KEEMIA- JA MATERJALITEHNOLOOGIA TEADUSKOND
TOIDUAINETE INSTITUUT
TEADUS- JA ARENDUSTEGEVUSE AASTAARUANNE 2012

1. Instituudi struktuur

Toiduainete instituut, Department of Food Processing
Instituudi direktor Raivo Vokk

- Toiduteaduse õppetool, Chair of Food Science, RaivoVokk
- Toidutehnoloogia õppetool, Chair of Food Technology, Toomas Paalme

2. Instituudi teadus- ja arendustegevuse (edaspidi T&A) iseloomustus

(NB! punktid 2.1- 2.6 täidab struktuuriüksus)

2.1 struktuuriüksuse koosseisu kuuluvate uurimisgruppide

2.1.1 teadustöö kirjeldus *(inglise keeles*);

Foodomics, a discipline that studies Food and Nutrition domains through the application of advanced omics technologies to improve the consumer's well-being, health, and confidence was combined with physical and sensory analysis. The studies were concentrated on the food quality related to processing of issues of rye sourdough bread, ice cream, yeast, lactic acid bacteria, spices and spice cured Baltic sprats etc. processing. The short overview of respective activities is given below.

Yeast group (*Dr. Ildar Nisamedtinov*). Yeast biomass contains a wide range of compounds with potentially bioactive/functional characteristics, which is why they are valued as ingredients for various food- and biotechnological applications. The major objectives in this project were to develop a comprehensive toolset for identification and quantification of functional compounds in yeast fractions, elucidate the mechanisms related to their functionality and develop the process techniques which enable to increase the concentration of these functional compounds in yeast fractions. The two major groups of bioactive compounds under study were the B-complex vitamins and small MW peptides.

Systems biology (*Dr. Kaarel Adamberg*). Development of systems biology methods (continuous cultivation, omics and modelling methods) to design more yield-efficient media and growth conditions for lactic acid bacteria and other microbial strains for biotechnological processes.

Food technology and flavor (*Dr. Katrin Laos*). Ice structuring proteins were shown to be effective in inhibition of ice re-crystallization in ice cream. *Flavor* (taste and aroma) is the most important feature of food products and the key for repeating purchase decisions. Therefore, the flavor management in combination with science-based food design is increasingly appealing to food industry. Freezing followed by thawing of fish prior to ripening of spice-cured sprats decreased the activity of enzymatic and microbiological processes, which influenced product main sensory quality attributes: hardness, general spiciness, and sourness. The systematic approach relating the consumer's expectations, preferences to the measurable flavor characteristics of products, raw

materials and processes on molecular level was introduced and used in studies of Baltic sprat ripening, formation of rye sourdough bread and Kama aroma.

Rye sourdough breads (*Dr. Inga Sarand***).** A desired and reproducible composition of the sourdough microflora is essential to produce the sourdough bread with a constant quality. The sourdoughs of different bakeries were compared to understand the effect of variations in technological conditions of sour dough cycle on microbial composition and to evaluate possible mechanisms of adaptation of the selected LAB. The establishment of microbial consortia in sourdoughs was studied in spontaneous rye sourdoughs during 56-day long propagation at different temperatures. Both classical culture dependent- and independent methods but also high-throughput sequencing were used in the work.

Pre- and probiotic research (*Dr. Signe Adamberg*). The studies on stability and dynamics of mixed cultures and natural microbial consortia were introduced 2011 – 2012 as an addition to pure culture studies in an upper gastrointestinal tract model. A novel algorithm to mimic environmental conditions of the large intestine with non-digestible prebiotic oligosaccharides as energy and carbon source were used in the GITS model. The following items were studied during 2012: 1) dynamics and stability of natural microbial consortia in fermented dairy products and cheese under conditions of the upper gastrointestinal tract; 2) metabolism of prebiotic oligosaccharides in lactobacilli and bifidobacteria, pure and mixed cultures under simulated gastrointestinal conditions; 3) novel oligosaccharides as putative substrates for gut bacteria.

2.1.2 aruandeaastal saadud tähtsamad teadustulemused *(inglise keeles)*.

