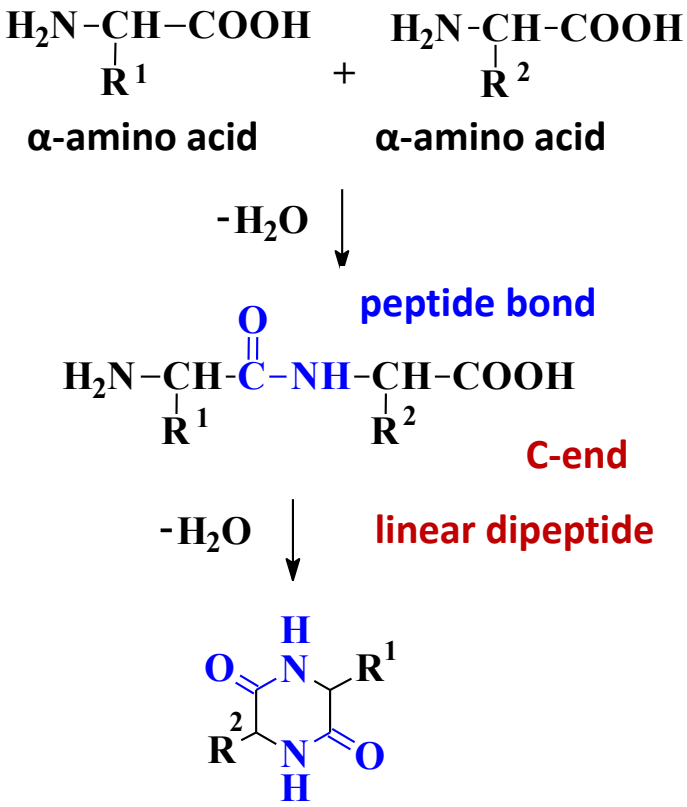
Peptides

structure

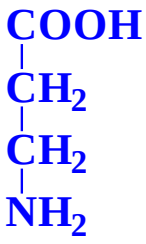
amino acids —**condensation**→ peptides



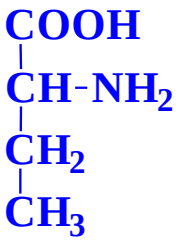
cyclic dipeptide (2,5-dioxopiperazine)

peptidic bond = amide bond

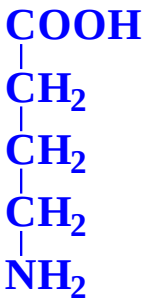
- uncommon binding of some amino acids distal COOH group of Glu = γ -peptide bond
- D-amino acids bound
- unusual amino acids bound



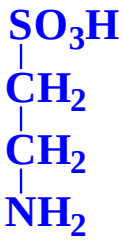
β-alanine



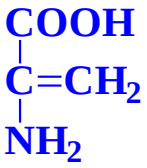
α-aminobutyric



γ-aminobutyric

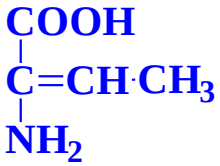


taurine



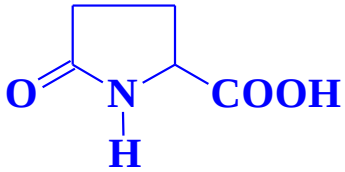
2-aminoacrylic

(dehydroalanine)



(*E*)-2-aminocrotonic

(dehydrobutyrine)



pyroglutamic

classification

number of bound monomers (amino acids)

- oligopeptides (2-10 amino acids)
- polypeptides, formerly: makropeptides (11-100 amino acids)

chain character

- linear
- cyclic

bond type

- homodetic (peptide bonds only)
- heterodetic (peptide bonds and other bonds) disulfides -S-S-
  esters (depsipeptides) -CO-O-R

bound components

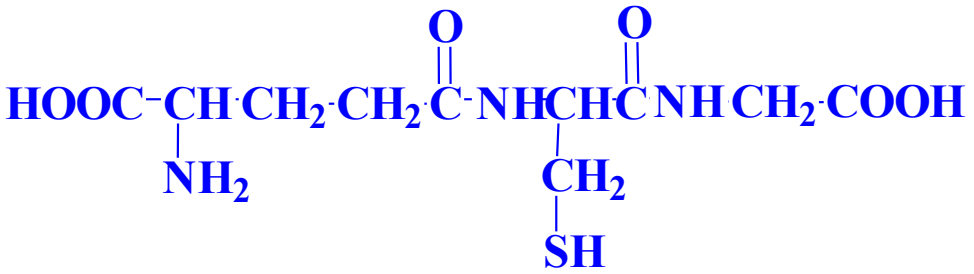
- homeomeric amino acids only
- heteromeric amino acids and other compounds

  nucleopeptides, phosphopeptides, lipopeptides, chromopeptides, glycopeptides, metalopeptides

nomenclature

glycylglycine	Gly-Gly	G-G

glycylalanine	Gly-Ala	G-A
alanylglycine	Ala-Gly	A-G
cyclic glycylglycine	cyklo-(Gly-Gly)	cyclo-(G-G)
glutathione	Glu	
	γ⌊Cys-Gly	ECG



occurence

- **product of metabolism**

  natural peptides

- **product of proteolysis**

  enzymatic and nonenzymatic

- **synthetic peptides** **artificial sweeteners**

product of metabolism

hormones

linear: secretine, insuline, thyroliberine ….

cyclic: vasopresine, oxytocine ….

antibiotics

products of lactic acid fermentation = bacteriocines

nisin (E234)

Streptococcus cremoris*, syn. *Lactococcus lactis* ssp. *lactis

preservative, stabilizing dairy fermented products

Abu = 2-aminobutyric acid ← **Glu**

Dha = 2-aminoacrylic a. (dehydroalanine) ← **Ser**, **Cys**

Dhb = 2-aminocrotonic a. (dehydrobutyrine) ← **Thr**

HoLys = 5-hydroxylysine

D = D-isomer

toxins

- bacteria: botulotoxins bacteria (*Clostridium botulinum*)
- fungi: fallotoxins, amatoxins (*Amanita phalloides*)
  cortinarins (*Cortinarius orellanus*)
- insects, reptiles, etc.

