Map Milling

Measure and control of mycotoxins, pesticides and acrylamide in grain milling sector

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Project acronym: MAP-MILLING

Project title: MEASURE AND CONTROL OF MYCOTOXINS, PESTICIDES AND ACRYLAMIDE IN GRAIN MILLING SECTOR

Instrument: Collective Research Project

Thematic Priority: Specific SME Activities

DFinal. Final Activity Report

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Publishable executive summary

Grain milling industry has detected one of their main problems in the final product quality assurance, is the control of raw materials and the control of their toxicity: mycotoxins (ochratoxin A, aflatoxins…), pesticides and formation of acrylamide.

Nowadays, agro-food SMEs are forced to use very laborious methodologies, with long preparation phases and high investments. These methods are slow and hardly applicable "in situ" or in processing line. Thus, there is a clear need to develop new analytical tools able to detect contaminants in an easy, reliable and economic way. Current analyses in laboratories are long and relatively expensive, so it disturbs flour production and increases final process costs. Grain Milling sector is very interested in developing and disseminating a new measurement technology at competitive market price.

"MAP-MILLING" PROJECT APPROACH

The **global objective** of the project is to increase the food safety in the grain processing/milling industry according to the new regulation and market demands, requiring a high quality products and demonstrated reliability.

Map Milling
Measure and control of mycotoxins, pesticides and acrylamide in grain milling sector

The **operative objectives** are:

- To design and develop **reliable, fast and economic measurement systems** and **minimisation strategies** focussing on the most common **pollutants** (mycotoxins, pesticides and acrylamide) on the grain milling/processing industry, both **in field and in processing** plant.
- To establish a **simple guide of "best procedures" and strategies** for decreasing levels of pollutants in food stuffs within the mentioned agro-food sub-sector.
- To develop a **risk management tool** that allows, by means of an easy and economic procedure, **to ensure** final product (flour) **quality and**

safety, integrating all the results obtained (measurement system results, correlation with official results and best procedures) in a **friendly and easy-to-use software tool** to be used by the milling companies.

- To **exploit and disseminate** the project **results amongst the SMEs** within the sector at different levels to help incorporate these new technologies.
- To analyse **other agro-food subsectors for technology transfer** and to develop additional market applications of project results.

PARTNERSHIP TO SUCCESS

The consortium is leaded by two entrepreneurial associations from very important agrofood regions (Aragon-Spain and Lazio-Italy) with experience on previous projects under FP 5 (**CEZ** and **ROME**). There are also 4 agro-food associations from 4 different countries (Spain - **AIAA**, Italy - **UNIONA**, Germany – **PRO AGRO** and Estonia - **AEFI**) covering regional and national scopes. Finally, Associations group is completed with a European Association (**DEMETER**) that represents milling industries in 12 EU countries.

The 3 SMEs represent different conditions in grain milling sector (Spain - **HARITASA**, Italy - **ROMANA** and Germany - **MUHLE**).

Finally, the Researchers have experience in European projects and show a great complementarity of skills: **AZTI** (food technology, food safety, pesticides), **IGV** (cereal grain industry, mycotoxins, acrylamide), **INTECHNICA** (Quality/Environment/Risk Management Systems) and **ITA** (Industrial applications, software and electronics, technology transfer and implementation).

WORK PERFORMED

MAP-MILLING project has completed the whole workprogramme of the project and major achievements obtained are the following:

- Identification of needs and requirements (appraisal and demands) of the potential end-users (grain-milling industries) for detection of pollutants in cereal samples
- Setting up of technical specifications for the measurement systems and preliminary analysis of measurement techniques (reference analytical methods, matrix effect studies…)
- Final research results and recommendations for acrylamide minimization.

- Design and development of improved measurement systems in ELISA format for some target pesticides (2,4-D, DDT and chlorphyriphos-methyl) in cereal products.
- Comparison of current and new Mycotoxins measurement systems (NIR, ELISA, visual detection, dip sticks…) and advice for quick, low cost and reliable prediction of DON and ZEA.
- Development of a Risk Management System for grain milling SMEs with a catalogue of key pollutants and the development of simple and easy understandable guidelines.
- Design and development of a open-source Software Tool integrating the results of the MAP-MILLING project.
- Preparation of 3 national conferences on Food safety in agro-food sector.
- Preparation of 1 European conference on Food safety in agro-food sector.
- Holding 3 Training Workshops at Regional level on project results for grain-milling sector.

FURTHER INFORMATION

Further information can be provided by contacting the MAP-MILLING's project technical coordinator:

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http://www.mapmilling.net/

Section 1 – Project objectives and major achievements

The **global objective** of the project is to increase the food safety in the grain processing/milling industry according to the new regulation and market demands, requiring a high quality products and demonstrated reliability.

The **specific objectives** are:

- To know the percentages of most relevant pollutants in flour's processing chain, both in the field and at the industrial company.
- To define processes to decrease the concentration of these pollutants, both in the field and in the food processing plant.
- To assure flours' high quality and safety for European consumers.
- To diffuse, exploit and actively disseminate results at regional, national and European level.

The **operative objectives** are:

- To design and develop reliable, fast and economic measurement systems and minimisation strategies focussing on the most common pollutants (mycotoxins, pesticides and acrylamide) on the grain milling/processing industry. These measurement systems must be easily applicable not only for the products in field but also for the products in processing plant.
- To establish a simple guide of "best procedures" and strategies for decreasing levels of pollutants in food stuffs within the previously mentioned agro-food sub-sector.
- To develop a risk management tool that allows, by means of an easy and economic procedure, to ensure final product (flour) quality and safety.
- To integrate all the results obtained (measurement system results, correlation with official results, best procedures, and risk management) in a friendly and easy-to-use software tool to be used by the milling companies.
- To exploit and disseminate the project results amongst the SMEs within the sector at different levels: awareness, training and technological advice to help incorporate these new technologies.
- To analyse other agro-food subsectors that could be objective of a possible technology transfer and to develop additional market applications of project results.

The specific objectives of the third reporting period within MAP-MILLING's project are the following:

- To develop quick, cheap and reliable techniques for measurement of main pesticides.
- To recommend a reliable method for fast detection of fusaria damaged grain kernels.
- To analyse and minimise the causes of acrylamide formation.
- To develop an easy-to-use software tool that integrates the whole approach proposed and validate it in the SMEs Core Group.
- To ensure an optimal utilisation of research results of the project and the transfer of knowledge within other interested agro-food sectors
- To carry out a diffusion of the results of the project at European level, both towards the grain milling sector and agro-food sector in general
- To provide advanced training of managers and technical staff of grain milling sector SMEs and Industrial Associations/Groupings on the knowledge produced within the project

During the project duration (15/06/2004 – 14/06/2007), major achievements of the MAP-MILLING project on each WP were the following:

- Analysis of grain processing industry specific needs (WP1):
    - Identification of needs and requirements (appraisal and demands) of the potential end-users (grain-milling industries) for detection of pollutants in cereal samples
    - Setting up of technical specifications for the measurement systems
- Preliminary analysis of measurement techniques (WP2):
    - Evaluation of the effect of sample matrix components on measurement processes
    - Setting up the reference analytical method for determination of 2 pesticides (2,4-D and DDT) and for mycotoxins
    - Development of suitable sample treatment methods for the types of cereal samples selected and procedures of assay to be carried out in the next workpackage (reference analytical methods, matrix effect studies…).
- Design and development of improved measurement systems for Pesticides and Mycotoxins (WP3):
    - Full list of recommendations for reducing the formation of Acrylamide taking into account:
        - Knowlage about the content of free asparagines
        - Link between free asparagine in cereals and acrylamide in bakery or extrusion products
        - Concrete advice for choosing raw material
        - Influence of other ingredients than cereals in cereal products for acrylamide formation

  - Research/commercial sources of antibodies for selected pesticides have been obtained.
  - ELISA for 2,4-D, chlorpyrifos-methyl and DDT has been developed and optimized with standard solutions.
  - Final Development and Optimization of ELISA format for detection of 2,4-D in cereal products
  - Final Development and Optimization of ELISA format for detection of chlorphyrifos-methyl in cereal products.
  - Good correlation between ELISA results and chromatographic results for detection of 2,4-D and chlorphyrifos-methyl in cereal products.
  - Comparison of Dipstick assays as direct rapid method for analysis of Fusariumtoxins (immunochemical method): RIDA® QUICK DON and REVAL® DON STRIP test.
  - Full report detailing the tested measurement systems for Mycotoxins, with an overall comparison between the studied methods and recommendation of the most interesting ones.
- Development of a Risk Management System for grain milling SMEs (WP4):
  - Development of a catalogue of key pollutants in the Grain Milling Industry
  - Development of the corresponding risk management system (definition of critical control points)
  - Development of simple and easy understandable guidelines.
- Integration and validation of pollutants control (WP5):
  - System Requirement Specification
  - System Analysis Software
  - Analysis of existing tools in the market
  - System Design Software
  - Implementation of the System
- Exploitation (WP6):
  - Discussion and conclusions about the best Exploitation Strategy to ensure an optimal utilisation of research results of the project and the transfer of knowledge within other interested agro-food sectors.
- Dissemination (WP7) and Training (WP8):
  - Preparation of 3 of the National Conferences (Spain, Italy and Estonia), with the related Training Workshops for the technical and managerial staff of the SMEs interested in the project results.
  - Preparation of the European Conference organised by DEMETER, aimed at all the interested parties in Europe (agro-food IAGs, food safety organisms, research bodies, public institutions, policy makers…).

- Intensive development of dissemination activities at different levels.
- Final development of the Multimedia Training Platform in the MAP-MILLING Web Site.

Section 2 – Workpackage progress of the period

Following points describe the progress of the project activities against the planned objectives and workplan, broken down into Workpackges:

WORKPACKAGE 1 "ANALYSIS OF GRAIN PROCESSING INDUSTRY SPECIFIC NEEDS"

The main objective of this workpackage was to ascertain the needs of grain milling industries and agrofood associations to establish the desired performance characteristics of the devices to be developed for analysis of pesticides, mycotoxins and acrylamide generators in cereals samples.

Task 1.1: Definition of end-users' specific needs:

For the registration of the specific needs of end-users, not only the knowledge of project partners was taken into account, but an extensive questionnaire was prepared and distributed among the Grain Milling companies through the distribution lists of the 7 IAGs. The responsible for the preparation of this questionnaire was CEZ, as project coordinator. CEZ was supported by the 4 RTD Performers in formulation of questions for each technical part. The structure of the questionnaire has the following points:

- General description of the sector and specific characteristics (CEZ)
- Technical specifications. Pesticides (AZTI)
- Technical specifications. Mycotoxins (IGV)
- General Framework for Risk Management System (INTECHNICA and ITA)

The objective of the questionnaire is to widely collect, in the Grain Milling sector and especially in the "Core Group" of SMEs, the exact end-user requirements.

The questionnaire was sent through different ways (postal service, e-mail, fax…) to:

- Enterprises members of the IAGs.
- Other interested enterprises of the sector.
- Core Group of SMEs (HARITASA, ROMANA and MUHLE)

At the same time, the project was again verbally described in phone calls in order to solve questions arisen from the questionnaire and to ask for the written reply.

The questionnaire was translated to national languages (Spanish, Italian, German and Estonian) because most people in the companies speak only little English.

General topic of the questionnaire was considered very important and interesting by the companies, but many of them commented that some questions were too complicated, so it was another barrier for them to answer. Thanks to the work performed by the 7 IAGs involved, a relevant number of answers (18 questionnaires during the 1st reporting period) were collected and it was the basis for the 2 deliverables foreseen within this Worpackage:

- D2 "Operational report about main pollutants in grain milling industry"
- D3 "Technical report describing end-user requirements".

Full explanation on methodology and results of the questionnaire is given on D2 and D3.

Task 1.2: Establishment of technical specifications:

The objective of this task was to address practical issues pertaining to the cereals samples and contaminants under investigation (pesticides, mycotoxins and acrylamide). The main responsible partners were AZTI and IGV.

A critical review of the state-of-the-art was conducted in order to validate the needs of end-users of the developed technology. These needs were explicitly obtained in task 1.1 and properly contrasted with Core Group of SMEs (HARITASA, ROMANA and MUHLE).

These specifications cover two main aspects:

- End-user requirements: to establish the desired performance characteristics of the developed measurement techniques (sensitivity, reliability, speed, format, type of measurements, etc.). This stage was performed with the support of IAGs and SMEs that were making their contributions.
- Dynamic range: The required dynamic range of the selected contaminants measurement microsystems was established through reference to regulatory bodies, established literature and discussions with end-users.

Direct result of this task is D3 "Technical report describing end-user requirements", which draft document was presented by AZTI and IGV during 2nd progress meeting hold in Nürnberg (Germany) on 28th January 2005. The document was finally delivered on 11th February 2005.

Task 1.3: Definition of general framework for Risk Management System:

According to the main guidelines provided by the European institutions regarding the food safety and the risk management in the food chain, MAP-MILLING consortium has established the main framework for the development of the Risk Management System.

Also thanks to the questionnaire collected among European grain milling SMEs and to the work mainly performed by INTECHNICA and ITA, in close cooperation with Core Group SMEs (HARITASA, ROMANA and MUHLE), the definition of this general framework was included in D2 "Operational report about main pollutants in grain milling industry", delivered on 11th February 2005.

Draft version of this part was also presented by INTECHNICA during 2nd progress meeting hold in Nürnberg (Germany) on 28th January 2005.

Deviations from the project workprogramme

According to the original workplan, there was a delay of 3 months in the Workpackage 1, mainly due to the following circumstances:

- Summer vacation period did not allow the consortium to work at full capacity till September'04 (month 3)
- Elaboration, translation, distribution and answers collection of the questionnaire of needs of Grain Milling Sector took more time than expected due to the additional support needed by SMEs to fill in the form.

This delay is showed in the timetable of section 3. The delay only affected the beginning of other workpackages.

List of deliverables

Deliverables resulting form this workpackage (D2 "Operational report about main pollutants in grain milling industry" and D3 "Technical report describing end-user requirements") was ready on month 7, presented in draft version in 2nd Progress Meeting and finally delivered on 11th February 2005.

Del. No.	Deliverable name	Workpackage No.	Date due	Actual/Forecast delivery date	Estimated indicative person-months (*)	Used indicative person-months (*)	Lead contractor
D2	Operational report about main pollutants in grain milling industry.	1	4	7	0,25	0,35	CEZ
D3	Technical report describing end-user requirements	1	4	7	0,35	0,35	CEZ

List of milestones

General delay in the WP affected the time milestones were achieved as follows:

Milestone No.	Milestone name	Workpackage No.	Date due	Actual/Forecast delivery date	Lead contractor
M1	Selection of relevant contaminants in grain milling sector	1	4	7	CEZ
M2	Preliminary study into operational and technical requirements	1	4	7	CEZ

WORKPACKAGE 2 "PRELIMINARY ANALYSIS OF MEASUREMENT TECHNIQUES"

The main objective of this workpackage was to establish the reference analytical methods for determination of proposed pesticides and mycotoxins and to develop suitable sample treatment methods for the types of cereal samples selected. The procedure to study acrylamide formation was also fine tuned.

Task 2.1. Search of various kinds of quick measurement techniques:

The first task to be performed was the detailed analysis of the different quick measurement techniques that were studied in the determination of pesticides and mycotoxins (research of published works and available measurement apparatuses). The main techniques evaluated were:

- Enzyme-Linked Immunosorbent assay (ELISA) techniques – Pesticides and mycotoxins.
- Near Infra-Red (NIR) and Fourier Transform Infra-Red (FTIR) techniques – Mycotoxins.
- Visual identification of fusaria damaged grain kernels – Mycotoxins.

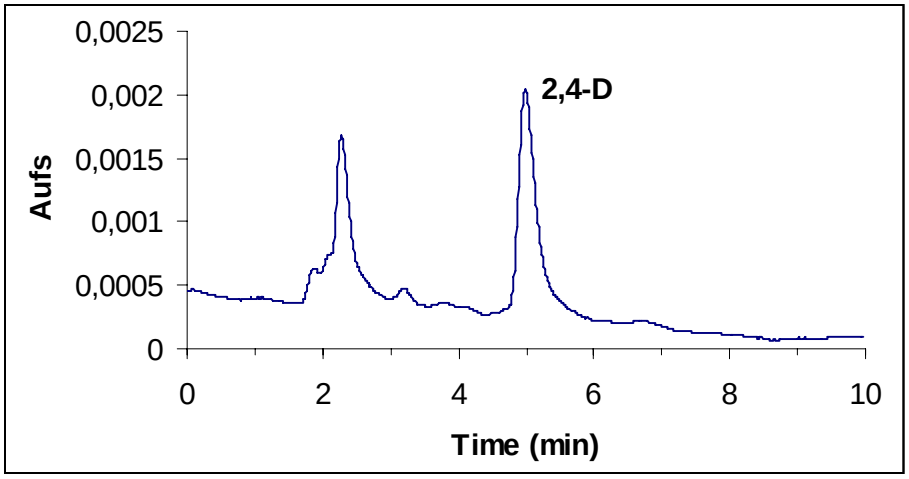
The work under this task was finished on time, being AZTI responsible of pesticides part and IGV of mycotoxins part.

