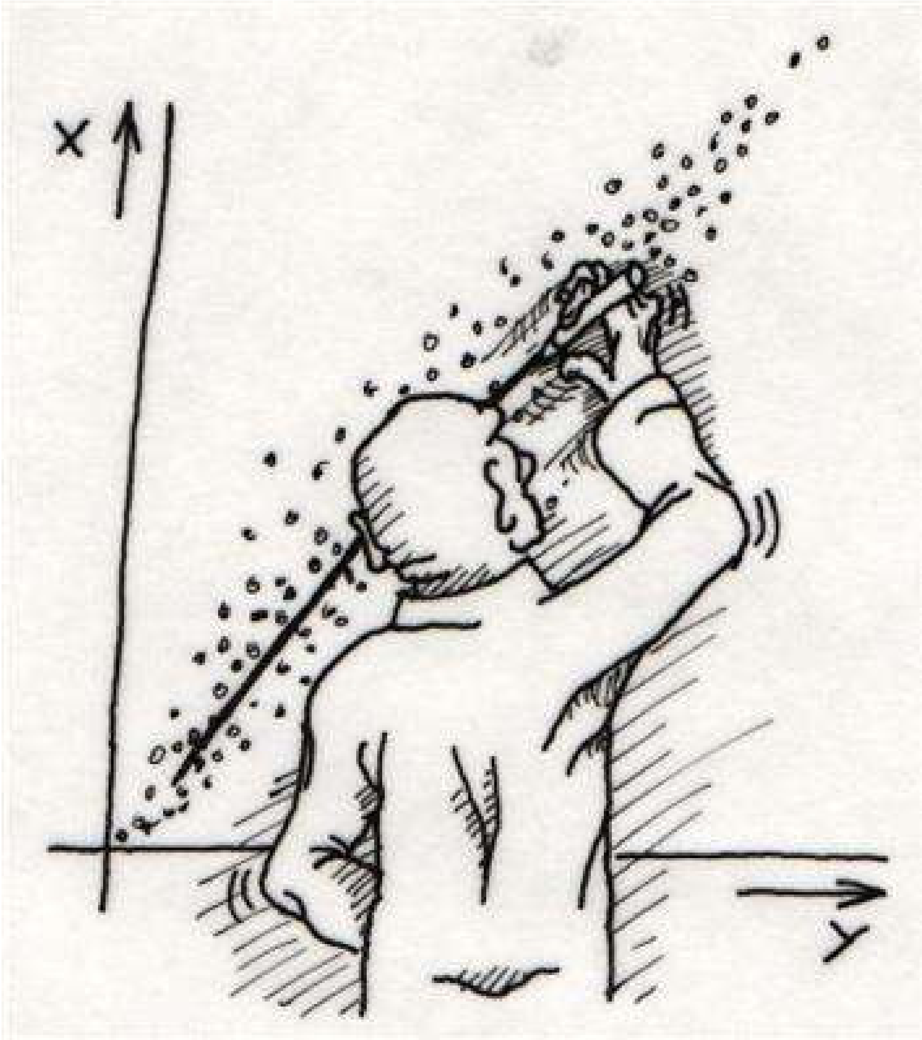
contact time and adsorption

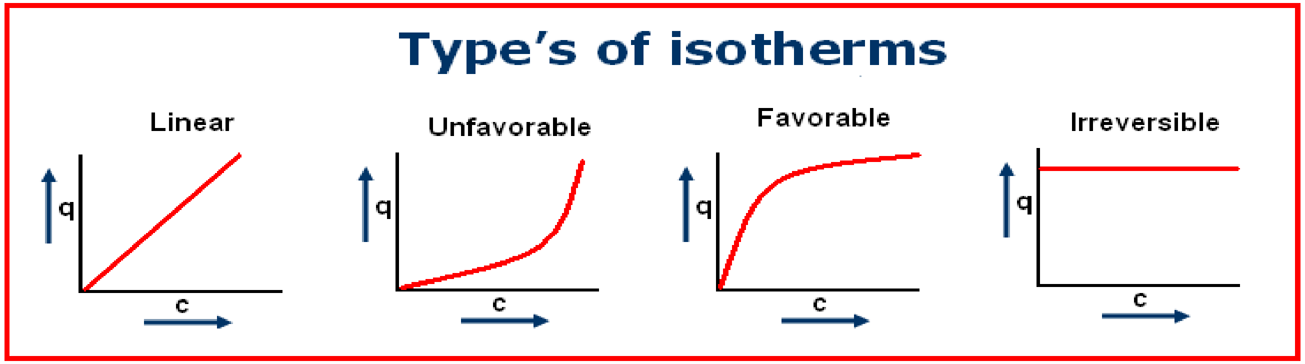




■ resin 1 Δ resin 2

Modelling





n=1 0<n<1 n>1