fallotoxins, amatoxinys

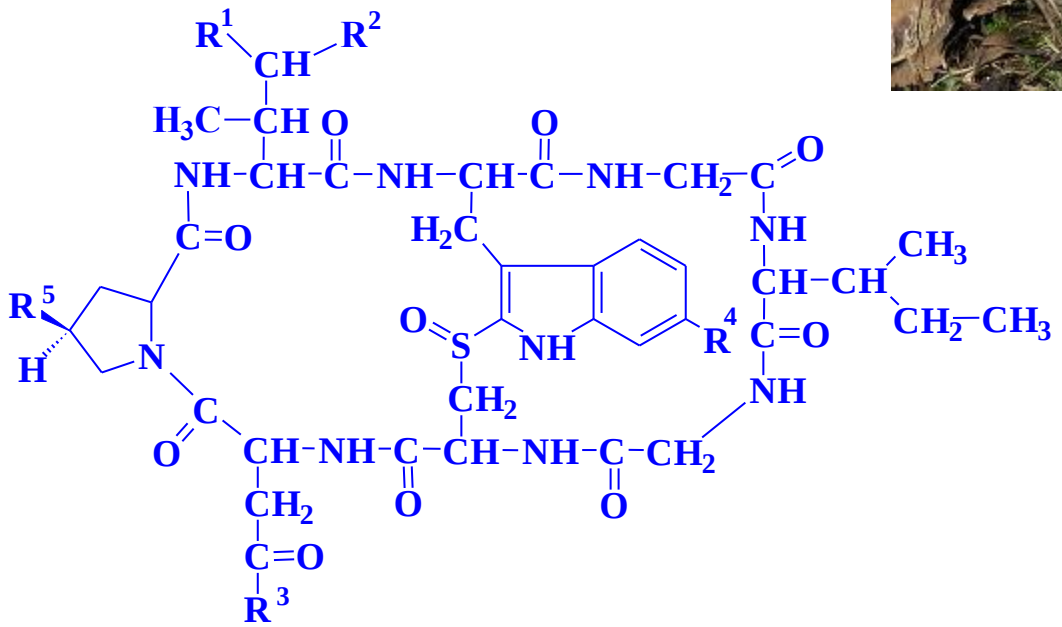
inhibition of enzymes metabolising nutrients (protein synthesis)

main toxic compounds

falloidin (about 100 mg/kg fresh mushroom)

α-amanitin (80 mg/kg, LD_{50} = 0,1 mg/kg)

β-amanitin (50 mg/kg, LD_{50} = 0,4 mg/kg)

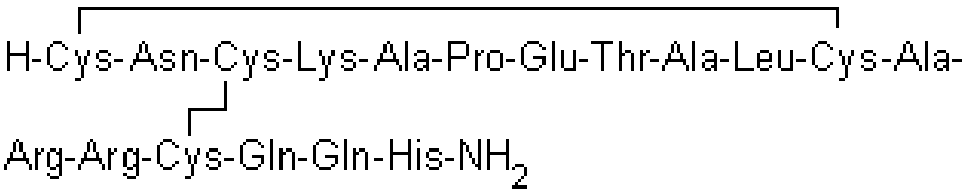


α-amanitin, R^1 = CH_2OH, R^2 = OH, R^3 = NH_2, R^4 = OH, R^5 = OH

β-amanitin, R^1 = CH_2OH, R^2 = OH, R^3 = OH, R^4 = OH, R^5 = OH

apamin

in the venom of bees *Apis mellifera* : peptide from 18 AA with 2 disulfidic bridges



melittin

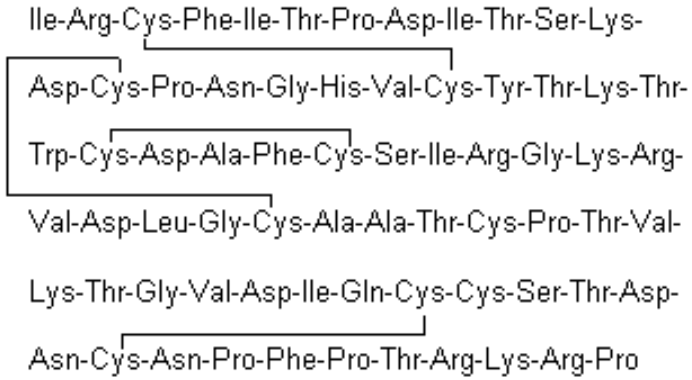
in the venom of bees *Apis mellifera* : peptide from 26 AA

H-Gly-Ile-Gly-Ala-Val-Leu-Lys-Val-Leu-Thr-

Thr-Gly-Leu-Pro-Ala-Leu-Ile-Ser-Trp-Ile-Lys-

Arg-Lys-Arg-Gln-Gln-NH_2

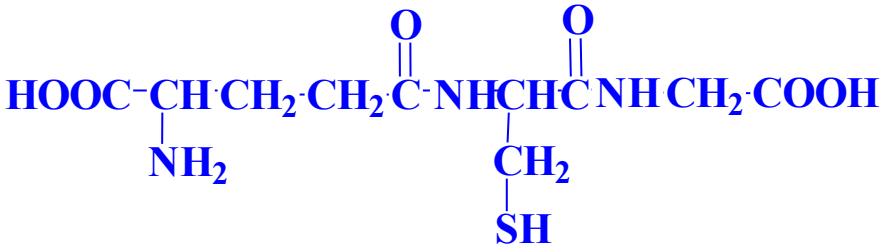
α–cobrotoxin



in the venom of the cobra*Naja naja kaouthia (siamensis)* : peptide from 71 AA with 5 disulfidic bridges

other important peptides

glutathion



γ-L-glutamyl-L-cysteinylglycine (γ-amidic bond)

reduced (G-SH) and oxidised form (G-S-S-G)

occurrence microorganisms, plants, animals

wheat	10-15 mg/kg
meat	300-1500 mg/kg

- protects the body against oxidative stress (involved in the removal of H_2O_2)

biochemistry

- participation in the detoxification(superoxiddismutasa, mitochondrial/Mn, cytosolic/Zn, Cu)
- transportation (transfer of amino acids into cells)
- substrate of peroxidases, glutathionreductase
  (maintaining the oxidation state of protein-SH)

$$\text{P-SH + P´-SH + ½ } O_2 \longrightarrow \text{P-S-S-P´+ } H_2O \text{ (autoxidation)}$$