Task 2.2. Establish of reference analytical methods:

The reference analytical methods were established by the two RTD performers in charge: AZTI and IGV. The aspects to be covered by this search were sample treatment and chromatographic method (HPLC).

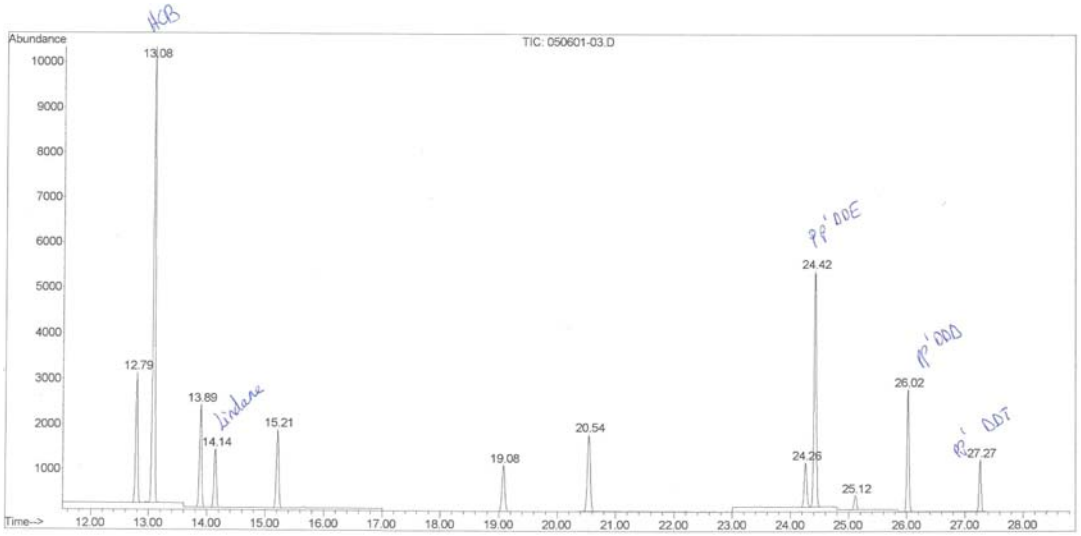
Once the targeted pesticides were selected under WP1, AZTI began to design reference analytical methods for their determination in cereal samples (wheat and flour). Two chromatographic methods were optimized from the beginning:

- HPLC method for the analysis of 2,4-D, chlorpyriphos-methyl and pirimiphos-methyl. With UV-Vis detection at different wavelengths the limits of detection are around 0.01 mg/L for all the compounds, below the maximum residue limit set by the EU. Next figure shows a chromatogram obtained from a standard solution of 2,4-D with 200 ppb with the best conditions achieved.



Standard solution of 2,4-D with 200 ppb detected by HPLC

- GC method capable of the determination of the isomers of DDT and their metabolites (DDD and DDE). With an ECD detector the limit of detection are below the EU MRL. With a mass spectrometer lower limits of detection below are achieved (0.005 mg/L). Figure below shows a chromatogram obtained from a standard solution with 50 ppb with the best conditions achieved.



Standard solution of DDT and their metabolites with 50 pp detected by GC_MS.

Good sensitivity was achieved with both methods as it is shown in the Deliverables.

The reference analytical methods for mycotoxin analysis (HPLC) were established by IGV. The analysis of mycotoxins (DON, zearalenone) was defined using spiked and/or certified samples. Information about quick

measurement techniques and publications on existing work was checked up. Main results were:

- Establishment of HPLC as reference method.
- First overview about advantages and disadvantages of the different measurement techniques

The ranking of measurement systems how they meet the claims is shown in the following table.

Method	advantage	disadvantage
HPLC	high correctness	high equipment costs time for measurement
ELISA	nearly quick	middle correctness middle price for equipment
FTIR	quick	high equipment costs low correctness
Visual detection	quick	low correctness

- First overview about test systems, offered on the market

Hersteller	Produktname
R-Biopharm AG	RIDASCREEN® Zearalenon
	RIDASCREEN® FAST Zearalenon
	RIDASCREEN® DON
	RIDASCREEN® FAST DON
Romer Labs	AgraQuant® Zearalenon
	AgraQuant® DON 48
Transia	Transia Plate Zearalenon
	Transia Plate DON
EURO-DIAGNOSTICA	ZEA
	DON
Tecnalab	Immunoscreen ZEA
	Immunoscreen DON „GOLD"
NEOGEN	Veratox® für Zearalenone
	Veratox DON 5/5
	Veratox DON HS

Task 2.3. Matrix effect studies:

AZTI was main responsible of this task, although IGV also supported them. The ideal ELISA is independent of sample matrix effects. In practice, this is unlikely and the effect of the sample on the performance of the assay must be investigated.

As wheat grain and flour are solid samples it is compulsory to use a solvent extraction in order to isolate the targeted pesticides with the minimum quantity of matrix components. After a literature search methanol was found to be the best solvent and it was selected to develop a proper sample treatment method.

Two different ways of extraction were evaluated: soaking the sample with a proper quantity of solvent and blending the sample with the solvent. The latter has achieved more extraction yield in the less amount of time, the matrix interferences being almost the same in both cases.

Task 2.4. Development of sample treatment methods:

Taking into account previous work performed, the project is establishing simple and low-cost sample preparation methods compatible with measurement procedures. Both AZTI and IGV worked on this task.

More specifically, a cleanup procedure of the extract obtained with methanol was found necessary in order to be able to perform a sensitive and selective determination of pesticides in cereal samples. In this sense, a number of simple methods can be employed to minimise any matrix effects of cereals samples. Adequate sample preparation by simple, rapid, low cost methods such as solid phase extraction, purge and trap or simple filtration may provide acceptable sample clean-up.

Solid phase extraction with C18 cartridges was tested obtaining good results. This technique removes a lot of matrix effects and it can also be adapted to consider analyte pre-concentration in order to achieve the necessary sensitivity.

After evaluating different extraction procedures (soaking, sonication, accelerated solvent extraction and blending) and solvents, blending with methanol has been found to be the best procedure for all the pesticides, obtaining good recoveries (around 90%) except for 2,4-D which has a strong affinity for the matrix. Even with exhaustive extraction procedures (with high temperatures and pressures, with caotropic agents) only around 50 % was extracted.

Main achieved results regarding pesticides are:

- Set up the reference analytical method (including a proper sample treatment method) for the determination of all target pesticides: 2,4-D, chlorpyriphos-methyl, pirimiphos-methyl and DDT (isomers and metabolites).
- Establish an appropriate sample treatment protocol for the extraction of the target pesticides from flour and grain that can be compatible with the immunoassays being developed

The "Report on preliminary research on existing works and techniques" was given as deliverable D5. Especially for utilization of FTIR-technique for investigation mycotoxin content current publications of other research group has been followed.

Dowell et al.[1] have measured Fusarium damaged wheat kernels by NIR. They have also tried to measure the content of DON and ergosterole in Fusarium damaged wheat kernels by NIR. For samples < 5 mg/kg the correlation even was not satisfying.

For trace analytics with regard to resolution and reproducability better suited than NIR seems to be Fourier transform IR spectrometry (FTIR).

Gordon et al.[2]-[3] were concerned with the development of a screening method for the detection of Aspergillus flavus fungi in maize by fourier transform infrared photoacoustic spectroscopy (FTIR-PAS) and by transient infrared spectroscopy (TIRS).

All newer publications on using FTIR for identification of moulds in cereals base on the works of Kos and co-workers. Kos et al.[4]-[5]-[6]-[7]-[8]-[9] were concerned with the development of a screening method for the detection of Fusarium fungi and DON on maize by Fourier transformation infrared spectroscopy with attenuated total reflection respective with photoacoustic technique.

[1] Dowell FE, Ram MS, Seitz LM (1999): Predicting scab, Vomitoxin, and ergosterol in single wheat kernels using near-infrared spectroscopy. Cereal Chem 76: 573-576
[2] Gordon SH, Schudy RB, Wheeler BC, Wicklow DT, Greene RV (1997): Identification of fourier transform infrared photoacoustic spectral features for detection of Aspergillus flavus infection in corn. Intern J Food Microbiol 35: 179-186
[3] Gordon SH, Jones RW, McClelland JF, Wicklow DT, Greene RV (1999): Transient infrared spectroscopy for detection of toxigenic fungi in corn: potential for on-line evaluation. J Agric Food Chem 47: 5267-5272
[4] Kos G, Lohninger H, Krska R (2005): Rapid Determination of Deoxynivalenol and Ergosterol in Wheat with Mid-Infrared Spectroscopy with Attenuated Total Reflection (IR-ATR) andChemometrics, Proceedings of the XI International IUPAC Symposium on Mycotoxins and Phycotoxins, May 17 - 21, 2004, Bethesda, MD, USA (submitted)
[5] Kos G, Lohninger H, Krska R, Griffiths P (2004): Rapid Screening of Fungal Infection on Corn using Mid-infrared Diffuse Reflection Spectroscopy (DRS) and Attenuated Total Reflection Spectroscopy (ATR): A Comparison Study, Analytical and Bioanalytical Chemistry 378: 159-166
[6] Kos G, Lohninger H, Krska R (2003): Development of a method for the determination of Fusarium fungi on corn using mid-infrared spectroscopy with attenuated total reflection and chemometrics. Anal Chem 75: 1211-1217
[7] Kos G, Lohninger H, Krska R (2003): Validation of chemometric models for the determination of deoxynivalenol on maize by mid-infrared spectroscopy. Mycotoxin Research 19: 149-153
[8] Kos G, Lohninger H, Krska R (2002): Classification of maize contaminated with Fusarium Graminearum using mid-infrared spectroscopy and chemometrics. Mycotoxin Research 18A: 104-108
[9] Kos G, Lohninger H, Krska R (2001): Using mid-infrared fourier-transform-spectroscopy with attenuated total reflexion (FT-IR/ATR)as a tool for the determination of Fusarium Graminearum on maize. Mycotoxin Research 17A: 102-106

We don't know newer publications on this subject from other working groups. FTIR was used for identification of moulds only – not for quantification of mycotoxins in µg/kg-range.

Recently in Norway FTIR was tested for characterization and identification of the mould penicillium Chrysogenum grown on agar and plaster boards[10]. Conclusion of the working group was: There are some difficulties, more work is needed.

This replies to the Mid-Term Review recommendation Nº 4 (To re-evaluate the suitability/capability of FTIR technique for determination of mycotoxins in grains and flours in line with latest development to be published in scientific literature). After many tests during WP3 on different alternatives (sample treatment, data analysis…) no good correlation as reported before could be obtained. That's why efforts were concentrated to more promising techniques. Additionally to planed tasks according to the proposal, IGV included in its working package the test of dipstick assays and investigated with more detail the correlation to foreign substances

Task 2.5. Sample collection:

This little task at the end of WP2 tries to ensure the best provision of test materials. Initially, the flour used by AZTI and IGV during the first experiments carried out was obtained directly from the market. The following experiments were carried out using products supplied by flour producing companies involved in the project (HARITASA, ROMANA and MUHLE). These necessary products were provided according to the specifications of D7 "Report on the design of assays".

Deviations from the project workprogramme

During 3rd Progress Meeting, it was decided to extend as much as possible this Worpackage, because from month 6 till month 12 (original end of WP2) there was little time to achieve the proposed objectives because it was not so easy to obtain the antibodies for target pesticides and there was a big amount of work involved in the preliminary investigations which was very important for the good performance of the final measurement system.

More realistic timetable considered to finish the main work at the end of July (month 13) and to deliver the final documents (D5 and D7) on October (month 16). This change did not affect the work of other workpackages and the rest of planning, as the WP3 and WP4 maintained the original timeline.

[10] Holme J, Jelle BP (2006): Infrared spectroskopy as atool for measurements of moulds. NBI Note 84, 2006 Building Technology. www.byggforsk.no/visVedlegg. aspx?vedleggID=482&dokumentID=1611

This delay is showed in the timetable of section 3. The rest of workpackages begun on date and were complying with scheduled workplan.

List of deliverables

Deliverables resulting from this workpackage (D5 "Report detailing preliminary investigations about existing works and techniques and the effect of cereal samples on the proposed measurement systems" and D7 "Report on the design of assays to be carried out during the whole project duration") were finished for 15th October 2005.

Del. No.	Deliverable name	Workpackage No.	Date due	Actual/Forecast delivery date	Estimated indicative person months (*)	Used indicative person-months (*)	Lead contractor
D5	Preliminary research on existing works and techniques and on the effect of cereal samples on the proposed measurement systems	2	8	16	0,2	0,25	IGV
D7	Report on the design of assays	2	12	16	0,3	0,35	AZTI

List of milestones

General delay in the WP affected the time milestones were achieved as follows:

Milestone No.	Milestone name	Workpackage No.	Date due	Actual/Forecast delivery date	Lead contractor
M3	Preliminary study into the effect of cereal samples on measures	2	8	12	IGV
M4	Estalishment of the reference analysitical methods	2	12	16	IGV

WORKPACKAGE 3 "DESIGN AND DEVELOPMENT OF IMPROVED MEASUREMENT SYSTEMS"

The main objective of this task was to design and develop measurement devices for sensitive, low costs and routine determination of pesticides in cereal samples at part per billion levels. In case of mycotoxins, the objective was to test present and usual measurement techniques used in cereal trade to extend their applicability regarding fast detection of fusaria damaged grain kernels and Fusaria toxin content. Regarding Acrylamide formation, the aime was to analyse and minimise the causes of this formation.

Task 3.1 Analysis of Acrylamide formation:

The analysis of Acrylamide formation was performed by IGV. In the study the link between free asparagine content of cereals, cereal milling products and the formation of acrylamid was investigated. In contrast to Heatox FP6-Project, MAP-Milling was focusing only on the very special topic of the asparagine in cereal products (which is not topic in Heatox) and their influence on acrylamide formation in baked goods. The aim was to give concrete advices for choosing raw material and processing cereals in mills.

The work of IGV under this task was mainly performed during last reporting period. Now as before, the minimization of acrylamide is an important item of the programme of quality assurance in cereal processing enterprises. This question is of special importance for products with a high signal value, e.g. gingerbread and crisp bread. For products with complex recipes even broader possibilities of recipe variations are given, whereas the possibilities for products only with a few components are rather limited.

Therefore, it was tested whether the acrylamide content of a product can be reduced by reducing the content of free asparagine in the grain raw material.

It was demonstrated that:

- There is a correlation between a low content of free asparagines in grain as raw material and the content of acrylamide in baked goods. A model baked product and crisp bread were used as proof.
- The content of free asparagine is decreasing with increasing extraction grade.
    - Bran has the highest content of free asparagine, followed by wholemeal. The lower the ash content of the flour the lower the content of free asparagine. The reduction of the bran content also will cause a reduction of the content of free asparagines.
- There is a clear difference between the content of free asparagine of the grain varieties.

  - Rice and maize have the lowest content of free asparagine, followed by maize, wheat, oats, rye.
  - Triticum durum shows less free asparagine than Triticum aestivum.
- There are variety differences in the content of free asparagine, but also scatterings within one variety because of different growing conditions. In some cases scatterings within one variety exceeds differences between the varieties. Therefore, the acrylamide content can be reduced by selecting varieties with low asparagine contents only if the grain varieties are supplied from contract growing, i.e. a varietal pure cultivation is guaranteed.
- Not all the acrylamide reducing capabilities on the basis of the reduction of free asparagines are applicable because undesirable changes of product properties can result.
- The addition of organic acids in concentrations not affecting sensory properties of the product is an effective and applicable way to reduce acrylamide in cereal products.
  - The acrylamide reducing effect of citric and ascorbic acid could be shown.
  - The successful application was shown in gingerbread in a laboratory scale and in crisp bread in an industrial scale.
- Single raw material variables show low acrylamide reducing capacity. For more effective reducing results it is suggested
  - To combine several effects derived from the raw material,
  - To combine effects derived from cereals as raw material with effects derived from other ingredients of the product,
  - To combine effects derived from raw material with technological parameters

The full description of tests, facts and figures is shown in the deliverable D11 "Study about Acrylamide formation and minimization".