Adsorption isotherm modelling

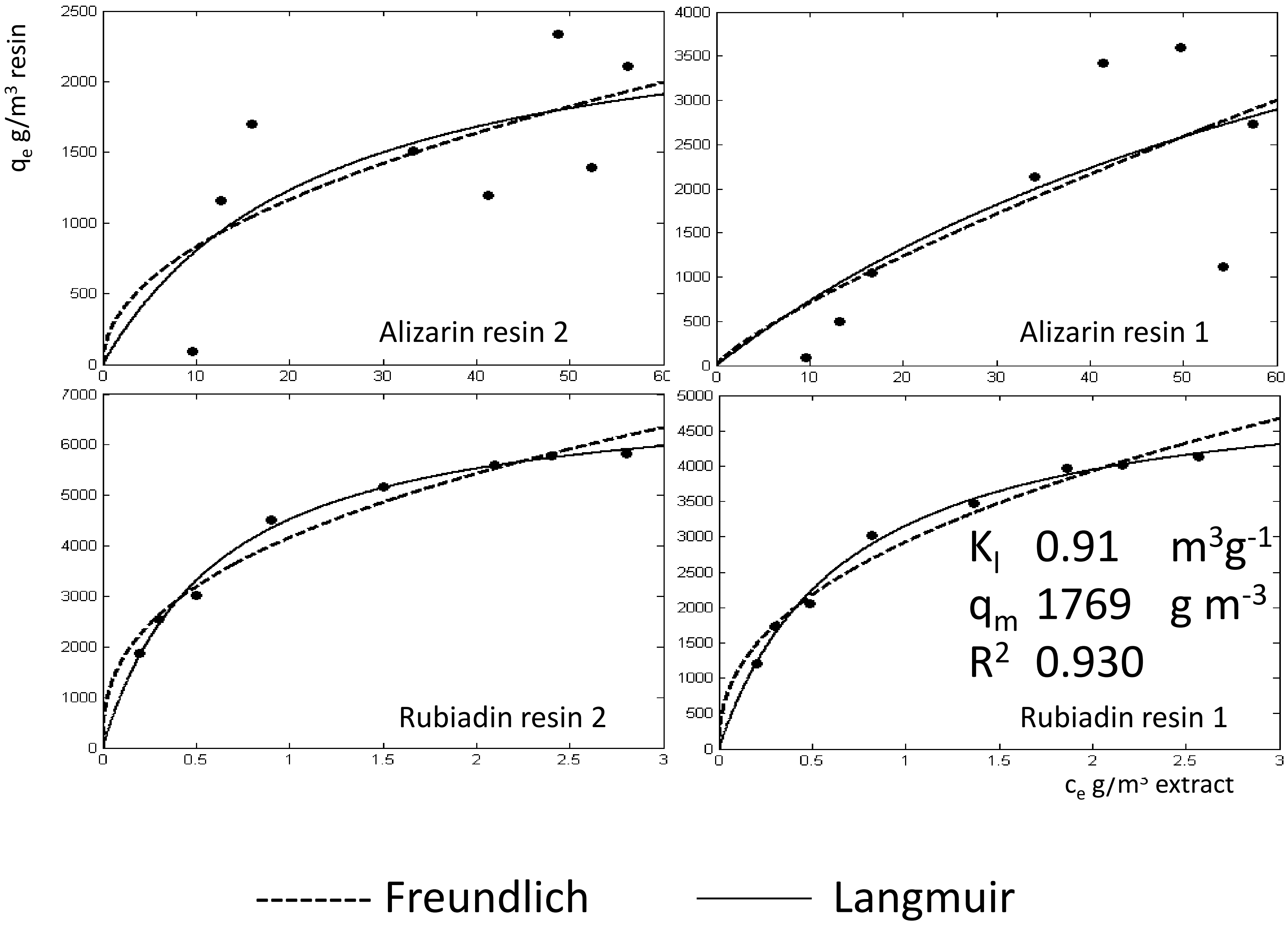
- Langmuir isotherm

$$q_e = \frac{q_m K_L C_e}{1 + K_L C_e} \qquad \frac{C_e}{q_e} = \frac{1}{q_m