Development of the method for determination of B-complex vitamins. A high throughput method for the simultaneous determination of the activities of vitamins B1, B2, B3, B5 and B6 using LC-MS and stable isotope internal standards was developed. A user-friendly assay kit, consist of isotope labeled vitamer standards and hydrolytic enzymes for liberation of B-complex vitamers from food matrix and for their conversion into the same forms as the internal standards was developed (PCT application submitted). Various certified reference materials were studied with the assay kit. The results suggest that the method is applicable for quantification of most B-complex vitamers. The method has been internally validated and used to analyze B-complex vitamins in yeast derivatives, rye sourdough bread, corn snacks, jams and sea buckthorn.

Demonstration of preference of adsorbtion of amino acids from peptides. The consumption patterns and preferences (in terms of amino acid content) of peptides of a *Lactococcus lactis* IL1403 culture using a complete synthetic medium supplemented with ^{15}N-labeled yeast hydrolysate was carried out. The results showed a sharp increase of ^{15}N-labeled amino acid content in biomass followed by a decrease during the later stages of the experiment. The latter was accompanied by a slight decrease of the specific growth rate of bacteria. On contrary to our expectations, the ^{15}N enrichment patterns of both essential and non-essential amino acids for *L. lactis* were similar suggesting that the incorporation of a particular amino acid is dependent on its availability in a readily assimilated (peptidic) form and not the organism's auxotrophy for it.

Demonstration of variability of sourdough consortia in different bakeries. Microbial consortia of the sourdoughs differed remarkably in the four bakeries. In several cases microbial composition of samples taken from the same bakeries at different times also varied. The studies showed that commercial starters, except those derived from same technological cycle, may not be competitive in the sourdough environment as they are replaced by more competitive species originating from raw materials and/or bakery environment. The instability of LAB population was followed by instability of yeast population. Succession of different yeast species were observed when determined by

different methods. New culture independent methods (DGGE, amplification of internal transcribed spacer, 28S rRNA pyrosequencing) were tested for evaluation of yeast populations. Surprisingly, highly recommended pyrosequencing correlated only partly with the results obtained by two other methods.

SILAC labelled method for determination of protein half-lives applied to calculate non-productive growth expenditures were introduced for cultures of *Lactococcus lactis* and *Escherichia coli*. Co-variation based statistical method was developed for the analysis of omics data and regulation coefficients between mRNA protein and flux were determined for *Lactococcus lactis*.

Development of multistrain (multispecies) probiotics containing multiple prebiotic substrates capable of synergistic growth after the acid and bile stress in upper gastrointestinal tract – cooperation with Lund University (paper submitted)

The inhibition of ice re-crystallization using ice structuring proteins. Ice formation during freezing affects the texture and sensory properties of the product. Initial results show that commercial ice structuring proteins from fish are promising additives to retard ice re-crystallization in ice cream: Concentrations as low as 0.01% ISP inhibit re-crystallization.

The influence of freezing rate and storage temperature on lactose crystallization in frozen systems. Our studies using a sucrose and lactose ice cream model show that freezing rate and storage temperature have strong effects on lactose crystallization. If the lactose content is high, freezing rate does not affect the results (>10% w/w). A difference is seen at low lactose concentrations, where quick freezing slowed the formation of lactose crystals. Low storage temperatures also inhibit the formation of lactose crystals.

Demonstrating the mechanism of bread staling. We propose that re-crystallization of starch amylopectin and amylose in a hydrated form causes a decrease in the availability of water during storage and the development of a dry mouth-feel during consumption. In rye sourdough bread this is less pronounced than in wheat bread because of matrix effects. Results of a descriptive sensory analysis of rye sourdough bread show that in comparison with wheat bread rye sourdough bread higher stability of quality during storage. This can be explained partly by a higher content of water in rye breads due to a higher water holding capacity, and partially by the influence of rye flour pentosans.