Task 3.2 Development of Pesticides Measurement System

The main objectives of this task were to develop quick, cheap and reliable techniques based on enzyme-linked immunosorbent assay (ELISA) for detection of pesticides selected at part per billion levels and to assess the performance of the assay with real samples (spiked with pesticides) at laboratory level.

Competitive Enzyme-Linked Immunosorbent assay (ELISA) formats for detection of pesticides selected (2,4-D, DDT and chlorpyriphos-methyl) were explored in initial phases of the project. In order to design and develop ELISA for detection of selected pesticides, antibodies were obtained from research and commercial sources and other immunochemical reagents as appropriate.

A serial of experiments was done in order to determine the working conditions of the pesticides selected in an ELISA format. The main objective during last period was to assess performance of the immunochemical methods on real samples. During the last period, cereal samples preparation methods were deeply investigated. Suitable working protocols were established with the aim of achieving the necessary detection limits for a given sample matrix. Furthermore, the developed immunochemical methods were correlated with reference methods.

Determination of 2,4-Dichlorophenoxyacetic Acid in cereal samples

An indirect enzyme immunoassay was developed. The best assay showed a limit of detection of 0.9 µg/L. The assay can be performed in about 2 hours.

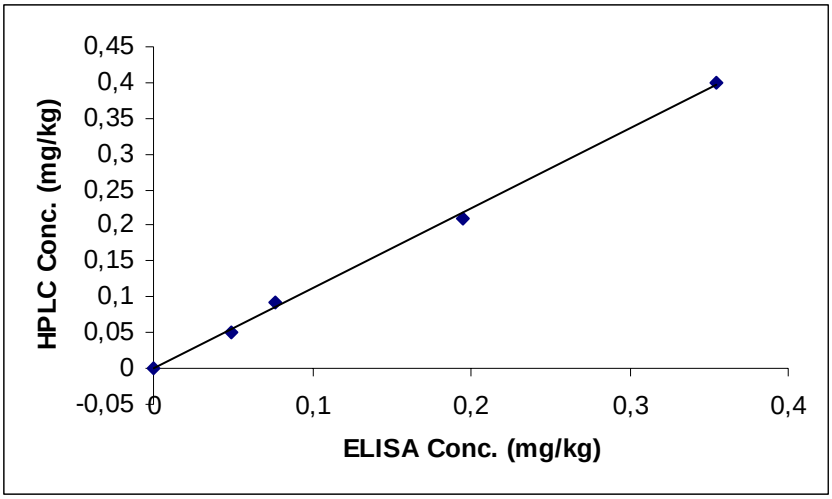
The final measurement procedure should be able to determine 2,4-D at least at 0.05 mg/kg (MRL set by EU) in cereal samples (wheat grain and wheat flour have been selected as the most relevant matrices). To achieve this goal, several approaches have been tested. In all of them a suitable solvent is needed to extract the target analyte and a clean up step of this extract had to be included in order to avoid the strong matrix effect that was disturbing the analysis in the ELISA procedure. In this sense, different techniques have been used: soaking, blending, sonication and accelerated solvent extraction for the extraction of 2,4-D from the cereal sample and solid phase extraction with various stationary phases for the clean up.

Methanol was the best solvent found for the extraction of 2,4-D although the recovery did not exceed 60%. Organic solvents have a strong influence on the interaction between the analyte and the antibody, so the clean up step was designed to remove the methanol and other compounds that might affect the immunoassay performance. Suitable cereal sampling protocol has been established.

Correlation with a reference method

An HPLC method has been optimized for validation purposes. Mass spectrometry has been chosen as detector to be able to quantify 2,4-D accurately at low levels of concentrations. Good correlation has been obtained (figure 1). This means that ELISA is performing as HPLC procedure and it can be a good method for 2,4-D evaluation in cereal samples.

The same samples fortified at different concentrations with the same sampling protocol were analysed by both methods, HPLC and ELISA. Each sample was analysed three times.



Slope: 1.111±0.026

r2: 0.9989

Figure 1. Correlation of ELISA with HPLC

Determination of DDT in cereal samples

An indirect enzyme-linked immunoassay ELISA was developed. The best assay showed a limit of detection of 0.14 µg/L. The assay can be performed in about 3 hours.

The final measurement procedure should be able to determine DDT at least at 0.05 mg/kg (MRL set by EU) in cereal samples (wheat grain and wheat flour have been selected as the most relevant matrices). To achieve this goal, as in the previous target pesticides, several approaches have been tested.

Methanol was the best solvent found for the extraction of DDT achieving good recoveries (around 90%). In this case, even after dilution of the extract there is still a strong matrix effect that makes impossible the measurement and more dilution is needed loosing the required sensitivity. Therefore a clean-up step by means of C18 cartridges was studied to remove the methanol and other compounds that might affect the immunoassay performance. Suitable sampling protocol has been established for standard solutions. However, when spiked samples were analysed the recovery achieved was less than 35%. Therefore other approaches are now being tested to avoid high dilution and to improve the recovery of the proposed method.

Chlorpyrifos Microplate ELISA

An indirect enzyme-linked immunoassay ELISA has been developed. The assay can be performed in about 3 hours. The best assay shows a limit of detection of 0.50 µg/L .

The factors determining the best immunoassay performance are summarized in table 1 and the corresponding standard curve is presented in figure 2. Under these conditions we have studied the reproducibility of the immunoassay for chlorpyrifos running six calibration curves in three-well replicates in six different days. The IC50 value is 10.41 nM, the dynamic range is between 1.02-9.78 µg/L and the limit of detection is 0.50 µg/L.

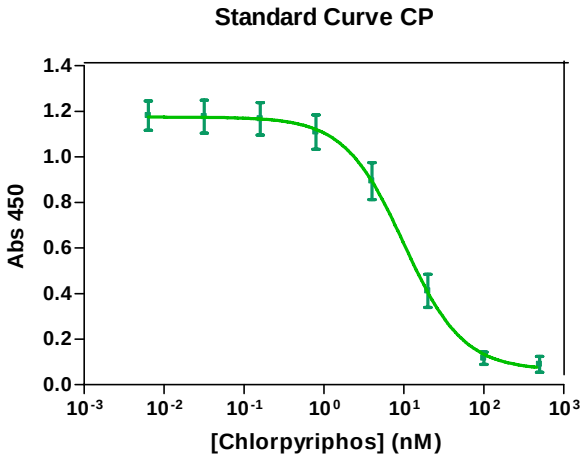


Figure 2. Standard Curve of chlorpyrifos

Table 1. Immunoassay features of Chlorpyrifos.					
$A_{max.}$	$A_{min.}$	IC_{50} (nM)	Slope	Linear range (nM)	LOD (nM)
1.18±0.15	0.08±0.08	10.41±3.54	-1.27±0.16	3.18-30.34 1.02-9.78 µg/L	1.57 0.50 µg/L

The data presented correspond to the average of 6 calibration curves run in 6 different days. Each curve was built using three-well replicates. The dynamic range is defined by the concentrations corresponding to 20 and 80% of the assay response at zero dose. The limit of detection (LOD) is the analyte concentration corresponding to 90% of the assay response at zero dose.

Determination of chlorpyrifos-methyl in cereal samples

The final measurement procedure should be able to determine chlorpyrifos-methyl at least at 3 mg/kg (MRL set by EU) in cereal samples (wheat grain and wheat flour have been selected as the most relevant matrices). To achieve this goal, as in the previous target pesticides, several approaches have been tested.

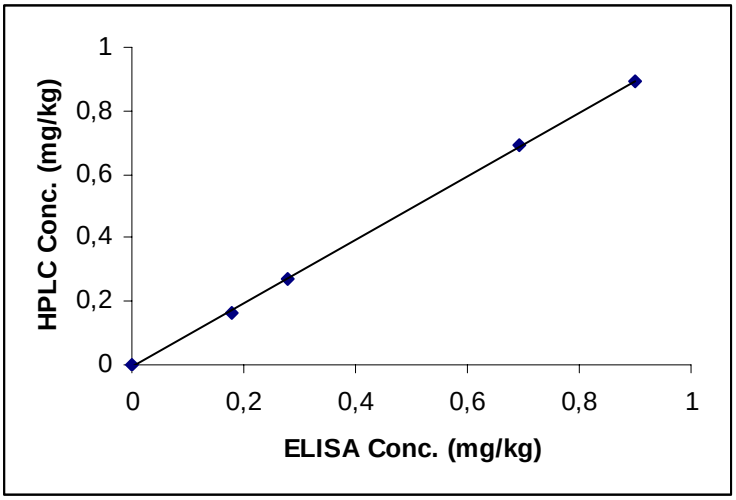
Methanol was the best solvent found for the extraction of chlorpyrifos-methyl achieving around 75 % of recovery. Because of the high sensitivity of the

ELISA in comparison to the MRL of this pesticide, the dilution of the extract could be enough to avoid the effect of methanol in the immunoassay. However, with this procedure there is still a strong matrix effect that makes impossible the measurement and more dilution is needed loosing the required sensitivity. Therefore a clean-up step was designed to remove the methanol and other compounds that might affect the immunoassay performance. Suitable sampling protocol has been established for detection of chlorppyrifos in cereal samples.

Correlation with a reference method

An HPLC method has been optimized for validation purposes. Good correlation has been obtained between immunoassay for determination of chlorpyrifos and HPLC procedure (figure 3). This means that ELISA is performing as HPLC procedure and it can be a good method for chlorpyrifos evaluation in cereal samples.

The same samples fortified at different concentrations with the same sampling protocol were analysed by both methods, HPLC and ELISA. Each sample was analysed three times.



Slope 1.002±0.011

r2 0.9996

Figure 3. Correlation of ELISA with HPLC

Achieved results in this task:

- Development of ELISA format for detection of 2,4-D in cereal products
- Development of ELISA format for detection of chlorphyrifos-methyl in cereal products.
- ELISA for 2,4-D, chlorpyrifos-methyl and DDT has been developed and optimized with standard solutions.

- Suitable sample preparation protocols have been established in order to detection of pesticides in cereal samples.
- Good correlation between ELISA results and chromatographic results for detection of 2,4-D and chlorphyrifos-methyl in cereal products.

The full description of activities under WP3.2 and its results is covered in the deliverable D13 "Testing of developed measurement system for Pesticides", that also includes a description of D15 "Prototypes of kit for detection of Pesticide residues".

Task 3.3 Comparison of Mycotoxins Measurement Systems

A very important part of the whole project is to give concrete advices for choosing raw material and processing cereals in mills according to there content of mycotoxins using quick and reliable measuring systems. The aim of the control is to check the accordance with maximal allowed limits of DON, given by EC regulation.

The limits for DON given by EC regulation 466/2001 are

- for raw cereal (despite hard wheat, oat): max. 1.250 µg/kg
- for raw hard wheat, oat: max. 1.750 µg/kg
- milled cereals max. 750 µg/kg.

IGV performed in a first stage the analysis of mycotoxin content of cereal samples (wheat, rye) from partners of cereal trade and food industry. Fusarium toxins (DON, zearalenone) were analysed through established HPLC methods. These results were used as reference for the rest of the work. Then, the relevant quick techniques will be performed:

- 1. Indirect rapid methods for analysis of Fusarium toxins
  - 1. Fourier Transform Infra-Red (FTIR) techniqes
  - 2. Visual identification of Fusarium damaged grain kernels
- 2. Direct rapid methods for analysis of Fusarium toxins (immunochemical methods)
  - 1. Competitive ELISA
  - 2. Dipstick assay

Fourier Transform Infra-Red (FTIR) techniqes

The testing of FTIR for applicability as quick method for mycotoxin measurement, included the collection of FTIR spectra, the calibration of FTIR – HPLC, the statistical check (full cross validation) and the optimisation (variation of sample preparation and of FTIR parameters, as well as the testing of different FTIR systems). First evaluation of the technique showed high differences between predicted values by FTIR and reference values by

HPLC, so it was concluded that there was not strict correlation between changes in grain compound caused by fusaria and mycotoxin content.

On a second stage, analysed data were checked again with help of Shimadzu calculation system. No better correlation as reported before was obtained.

So two preconditions for suggestion of the system were not given: costs and reliability. That's why effort was focused on more promising techniques. Additionally to planed tasks according to proposal, IGV included to its working package the test of dipstick assays and investigated more detailed the correlation to foreign substances.

Visual identification of Fusarium damaged grain kernels

Visual identification has the advantage of low costs and quick measurement. That's why it is very common to use. But the question was the reliability of the prediction of mycotoxins.

On the basis of investigation of 60 samples of wheat with DON concentration in a range of 1,4 to 34,3 µg/kg the correlation to fusarium-contaminated kernels was checked.

The DON correlation coefficient between HPLC reference values and content of visual selected fusarium contaminated kernels depends on the concentration range of DON and the special fraction of fusarium contaminated kernels.

Concentration range DON (mg/kg)	R in red coloured kernels	R in shrunken kernels, white coat	R in total fusarium contaminated kernels
1 - 16	-0,19	0,84	0,76
4 - 16	0,1	0,93	0,91
1 - 3,5	-0,01	0,52	0,22

As shown in the above table the following conclusions can be made:

- Highest reliability is given by counting the fraction of shrunken kernels, white coated in high DON concentrations > 4 mg/kg
- In high DON concentrations > 4 mg/kg also counting of total fusarium contaminated kernels gives acceptable correlation to DON reference values
- Including lower concentrations in total range of DON 1- 16 mg/kg shrunken kernels, white coated kernels give an acceptable approximation, lower by total fusarium contaminated kernels

- In low concentrations (max 4mg/kg), which is closed to practical questions, counting total fusarium contaminated kernels gives quite insufficient correlation to DON reference values
- Counting shrunken kernels, white-coated kernels in low concentrations (max 4mg/kg) improves the approximation, but the correlation coefficient is very low.
- Insufficient reliability is given by counting only the fraction of red coloured kernels; independent from the concentration range of DON

In summary is to conclude, that the prediction of DON contamination by counting foreign matter is only possible in very high DON concentrations and best by counting shrunken, white coated kernels. Checking the abidance of DON limits can only by done as very raw estimation and only by counting

Competitive ELISA

ELISA (enzyme-linked immunosorbent assay) is a relatively expensive chromatographic method. It's an immunochemical method with high specifity. The quantification of the toxin is possible by calibration.

Measurement of DON by ELISA can be assessed as recommendable screening method. It fulfils some requirements of the project approximately. It is still a relatively quick method, but it's necessary to buy equipment. The reliability is less than HPLC. In International Proficiency Test DON results obtained by ELISA the satisfying was 63%.

This task answers Mid-Term Review recommendation Nº 2 (To adapt and validate mycotoxins commercially kit tests based on ELISA principles).

Dipstick assay

Dipsticks work similar like immobilized ELISA system on the basis of antigen-antibody reaction as. We checked 37 samples of wheat with a DON content of 0,2 to 6,6 mg/kg. That means the check was carried out in concentration range, which is relevant for practical use to evaluate abidance of limits. We tested correlation

- DON, measured by HPLC,
- competitive ELISA and
- dipstick assays.

In the test were included two commercial available systems:

- R-Biopharm AG (RIDA®QUICK DON)
- NEOGEN Europe Ltd. (REVEAL® DON Strip test)

It could be shown that dipsticks are the most advisable DON-quick-measurement-system at this time. The advantages are:

- Quick: preparation steps are only:
  - milling, separation, 5-min reaction
- Low cost
  - buying dipsticks
  - and reader according to the offer of NEOGEN
- approximately reliable

The two systems (R-Biopharm and NEOGEN) differ in the kind and claim of getting results:

- R-Biopharm AG (RIDA®QUICK DON), kind and claim of getting results:
  - Visual detection, qualitative results:
    - Test line visible = negative = free of DON or only low concentration < 1ppm
    - No test line = positive = 1ppm DON or more
- NEOGEN Europe Ltd. (REVEAL® DON Strip test), kind and claim of getting results
  - Visual detection, qualitative results or by AccuScan reader, semi quantitative results:
    - 0 = 0...500µg/kg
    - 0,5 = 500...1000µg/kg
    - 1,0 = 1000...2000µg/kg
    - 2,0 = > 2000µg/kg

Both tests are suitable to select DON-containing samples. They are applicable also in the concentration range, which is relevant for practical use. This is the most important advantage compared with visual measurement of foreign matter.