K_L} + \frac{C_e}{q_m}$$

- Freundlich isotherm

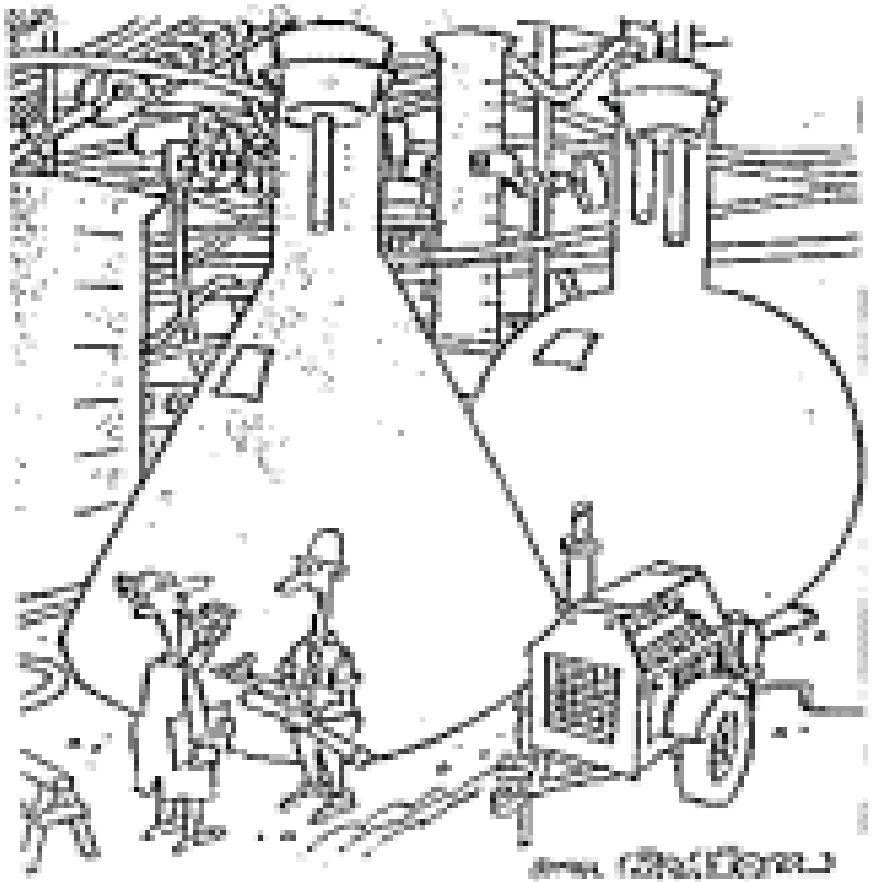
$$q_e = K_F C_e^{1/n} \qquad \log q_e = \log K_F + \frac{1}{n} \log C_e$$