$$\text{P-S-S-P´ + G-SH} \longrightarrow \text{P-SH + G-S-S-P´}$$

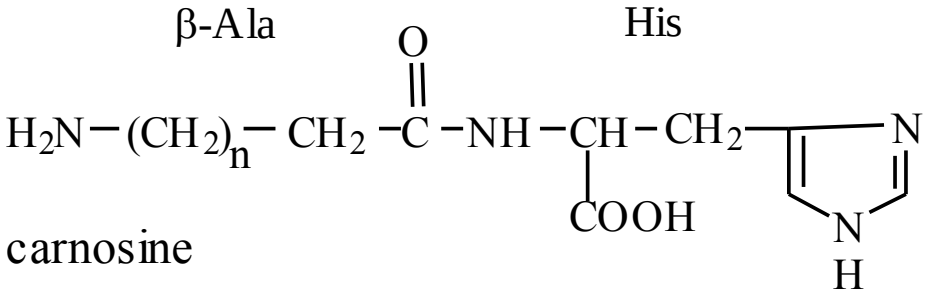
$$\text{G-S-S-P´ + G-SH} \longrightarrow \text{P´-SH + G-S-S-G}$$

$$\text{G-S-S-G + 2 H} \longrightarrow \text{2 G-SH}$$

β-alanylhistidine dipeptides

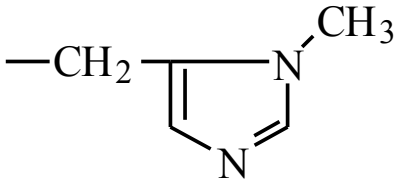
biochemistry

- **participation in the contraction of skeletal muscle**
- **organoleptic properties (taste umami)**

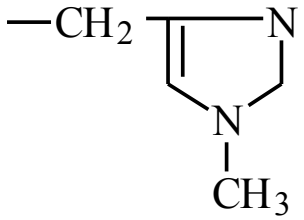


carnosine

anserine (n = 1), R =



balenine (n = 1), R =



analytics

- **criterion for the determination of meat origin in meat products (e.g. chicken meat in pig meat products)**

product of proteolysis

spontaneous proteolysis (autolysis)

desirable meat maturation (consistency, aroma), production of yeast autolyzates (additives)

undesirable

intentional proteolysis

cheese production (desirable consistence, aroma)

malt production (stabilisation of beer foam by hydrofobic polypeptides from proteins barley and yeast)

protein hydrolyzates production

enzymatic: soya sauce

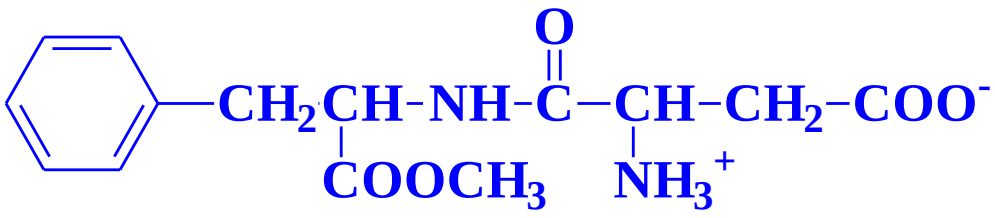
acid: hydrolysed vegetable protein

synthetic peptides

artificial sweetner Aspartam (Asp-PheMe)

sweetness **Aspartam** **100-200**

sacharosa **1**



currently - in most light drinks (such as Coca Cola Zero) and food, table-top sweeteners

- **E951**

Proteins

polymers of amino acids

> 100 amino acids

M_r ~ 10.000 - miliony Da

organized structures

(constitution and conformation optimal for their functions)

main nutrients

peptide bonds

other bonds

- disulfide -S-S-
- ester
- amide

other compound besides amino acids (physical or chemical bond)

- water
- inorganic ions
- lipids, saccharides, nucleic acids, colour compounds (pigments)

classification

according to origin

- **animal** (meat, milk, eggs)

  50-60% proteins of diet

- **vegetal** (cereals, legumes, fruits, vegetables, root crops)

  40-50% proteins of diet

- **nontraditional** (algae, microorganisms)
  - yeasts (*Candida*)
  - algae (*Chlorella*)
  - bacteria
  - protein concentrates (proteins=50% of dry matter)
  - protein isolates (proteins=90% of dry matter)

classification according to function

- structural (components of cells, e.g. collagen)
- catalytic (enzymes, hormones)
- transport (tranport of compounds, e.g. myoglobin)
- motoric (muscle proteins, e.g. actin, myosin)
- defensive (imunoglobulins, lectines)
- storage (ferritin)
- sensory (rhodopsin)
- regulatory (histones, hormones)
- nutritional (source of essential amino acids, source of nitrogen, material for tissues)

according to structure (presence of nonprotein components)

simple

- globular, spheroproteins (albumins, globulins, ….) soluble in water
- fibrilar (fibrous), scleroproteins (collagens, keratins, elastins) insoluble

conjugated

- nucleoproteins (nucleic acids)
- lipoproteins (neutral lipids, phospholipids, sterols)
- glykoproteins (saccharides)
- phosphoproteins (phosphoric acid)
- chromoproteins (porphyrins, flavins)
- metaloproteins (bound metals)

according to solubility

soluble

- albumins
  milk: lactalbumin
  egg white: ovoalbumin, conalbumin
  wheat: leucosin
- globulins
  meat: myosin, actin
  milk: lactoglobulin
  egg: ovoglobulin
- gliadins or prolamins (high amount of bound Pro and Gln, chybí Lys)
  wheat: gliadin
  barley: hordein
  corn: zein
- glutelins
  wheat: glutenin
  rice: oryzenin

- **protamins** basic

  fishes (cyprimin, salmin, klupein, skombrin)