In the range of EC limit the dipsticks some times predict higher DON level. That means, this samples should be selected for further control by HPLC, but the user is on the assured side. In our test we found differences in there reliability.

- R-Biopharm AG (RIDA®QUICK DON): In the evaluation of results we assessed the result as wrong, if
  - dipstick shows test line and HPLC shows more than 1 ppm
  - dipstick shows no test line and HPLC shows less than 1ppm

  When a weak line is visible, the DON-content near EC-limit is predicted.

This result was evaluated as right, but should be repeated by HPLC to decide the abidance of limits.

According to this assessment 37% of results were evaluated as wrong.

- NEOGEN Europe Ltd. (REVEAL® DON Strip test): In the evaluation of results we assessed the result as wrong, if
  - According to criteria of Neogen
  - According to the criteria of R-Biopharm

  According to criteria of Neogen 20% of results were assessed as wrong.

  According to criteria of R-Biopharm only 3% were assessed as wrong.

Finally, IGV performed a complete analysis considering the market costs of each solution:

In the field of indirect rapid methods, the **Fourier Transform Infrared Spectrometry (FTIR)** with wheat flours and the **visual assessment** with whole wheat kernels was tested. The two methods are not suited as rapid methods.

It was not possible to predict for unknown samples correct DON contents with all the FTIR calibration models which have a good correlation between HPLC and FTIR spectra. This was indicated already in the cross validation of the calibration when the correlation significantly worsened. Obviously there is no direct connection between the change of the components of the grain kernel (carbohydrates, proteins, fat) by moulding and the DON content. A strong moulding is not synonymous with a high DON content. It is possible that changes of the components may occur but no or only very little DON is developed, and vice versa. Other fungi which do not develop DON could have caused the changes of the components.

Costs: FTIR spectrometer, Shimadzu 23, 500 €, Bruker 31, 950 €.

Due to frequent requests of farmers, grain-chandlers and millers, the suitability of the visual assessment (analysis of impurities) was selected to be tested. Only strongly DON-contaminated wheat samples, which generally are not usual in milling industry, had a relatively good correlation between the content of shrunken kernels with a whitish layer and the DON content (45 samples with DON contents of 4-35 mg/kg, correlation coefficient r = 0.873). Just in the concentration range which is of interest to the milling industry, the correlation was bad (15 samples with DON contents of 0.5-2 mg/kg, correlation coefficient r = 0.521). The same holds true to the fraction with kernels of red discoloration (r = 0.012) and the total content of Fusarium damaged kernels (r = 0.216). That means that the visual assessment – although frequently used in practice – is not suited as rapid method for the acceptance or refusal of a wheat batch.

Costs: -

Regarding direct rapid methods for analysis of Fusarium toxins (immunochemical methods), IGV mainly tested ELISA and the dipstick assay:

ELISA now is a well established method in many enterprises of food industry. There was a very good correlation of the tested enzyme immunoassay Veratox® 5/5 DON and HPLC (correlation coefficient 0.984). The method can be recommended for raw material control. The measured results are storable. If the measured data should be stored permanently they can be transferred to a PC equipped with the corresponding software.

Costs test kit: 260 € (48 cavities)

Costs Neogen photometer incl. required attachments (starter kit basic packet): 4550 €

The ELISA test kit is offered also for zearalenone (Veratox® 5/5 zearalenone), but was not tested within this project.

The dipstick assay is a new method which till now is not generally known, but more and more growing in interest. Owing to their relevance and the growing interest in the suitability of this test, dipstick assays were selected to be included into the test programme. This rapid method of evaluation whether the limit value is exceeded or fallen below has the advantage that the result is achieved very quickly and without extensive use of instruments. The decision can be made visually (e.g. R-Biopharm RIDA®QUICK DON strip test) and more objectively by a scanner (for instance NEOGEN REVEAL® DON strip test). The dipstick assays, however, are applicable only for DON, till now.

The R-Biopharm test allows a five-minute mycotoxin assessment (above or below a limit value) after a 5 minute sample preparation. The correlation with HPLC was bad in case of RIDA QUICK DON strip test (correlation coefficient r = 0.500). The decision that the limit value was exceeded, was made in this test for many samples with low DON contents (64 % incorrect positive results) because the visual evaluation is difficult and not always definite. The test functions only for wheat; rye is not measurable (because of the slimy substances which adversely affect the flow behaviour). The test is not recommended for raw material control. It is intended to improve the possibilities of evaluation.

Costs of test kit: 209 € (25 test stripes).

The NEOGEN test allows a five-minute semiquantitative mycotoxin measurement after a 5 minute sample preparation. The correlation with HPLC was good in case of REVEAL DON strip test (correlation coefficient r = 0.919), where the decision is made more objectively by a scanner. The test is recommended for raw material control, however only for wheat.

Costs of test kit: 195 € (25 test stripes)

Costs AccuScan reader: 1495 € (AccuScan starter kit: 1995 €)

150 measuring results can be stored in the AccuScan, and then the datasets are transferred. For a permanent storage of the data they can be transferred to a PC equipped with the corresponding software.

At the end of the project, a new technique was considered: **Fluorescence polarization immunoassay.** This assay is known in the field of immunochemical methods, but not available commercially in Germany till now for mycotoxin analysis. By cooperation with the AOKIN AG in testing a system during the preparation phase a test developed for zearalenone was tested. The correlation with HPLC was good (correlation coefficient r = 0.956). The test allows a five-minute quantitative mycotoxin determination in the ppb range after a sample preparation of 5 minutes. The company intends in future also to offer a test for DON.

The spectrometer required for the test can be controlled in any computer. Corresponding software is used for data storage and automatic data evaluation. All the required calibration curves are stored for a dynamic range from 1 to 1000 µg/kg zearalenone. The measuring results, too, are storable on the PC for the control of the instruments.

Costs: As the test is under development the costs for the spectrometer and the test kit not yet are defined.

The next page shows a table that compares the studied measurement systems according the most interesting criteria. It initially shows that the most interesting method are the dipstick assay of Neogen, but it only gives an approximation and each specific case should take fully into account the comments in the final deliverable D14.

Method	Exact quantific.	Control of legal limit	Rapidity	Easy handling	Low costs	Summary
HPLC	(10)	(10)	(2)	(0)	(0)	reference
ELISA	8	8	6	6	6	34
Dipstick assay R-biopharm	2	4	10	8	9	33
Dipstick assay neogen	4	7	10	10	8	**39**
Fluoresc. polarization	8	8	6	6	5	33
FTIR	0	0	4	4	2	10
Fusarium damaged kernels	0	0	6	6	10	22

The full description of activities under this task and its results is covered in the deliverable D14 "Report detailing the tested measurement systems for Mycotoxins".

Deviations from the project workprogramme

There were not significant time deviations regarding the Work Programme and the work performed under this workpackage was delivered on time, except with the additional time required for Task 3.1. This workplan is showed in the timetable of section 3.

List of deliverables

Deliverables resulting from this workpackage (D11, D13, D14 and D15) were submitted in the previous progress report (D16).

Del. No.	Deliverable name	Workpackage No.	Date due	Actual/Forecast delivery date	Estimated indicative person-months (*)	Used indicative person-months (*)	Lead contractor
D11	Study about Acrylamide formation and minimization	3	24	28	0,7	0,7	IGV
D13	Testing of developed measurement system for Pesticides	3	28	28	0,4	0,4	AZTI
D14	Assessment of the tested measurement systems for Mycotoxins	3	28	28	0,4	0,55	IGV
D15	Prototypes of kit for detection of Pesticide residues	3	30	30	1	1,1	AZTI

List of milestones

Milestones of WP3 have been achieved as planned in the last period.

Milestone No.	Milestone name	Workpackage No.	Date due	Actual/Forecast delivery date	Lead contractor
M6	Selection of optimal measurment formats	3	28	28	AZTI
M7	Characterisation of final prototype of measurement kit	3	32	32	AZTI

WORKPACKAGE 4 "DEVELOPMENT OF A RISK MANAGEMENT SYSTEM FOR GRAIN MILLING SMEs"

The general objective of this workpackage was to develop a Risk Management System to guarantee quality and to increase safety of agro-food products in the grain milling industry.

Task 4.1. Definition of main catalogue.

The first task to develop the Risk Management System was the assessment of main pollutants in grain milling industry. This assessment was developed by INTECHNICA on the basis of the work carried out in the WP1 "Analysis of Grain Processing Industry Specific Needs".

Based on the results of the questionnaires and the knowledge of the consortium the key pollutants for the grain milling industry as well as the legislative, the standard oriented and the grain oriented framework have been assessed. In Deliverable 6 "Catalogue of the Key Pollutants in the Grain Milling Industry" the results of these assessments were reported. Assessments were performed for pesticides, mycotoxins and acrylamide according to the workplan.

- Pesticides are used by farming to control insects, diseases and weeds. There are strict regulations governing the use and sale of pesticides.
- Mycotoxins occur naturally in grain. The formation depends on whether and storage conditions like temperature, humidity etc.
- Acrylamide will be built during heating, baking, frying etc. of starch rich food. The amount of the formation depends on the frying conditions. Acrylamide will be formed at temperatures above 120ºC.

Full description of results is provided in deliverable D6 "Catalogue of the Key Pollutants in the Grain Milling Industry".

Task 4.2. Assessment and indication of critical control points.

Based on information elaborated and reported earlier in the project (Task 4.1), the assessment of critical control points started.

Therefore a questionnaire was developed to assess the state of the art and the problems of the SMEs. It was distributed to the consortium partners. Furthermore, Demeter distributed it to its contract partners in order to increase the number of participants.

The milling process was separated into single process steps and illustrated. In the questionnaire relevant critical control points for the milling process steps

were supposed and the state of the art in the SME milling companies was assessed with the collaboration of HARITASA, MÜHLE and ROMANA.

Critical control points were indicated based on the results of the survey performed and cheap and reliable inspection processes assessed in WP3 by AZTI and IGV. It was shown that there are three relevant Critical Control Points in the Milling Process:

- Income control (incl. choose of delivers, cultivars etc.)
- Storage control
- Product control

Critical control points were indicated based on:

- the results of the survey performed (needs and wishes of SME)
- cheap and reliable inspection processes assessed in WP 3 by AZTI and IGV
- scientific state of the art

This task finished on September 2005 (delay of 2 months regarding the original plan), due to the difficulty of collaborating with SMEs during summer vacation period. It did not affected the achieving of WP objectives on time.

Task 4.3. Development of the Risk Management System.

According to the work performed in Task 4.2 the Risk Management System (RMS) designed for the SME's of milling industry was developed. The system was done mainly by INTECHNICA, and ITA was supporting them in its design in order to ensure the optimum coordination between the theoretical RMS and the final system to be implemented in the SMEs. The developed RMS consisted of:

- Critical control points in the system and corresponding limit values according to income control, process control and product control.
- Requirements for good hygiene practice according to employee hygiene, facility and working room hygiene, waste management and water supply.
- Organisational structure according to responsibilities, required training, communication processes and supplier control
- Analytical requirements
- Control and prevention methods and emergency requirements

Guidelines of the Risk Management System and best practices were developed. The work performed paid attention to provide guidelines well illustrated and easy understandable, in order to use the content of the guidelines as training material in WP8.

The guidelines were submitted by February 2006. The status of the guidelines at the date of submission was version 2. Nevertheless, the guidelines were adapted and fine-tuned later on in order to make them understandable for the needs of a SME' s workstaff (more illustrations…).

Furthermore the results of the research and development according to the control of acrylamide had to be included, because this was not ready by the submission date.

According to the work plan of MAP-Milling the development of reliable, quick and cheap analysis methods was restricted to DON in the case of mycotoxins and to 3-4 pesticides in the case of pesticides.

Nevertheless there is no doubt that in case of pesticides tests to more pesticides than 3 or 4 have to be performed. DON could be seen as the key mycotoxin, but there are limits for more mycotoxins which have to be kept. In the case of a mycotoxin which accompanies DON like ZEA it can be said that ZEA limit is kept, if the DON limit is kept. But there are other mycotoxins, e.g. aflatoxins which have to be tested anyway.

Furthermore another big challenge for the risk management system and the software tool was that there were usually different limit values for:

- Different commodities (usually different limit values for rye, wheat, durum wheat, maize etc.)
- Different products (e.g. unprocessed grain has usually higher limit values than processed products or infant food)

The software tool to be developed had to be easily modified so that further tests and their test results can become included. The guidelines contained the limits of all relevant mycotoxins for all commodities and types of products. In case of pesticides there is a database of the EU Commission which contains all limits for pesticides. The guidelines contain the link to this database.

Nevertheless, it seems to be necessary that Milling SMEs will become supported in modification and installation of the software tool to their specific needs. This support has to be given by the foreseen cascade training procedures. If cascade training is not sufficient, additional support has to be performed by the IAG's.

The following measures were added at the end:

- Implementation of the R & D results of IGV according to acrylamide
- Implementation of new developments of new quantitative and semi-quantitative analytical procedures (Mycotoxins and Pesticides) according to the work of IGV and AZTI

- Implementation of new legal and other requirements
- Improvement of the guidelines' understandability by further pictures and graphics and illustrations

By preparation of training material, INTECHNICA developed modular material:

- Basic knowledge for every SME worker, very easy understandable
- Special knowledge for workers with special tasks according to food hygiene.

Deviations from the project workprogramme

Because of the slight delay of finish of work package 1, the work for work package 4 started with a slight delay because the work was based on the results of work package 1.

Nevertheless, deliverable 6 was submitted in due time. With regards to the content of the work package 4 there are no deviations so far.

List of deliverables

The catalogue of pollutants (D 6) was submitted in April 2005. According to Commission Regulation 856/2005 for the mycotoxins Deoxynivalenol (DON), Zearalenon (ZEA), Fumonisins and the T-2- and HT-2-Toxin new limit values were set in June 2005. Limit values became effective by July 1 st , 2006. Furthermore the commission regulation (No.) 123/2005 deals with Ochratoxin A in foodstuff and changes regulation 466/2001 and its amendments. There were changes to limit values in dried grapes, roasted coffee, whine and grape juice. For cereals all limit values and references given in D 6 were still active.

Due to the new regulations it was necessary to report these changes in mycotoxin legislation in an addendum to D 6. The addendum to D 6 gave the new limit values for the mycotoxins Deoxynivalenol (DON), Zearalenon (ZEA), Fumonisins and the T-2- and HT-2-Toxin for cereals and cereal products. It was performed and submitted by the end of June 2005.

Both D 6 "Catalogue of pollutants" and Addendum to D 6 "Catalogue of pollutants" were published in the public area of the MAP-Milling homepage www.mapmilling.net.

Del. No.	Deliverable name	Workpackage No.	Date due	Actual/Forecast delivery date	Estimated indicative person months (*)	Used indicative person-months (*)	Lead contractor
D6	Catalogue of pollutants in the grain milling industry	4	10	10	0,7	0,6	INTECHNICA
D10	Guidelines of the Risk Management System and best practices	4	20	20	1	1	INTECHNICA

List of milestones

Milestone associated to this WP was slightly delayed due to Task 4.2, as the table below shows:

Milestone No.	Milestone name	Workpackage No.	Date due	Actual/Forecast delivery date	Lead contractor
M5	Definition of control points and test to guarantee safety	4	13	15	INTECHNICA

WORKPACKAGE 5 "INTEGRATION AND VALIDATION OF POLLUTANTS CONTROL (INDUSTRIAL SCALE)"

The general objective of this workpackage was to validate and integrate the foreseen approach for grain milling industries on detection and control of Mycotoxins and Pesticides, and minimization of Acrylamide formation in the baking process.

The Specific objectives of this WP were:

- To develop an easy-to-use software tool that integrates the whole approach proposed.
- To validate technically the measurement systems developed at industrial level.
- To integrate the entire project results.

Once the RMS document was closed as version 1.0, and since ITA participated in Task 4.3, acquiring all the knowledge needed to implement the tool, then, Task 5.1 started in March.

The SW Tool has been developed under the Open Source philosophy, so no licenses are needed to be paid, and then the cost is totally free. Additionally, since the tool is open, then It may be continued and evolved by anyone else after the project ends. The tools will use MySQL, Hibernate and Java.