ISOTHERMS



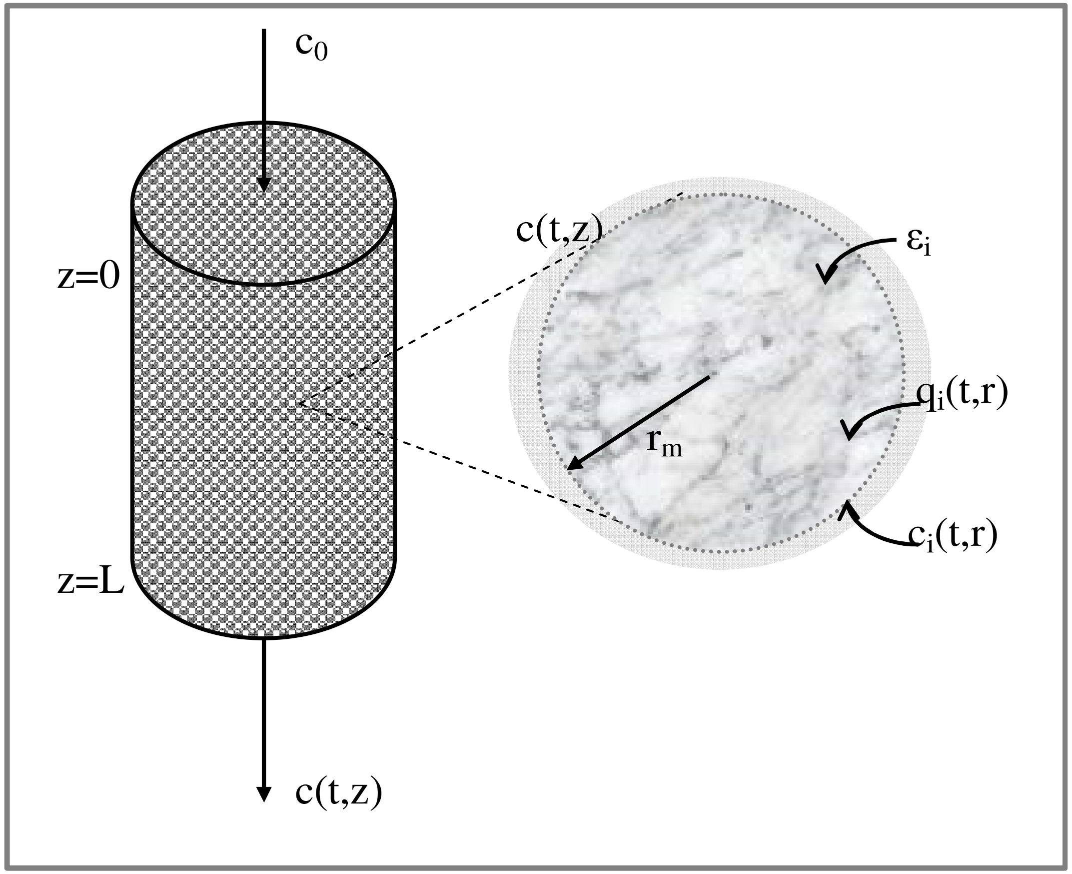
q_e g/m³ resin
Alizarin resin 2
Alizarin resin 1
Rubiadin resin 2
Rubiadin resin 1
K_l 0.91 m^3g^{-1}
q_m 1769 g m^{-3}
R^2 0.930
c_e g/m³ extract
--------- Freundlich
———— Langmuir

Scale-up



"Got a few problems going from lab scale up to full-scale commercial."

Model of the affinity packed bed column



1: mass balance in fluid: convection (flow), dispersion and mass transfer

2: mass balance in the fluid of the particle

3: Interaction of rubiadin and affinity adsorbent, 2nd order.

R + S ⟺ RS

P. Guerrero-German, R.Ma. Montesinos-Cisneros, A. Tejeda-Mansir; Simulation of frontal protein affinity chromatography using MATLAB; J. Chem. Eng. Process Technol., 3(3) (2012); 6p.