Spice-cured sprats ripening, sensory parameters development and quality indicators. A descriptive sensory analysis method for Baltic sprat was created and results of this survey are related to compositional analysis. Baltic sprats in different catches during year have distinct sensory properties, which can influence product quality. The most important attributes which set apart spice-cured sprats via ripening time were: hardness, humidity, general spiciness, sour taste, and rancid flavour. A comparative acceptance study of spice-cured sprat products in Estonia and Thailand shows that products scored lower in the new market.

2.2 Uurimisgrupi kuni 5 olulisemat publikatsiooni läinud aastal.

Arike, L.; Valgepea, K.; Peil, L.; Nahku, R.; Adamberg, K.; Vilu, R. (2012). Comparison and applications of label-free absolute proteome quantification methods on *Escherichia coli*. Journal of Proteomics, 75(17), 5437 - 5448.

Adamberg, K.; Seiman, A.; Vilu, R. (2012). Increased Biomass Yield of Lactococcus lactis by Reduced Overconsumption of Amino Acids and Increased Catalytic Activities of Enzymes. PLoS

ONE, 7(10), e48223

Orumets K, Kevvai K, **Nisamedtinov** I, Tamm T, **Paalme T.** (2012) YAP1 over-expression in *Saccharomyces cerevisiae* enhances glutathione accumulation at its biosynthesis and substrate availability levels. Biotechnol J. 2012 Apr; 7(4):566-8. doi: 10.1002/biot.201100363. Epub 2011 Nov 21.

Mihhalevski, A; Nisamedtinov, I; Hälvin, K; Ošeka, A; Paalme, T. Stability of B-complex vitamins and dietary fibre during rye sourdough bread production. Journal of Cereal Science, 2013 57(1), 30 - 38.

Hälvin K, Paalme T, Nisamedtinov I. METHOD AND ANALYTICAL KIT FOR SIMULTANEOUS QUANTIFICATION OF B-COMPLEX VITAMIN CONTENT IN FOOD. WO/2013/010553. **International Application No.:** PCT/EE2012/000004

2.3 Loetelu struktuuriüksuse töötajate rahvusvahelistest tunnustustustest.

2.4 Loetelu struktuuriüksuse töötajatest, kes on välisakadeemiate või muude oluliste T&A-ga seotud välisorganisatsioonide liikmed.

Nisamedtinov, Ildar: member of scientific committee of European Association for Specialty Yeast Products

2.5 Aruandeaasta tähtsamad T&A finantseerimise allikad.

Sihtfinantseerimine T090 ja SA Eesti Teadusfond grandid

2.6 Soovi korral lisada aruandeaastal saadud T&A-ga seotud tunnustusi (va punktis 2.3 toodud tunnustused), ülevaate teaduskorralduslikust tegevusest, teadlasmobiilsusest ning anda hinnang oma teadustulemustele.

2.7 Instituudi teadus- ja arendustegevuse teemade ja projektide nimetused *(Eesti Teadusinfosüsteemi, edaspidi ETIS, andmetel)*

- Haridus- ja Teadusministeerium

– sihtfinantseeritavad teemad:
T090, Toidu süsteemibioloogia ja füüsika, Paalme Toomas (2008 – 2013)

– baasfinantseerimise toetusfondist rahastatud projektid (sh TTÜ tippkeskused):
– riiklikud programmid:

- Teiste ministeeriumide poolt rahastatavad riiklikud programmid:

- Uurija-professori rahastamine:

- SA Eesti Teadusfond/Eesti Teadusagentuur

– grandid:
ETF8165, *Escherichia coli* atsetaadi metabolismi kvantitatiivne kirjeldamine bioprotsesside optimeerimiseks, kasutades aktselerostaatseid paralleelkultivatsioonisüsteeme , Adamberg Kaarel (2010 – 2013)

ETF9189, B-kompleks vitamiinide metaboloomi analüüs toidu tootmise ahelas, Paalme Toomas (2012 – 2015)

ETF9417, Piimhappebakterite ja pärmide mitmekesisus ja stabiilsus rukkitaigna uuendamisel, Sarand Inga (2012 – 2015)

– ühisgrandid välisriigiga:
– järeldoktorite grandid (SA ETF ja Mobilitas):
– tippteadlase grandid (Mobilitas):