Then, the SW tool must be adapted to the CCPs established within the MAP-MILLING project and must follow the Risk Management System design also in this project. Additionally to these points, the proposed software tool also provides other interesting innovation points:

- Distributed architecture (1). We have seen that it requires a high effort to the SMEs to be continuously updated about the limits of the pollutants according to different legislation regulations. Then, the system has been designed so that there may be a central point where one "Association" updates those parameters and then, automatically or under the SMEs request, then those parameters are updated in every SME.
- Distributed architecture (2). The distributed concept also applies to the fact that the tool has been design in a distributed way, with the three layers of User Interface, Control and Database. There may be modifications in the User interface or create different User Interfaces without affecting the control or the Database pieces of code and the other way round. In that sense the system is adapted to different SME size, for example, there may be a unique point in the company to introduce all the data or there may be different physical places where different data is introduced, and moreover, the it may be configured that the data

introduced in different places may be different. This is related to the fact that it is also a user profile configuration, so that one user may only have access to some specific parts of the application.

- The SW Tool will be independence of the platform: Linux, windows, others. At least the control and database components.
- It will be a friendly and easy-to-use tool, taken into account the profile of people who is going to use it.
- The tool has been design so that it will be a multi-language tool.

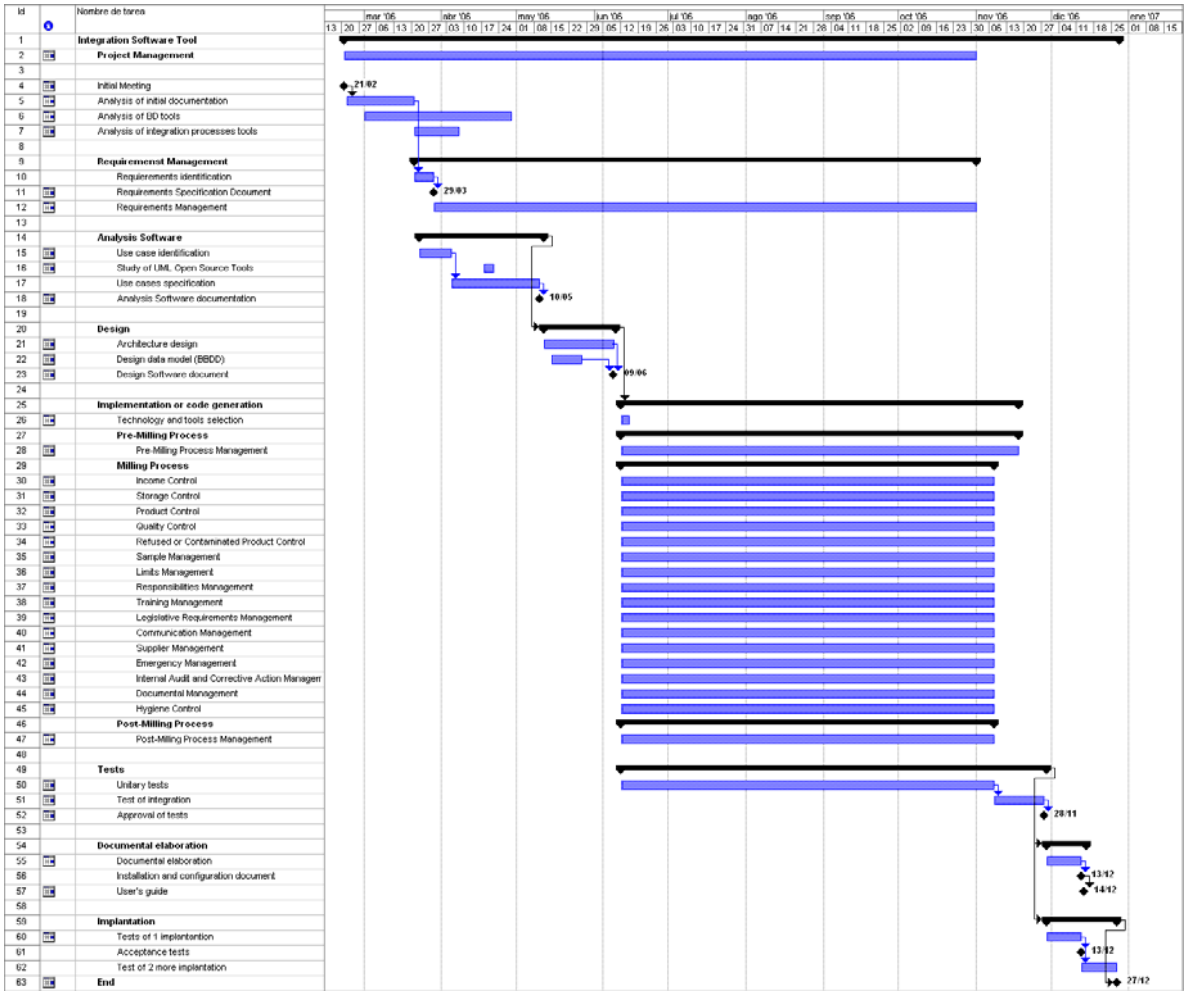
The first step done was to create an internal management for creating the tool. The objectives of this first activity were:

- To establish and maintain plans that define MAP-Milling project activities.
- To Monitor and Control the project's progress.
- To manage requirements changes.

With those task in mind, the following tasks were established:

- Developing the project plan.
- Getting commitment to the plan.
- Maintaining the plan.
- To take appropriate corrective actions when the project's performance deviates significantly from the plan.

The output of this activity was a complete planning of the project, with a detailed description of the activities to implement and a concrete timing of them, so that a strict control may be carried out and potential deviation may be predicted. The specific planning for WP5 tasks is described in the next page:



It may be seen in the previous planning that there was established a Work Breakdown Structure with the typical phases. And according to the timing, the following activities were achieved:

- System Requirement Specification
- System Analysis Software
- Analysis of existing tools in the market
- System Design Software
- Implementation of System
- Tests of System
- Development of the associated documentation of the project
- Pilot deployments

Task 5.1. Development of the integrating Software Tool

Then, the first activity already performed was the System Requirements Specification. The objectives of this activity were:

- To analyze the documents provided by Intechnica

- To identify the system requirements
- To manage requirements changes

Then, the following tasks were established:

- To study the Risk Management System in the milling industry designed in WP4, as well as other initial documentation of project
- To defined the high level requirements of the system:
  - To determine functional characteristics
  - To determine non-functional characteristics of the tool such as performance, operational, resources, verification, acceptance test, security, portability, reliability requirements...
  - To determine other aspects as constraints of the system, user characteristics, operating environment, assumption and dependences, etc.

And the output of this activity is the "Requirement Specification Document". These are of those requirements.

ITA had **the constrain** to integrate a food safety control in the processes management at a very low cost and without the necessity to have an advanced training in new technologies. That is the reason why we have chosen open source technologies since there is no cost to pay for licenses. Regarding the **User characteristics**, the software tool will be destined to users whom they do not have obligation to have computer knowledge. Then, a friendly and easy-to-use tool will be provided. Regarding the **operating environment**, there were no special restrictions, namely, it does not have the need to be developed with specific framework (tools, concrete database managers...), and that is the reason why we are going to develop it multi-platform, so that it may be deployed over windows, or linux or others. Regarding the **verification requirements**, the tool will be made tested and proved with the aim to fit the procedures and to make them effective. Regarding the **maintenance requirements**, the tool must be extended and easy to maintain. Some additional requirements consider the fact that as the countries represented within the consortium are especially significant in agro-food, the software tool must be multi-language. And the main requirements are the **functional requirements**. The functional requirements are structured related to the three phases of the process: pre-milling, milling and post-milling. A detailed description is provided in the document referred before.

Then, the next activity is the Software System Analysis. The objective of this activity is to obtain a more detailed specification of the software tool and to identify inconsistency, ambiguities, duplicity or lack of information. The selected method to implement the system analysis is thought he know use cases definition and the express them in the following ways: Description in

natural language, Use of UML use case diagrams, Use of UML sequence diagrams, Use of UML activity diagrams. Indeed, theses are the main tasks of this activity. The output is the Analysis Software Document. The summary of the analysis is divided into the following packages: Pre-Milling Process Management, Milling Process Management and Post-Milling Process Management.

It has also been performed an study Analysis of existing tools in the market in order to study existing tools in the market in order to use them in the development of the software tool, considering the requirements expressed in the requirements document (low cost -> open source, friendly User Interface, …).

The tasks performed have been:

- Study of tools in relation to Object/Relational persistence and query service (like Hibernate)
- Study of process integration tools (like JDBM)
- Study of UML open source tools (Omondo's plugin EclipseUML, argoUML,…)

And the output has been to select mysql as database, Hibernate to connect the database and the control part, and java to develop the control part. The UI will be coded in java or .net.

The next activity is the Software System Design. The main tasks of this activity are:

- Architecture design
- Design State Machine
- Infrastructure: scenarios definition
- Interface Prototype
- Design data model (BBDD)
- Design class model

And the output is the Design Software Document. A draft of the system architecture is shown in the previously mentioned document, as well as a detailed description of the State Machine Framework. This infrastructure facilitates the development of distributed systems, conceived as blocks which interchange information though asynchronous messages. The internal structure is composed of:

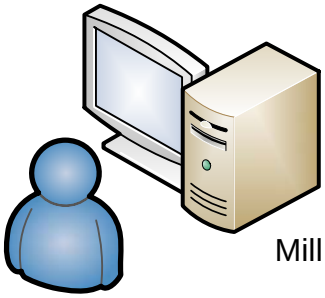
- Activator: Activation patron of the infrastructure.
- Log: registry of all the actions by levels (debug, error, warning, trace, trace all) in order to facilitate the monitoring of the tasks and the real time treatment from remote web access.

- Sender: responsible to send all the encapsulated information through different protocols.
- Server. Responsible to receive requests and answers in several formats and convert them in messages.
- Manger. System core responsible to store all the pending requests and redirect them to the state machines.
- MessageList: producer-consumer with all the pending tasks.
- SMList: patron to process all the messages as a sequence of actions.
- Committed: cache patron which stores part of the persistent information in memory in order to improve the system performance.

The system may be configured to work with different system architectures, depending on the size of the SME.

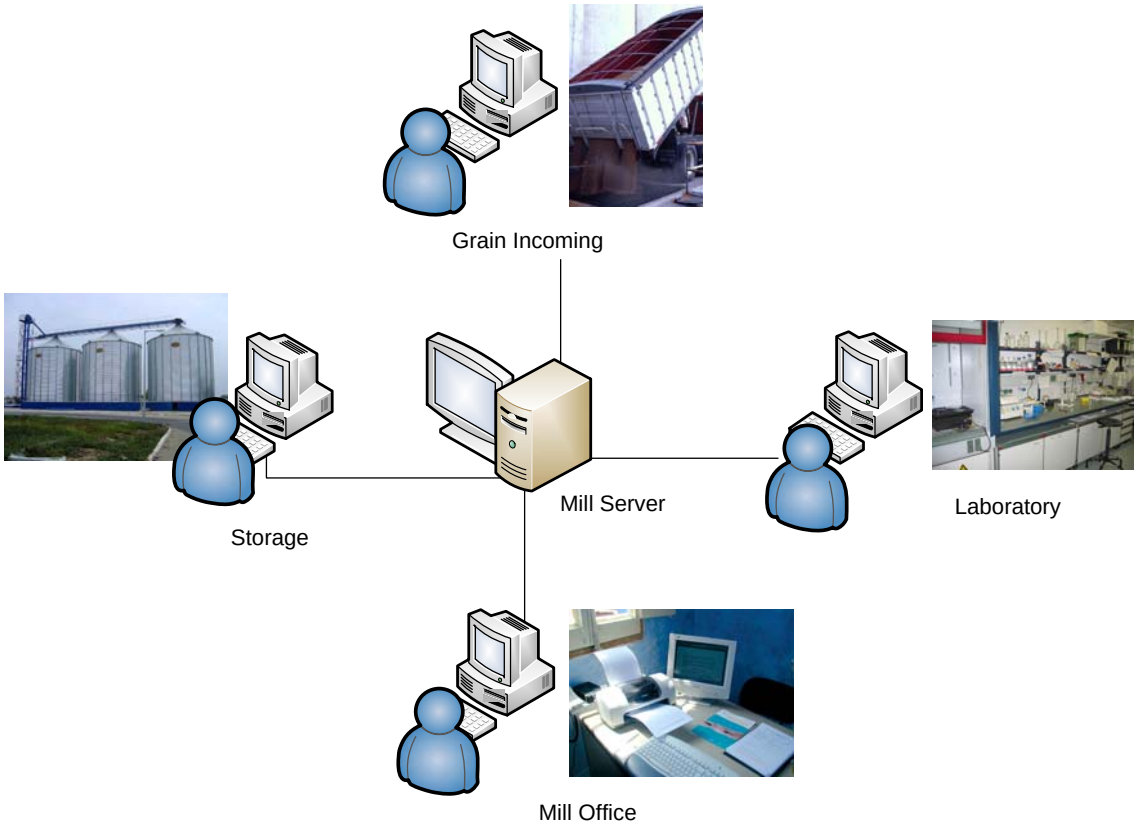
- There is only one PC and all the data is introduced through that PC.

Mill Office



Mill Server

- There are several points in the SME where the data may be introduced.

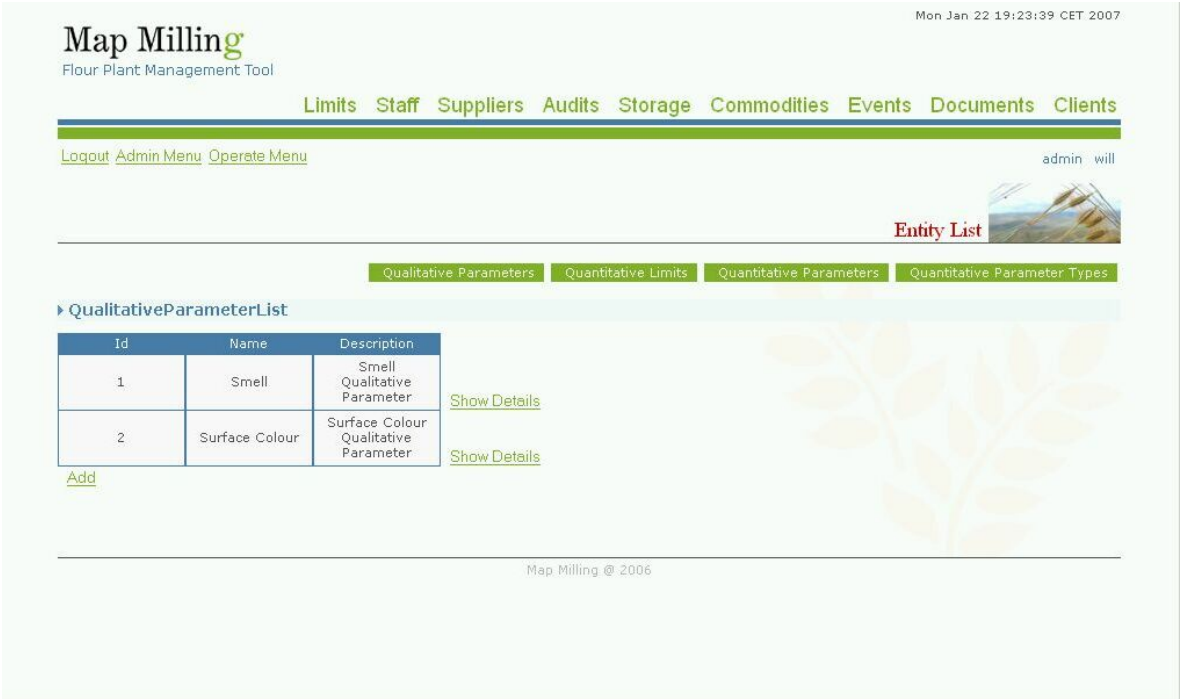


With the following arquitecture, it will be possible, that all legislative requirements are centralized in a server. In that way, when the responsible milling company or association finds changes to legislative requirements, it will inform to the rest of the milling companies about the new changes.

The next work performed was the interface prototype. It has been designed considering all the requirements, and it has been defined the two kind of user's profile:

- Operator, related to CCPs (Incoming, Storage, Product, Quality) and warnings.
- Administrator, related to management (Responsibilities, Limits, Training, Communication, Supplier, Emergency, Audits, Documental, Hygiene, Legislative)

All the screens were produced from a functional point of view, and the final aspect of them was worked out in the final phase of the project. Below one example of a profile is shown:



ITA has developed the Map-Milling Software Tool (D17), that may be found and downloaded from the Official Map-Milling Web site http://www.mapmilling.net/downloads.htm .

