DoReMi - Low Dose Research towards Multidisciplinary Integration

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1 Executive summary

Humankind is exposed to low levels of ionising radiation from natural sources. Other low dose exposures can be received from medical and industrial uses of radiation, as well as from high altitude air travel. For the general public natural background and medical sources predominate. As even these low doses can be potentially damaging to health it is important to understand the risks radiation poses at all exposure levels but especially at the low levels that predominate, and where direct evidence of health effects are masked by the high incidence of sporadic diseases. An improved understanding of the risk at low doses is important for the further development of an effective and transparent system of radiation protection.

The aim of the DoReMi consortium has been to promote the sustainable integration of low dose risk research in Europe, in order to facilitate efforts to resolve the key policy questions identified by the 'High Level Expert Group (HLEG) on Low Dose Risk Research' (www.hleg.de). These questions are the shape/s of cancer dose-risk relationship/s, variation in risk between individuals, differences in tissue sensitivities for cancer, effects of radiation quality, risks from internal exposures and the risks of non-cancer effects. The research activities of DoReMi have focused on the research areas identified by the HLEG as being the most promising in terms of resolving the stated key policy questions. DoReMi has provided an operational tool to continue the development of the MELODI platform (Multidisciplinary European Low Dose Risk Research Initiative) that represents the major national bodies and research programmes with a long term commitment to low dose risk research in Europe. The Joint Programme of Activities (JPA) of DoReMi has included: (i) a Joint Programme of Research (JPR) covering the research priorities (key questions) outlined above and including the sharing and updating of existing infrastructures; (ii) a Joint Programme of Integration (JPI) to promote sustainable integration between the key players in Europe; and (iii) a Joint Programme for the Spreading of Excellence (JPSE), covering in particular knowledge management, training & mobility and the communication of significant DoReMi findings to stakeholders and policymakers. The Joint Programme of Research addressed three main topics: the shape of dose response curve for cancer, effects of individual susceptibilities and the risks of non-cancer effects. Radiation quality, internal exposures and tissue sensitivities were addressed as cross cutting themes within these main research areas. The research activities have taken a multi-disciplinary approach, including interfacing with the broader (i.e. non-radiation) biological, toxicological and epidemiological research communities. A substantial proportion of the activities of DoReMi were dedicated to the joint programme of research as DoReMi took the lead towards sustainable integration of low dose risk research in Europe. In the longer term this will aid the resolution of the key policy questions in radiation protection.

2 A summary description of project context and the main objectives

The aim of the DoReMi consortium has been to promote the sustainable integration of low dose risk research in Europe, in order to facilitate efforts to resolve the key policy questions identified by the 'High Level Expert Group (HLEG) on Low Dose Risk Research' (www.hleg.de). DoReMi has provided an operational tool to continue the development of the MELODI platform (Multidisciplinary European Low Dose Risk Research Initiative) that represents the major national bodies and research programmes with a long term commitment to low dose risk research in Europe.

Strategic planning of DoReMi activities were carried out in close collaboration with MELODI. The long term Strategic Research Agenda (SRA) for European low dose radiation risk research has been developed by MELODI. DoReMi formulated research priorities in the Transitional Research Agenda (TRA) that focused on objectives that were feasible to achieve within the 6-year lifetime of the project and that are in areas where stimulus is needed in order to proceed with the longer term strategic objectives of the SRA.

Since the beginning of the DoReMi Network of Excellence in January 2010, there has been rapid progress in establishment of a European research platform to focus on questions of low dose risk. DoReMi continued the initial work of HLEG by contributing to the development of the long-term SRA of MELODI, and by establishing the more detailed shorter-term DoReMi TRA. The research agendas provided by MELODI and DoReMi have helped to identify priorities for low dose risk research not only by the organisations involved but also in national, European and global contexts. The planned enhancement of the DoReMi network through the calls for partners with new expertises resulted in the inclusion of 24 new beneficiaries. This enhanced the competence of the consortium in several key areas, by integrating research experts in biomarker identification, immunological/inflammatory pathways, and the effects of chronic low dose exposures, cataractogenesis, vascular effects, stem cells, epigenetics, novel mechanisms of genome reorganisation, as well as retrospective dosimetry. The three DoReMi competitive calls attracted proposals from 89 different organisations in 25 countries (including 21 European MS).