  ↓ ↓ ↓ ↓

  carp salmon herring mackerel

- **histons** basic

  blood: hemoglobin and myoglobin

insoluble

epithelial tissue — **skin**

supporting (connective) tissue — **cartilage, bone**

muscle tissue

} **collagen, elastin, keratin**

according to state

- native (natural, they have various biological functions)
- denaturated
- modified (additives)

according to nutritive value

- nutritionally balanced
  (essential amino acids in optimal amount)

  egg and milk proteins
- almost nutritionally balanced
  (slight shortage of some essential amino acids)

  muscle proteins of animals
- nutritionally disbalanced
  (rough shortage of some essential amino acids)

  all vegetables, from animals - connective tissues

origin of proteins in diet	(%)
meat, fishes	27
cereal products	20
milk, cheeses	20
eggs	5
potatoes, fruits, vegetables	4
animal	50 - 60
vegetal	40 - 50

protein content in some foods of animal origin

food	content (%)	average (%)
meat, meat products		
beef	13 - 27	20.8
pork	9 - 20	15.5
veal	18 - 28	21.8
sausages	13 – 28	20.8
poultry	13 – 24	21.1
chicken		20.5
duck		16.1
fishes	16 – 29	18.7
milk, milk products		
cow milk	3,0 - 3,4	3.2
quark	18 – 21	19.4
soft cheeses	12 - 20	15.0
hard cheeses	24 – 41	24.8
butter	0.4 – 0.6	0.5
chicken eggs		
whole eggs		13.0
white		11.0
yolk		17.0

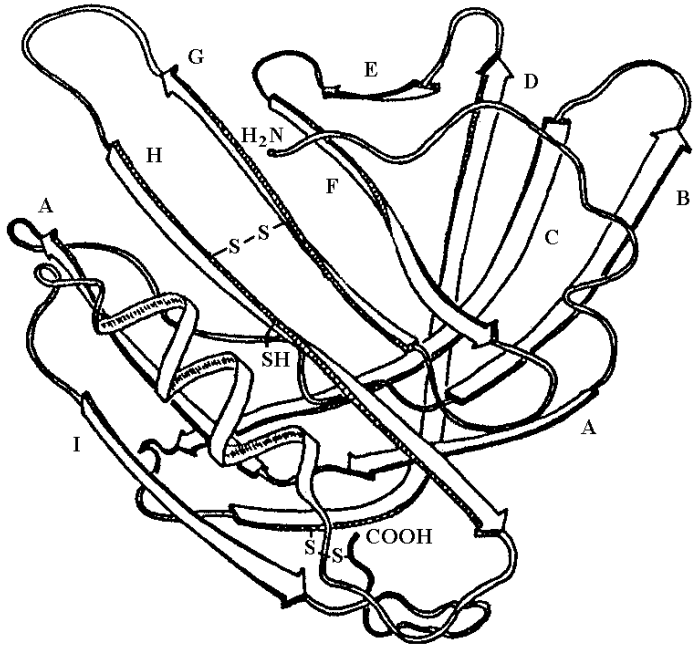
protein content in some foods of vegetal origin

food	content (%)	average (%)
cereals, cereal products		
wheat flour	8-13	10.1
rye flour	5-12	9.6
rice		7.5
bread	4.7-11.6	6.7
bílé pečivo	7.3-9.7	8.5
pastry	3.5-7.8	5.6
pasta	9.8-12.5	11.8
legumes, oilseeds, nuts	21-45	24.2
soybeans		44.7
poppy		19.5
potatoes		2.0
vegetables		
Brassica	0.7-1.8	1.4
leafy	1.3-3.9	2.6
Root	1.0-3.3	2.0
Fruits		
fresh	0.3-1.5	1.0
dried	1.4-4.0	2.3
other food		
mushrooms		2.6
chocolate	4.9-8.1	6.8

structure

β-lactoglobulin of cow milk

162 amino acids, 2 disulfidic bridges (Cys 66-Cys 160, Cys 106-Cys 119), free thiol group (Cys 121)



cylindrical structure called β-barrel
9 fibers of pleated sheet (A – I)
fiber connection H – I = α-helix (AA 130-140)

physiology and nutrition

minimum need of nutritionally balanced proteins: 0.5 – 0.6 g.kg^{-1}

recommended amount: 1.0 – 1.2 g.kg^{-1} (not digested optimally)

up to ~ 2,4 g.kg^{-1} during growth, reconvalescents etc.

- **coverage of total requirement of energy:~ 10 energy % (E%)**

nutrition value (biological)

total intake

composition of amino acids

availability of peptides bound by digestive enzymes

other factors

formerly

- **BV (Biological Value, g of P retained by body / 100 g P in food)**
- **NPU (*Net Protein Utilization*)**
- **PER (*Protein Efficiency Ratio*)**

today

- **AAS (*Amino Acid Score*),**

$$AAS(\%) = \frac{100\ A_i}{A_{si}}$$

A_i = content of esential amino acid in protein

A_{si} = content of esential amino acid in standard (reference) protein

imaginary protein with an optimal composition of the essential amino acids(AAS = 100%)

- **EAAI (*Essential Amino Acid Index*)**

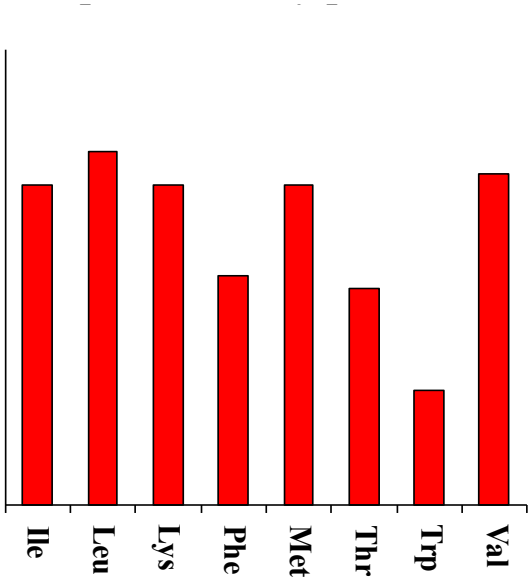
$$EAAI = \sqrt[n]{\frac{100A_1}{A_{S1}} \cdot \frac{100A_2}{A_{S2}} \cdot \quad \cdot \frac{100A_n}{A_{Sn}}}$$