Task 5.2. Industrial validation of the measurement systems

It has also been finished the System Implementation. The objectives were to generate the code of the different functional packages derived of analysis and design of system and to create the final interface design

The task was to develop the packages identified in the analysis and design of system:

- Pre-Milling Process Management
- Milling Process Management (Income Control, Storage Control, Product Control, Quality Control, Refuter or Contaminated Product Control, Sample Management, Limits Management, Responsibilities Management, Training Management, Legislative Requirement Management, Communication Management, Supplier Management, Emergency Management, Internal Audit and Corrective Action Management, Documental Management, Hygiene Control)
- Post-Milling Process Management

Therefore the output was the Software tool's executable and source code of different work packages. And then, the last activities performed till the end of the tasks (February'07) were:

- System Tests. The objective is to carry out a test plan of system, performing unitary tests of each one of the work modules that has been developed and an integration tests between the different modules. The output is the approval of executed tests.
- Provide all the associated documentation already indicated.

Task 5.3. Final integration of technologies and systems

Task 5.3 started in November with the collaboration with Haritasa. This SME has provided an important feedback in the system requirements and features from a practical point of view, minimizing the deviation from the expected results from SMEs.

An Installation Manual was produced which shows how the software tool may be installed in a very easy step by step way and ITA has also provided a User Manual which shows how every feature of the tool may be used.

The integration and validation process is highly coupled with the analysis, design, development and test of the tool since it was adopted an interactive process. ITA has continuously taken into account the final user perspective and opinions, and indeed, HARITASA, one SME participating in the project, participated in all the phases of the development of the tool. In that way, the possible deviations from what the finals user expects to what they really obtain at the end of the development is minimized to zero.

Following that strategy, ITA visited several times the facilities of HARITASA (Harinera de Tardienta, S.A.) and held periodic meetings to show the progress made in order to obtain feedback from a final user perspective which is definitely very important.

Once the software tool, the documentation, the installation manual and the user guide were fined-tuned with the continuous feedback from Harinera de Tardienta S.A., then, all these results were provided to the National Associations, which have distributed these results among their SMEs and in particular to those which have participated in the project (Romana Macinazione S.r.L. in Italy and Mühle Müschen in Germany) which have finally validated the high quality of the results and have integrated the software in their industries. ITA has provided support to all the SMEs which have requested it.

Deviations from the project workprogramme

There was only a deviation of 1 month in order to finish the final integration of the tool in the 3 expected SMEs, so task 5.3 finished in March'07.

This plan is showed in the timetable of section 3.

List of deliverables

Deliverables resulting form this workpackage (D17 and D18) have been submitted in month 32 and 33. Both are included in one report, since D18 is the report of the integration and validation of the Software Tool made in D17 (Software).

Del. No.	Deliverable name	Workpackage No.	Date due	Actual/Forecast delivery date	Estimated indicative person-months (*)	Used indicative person-months (*)	Lead contractor
D17	Software tool integrating the approach of R.M.S. & pollutants control	5	32	32	1	1	ITA
D18	Report detailing the integration and validation	5	32	33	0,75	0,8	ITA

List of milestones

General milestone of the WP was achieved as foreseen.

Milestone No.	Milestone name	Workpackage No.	Date due	Actual/Forecast delivery date	Lead contractor
M8	Succesful integration of control technologies	5	32	33	ITA

WORKPACKAGE 6 "EXPLOITATION OF PROJECT RESULTS"

The general objective of this Workpackage was to ensure an optimal utilisation of research results of the project and the transfer of knowledge within other interested agro-food sectors.

All deliverables related with this WP (D20 "Documentation of request for patent" and D21 "Analysis of other agro-food sectors for technology transfer") were foreseen for the last period.

Task 6.1. Protecting the knowledge

As a previous step to the final dissemination of the project's results, the co-ordinator (CEZ) proposed a discussion regarding the possible exploitation of Project Results, to ensure optimum use of project results. This debate was mainly held on the 6th progress meeting in Sukarrieta (Bilbao), once the Project Results were fully developed and clear.

According to the RTD Performers, the main results from MAP-MILLING project are not patentable:

- ELISA toolkit for pesticides detection (2,4-D, DDT and chlorphyriphos-methyl)
- Knowledge on Mycotoxins measurement systems.
- Knowledge on minimization of Acrylamide formation from the source.
- Tailored Risk Management System for grain milling SMEs on a open-source Software Tool.

Thus, the IAGs will own the project results, but their exploitation strategy will be focused in the distribution among interested SMEs in the Grain milling sector. In a first step, this distribution was done through the participating IAGs and associated companies, and later to other associations that could reach additional companies in the sector.

The Core Group of SME's benefited from an early use and deep knowledge of the project results and the RTD Performers had preferential access to knowledge generated within the project in order to perform further research in their research areas.

Due to these reasons, there was no documentation to be submitted as request for patent within the deliverable D20 "Documentation of request for patent".

Task 6.2. Analysis of technology transfer

As project co-ordinator, CEZ and its OTRI (Office for Transfer of Research Results) led and coordinated the analysis of possible technology transfer processes in order to spread the project results to other agro-food sectors that could be interested in the control of mycotoxins and pesticide residues.

There was also discussion on the new ways of research to advance on the project results, both in the pollutants detection and minimization and in the Management Systems for SMEs

The results of this debate were summarised in D21 "Analysis of other agro-food sectors for technology transfer"

Deviations from the project workprogramme

There has not been any deviation in the development of this WP, so it has been finished on time at the end of the project. This plan is showed in the timetable of section 3.

List of deliverables

Deliverables resulting form this workpackage (D20 and D21) were submitted at the end of the project, but only D20 has no sense as the results obtained are not patentable.

Del. No.	Deliverable name	Workpackage No.	Date due	Actual/Forecast delivery date	Estimated indicative person-months (*)	Used indicative person-months (*)	Lead contractor
D20	Documentation of request for patent	6	36	36	0,35	0,2	CEZ
D21	Analysis of other agro-food sectors for technology transfer	6	36	36	0,2	0,25	CEZ

List of milestones

General milestone of the WP was achieved at the end of the project.

Milestone No.	Milestone name	Workpackage No.	Date due	Actual/Forecast delivery date	Lead contractor
M10	Exploitation agreements on new technology	6	36	36	CEZ

WORKPACKAGE 7 "DISSEMINATION"

The general objective of this Workpackage was to carry out a diffusion of the results of the project at European level, both towards the grain milling sector and agro-food sector in general. The dissemination included also the European citizens.

All deliverables related with this WP (D22 "Diffusion materials used in the 5 conferences", D23 "Articles and list of publications used", D24 "MAP-MILLING Web Site" and D25 "Multimedia training platform") were foreseen in the last period, but the Web Site (D24) was already working from the month 12.

Task 7.1. Diffusion towards grain milling sector

This task consisted in the Preparation of 4 national conferences for grain milling SMEs and SMEs associations concerning food safety, main pollutants, new normatives and project results.

These conferences included dedicated training workshops, which are fully described in the next WP (WP8 Training). 3 national conferences were held on Spain (CEZ), Italy (UNIONA) and Estonia (AEFI). The fourth one in Germany was responsibility of PRO-AGRO, but they refused to organize it because of "…the rearrangement of the structure of our organisation and personal limitations…", according to the letter sent by them at the end of the project. This made that no other partner could perform this work on due time, so the final objective of the Task was only partially achieved.

The conferences were focused on grain milling sector, but other interested sectors also attended.

The responsible IAGs worked on the logistics required and the possible dates to hold the different conferences in order to coordinate all the necessary resources, and some of them were organised within other established events in the agro-food sector. The participating SMEs and RTD Performers were also represented in the events.

SPAIN

CEZ and AIAA, organised the Spanish Conference and Training Workshop hold in Zaragoza on 30th May'07. There were around 50 people, representing all the milling companies of Aragon and some of the neighbour regions, as well as some agrofood laboratories.

It's considered quite positive the fact that the conference invited some speakers from outside of the consortium (public administrations, agrofood associations and local researchers) in order to get a critical mass and complement the approach of the MAP-MILLING Project.

Organisers also highlights that the work of pre-dissemination of the Conference in order to confirm attendants, was quite hard, and it was very important to refine the specific people to be contacted within each organisation.

We would like to emphasize that in the National Conference in Spain, they were present maximums responsible for the Autonomic Authorities in Agro-Food matter, for Food Safety and Innovation, like the Advisor of Agriculture and Food of the Government of Aragón, the General Director of Investigation of the Government of Aragón and the General Director of Public Health of the Government of Aragón; in addition to the main representatives of the Spanish partners of the project (President of AIAA, General Secretary of CEZ and Director of the ITA).

Also it were invited Spanish experts in Mycotoxins and Acrylamide, that hadn't participated in the Map Milling Project, but their studies and investigations also were interesting for the Food Companies.

Dissemination activities included the elaboration of Notes of Press, contacts and attention with different Mass media (Press, Radio and TV).

ITALY

ROME and UNIONA) organised the Italian National Conference and Training Workshop. This event was held on 20th June'07, and it was in the same place and just after the Stock Exchange of cereal sector companies. The Agency of the Chamber of Commerce of Rome: "Romana Mercati" was contacted. This Agency is specialized on food products and rules the Roman Stock Exchange for cereals (Borsa Merci di Roma "BMR"). Every week the BMR fix the list prices of the cereals and all the major mill industries of the regions of central Italy attend the session.

Thus, the National Conference was organized inside the BMR, the same day of the session for the cereals list price, in order to have a better audience. Organisers also contacted the Chamber of Commerce of Rome, the Union of Chamber of Commerce of Latium and the Latium Region in order to have their cooperation in the dissemination of the results of the project regarding the SW tools.

Organisers made a research to elaborate a mailing list and the formal invitation was sent to laboratories, SME, public administrations; while Romana

Mercati sent the information and the invitation to their data base of companies.

The dissemination of the information about the conference begun on April 2007, during the fair "Cibus Roma" for food industry. We visited the fair and informed the companies attending the show that on next June we will organize the MapMiling national conference to inform the food industry about the results of the project and about the SW tool that the companies could download from the web site of the MapMilling project. We focused our contact on companies producing wheat and cereals derivates.

ESTONIA

AEFI hold the Estonian Conference and Training Workshop in Tallin on 6th June'07. The attendants to the Conference were mainly technicians from agrofood laboratories, some milling industries and 2 representatives from the Estonian Government.

There were an intense feedback from attendants in order to make further questions after the sessions organised, and Mr. Volker Troebs (INTECHNICA) as one of the trainers has been replying questions in weekly batches.

In general, the feedback from the attendants was quite positive. Media used to publicise the event: most of the information was sent via emails and information was given on several seminars, which had been organised by the Association. Information about the event was publicised on the web page of the Association of the Estonian Food Industry (www.toiduliit.ee)

Presentations of the Event and training materials (handouts, some also translated into Estonian, are attached), information about the project results is publicised on the web page of the Association of the Estonian Food Industry (http://toiduliit.ee/Tegevused/Projektid/)

The different materials associated with these Conferences were compiled in the D22.

Task 7.2. Diffusion towards agro-food sector

The diffusion to the agro-food sector in general will be done through the following activities:

- Preparation of a European Conference. This conference had a similar structure than the national events, but it was mainly focused on the European agro-food sector. The event was organised by DEMETER and all the interested parties in Europe were invited (agro-food IAGs, food safety organisms, research bodies, public institutions, policy makers…). CEZ also

provided a strong support in the organisation of this event as Project Coordinator. The conference was held in Brussels on 5th June'07.

The attendance to the Conference was not as big as desirable, specially taking into account the number of contacts performed during the previous weeks. Maybe the fact that the Conference issues were quite specific undermined the participation of more people. The technical character of the project made the issues so specific. Another reason to explain the attendance level could be the quality of the list of addresses, the right organisations were contacted, but maybe the correct people within these organisations were not directly invited.

In any way, the feedback from the attendants during the last part of the Conference was very positive, and additional information was asked by many of them regarding the project results: rapid methods for determination of contaminants and the software tool for Risk Management System.

- Apart from the Conference, many articles and press releases related to the project have been included both in the main technical journals of each country and in periodical publications of IAGs at local, regional and national level. The full consortium has worked on a list of publications and different medias to be used for the releasing of 3 different kind of materials:
    - Scientific articles.
    - Technical articles.
    - General diffusion articles.

Some of the main diffusion actions performed are:

- Elaboration of a special Bulletin informing about the celebration of the National Conference in Spain and the presentation of the Project. The Bulletin was spread more than 500 companies of the Milling sector at regional and national level
- Scientific Oral Presentation: Argarate N., Barranco A., Arestin M. and Alfaro B. Determination of chlorpyriphos methyl in wheat samples by monoclonal enzyme immunoassay. IXth International Conference of AgriFood Antibodies. Norway, 10-13/09/2007
- Dissemination of results of Map-Milling project: http://www.azti.es/ Presentación de resultados del proyecto Map-Milling sobre detección y control de contaminantes en el sector de derivados de cereales.
- Presentation of project results at demeter milling working group (may & autumn 2007) & working group for processing of national association of organic food industry (autumn 2007)

- Article in Lebendige Erde (summer 2007) & Demeter-International-Newsletter (summer 2007)
- Information & diffusion material by phone and post to organisation of german mills (june 2007)
- Information & diffusion of material by visiting german SMEs (july - december 2007)
- Interview of HARITASA in the local newspaper "El Heraldo de Aragón" (Article published: 5 June 2007 (see copy))
- Brief presentation in the Mühlenchemie Symposium (Date: 13,14,15 June 2007; Place: Hamburg; Targets: Millers from Asia, Australia, Africa, South America, Europe
- Intechnica Homepage (www.intechnica.de) Short description of the project with link to www.mapmilling.net
- Detailed publication of project and its results in technical literature "Mühle und Mischfutter", Verlag Moritz Schäfer, Detmold, Publication probably in August or September issue (approx. 20 pages). Also published as pdf-download on www.intechnica.de
- Short publication of project and its results (1/2 page) in newsletters of associations of milling companies, chamber of crafts, etc.
- Presentation at the annual conference 2007 of the "Bavarian Association of Milling companies" on June 23rd, 2007
- The regional newspapers with the highest circulation in Aragon – Spain (Heraldo de Aragon and El Periódico) have informed about all the related activities and research performed. Several articles have been published showing not only the progress and research of the regional activities but all the research made by the project.
- ITA together with Harinera de Tardienta S.A. has made a TV story which has been shown in the regional TV showing the results of the project in the Harinera de Tardienta S.A. facilities.
- The dissemination of information about the SW tools begun during the SIAL fair of Paris on October 2006.
- On May 2007, some Italian SME industries (mill and cereal industry) were visited in order to better explain the project and the great opportunity that the SW tool will offer them
- The information about the project will be put on the WEB site of Federlazio with the link to the MapMilling web site.
- On October 2007 the Consorzio ROME will attend the most large food fair in Europe: Anuga fair of Koln, Germany. They will visit the stand of Italian companies of mill and cereal sector in order to distribute the information about the MapMilling project results and the training SW tool material, translated in Italian, on a CD.

The different materials associated with these Dissemination Activities we compiled in D23.

Task 7.3. General diffusion at European level

The more general diffusion has the objective of building awareness about project results, both in the consumers and in the economic actors of the agro-food sector. The main actions performed are the following:

- Creation of a free Web Site (accessible to all parties in EU) compiling the results and setting a network to exchange experiences about the food safety. The Web Site is already working since 2 years ago and is available at: http://www.mapmilling.net/
- Additionally, the consortium has developed a multimedia training platform for computer assisted training about the project issues and their direct application in all the countries. It has been supported in the already created Web Site.

Deviations from the project workprogramme

The fourth National Conference to be hold in Germany was responsibility of PRO-AGRO, but they refused to organize it because of "…the rearrangement of the structure of our organisation and personal limitations…", according to the letter sent to the coordinator at the end of the project. This made that no other partner could perform this work on due time, so the final objective of the Task was only partially achieved.

The schedule of the workplan remained unaffected and it is showed in the timetable of section 3.

List of deliverables

Deliverables resulting form this workpackage (D22, D23, D24 and D25) were submitted in the last Period (month 36).

Del. No.	Deliverable name	Workpackage No.	Date due	Actual/Forecast delivery date	Estimated indicative person-months (*)	Used indicative person-months (*)	Lead contractor
D22	Diffusion materials used in the 5 conferences	7	36	36	0,2	0,25	DEMETER
D23	Articles and list of publications used	7	36	36	0,35	0,4	DEMETER
D24	MAP-MILLING Web Site	7	36	13	0,5	0,6	DEMETER
D25	Multimedia training platform	7	36	36	0,8	0,65	CEZ

List of milestones

General milestone of the WP was achieved at the end of the project.