A substantial proportion of the activities of DoReMi were dedicated to the joint programme of research as DoReMi took the lead towards sustainable integration of low dose risk research in Europe. In the longer term this will aid the resolution of the key policy questions in radiation protection. DoReMi implemented research programs addressing the three key research areas: shape of dose-response curve for cancer, individual radiation sensitivity for cancer and non-cancer effects. All RTD activities also addressed the cross-cutting issues of radiation quality, tissue sensitivity and internal emitters. The research activities have taken a multi-disciplinary approach, including interfacing with the broader (i.e. non-radiation) biological, toxicological and epidemiological research communities. Several workshops were convened to develop strategies that focused on the most promising lines of research for the three areas. Experimental programs were launched and amended in all three areas and a total of 27 new tasks were amended in the project portfolio via the calls. The RTD approaches have been closely coordinated through discussions on needs for research infrastructures and analytical platforms, as well as targeted stimulation of training and education of next-generation researchers at the European level.

The programme on shape of dose response for cancer had two overarching objectives: (i) to improve knowledge of low dose/dose rate radiation cancer risk in humans, and (ii) to improve low dose/dose rate risk projection models based on knowledge of the processes that drive carcinogenesis. WP5 was structured into ten tasks, expanding the project portfolio from original five. Dose and dose-rate response relationships of cancer-related processes were examined and special attention was paid in cellular and tissue processes that could modify the cancer outcome (non-linear responses), such as senescence (aging), non-targeted and systemic effects and effects on immune system. The different steps and in radiation carcinogenesis were addressed, ranging from initial events, stem cells and molecular pathways to preneoplastic changes and cancer among Czech and UK radiation worker cohorts.

Another important objective has been to provide a scientific basis for decision-making on the inclusion of individual sensitivity as a modifier of risk at low doses. Current risk models use LNT to extrapolate from high to low doses. By convention this assumes an equal risk distribution amongst all members of an exposed population. Consequently, the risk assessments delivered by the models can only represent the average risk across the population. Despite inbuilt safety margins the present state-of-the-art knowledge does not allow us to assume that exposed individuals with a greater natural predisposition due to age, gender, genetic background or interactions of these variables with lifestyle and/or environment are effectively protected. Consequently, we have striven to understand how, and to what extent, individual susceptibility influences the carcinogenic effects of low doses and low dose rates.

In a context of a lack of epidemiological evidence for the non-cancer effects at low-dose exposures, and recognizing that (i) multi targeted biological effects observed in chronically exposed experimental models with internal emitters are quite puzzling; and (ii) no convincing mechanistic explanations are available that can account for the findings observed, the overarching strategic objective of the DoReMi WP7 (non-cancer effects) was to implement a long-term, integrated approach involving several disciplines, namely, epidemiology, radiobiology, immunology and toxicology, for the purpose of risk evaluation for radiation-induced non-cancer effects. Therefore the scientific objectives were: (i) to design well-controlled molecular epidemiology studies having in prospect the identification of markers of the initial steps of low-dose radiation-induced non-cancer health effects, the record of biological non-radiation "risk factors" connected to diseases under study, and the monitoring of pertinent blood biomarkers of biological radiation effects; (ii) to promote, in the field of experimental radiobiology and radiotoxicology, high-throughput technologies (e.g. "omics") and systems biology approaches that would be expected to better describe the complexity of low-dose radiation-induced tissue level responses; (iii) to challenge the classical molecular DNA strand break paradigm in search for the mechanisms behind non-cancer effects, and in this way to promote research activities in the field of cell physiology (i.e. cell senescence, long term cell phenotypic changes), immunology and the radiobiology of intercellular communications and signaling; (iv) to differentiate radiation-induced tissue and cell responses due to adaptation to the radiation stress, from true adverse alterations, involved in pathological processes; (v) to support research on mechanisms of radiation action, adopting a multi-scale/system biology approach and putting the emphasis on the relationship between initial stochastic track structures of low and high LET radiations, early chemical/biological processes and long term pathophysiological effects such as inflammatory response; and finally (vi) to assess whether there are scientific arguments for replacing the classical "threshold" paradigm for non-cancer effects with the "non–threshold" paradigm.