content of essential amino acids in the standard protein

(g based on 16 g nitrogen) and the daily requirement of these amino acids

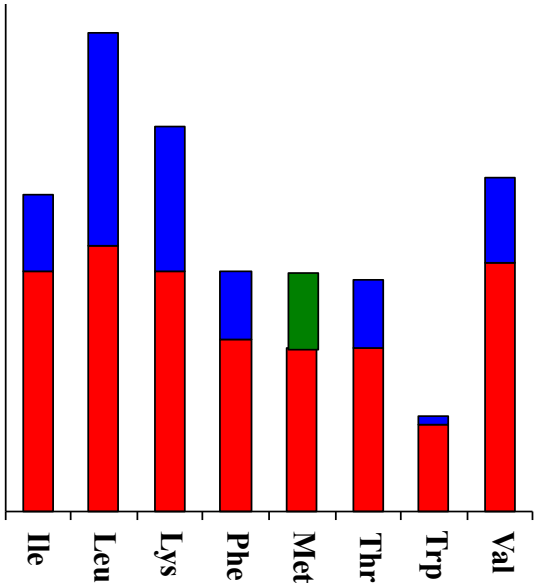
amino acid	protein FAO/WHO	daily requirement (g)
Val	5.0	11-14
Leu	7.0	11-14
Ile	4.0	10-11
Met and Cys	3.5	11-14
Thr	4.0	6-7
Lys	5.4	9-12
Phe and Tyr	6.1	13-14
Trp	1.0	3-3.5
total	36.0	

standard (reference) protein



protein FAO/WHO

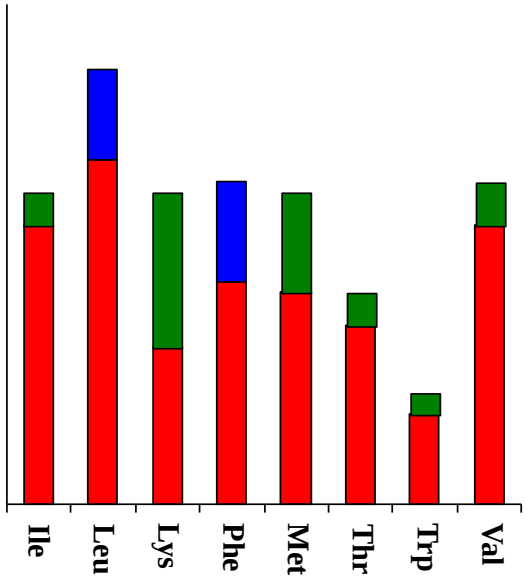
cheese



limiting: Met

milk protein

bread



limiting: Ile, Lys, Met, Thr, Trp, Val

wheat protein

physical-chemical properties

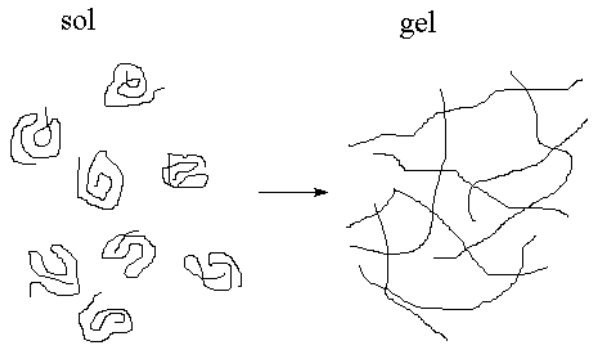
solubility, hydratation, swelling

- molecules hydrated
- macromolecular polyions, polyamfolytes
- globular = colloidal dispersions, colloids (1-1000 nm)

  dispersive system (dispersion): sols, gels

- micellar (associative) colloids, aggregates of molecules

  e.g. α-, β- and κ-caseins in milk



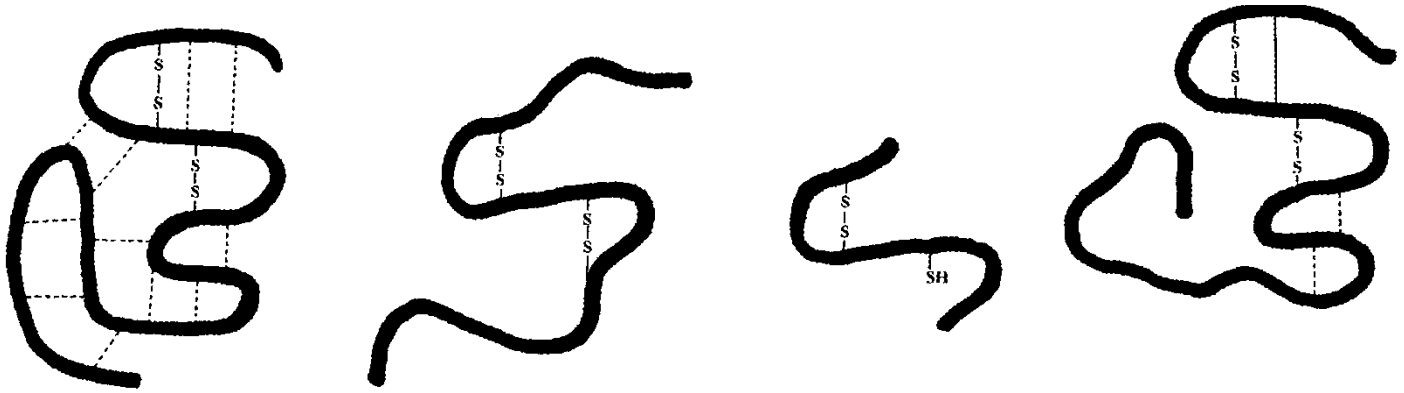
denaturation

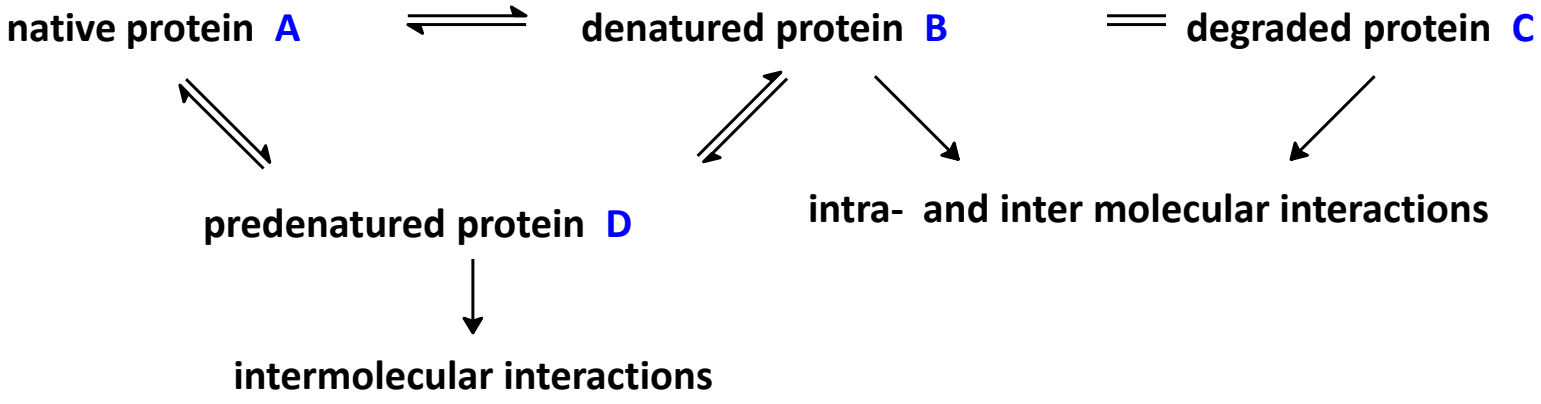
protein structure changed to less organized