Milestone No.	Milestone name	Workpackage No.	Date due	Actual/Forecast delivery date	Lead contractor
M11	Wide diffusion and dissemination at European level	7	36	36	DEMETER

WORKPACKAGE 8 "TRAINING"

The general objective of this Workpackage was to provide advanced training of managers and technical staff of grain milling sector SMEs and Industrial Associations/Groupings on the knowledge produced within the project.

Task 8.1. "Cascade" training workshops

The consortium organised a series of workshops in each region addressing pollutant new measurement techniques and the risk management approach, taking the advantage of the National Conferences described in the previous WP. During the last part of the project, the following activities were performed:

- The RTD developers elaborated the training materials to be used in the workshops. The use of additional experts from outside the consortium was assessed, and in some cases was finally performed.
- There were 3 training workshops organised by the following organisations in: Lazio – Italy (ROME), Aragon – Spain (AIAA) and Estonia (AEFI). The 4th one was supposed to be organised in Brandenburg – Germany by PRO AGRO, but as it has been explained before, this partner finally refused to carry out this task
- The responsible IAGs worked on the logistics required and the possible dates to hold the different workshops, according to the specific characteristics of each region/country. The training workshops were mainly hold in afternoon sessions during the same day of the National Conferences. It helped to gain critical mass and to increase the impact of the dissemination and training actions.
- Who have attended the workshops apart from organising IAGs?: RTD as main trainers, MAP MILLING SME's staff. They have contributed with their own experience on how to implement the results of the project in others SME's.
- To Whom have been addressed?: Workshops were aimed at training managers and technical staff of other national SME's and IAG's.
- How was the training methodology?: Cascade Training. To Train the trainers. Attendants will adapt the workshop to their particular needs and later will establish a different level of training.

The different RTDs prepared the materials for their field of expertise. The materials in English were included as D19.

Deviations from the project workprogramme

The only deviation was the final refusal of PRO AGRO to organise the German Training Workshop.

The general schedule of the WP remained unaffected and it is showed in the timetable of section 3.

List of deliverables

Deliverable resulting form this workpackage (D19) was submitted at the end of the Project.

Del. No.	Deliverable name	Workpackage No.	Date due	Actual/Forecast delivery date	Estimated indicative person-months (*)	Used indicative person-months (*)	Lead contractor
D19	Training materials used in the workshops	8	33	33	0,25	0,25	CEZ

List of milestones

General milestone of the WP was partially achieved (only 3 training workshops) in the project.

Milestone No.	Milestone name	Workpackage No.	Date due	Actual/Forecast delivery date	Lead contractor
M9	4 training workshops about project results	8	33	36	CEZ

Section 3 – Consortium management

This section should summarise the status of the project and its management activity.

WORKPACKAGE 9 "PROJECT MANAGEMENT"

The main objective of this workpackage was to make possible the adequate co-ordinating organisation of the project, necessary to assure and/or establish:

- Communication flow
- Project follow-up (project progress control and planning)
- Decision making procedures
- Networking and interaction with third parties (projects, innovation actors, institutions…)

Kick-off Meeting (Zaragoza – 24th June, 2004)

The Kick-off Meeting of MAP-MILLING's project was held in 24th June 2004 on the offices of the Coordinator (CEZ) in Zaragoza (Spain). This meeting was the starting point of the proposal, where partners met together, know each other and committed to perform their best to achieve MAP-MILLINGs objectives.

All the partners but two attended or were represented in the meeting. ROME due to illness of Mr. Scipioni and ROMANA due to last minute changes in flights.

The working meeting was held on Thursday 24th June, and ITA organised a guided visit through their facilities for the next day (Friday, 25th June).

The agenda of the meeting and its minutes were included in deliverable D4.

D1. Project Presentation

The project prepared a brief project presentation in English of three pages describing the main aspects of the project in a style which is accessible to the non-specialist, avoiding technical language, mathematical formulae and acronyms as much as possible. This document was attached to deliverable 4.

Pre-payment distribution

Once the pre-payment from the Commission was received by the coordinator, all the partners of the consortium was asked to fill in a Financial Identification Sheet with the details of the bank account where they wanted to receive their EC funding.

Each partner signed, stamped and sent its form, being collected by CEZ financial offices (Mr. Lalaguna).

According to the specific instructions of the European Commission regarding the pre-payment of MAP-MILLING partners, all partners received their funding in advance, except DEMETER and MUHLE (as they do not established the bank guarantees required by the European Commission).

Change of address of AEFI

Official address of AEFI changed after the kick-off meeting, so it required some administrative procedures to change the contract (new CPF's etc.). The new address is:

Street name and number: SAKU 15

Postal Code: 11314

TALLINN ESTONIA

2nd Meeting (Nürnberg – 28th January, 2005)

2nd Meeting of MAP-MILLING's project was announced and prepared for 28th January 2005. This meeting was held on the facilities of INTECHNICA in Nürnberg (Germany).

This meeting was mainly technical, so the attendance of Project Coordinator and all RTD Performers is compulsory. Rest of partners will provide their contributions to the meeting through the Project Coordinator. The agenda of the meeting was described in deliverable 4.

Minutes of this meeting was sent to MAP-MILLING's Projetc Officer, including all the presentations and materials prepared to be discussed.

3rd Meeting (Rome – 26th May, 2005)

3nd Meeting of MAP-MILLING's project was prepared for 26th May 2005. This meeting was held on the offices of Consorzio ROME in Rome (Italy).

This meeting was very important to prepare properly the first reporting period, from the technical point of view and especially regarding the economic justification (Forms C and audit certificates). Thus, the attendance of all the partners was completely necessary. All partners attended direct or indirectly (HARITASA, PRO AGRO and MUHLE) the meeting.

The agenda of the meeting is included below:

ROME (26th May, 2005)		
Agenda Item	**Remark**	**Time**
PART 1	***INTRODUCTION***	
Welcome	Introduction and welcome by host organisation – (CONSORCIO R.O.M.E.)	09:00 – 09:05
Review	Review of project objectives, management, partner roles and interactions – (CEZ)	09:05 – 09:15
Project situation	Brief presentation of progress status and situation: – D2 "Operational report about main pollutants in grain milling industry" – D3 "Technical report describing end-user requirements" – D6 "Catalogue of pollutants in the grain milling industry"	09:15 – 10:00

PART 2	***TECHNICAL SESSION***	
Discussion on Workpackage 2	Discussion on final revision of Workpackage 2, and of Deliverables 5 and 7.	10:00 – 10:20
Progress on Workpackage 3 and plan for next period	Presentation of WP3 status ("Design and development of improved measurement systems") and planning of future actions: – Part I: Pesticides (AZTI) – Part II: Mycotoxins and Acrylamide (IGV)	10:20 – 11:00
COFFEE BREAK		11:00 – 11:15
Progress on Workpackage 4 and plan for next period	Presentation of WP4 status ("Development of a Risk Management System for grain milling SMEs") and planning of future actions: – RMS (INTECHNICA) – Implementation on grain milling SMEs (ITA)	11:15 – 11:45
PART 3	***ADMINISTRATIVE SESSION***	
Technical progress report	Reporting requirements of every partner – (CEZ) Schedule for achieving the deadline (30th June) – (CEZ)	11:45 – 12:20
Financial report	Revision and questions about Draft Cost Statements - (Every partner) External audit requirements - (CEZ) Schedule for achieving the deadline (30th June) – (CEZ)	12:20 – 13:00
WORKING LUNCH		13:00 – 15:00
PART 4	***FINAL REMARKS & CONCLUSIONS***	
Coordination	Coordination with other projects – (CEZ) Web Site & MAP-MILLING logo – (CEZ)	15:00 – 15:30
Last agreements	Conclusions and specific actions for the coming period Date/place for the next meeting	15:30 – 16:00

Minutes of this meeting was sent to MAP-MILLING's Projetc Officer, including all the presentations and materials prepared to be discussed. The minutes were included in the Annexes of previous reports.

Web Site & MAP-MILLING Logo

During 3rd meeting, CEZ presented the three initial logos for MAP-MILLING project proposed by the graphic designers to the coordinator. These logos were explained to the partners and the consortium chose one of them by a vote. The final version is used in the cover page of this report and is shown in the figure below.

A draft version of the future Web Site of MAP-MILLING project also was presented in this meeting. Different requests were received from the partners, and final version of the Web Site has been launched during July. The main objective of the Web Site will be to provide information about the project activities to other stakeholders and to inform about results obtained. The address is:

www.mapmilling.net

Workshop – Reporting requirements for FP6 contracts (Brussels – 3rd June, 2005)

CEZ attended the workshop organised by the Research and SMEs unit (M4) and the Administration and Finance unit (M5) on the reporting requirements for MAP-MILLING project.

The meeting dealt with issues such as:

- activity report
- management report
- project deliverables
- financial statements
- audit certificates
- report review and acceptance
- electronic management of contracts
- deadlines

It was a 1 day meeting that took place on 3 June in Brussels, and guidelines provided will facilitate the reporting requirements and further acceptance of the reports by the Commission.

Approval of reports of the 1st Reporting Period

The coordinator received during December 2005 the formal "review Report" from the Commission with the Overall Assessment of the project and the main calculation of the 2nd pre-financing. The general assessment of the 1st reporting period was "Acceptable project: The project has achieved most of its objectives and technical goals for the period with relatively minor deviations".

The comments made both by RTD M4 and RTD M5 in this review will be discussed in the 4th Progress Meeting and main recommendations will be followed for the management of the project in the next reporting periods.

2nd Pre-financing distribution for the 2nd reporting period

The 2nd pre-payment from the Commission was received by the coordinator on December 2005.

According to the specific instructions of the European Commission regarding the pre-payment of MAP-MILLING partners, all partners have received their funding for the 2nd period in accordance with the contract provisions and the decisions taken by the consortium, except DEMETER and MUHLE (as they do not established the bank guarantees required by the European Commission).

Apart from this, DEMETER will receive the payment according to their costs accepted and audited during the 1st reporting period.

Change of representative of AEFI

The authorised representative person of AEFI, which has signed until last November all the documents, has left the association and a new person is taking her place (Mrs. Sirje Potisepp). The European Commission services were fully informed about this change and MAP-MILLING project sent the following documentation for the proper amendment of the contract:

- Copy of the legal document that authorises to Mrs Potisepp to represent AEFI since 5th December 2005 (Minutes of the meeting of the AEFI's Steering Committee).
- Brief translation to English of the document.

4th Meeting (Potsdam – 12th January, 2006)

4th Meeting of MAP-MILLING's project was scheduled for 12th January 2006. This meeting was organized by the partner IGV in Potsdam (Germany). This

meeting was mainly devoted to the most technical aspects of the project, so the participation of all the RTD Performers was crucial, since it helped to prepare and perform last part of research in order to fulfil the project objectives. The agenda of the meeting is included below:

POTSDAM (12th January, 2006)		
Agenda Item	**Remark**	**Time**
PART 1	***INTRODUCTION***	
Welcome	Introduction and welcome by host organisation – (IGV)	09:00 – 09:15
Review	Review of project management, comments and recommendations from the Commission– (CEZ)	09:15 – 09:30
PART 2	***TECHNICAL SESSION***	
WP 2: Preliminary Analysis of measurement techniques	Presentation of deliverables D5 and D7: – Pesticides D5 – D7 (AZTI) – Mycotoxins D5 – D7 (IGV)	09:30 – 10:15

POTSDAM (12th January, 2006)		
Agenda Item	**Remark**	**Time**
WP3: Design and development of improved measurement systems	Progress achieved and future activities till month 24 (15th June 2006): – AZTI – Development of Pesticides Measurement System – IGV – Development of Mycotoxins Measurement Systems + Analysis of Acrylamide formation	10:15 – 11:00
COFFEE BREAK		11:00 – 11:15
WP4 – WP5: Development of a RMS for grain milling SMEs & Integration	WP4: Progress achieved and future activities till month 20 (15th February 2006) – INTECHNICA WP5: Future activities till month 24 (15th June 2006) – ITA	11:15 – 12:30
LUNCH		12:30 – 14:30
PART 3	***ADMINISTRATIVE SESSION***	
D9. Mid-Term Assessment Report	Presentation and discussion of the draft D9. Mid-Term Assessment Report – CEZ	14:30 – 15:30
Coordination	Coordination with other projects – (CEZ)	15:30 – 15:45
Last agreements	Conclusions and specific actions for the coming period Date/place for the next meeting	15:45 – 16:00
VISIT IGV FACILITIES		16:00

Mid-Term Review (Brussels – 26th-27th April 2006)

The Commission Services organized a mid-term review on Collective Research Projects in Brussels on 26th – 27th April, 2006. The review included an assessment of MAP-MILLING project by external experts.

The review was attended by 4 representatives of the project: Mr. Cester (CEZ - Project general coordinator), Mr. Díaz (CEZ – Project technical assistant), Mr. Barranco (AZTI – RTD's coordinator) and Mr. Idareta (CEZ – Project administrative assistant).

Additional information was required for the review. A 6-month management report on the MAP-MILLING project for the period from 15/06/2005 to

15/12/2005, including a detailed breakdown of person months and costs, was submitted in order to better assess the status of the project.

On early June 2006, CEZ received the consolidated midterm review report of the two independent experts of MAP-MILLING project. Regarding the implementation of these recommendations, MAP-MILLING project proposed the following initial measures:

- Appointment of responsible partner and person for implementation of each recommendation (CEZ - Mr. Nacho Díaz: 7, 8, 10 and 11; AZTI - Ms. Begoña Alfaro: 1 and 3; IGV - Ms. Monica Springer: 2, 4, 5 and 6; INTECHNICA - Mr. Volker Troebs: 9; ITA - Mr. Javier Val: 12). See the list in Section 1 for reference on recommendations numbering.
- Development by each responsible of specific implementation measures.
- Presentation to the consortium in the next Progress Meeting (22nd June, Tallin, Estonia) and agreement among all the partners.
- Continuous evaluation of implementation measures in following progress reports

5rd Meeting (Tallin – 22nd June, 2006)

5th Meeting of MAP-MILLING's project has been prepared for 22nd June 2006. This meeting was hold on the offices of AEFI in Tallin (Estonia).

This meeting was very important to prepare properly the second reporting period, from the technical point of view and the economic justification. It also helped to highlight main RTD results of the project and to discuss the important work to be performed by the IAGs in the last year of project. Thus, the attendance of all the partners was completely necessary. The agenda of the meeting is included in the next page:

TALLIN (22nd June, 2006)		
Agenda Item	**Remark**	**Time**
PART 1	***INTRODUCTION***	
Welcome	Introduction and welcome by host organisation – (AEFI)	09:00 – 09:15
Review	Review of project management, comments and recommendations from the mid-term Review – (CEZ)	09:15 – 09:45
PART 2	***TECHNICAL SESSION***	
WP3: Design and development of improved measurement systems	Progress achieved and future activities till month 30 (15th December 2006): – AZTI – Development of Pesticides Measurement System – IGV – Development of Mycotoxins Measurement Systems + Analysis of Acrylamide formation	09:45 – 11:00
COFFEE BREAK		11:00 – 11:15
WP4 – WP5: Development of a RMS for grain milling SMEs & Integration	WP4: Progress achieved – INTECHNICA WP5: Progress achieved and future activities till month 30 (15th December 2006) - ITA	11:15 – 12:30
PART 3	***ADMINISTRATIVE SESSION***	
Coordination	Coordination with other projects – (CEZ) Exploitation of results: protection and dissemination of results – (CEZ)	12:30 – 12:45
Technical progress report & Financial report	Reporting requirements of every partner – (CEZ) Revision and questions about Cost Statements - (Every partner) Schedule for achieving the deadline (30th July) – (CEZ)	12:45 – 13:30
Last agreements	Conclusions and specific actions for the coming period Date/place for the next meeting	13:30
LUNCH		

5th Meeting (Tallin – 22nd June, 2006)

5th Meeting of MAP-MILLING's project was scheduled for 22nd June 2006. This meeting was organized by the partner AEFI in Tallin (Estonia).

This meeting was very important to prepare properly the second reporting period, from the technical point of view and the economic justification. It also helped to highlight main RTD results of the project and to discuss the important work to be performed by the IAGs in the last year of project. Thus, the attendance of all the partners was completely necessary.