The availability of suitable infrastructures for performing low dose risk research was specifically addressed by DoReMi. Experimental radiation research is highly dependent on the availability of appropriate radiation sources that are reliable, capable of delivering a range of radiations, are robust and accurate. Low dose research also needs access to well defined epidemiological cohorts, reliable databases and biobanks and as well the appropriate platforms for analysis. After the initial mapping of infrastructures and their availability, DoReMi has provided access to several new infrastructures that will enhance the European capabilities in addressing scientific questions relevant for low dose risk.

DoReMi also addressed Education and Training needs in low dose risk research and explored the best way of organizing an Integrated Training and Education Network. The need for a European MSc in radiobiology as an entry point for students into the low-dose research area was highlighted and, subsequently, a new MSc course was stared in Munich. A very successful line of action was the

organization of focused 1-3 week courses by DoReMi partners: altogether 39 courses were held and about 500 students trained during the DoReMi lifetime.

Dissemination of information on ongoing low dose risk research to the general public, the scientific community, policy makers and stakeholders has been an important part of DoReMi networking activity. More information on DoReMi activities can be found at the DoReMi's own website (http://www.doremi-noe.net/), which is still operational for some time after the end of the project. Through the website, DoReMi has promoted interdisciplinary interaction and increased European integration of research as well as facilitated the spreading of knowledge. The information and materials with sustainable value have been transferred to http://www.melodi-online.eu/DoReMi/home.html, which is kept up to date also in the future.

3 The objectives and main achievements of the period by each WP

3.1 WP2 Structuring of MELODI

This work package has been lead by Dietrich Averbeck, IRSN/CEA, France.

High Level Expert Group (HLEG) on Low Dose Risk Research recommendations were the basis for WP2 establishing MELODI as an open multidisciplinary platform in Europe with funding bodies and research organisations that are significantly involved in low dose risk research. The main objective was to set up an appropriate governance structure promoting low dose risk research in Europe using a long term sustainable and fully integrated approach. For this, the attraction and integration of adequate partners as well as the setting-up and regular updating of a Scientific Research Agenda, i.e. the Transitional Research Agenda (TRA) (for the 6 years of the DoReMi project) based on HLEG premises were essential. WP2 was also actively contributing to the long-term Strategic Research Agenda (SRA) of MELODI. Care was taken that not only scientific (WP5-7 related) aspects but also education and training (WP3) and infrastructure (WP4) –related aspects were considered in the TRA and SRA. Possible barriers of integration were to be overcome by providing suitable operational structures for the leadership and management of low dose risk research. Coordinated efforts with European and national funding were made to set-up effective mechanisms for providing the necessary financial support and sustainability of low dose risk research in Europe. This ensured that research groups performing multidisciplinary research with excellent scientific track records could be supported and integrated.

Scientifically speaking, the long term objective was to find answers to the key questions previously formulated by the HLEG: how robust is the current system of radiation protection and risk assessment, and how can it be improved? The DoReMi project quite effectively developed the notion of a European Research Area (ERA) in low dose risk research, also promoting education and training and suitable infrastructures, overall networking and interaction. In addition, the dissemination of low dose research concepts, methodologies and results was greatly promoted through the Joint Programme of Spreading Excellence (including networking, the organisation of workshops and DoReMi Website www.doremi-noe.net. This also supported the development and maintenance of an integrated Training & Education network (collaboration with WP3) and infrastructure network (collaboration with WP4). Links to other relevant international (US, Japan, Russia) and European low dose risk and radiation protection programmes were also established. Moreover, the original HLEG concept was

widened and new opportunities identified that facilitated sustainable integration of national, bi- and multilateral research programmes within an agreed Strategy Research Agenda (SRA) in the MELODI context. This included seeking an interface with stakeholders, investments in key infrastructures, coordinating overall knowledge management ensuring consistent research methodologies and education & training.