PARTIAL DIFFERENTIAL EQUATIONS

$$\varepsilon A \Delta z \frac{dc_z}{dt} = (Fc_z - Fc_{z+\Delta z}) - (\varepsilon AN_z - \varepsilon AN_{z+\Delta z\,z}) - (1-\varepsilon) A \Delta z \frac{3}{r_m} N\Big|_{r=r_m}$$

$$N = -D_L \frac{dc}{dz}$$

$$N\big|_{r=rm} = k_f (c - c_i)$$

$$\frac{\partial c}{\partial t} = D_L \frac{\partial^2 c}{\partial z^2} - \frac{F}{\varepsilon A} \frac{\partial c}{\partial z} - \frac{3}{r_m} \frac{(1-\varepsilon)}{\varepsilon} k_f (c - c_i)\Big|_{r=r_m}$$

$$\varepsilon_i A_r \Delta r \frac{dc_i}{dt} + (1-\varepsilon_i) A_r \Delta r \frac{dq_i}{dt} = A_r N_r - A_{r+\Delta r} N_{r+\Delta r}$$

$$\varepsilon_i \frac{\partial c_i}{\partial t} + (1-\varepsilon_i) \frac{\partial q_i}{\partial t} = \frac{1}{A} \frac{\partial (AN)}{dr}$$

$$N = -D_R \frac{dc}{dr}$$

$$\varepsilon_i \frac{\partial c_i}{\partial t} + (1-\varepsilon_i) \frac{\partial q_i}{\partial t} = D_R \left(\frac{\partial^2 c_i}{\partial r^2} - \frac{2}{r} \frac{\partial c_i}{\partial r} \right)$$

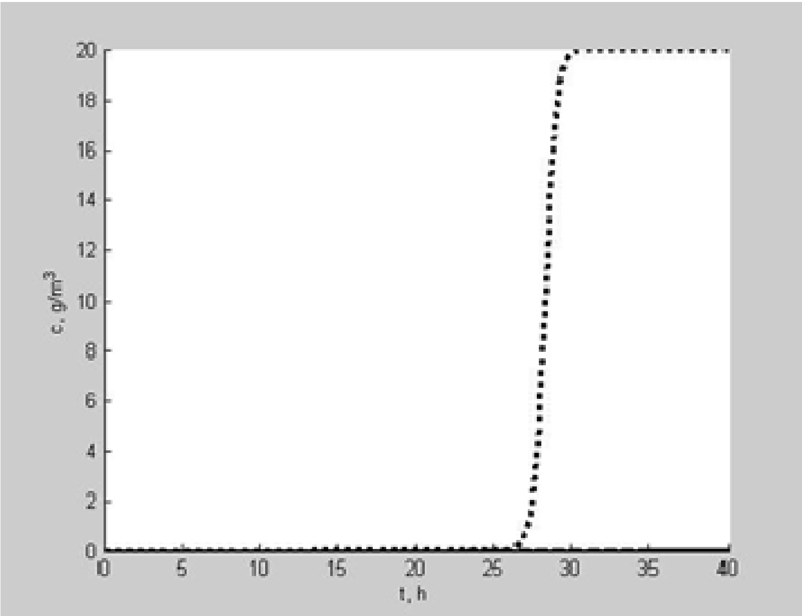
$$R + S \Longleftrightarrow RS$$

$$\frac{\partial q_i}{\partial t} = k_{ads} c_i (q_m - q_i) - k_{des} q_i$$

Scale-up

Flow= 5000 L per 24 h (208 L h^{-1})

C_{in}= 20 g m^{-3}



Based on Matlab:

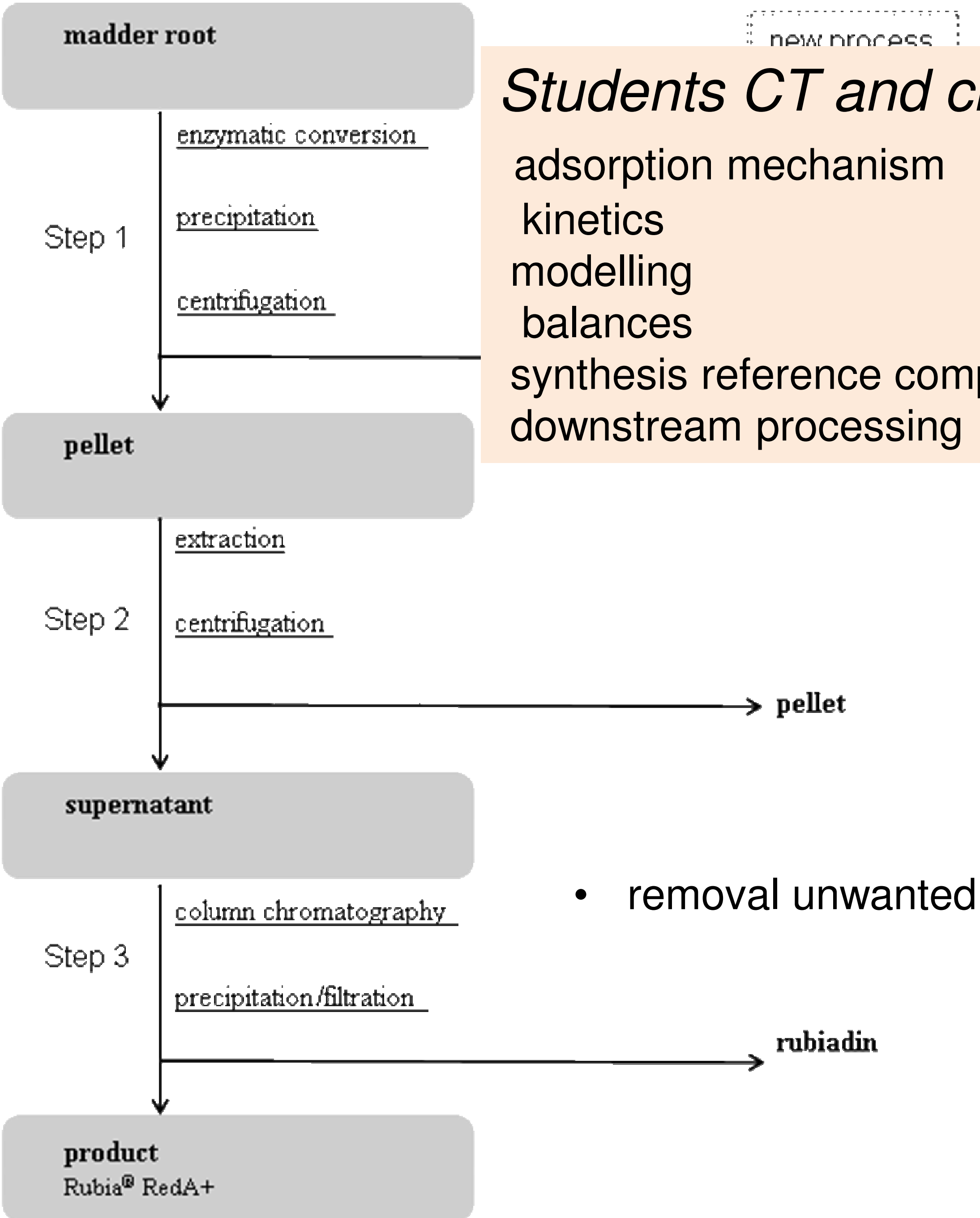
V= 0.351 m^3

$t_{br\ 10\%}$ = 27 h.

Based on experimental:

V= 0.170 + 0.170 =0.340 m^3

$t_{br\ 10\%}$ = 24 h.



Students CT and chemistry

adsorption mechanism
kinetics
modelling
balances
synthesis reference compounds
downstream processing

- removal unwanted anthraquinones

Isolation of secondary metabolites

Natural Colours as a product

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Thanks to:

- research members
- students
- companies
- TU/e and WUR
- audience

Thanks for your attention

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