The agenda of the meeting was included in the last progress report. Minutes of this meeting were sent to MAP-MILLING's Project Officer, including all the presentations and materials prepared to be discussed.

6th Meeting (Sukarrieta – 25th January, 2007)

6th Meeting of MAP-MILLING's project was scheduled for 25th January 2007. This meeting was organized by the partner AZTI in Sukarrieta (Spain).

This meeting was mainly devoted to the approval of last technical aspects of the project, so the participation of all the RTD Performers is crucial, and to define in detail the Dissemination and Training Activities to be performed by the consortium in the last part of the project.

The agenda of the meeting was also included in the last reporting. Minutes of this meeting were sent to MAP-MILLING's Project Officer, including all the presentations and materials prepared to be discussed.

7th Meeting (Brussels – 8th June, 2007)

7th Meeting of MAP-MILLING's project was scheduled for 8th June 2007. This meeting was organized by the partner CEZ in Brussels (Belgium), and the facilities of the regional government of Aragon in this city were used for the event.

This meeting was very important to prepare properly the final technical and financial reports, from the technical point of view and the economic justification. It also helped to discuss about the main RTD results of the project and to evaluate and assess the important work to be performed by the IAGs in the last year of project. The Project Officer of the project (Ms. Marta Iglesias) was also invited and was attending the last part of the meeting.

The agenda of the meeting is included below:

BRUSSELS (8th June, 2007)		
Agenda Item	**Remark**	**Time**
PART 1	***INTRODUCTION***	
Welcome	Introduction and welcome by host organisation – CEZ	09:00 – 09:15
Review	Review of project management and general comments – CEZ	09:15 – 09:30
PART 2	***TECHNICAL SESSION***	
WP7 – WP8: Dissemination and training	Comments on the Spanish Conference and Training Workshops – AIAA Comments on the European Conference – DEMETER Questions and discussion about the rest of National Conferences and Training Workshops - CEZ and rest of Consortium (discussion)	09:30 – 11:00
COFFEE BREAK		11:00 – 11:15
PART 3	***ADMINISTRATIVE SESSION***	
WP9: Technical progress report & Financial report	Extension of the project to cover last activities - CEZ Technical Reporting requirements of every partner (Final Report) – CEZ Revision and questions about Last Cost Statement - Every partner Schedule for achieving the deadlines – CEZ	11:15 – 13:15
Last agreements	Conclusions and specific actions for the coming period	13:15 – 13:30

Web Site updating

During the whole project duration, CEZ was in charge of updating the information shown in the MAP-MILLING's Project Web Site (www.mapmilling.net). As soon as the different reports were approved by the Commission, they were uploaded and were accessible to everybody.

The Web Site also became an excellent mean to disseminate different events, news and relevant information to the Grain Milling and Processing sector.

During the last period, the Web Site took all the intended size and impact thanks to all the materials resulting from Project Results, events organisation and dissemination activities performed.

Coordination activities

According to the contract negotiations, MAP-MILLING project was committed to establish a good and constructive collaboration with other projects that deal with complementary problems in food safety issues (e.g. MYCOSENS, PESTISENS, Mycotoxin Prevention Cluster, HEATOX…). This guaranteed synergies among the projects and optimum allocation of research resources.

This collaboration was based on regular contacts between MAP-MILLING's Project Co-ordinator (Mr. Cester – CEZ) and the co-ordinators of the other projects. The results and conclusions of these contacts were reported in each six-monthly technical report. Project Workprogramme was revised also according to these interviews.

During 3rd Progress Meeting, the coordinator explained the specific coordination actions taken during the initial period:

- Identification of existing projects on the MAP-MILLING strategic issues. Contact list updated every 6 months. The table below shows the table.

PROJECT		**PROJECT COORDINATOR**	**CONTACT DETAILS**
Name (Number)	**Period**	**Organization**	**Address**
MYCOSENS (QLK1-CT-2001-70556)	2001-09-01 till 2003-08-31	ADGEN LTD	Newes Gate (KA6 5HW) Auchincruive – Ayr (UNITED KINGDOM)
PESSTISENS (QLK1-CT-2002-70829)	2002-05-01 till 2004-04-30	EGI - Sociedade de engenharia e gestao da qualidade industrial, lda	Praceta Madre Deus, 52 (4405-195) Canelas- V. N. GAIA (PORTUGAL)
OTA PREV (QLK1-CT-1999-00433)	2000-02-01 till 2003-10-31	NATIONAL FOOD ADMINISTRATION	622 (751 26) Uppsala (SWEDEN)
DETOX FUNGI (QLK1-CT-1999-01380)	2000-02-01 till 2003-07-31	NATIONAL RESEARCH COUNCIL OF ITALY	Ufficio Entrate 7 Piazzale Aldo Moro 7 (00185) ROMA (ITALIA)
CONTROL MYCOTOX FOOD (ICA4-CT-2002-10043)	2003-01-01 till 2005-12-31	Centre de cooperation internationale en recherche agronomique pour le developpement - CIRAD	Avenue Agropolis (34398) MONTPELLIER (France)
MYCOTOXIN PREVENTION CLUSTER	Projects 3, 4 and 5	Applied Mycology Group, Biotechnology Centre, CRANFIELD UNIVERSITY	Silsoe (MK45 4DT) Beds. (UNITED KINGDOM)
HEATOX (CT-2004-506820)		Applied Nutrition and Food Chemistry - Center of Chemistry and Chemical Engineering LUNDS UNIVERSITY	Getingevägen 60 (221 00) Luna (Sweden)
GRAIN LEGUMES (CT-2004-506223)		BBSRC John Innes Centre	Norwich Research Park – Colney (NR4 7UH) Norwich (UNITED KINGDOM)

- Initial contact between CEZ and the other Project Coordinators. It was sent to all them a letter offering public information and asking for a regular contact.

- Exchange of information: exchange of public deliverables and other materials. CEZ sent at least Map Milling Project presentation (Deliverable D1) to all projects and exchange has been established with MYCOTOX Project.

MAP-MILLING fostered the exchange of information on the research lines performed by the project, but the level of reciprocity was not as good as initially expected, as only one project sent relevant information about their activities.

On the other side, final work of the project required of specific collaborations with other European research groups in order to optimise the progress in some fields as:

- Veterinary Research Institute, Brno, Czech Republic (Dr. Fránek): monoclonal antibody clone E2/G2 used in the assay of 2,4-Dichlorophenoxyacetic Acid.
- Universidad Politécnica de Valencia, Spain: specific monoclonal antibodies (LIB-DDT5.25) for the group of organochlorine insecticide DDT and the insecticide chlorpyriphos

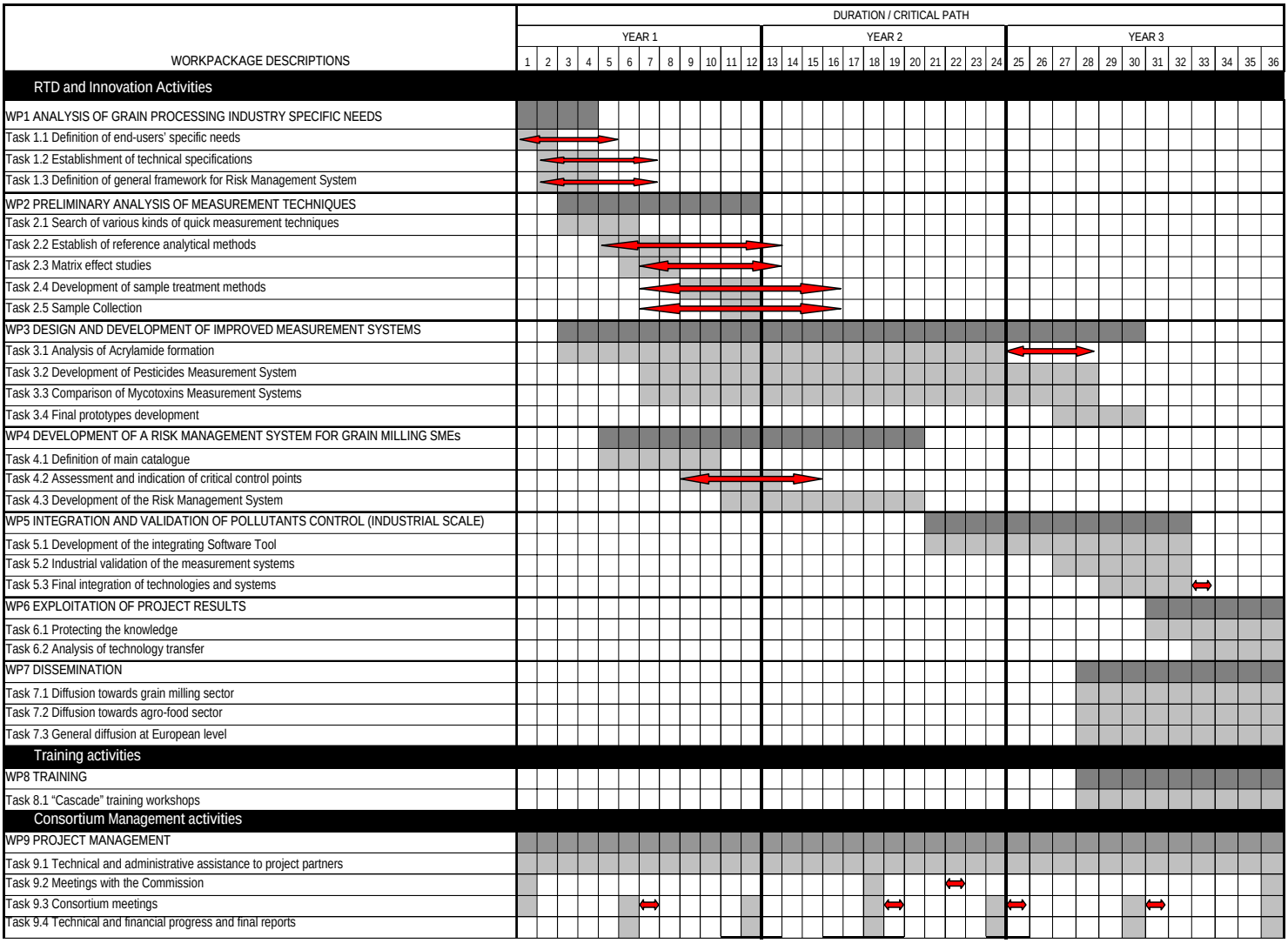
In January 2006, the project identified other research group that MAP-MILLING project will try to approach for establish collaboration. The foundation by the International Association for Cereal Science and Technology (ICC) has created an ICC Task Force on Mycotoxins and Sampling. Goal of the task force is the development of harmonised methods for screening methods, sampling, sample preparation and detection of mycotoxins in cereals.

First step of this Task Force is investigation of the status quo and identification of gaps (until Sep 2006). Please find more information on the ICC website at http://www.icc.or.at/task/summary.pdf. The partner IGV was responsible of contacting the Task Force and designing common activities for the upcoming events to be organised by this group.

First step was investigation of the status quo and identification of gaps (until Sep 2006). The exchange of information was intensified in the last period through the partner IGV, and the project was having contacts with them in order to improve the collaboration by joining common events and conferences.

Project Timetable and Status

The table below shows the progress of the project against the planned timetable, marking with red arrows main time deviations on specific tasks. This deviations and the status of each WP have been further explained in Section 2.



Section 4 – Other Issues

Contributions of IAGs, RTDs Performers and SMEs Core Group

During the project duration, all partners of MAP-MILLING project were involved in the different activities. These were their overall contributions:

- Project Coordinator: the specific work of CEZ, as Project coordinator has been focused in the WP 9 "Project Management" activities as:
    - Pre-financing distribution
    - Progress Reports
    - Preparation of Progress Meetings
    - Mid-Term Review
    - Coordination activities with other projects
    - Web Site updating
- IAGs: their work has been concentrated on WP 1 "Analysis of grain processing industry specific needs", on WP6 "Exploitation of Project Results", WP7 "Dissemination", WP8 "Training" and WP9 "Project Management", in the activities as:
    - Preparation and agreement of the Questionnaire.
    - Sending of questionnaires to target companies in each influence area.
    - Translation from English into national languages.
    - Contacting some companies by phone or through interviews.
    - Collection and initial analysis of answers received.
    - Contributing to the contents of D2 and D3.
    - To establish the best exploitation strategy for the Project Results and knowledge generated.
    - To explore other agro-food sectors that could be interested on a possible technology transfer of the project results.
    - To organise the National Conference (Spain, Italy and Estonia).
    - To organise the European Conference.
    - To develop specific dissemination activities on Project Results at different levels.
    - To define and establish the Multimedia Training Platform.
    - To organise the Training Workshops (Spain, Italy and Estonia)
- RTDs Performers: they have participated on all WP active in the 36 months with activities as:
    - Preparation of technical questions of the questionnaires according to their role in the project.

- Final analysis of answers received
- Preparation of main contents and conclusions of D2 and D3 (according to their role).
- Preliminary analysis of measurement techniques including establishing of reference analytical methods, matrix effect studies and design of assays.
- Initial development of pesticides measurement systems.
- Initial comparison of mycotoxins measurement systems.
- Definition of main catalogue of pollutants.
- Analysis and minimisation of the causes of acrylamide formation.
- Development of an efficient Risk Management System based on the catalogues of toxic substances and the developed quick and reliable tests for pollutants.
- Full list of recommendations for reducing the formation of Acrylamide.
- Development of ELISA format for detection of 2,4-D and chlorphyrifos-methyl in cereal products
- Optimization and good correlation between ELISA results and chromatographic results for detection of 2,4-D and chlorphyrifos-methyl in cereal products.
- Full report detailing the tested measurement systems for Mycotoxins, with an overall comparison between the studied methods and recommendation of the most interesting ones.
- Final development of the Software Tool on open-source integrating the project results
- Development of all the training materials.

- SMEs Core Group: SME's tasks in the current period concerns mainly to activities under WP1, WP2, WP4, WP5 and WP8:
  - Agreement of the Questionnaire.
  - Collaboration in diffusion of Questionnaire.
  - Detailed answer to the Questionnaire.
  - Validation and contribution to the results and conclusions obtained in D2 and D3.
  - Preparation and planning of samples needed for assays of RTD Performers.
  - Collaboration in the general framework of RMS and Guidelines.
  - Collaboration in the definition of Software Tool requirements.
  - Validation and assessment of the whole approach integrated in the final industrial solutions.
  - Collaboration in the Training activities performed in the different countries

Extent to which work of RTDs has benefited of large communities of SMEs

The food and drink industry is a leading industrial sector in the EU, with an annual production worth almost 600 billion €, or about 15% of total manufacturing output. EU is the world's largest producer of food and drink products. The food and drink industry is the third-largest industrial employer of the EU with over 2.6 million employees, of which 30% are in Small and Medium Enterprises (SMEs).

There are more than 26,000 companies, and in general terms, more than 90% of the food and drink companies are SMEs. The subsector of grain processing/milling industry in EU has a production of approximately 21 billion euros and an added value of 4 billion euros, in approximately 2,500 companies.

These are the main benefits that will be obtained for the SMEs:

- Increase in Knowledge: the main pollutants that can be encountered in the process, what are critical points on this process regarding product safety, and what the most appropriate treatment is. Risk management system will modernise obsolete management models of the sector.
- Increase in Technology: both in pollutant measurement and in treatment systems. The project will have a special impact in technological aspects, as agro-food SME's show in general very traditional models of economy, with little technological intensification.
- Increase in Competitiveness: providing an answer for the market with products of a reliable and controlled quality. The project will result in a reduction of costs (of time and money) in the trials usually made in external laboratories. It will also allow SME's to take decisions on real time, improving efficiency of the whole process.

The current project will have a direct impact in the European SMEs belonging to cereal flours sector, and the rest of the agro-food sector will take advantage of the experience obtained in the project by means of the dissemination phase and knowledge transference.

Balance of work between RTD Performers and IAGs/SMEs

As it was foreseen in the initial proposal, the balance of work between RTD Performers and IAGs/SMEs is slightly concentrated (52% of costs) in favour of RTD's, as IAGs has mainly participated in all the exploitation, dissemination and training activities under WP6, WP7 and WP8 and SMEs has contributed in WP1, WP2, WP4 and WP5.

On the other side, RTD Performers have used strongly their resources during the first and second year in order to achieve the results and objectives of the project as soon as possible for beginning the training and dissemination activities during third year (in charge of IAGs). The use of RTD Performers resources during third year was significantly reduced, as the in order to complete and achieve the final results and objectives of the project on due time, so the training and dissemination activities can be performed during third year.