- Ettevõtluse Arendamise SA

– eeluuringud:
– arendustoetused:

- SA Archimedesega sõlmitud lepingud

– infrastruktuur (nn „mini-infra“, „asutuse infra“):
– Eesti tippkeskused:
– riiklikud programmid:
–

AR12170, Biotehnoloogia, Design and application of novel levansucrase catalysts for the production of functional food ingredients (Functional Food Ingredients), Signe Adamberg (1.10.2012 - 31.08.2015)

– muud T&A lepingud:

- SA Keskkonnainvesteeringute Keskusega sõlmitud lepingud:
- Siseriiklikud lepingud:
- EL Raamprogrammi projektid:
- Välisriiklikud lepingud:

2.8 Struktuuriüksuse töötajate poolt avaldatud eelretsenseeritavad teaduspublikatsioonid *(ETIS klassifikaatori alusel 1.1, 1.2, 1.3, 2.1, 2.2, 3.1, 3.2, 3.3, 4.1 ja 5.1).*

1.1

Vokk, R.; Kutsar, L.; Veskus, T.; Quantick, P. (2012). Quantitative microbial risk assessment in Estonian dairies. Journal of Environmental Science and Health Part A-Toxic/Hazardous Substances & Environmental Engineering , 130 [ilmumas]

Timberg, L.; Koppel, K.; Kuldjärv, R.; Paalme, T. (2012). Spice-cured sprat ripening and sensory properties development. Journal of Aquatic Food Product Technology, xx - xxx. [ilmumas]

Timberg, L.; Koppel, K.; Kuldjärv, R.; Chambers IV, E, Soontrunnarudrunggsri, A; Suvonsichon, S; Paalme T. (2012) Seasoned sprat product acceptance in Thailand and Estonia, Journal of Aquatic Food Product Technology xx – xxx [ilmumas]

Hälvin, K; Paalme, T; Nisamedtinov, I (2012). Comparison of different extraction methods for simultaneous determination of B-complex vitamins in nutritional yeast using LC/MS-TOF and stable isotope dilution assay. Analytical and Bioanalytical Chemistry, xxx - xxx. [ilmumas]

Mihhalevski, A; Nisamedtinov, I; Hälvin, K; Ošeka, A; Paalme, T (2013). Stability of B-complex vitamins and dietary fibre during rye sourdough bread production. Journal of Cereal Science, 57(1), 30 – 38

Viiard, E.; Mihhalevski, A.; Rühka, T.; Paalme, T.; Sarand, I. (2013). Evaluation of the microbial community in industrial rye sourdough upon continuous back-slopping propagation revealed Lactobacillus helveticus as the dominant species. Journal of Applied Microbiology, 114(2), 404 - 412.

Arike, L.; Valgepea, K.; Peil, L.; Nahku, R.; Adamberg, K.; Vilu, R. (2012). Comparison and applications of label-free absolute proteome quantification methods on Escherichia coli. Journal of Proteomics, 75(17), 5437 - 5448.

Adamberg, K.; Seiman, A.; Vilu, R. (2012). Increased Biomass Yield of Lactococcus lactis by Reduced Overconsumption of Amino Acids and Increased Catalytic Activities of Enzymes. PLoS ONE, 7(10), e48223

Mihhalevski, Anna; Heinmaa, Ivo; Traksmaa, Rainer; Pehk, Tõnis; Mere, Arvo; Paalme, Toomas (2012). Structural changes of starch during baking and staling of rye bread. Journal of Agricultural and Food Chemistry, 60(34), 8492 - 8500.

Sumeri, I.; Adamberg, S.; Uusna, R.; Sarand, I.; Paalme, T. (2012). Survival of cheese bacteria in a gastrointestinal tract simulator. International Dairy Journal, 25, 36-41

Orumets, K; Kevvai, K; Nisamedtinov, I; Paalme, T (2012): YAP1 over-expression in *Saccharomyces cerevisiae* enhances glutathione accumulation at its biosynthesis and substrate availability levels BIOTECHNOLOGY JOURNAL **7** (**4)** Pages: **566-568**

3.2

Pitsi, Tagli; Oja, Leila (2012). 2010/2011õppeaasta koolipuuviljakava hindamine. Raport.