conformational changes: reversible

irreversible

often simultaneously coagulation (due to aggregation of molecules)





physical factors

temperature, pressure, ultrasound, irradiation

chemical factors

salts, pH , surface active compounds

consequences

- **better access to digestive enzymes**
- **denaturation of antinutritive factors, toxic compounds**

  (inhibitors of proteases, amylases, lectins, decomposition of toxic compounds)
- **inhibition of undesirable enzymes and microorganisms**

meat, meat products, poultry, fish

4 main types of tissue

- **epithels**
- **connective tissue**
- **muscle tissue**
- **neural**

meat **definition**

parts of warm-blooded animals in natural or processed state

mainly skeletal muscle tissue

components (pork meat)

- water (30 – 72%)
- proteins (9 – 20%; average 15%)
- fat (1.5% and more)
- minerals (~1%)
- vitamins
- glycogen (1 – 2%)
- sugar phosphates and free sugars (0.05 – 0.2%)
- free amino acids (0.1 – 0.3%)
- taurine(0.02-0.1%)
- quaternary ammonium compounds
- choline (0.02-0.06%)
- lactic acid and other acids (0.2 – 0.8%)
- purines and pyrimidines (0.1 – 0.25%)

muscle proteins

proteins	%
myofibrilar proteins	60.5
myosine	29
actine	13
connectine	3.7
tropomyosine	3.2
troponine (C, I, T)	3.2
actinine (α-, β-, γ-)	2.6
myomesine, desmine etc.	5.8
sarkoplasmatic proteins	29.0
enzymes	24.5
myoglobine	1.1
hemoglobine etc. (extracelular proteins)	3.3
structural proteins	10.5
collagen	5.2
elastine	0.3
mitochondrial proteins	5.0

reactions post mortem

- ATP - produced by anaerobic glycolysis from glycogen (until present)

  → lactic acid → decrease of pH from 6.8 to < 5.8

- inhibition of glycolytic enzymes

- Ca^{2+} still evokes the reaction of actin with myosin

  ATP is missing → postmortem stiffening

  (*rigor mortis*)

cattle: 10 - 24 h

pig: 4 - 18 h

chicken: 2 - 4 h

influence on meat quality

- after slaughter dry, good water-binding
- in rigor mortis moist, firm, little water-binding

ripening of meat

- splitting of actomyosin by endogenous proteases (mostly cathepsins)
- splitting of collagen by collagenases

  moderately bound water

defects of meat

- DFD (dry-firm-dark) a DCB (dry-cutting-beef)

  dark, high water holding capacity, low stability, remotion of lactic acid during blooding, pH~ 6

- PSE (pale-soft-exudative)

  pale, low water holding capacity, grey-green surface, increased glycolysis stimulated by hormones, pH~ 5.6

changing during processing

~35° C association of sarcoplasmatic proteins, decrease of water holding capacity, increase of firmness

~45° C: visible changes, shrinkage =denaturation of myosin

~50-55° C: denaturation of actomyosin

~55-65° C: denaturation of sarcoplasmatic proteins,

→ association, gel structure

~60-65° C: changes of collagen conformation (shrinkage by 1/3 - 1/4)

~80° C: oxidation SH-groups

~90° C: gelatinisation of collagen (loosening of tropocollagenic fibres, sol, gelatine)

~100 ° C: elimination NH_3, H_2S, other compounds

→ aroma formation, changes in colour

milk and milk products

content of nutrients in milk

component	%			
	cow	goat	ovine (sheep)	human
total proteins	3.2	3.2	4.6	0.9
caseins	2.6	2.6	3.9	0.4
whey proteins	0.6	0.6	0.7	0.5
fats	3.9	4.5	7.2	4.5
saccharides	4.6	4.3	4.8	7.1
minerals	0.7	0.8	0.9	0.2

complex disperse system ⇒ **white colour**

globular whey proteins: colloidal disperse

casein molecules: micellar disperse

fat: fat globules (microsomes, 0.1-10 µm): emulsion

particles of lipoproteins: colloidal suspension

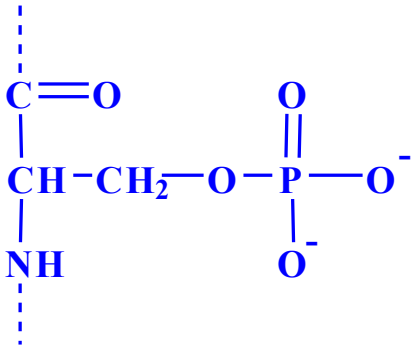
low molecular weight compounds (lactose, amino acids, minerals, vitamins): solution

proteins	%	content in $g.dm^{-3}$
caseins total	80	25.6
α_s-casein	42	13.4
β-casein	25	8.0
γ-casein	4	1.3
κ-casein	9	9.2
whey proteins total	20	6.4
α-lactalbumin	4	1.3
serum albumin	1	0.3
β-lactoglobulin	9	2.9
imunoglobulins	2	0.6
polypeptides (proteoses, peptones)	4	1.3

caseins

α-caseins = phosphoproteins, α_{S1}, α_{S2} (4 genetic modifications, B)