2.9 Struktuuriüksuses kaitstud doktoriväitekirjade loetelu *(NB! struktuuriüksus lisab struktuuriüksuse töötaja juhendamisel mujal kaitstud doktoriväitekirjade loetelu)*

Kristel Kaseleht, toiduainete instituut
Teema*: Identification of Aroma Compounds in Food Using SPME-GC/MS and GC-Olfactometry* (Aroomiühendite määramine toidus kasutades SPME-GC/MS ja GC-Olfaktomeetriat)
Juhendajad: prof Toomas Paalme ja prof Erich Leitner
Kaitses: 25.05.2012
Omistatud kraad: filosoofiadoktor (keemia- ja materjalitehnoloogia)

Kerti Orumets, toiduainete instituut
Teema: *Molecular Mechanisms Controlling Intracellular Glutathione Levels in Baker's Yeast Saccharomyces Cerevisiae and a Random Mutagenized Glutathione Over-Accumulating Isolate* (Rakusisesed glutatiooni taset kontrollivad molekulaarsed mehhanismid pagaripärmis Saccharomyces cerevisiae ja selle juhuslikul mutageneesil saadud glutatiooni üleakumuleerivas isolaadis)
Juhendaja: dotsent Ildar Nisamedtinov
Kaitses: 16.11.2012
Omistatud kraad: filosoofiadoktor (keemia- ja materjalitehnoloogia)

Liisa Arike, toiduainete instituut
Teema: *Quantitative Proteomics of Escherichia Coli: from Relative to Absolute Scale* (Kvantitatiivne Escherichia Coli proteoomika: relatiivsetest numbritest absoluutsete kogusteni)
Juhendajad: vanemteadur Kaarel Adamberg, prof Raivo Vilu ja vanemteadur Lauri Peil
Kaitses: 22.11.2012
Omistatud kraad: filosoofiadoktor (keemia- ja materjalitehnoloogia)

Anna Mihhalevski, toiduainete instituut
Teema: *Rye Sourdough Fermentation and Bread Stability* (Rukkitaigna hapendamine ja leiva vananemine)
Juhendaja: prof Toomas Paalme
Kaitses: 29.11.2012
Omistatud kraad: filosoofiadoktor (keemia- ja materjalitehnoloogia)

Loreida Timberg, toiduainete instituut
Teema: *Spice-Cured Sprats Ripening, Sensory Parameters Development and Quality Indicators* (Vürtsikilu valmimine, sensoorsete omaduste kujunemine ja kvaliteediindikaatorid)
Juhendaja: prof Toomas Paalme
Kaitses: 7.12.2012
Omistatud kraad: filosoofiadoktor (keemia- ja materjalitehnoloogia)

David Schryer, küberneetika instituut,
Teema: Metabolic flux analysis of compartmentalized systems using dynamic isotopologue modelling,
Juhendajad: prof. Marko Vendelin, Dr. Pearu Peterson, prof. Toomas Paalme
Kaitses**:** 12.04.2012
Omistatud kraad: filosoofiadoktor (tehniline füüsika)

2.10 Struktuuriüksuses järeldoktorina T&A-s osalenud isikute loetelu *(ETIS-e kaudu esitatud taotluste alusel)*

2.11 Struktuuriüksuses loodud tööstusomandi loetelu

PCT/EE2012000004
Method and analytical kit for simultaneous quantification of B complex vitamiin content in food
Taotlus esitatud: 17.07.2012
Autorid: Kristel Hälvin, Toomas Paalme, Ildar Nisamedtinov
Omanikud: TTÜ, AS Toidu- ja Fermentatsioonitehnoloogia Arenduskeskus

3. Struktuuriüksuse infrastruktuuri uuendamise loetelu (summa

PV007441, Antioksüdatiivsete omaduste määramise seade , 28.08.2012 (26 040,00)
PV007245, Viskosimeeter 15.12.2012 (10281,94)