β-caseins = phosphoproteins



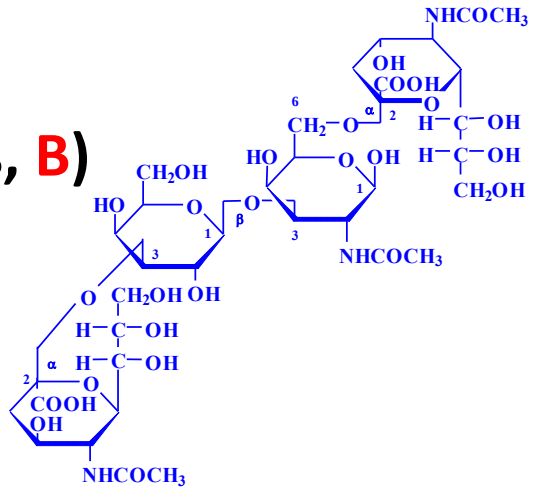
phosphoserine

γ-caseins = products of β-caseins degradation

κ-caseins = glycoproteins (2 genetic modifications, B)

saccharides = tetra-, tri-, di-, mono-

GalNAc, Gal, NeuAc, bound on Thr



caseins

α_S-, β-, κ-caseins aggregated into micelles and submicell(> 5 °C)

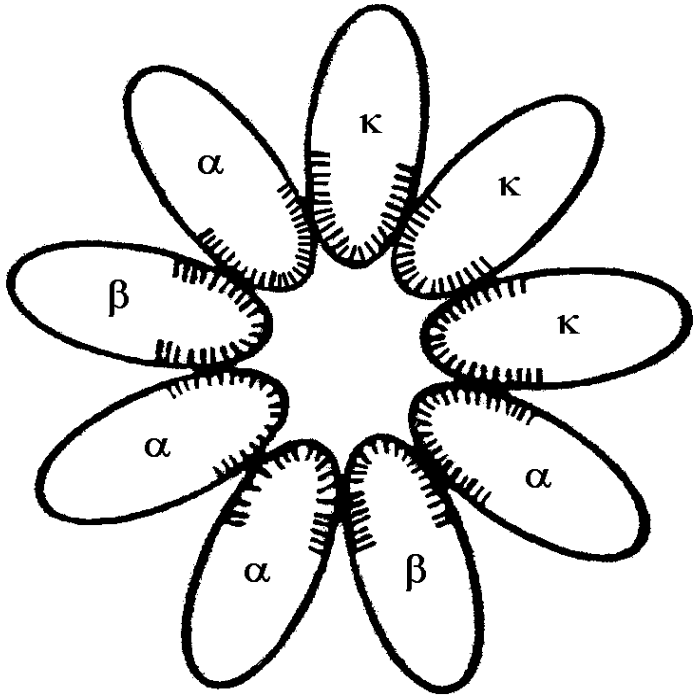
molecules of caseins → submicells → micells

- submicells form rotational ellipsoid (25-30 molecules)

nonpolar parts into centre (hydrofobic interactions)

polar parts (phosphoserine) α_S- a β-caseins interact with ions Ca^{2+}, oligosaccharides κ-caseins with water

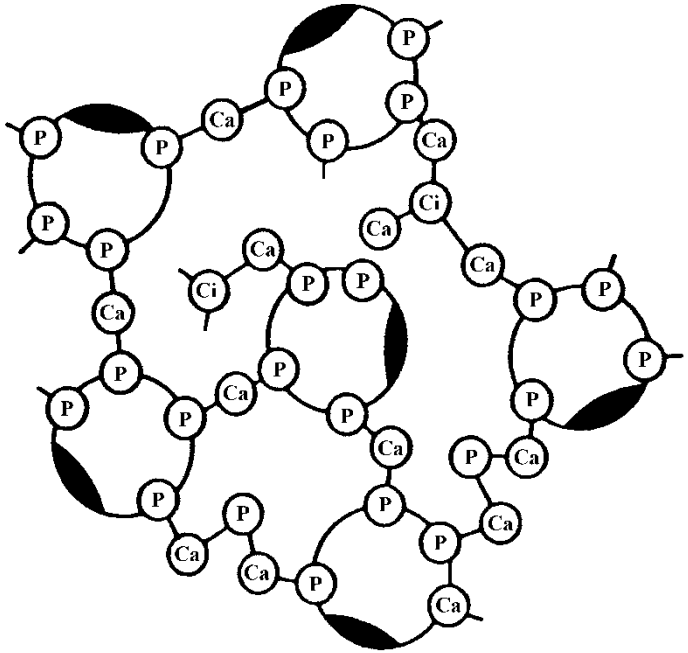
cross section of submicell
(dashed hydrophobic part)



- **from submicells formed micells through phosphate (phosphoserine) groups of α_s-caseins, β-caseins and Ca^{2+} ions, directly or through free phosphates and citrates**

micells of cow´s milk = 20000 molecules caseins

average = 50-300 nm, sum = 1.10^{12} in 1000 ml



42-50% α-caseins , 30% β-caseins, 15-26% κ-caseins

changes during storage and heat processing

- agglomeration of fat globules in fresh milk, ~ macroglobulin
- whey proteins thermolabile, denaturation, caseins thermostable, practically do not denaturate

pasteurisation

72-74 °C (20-40 s): denatured appr. 50-90% whey proteins
most of enzymes are inhibited

> 75 °C: reduction of disufide bonds
H_2S elimination (β -lactoglobulin)
sulfide, disulfide, taste changes (Met)
thiamine degradation
lactones and methylketones formation

sterilization (UHT)

140 °C (4 s) denatures 100% of proteins
reaction of whey proteins with lactose
loss of Lys (Maillard´s reaction)

Lys losses (Maillard reaction)

aroma compounds

fresh and pasteurised milk

cca 400 aroma compounds (1-100 mg/kg)

precipitation and proteolysis of caseins

milk: pH 6.5-6.75

precipitation of caseins: pH 4.6 **(due to microorganisms)**

hard cheese

- **microorganisms (lactic acid), acidification (pH 5.5)**

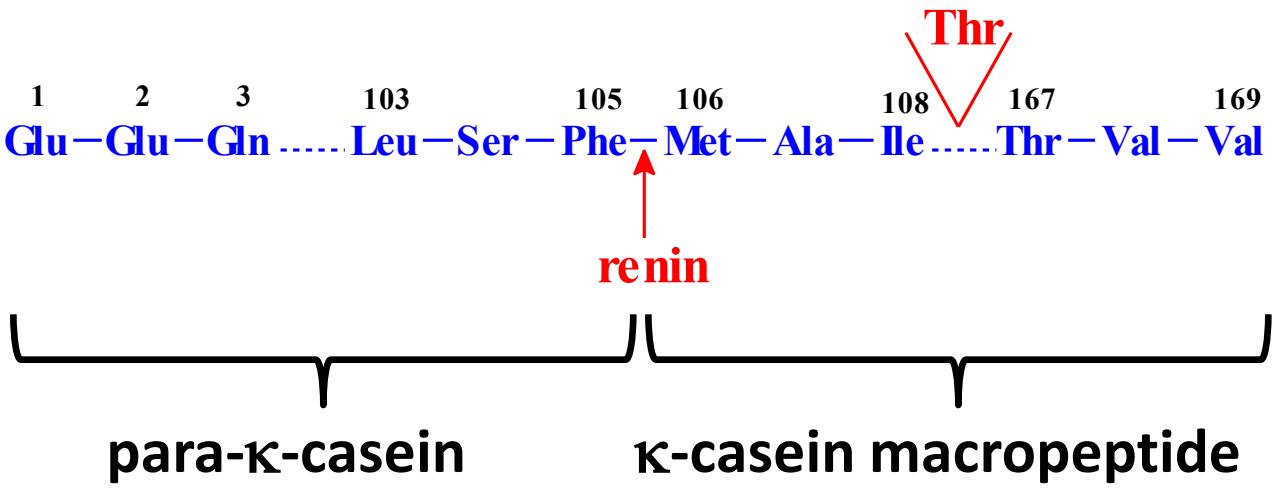
  proteolytic enzym rennin (chymosin)

  specific hydrolysis of κ-casein on 2 parts:

  ***para*-κ-casein = hydrofobic part κ-casein (part of micell)**

  κ-caseinmacropeptide = hydrophilic part

κ-casein



stay as part of micell

go to whey

coagulation (storage→ firmness, whey centrifugation, addition of NaCl, ripening (at Emmental cheese conversion of lactic acid → propionic acid + CO_2), proteolysis, lipolysis → hard cheese

soft cheese, youghurt

precipitation, low pH (fermentation of lactose, lactic acid), partial coagulation of caseins, micells association (gel structure)

other products

unsoluble acid casein

sweet casein (precipitated by enzymes)

caseinates (soluble:Na, K, NH4; dispersable: Ca, Mg)

soluble coprecipitates

whey

eggs

white egg protein 53%, yolk 47%

nutrient content in chicken eggs

component	%		
	shell	white	yolk
proteins total	3.3 [1)]	10.6	16.6
fat	-	0.03	32.6
saccharides	-	0.9	1.0
minerals	95.1 [2)]	0.6	1.1
water	1.6	87.9	48.7
% of total weight	10.3	56.9	32.8

[1)] complex of proteins with mucopolysaccharides in the ratio 50:1

[2)] $CaCO_3$ with small amount of $MgCO_3$ and phosphates

composition of the protein chicken white and yolk of eggs

proteins	%	content in g.kg $^{-1}$
proteins of egg white total	100	106
ovalbumin	54	57
konalbumin (ovotransferrin)	12	13
ovomucoid	11	12
lysozym (globulin G_1)	3.5	4
globulin G_2	4	4
globulin G_3	4	4
ovomucin	1.5	2
ovomacroglobulin	0.5	< 1
ovoinhibitor	0.1	< 1
avidin	0.1	< 1
proteins of yolk total	100	166
lipovitellin (HDL [1)])	36	60
fosvitin	13	21
LDL [1)]	1	2
lipovitellenin (LDL [1)])	16	26
livetin	27	45

1) HDL = *High Density Lipoprotein*
2) LDL = *Low Density Lipoprotein*

proteins of egg white

~ 40 proteins (globulins, glycoproteins and phosphoproteins)

- **enzymes (lysozyme, activity of N-acetylmuramides, murein, cell walls of bacteria)**
- **protein components of enzymes (flavoprotein/riboflavin, avidin binds biotin)**
- **proteases inhibitors (ovomucoid, ovoinhibitor)**

consequences

- **viscosity and gel consistence of egg white: ovomucoid a ovomucin**
- **foam stabilisation of whipped egg white: ovoglobulins G_2 a G_3**
- **antimicrobial effects: lysozyme (ovoglobulin G_1)**
- **antinutritive effect: avidin**

egg yolk proteins

emulsion of fat in water
1/3 = proteins
2/3 = lipids

glyco-, lipo-, glycophospho- a glycophospholipoproteins

granules: lipovitellin a phosvitin
plasma: lipovitellenin a livetin

changes during storage and processing

- **partial denaturation of egg white protein during whipping**
- **heat denaturation**
- **57°C: beginning**
  60-65°C most of proteins denaturate (ovomucoid does not)
  65-70°C most of yolk proteins denaturate (phosvitin not)

cereals and pseudocereals

basic cereals chemical composition

cereal	water	proteins	lipids	starch	minerals
wheat	13.2	11.7	2.2	59.2	1.5
rye	13.7	11.6	1.7	52.4	1.9
barley	11.7	10.6	2.1	52.2	2.3
oat	13.0	12.6	5.7	40.1	2.9
rice	13.1	7.4	2.4	70.4	1.2
corn	12.5	9.2	3.8	62.6	1.3

wheat proteins

flour: 7-13 (up to 15) % proteins

- 15% albumins (soluble in water) leukosin
- 7% globulins (0.4 M-NaCl) edestin
- 33% prolamins (70% ethanol) gliadin
- 46% glutelins (rest) glutenin

strong flour = bread flour (12-14%)

(dough elastic, firm, essential intensive mixing during preparation, hinder CO_2, air, voluminous products)

weak flour = production of biscuits, pastries (< 10%)

dough - with water starch based adhesive and viscoelastic material - gluten

2/3 water + 1/3 hydrated glutelins (viscoelastic properties, 3D-grid)

and gliadins (elasticity)

products without gluten: limit < 100 mg gliadins/kg (dry matter)

alergenic celiac disease (0.05% children in Europe)

changes of epithelial gut cells, improper resorption of nutrients

responsible: prolamine fraction of wheat, rye, barley

sequences: Pro-Ser-Gln-Gln and Gln-Gln-Gln-Pro

utilisation of nontraditional protein sources

texturised plant proteins, protein rich products