From the start, the objectives were to attract as much as possible partners interested in low dose health risk research to take an active part in a Joint Programme of Integration (JPI) and to include as much as possible all partners in (1) consolidating the Joint Programme of Research on the basis of discussions between partners and the broader scientific community during dedicated DoReMi and MELODI meetings, (2) identifying important research needs and promoting forthcoming new research lines, (3) regular updating of the DoReMi TRA through DoReMi statements and the DoReMi Roadmap, and actively contributing to the development and updating MELODI SRA and MELODI statements and the MELODI Roadmap, (4) defining research priorities in the DoReMi TRA derived DoReMi statements and MELODI SRA derived statements as important pre-requisites for the preparation of specific calls, and (5) contributing to the launching of competitive calls for integrative RTD projects coordinating EC and national funding and internal calls allowing attraction and integration of new partners to DoReMi and MELODI. Throughout this ensured the preparation, evaluation, prioritisation, integration and coordination of multidisciplinary research projects in the 6 year framework of DoReMi and for the long term research initiative MELODI.

The development of the TRA and the SRA and their regular updating were the most important tasks of DoReMi WP2 to provide the scientific background and tools for the different work packages (WPs) and MELODI activities.

Establishment of scientific research agendas for DoReMi and MELODI

The first DoReMi TRA (long version) was provided in September 2010 and then regularly updated by establishing shorter versions, i.e. the so-called DoReMi TRA Statements (version 1, September 2011, version 2, March 2013 and version 3, October 2013), and the DoReMi Roadmap for the different WPs (all versions are available at http://www.doremi-noe.net/transitional_research_agenda.html). The short statements (derived from the DoReMi TRA and the MELODI SRA) played an essential role for the preparation of internal and external calls in summarizing the main issues and focusing on the research priorities. Based on DoReMi outcomes, the final DoReMi TRA highlighting the DoReMi outcomes and research perspectives beyond DoReMi has been prepared for the end of the project.

The DoReMi TRA and statements have facilitated the establishment of MELODI SRA and statement research priority setting that have been crucial prerequisites for the preparation of EU calls. The short term DoReMi TRA proposed and/or reinforced many new research lines including immunological and medical diagnostic and therapeutic aspects, stem cell research, molecular epidemiology, modelling and systems biology as well as training & education activities and infrastructure availability, use and sustainability. The updated versions of the long term SRA and the Roadmap, since 2014 prepared with the MELODI SRA Working Group, have been essential for the organisation of future calls in the framework of OPERRA and the forthcoming EJP initiative in the framework of Horizon 2020. They also opened the way for new scientific initiatives and close cooperation with ALLIANCE, EURADOS, NERIS and EUTERP including radioecological, dosimetric, emergency management and medical aspects of research in radioprotection, respectively. Direct participation of DoReMi in the structuring of MELODI

and beyond has been also evidenced by the establishment of permanent links and active participation in the set-up of new MELODI structures such as the Scientific Committee, and the working groups on SRA and Roadmap updating, Education & Training and Infrastructures. All this contributed to highly increase the level of integration of new members and scientific research lines in coordinated low dose risk research and radiation protection in Europe.

The final Transitional Research Agenda (TRA) from DoReMi covers research strategies and scientific outcomes of DoReMi and perspectives for the future of low dose radiation health risk research and thus feeds in to the MELODI SRA. With this, altogether, the DoReMi TRA and statements as well as the MELODI SRA continue to have a strong bearing on the preparation of future EU calls in the domain of research on low dose risk and general radiation protection.

By definition, DoReMi WP2 served very much in establishing MELODI and ensuring permanent links between DoReMi and the MELODI Association. In fact, since various DoReMi consortium members became also members of the MELODI, the input of DoReMi in the structuring of MELODI and in all MELODI activities was quite strong from the start and increasing during the years. Lately, MELODI has been slightly restructured and since 14 January 2014 MELODI working groups on the SRA, Education & Training and Infrastructure have been established. WP2 is still actively involved in the MELODI SRA WG (the WP2 leader Dietrich Averbeck as invited external expert), and many DoReMi consortium partners are actively involved in all three WG. In addition, MELODI created an extended Scientific Committee where the DoReMi WP2 leader (D. Averbeck) has become an active member. First meetings of the WG have already taken place, and a new version of the MELODI SRA (a MELODI statement and the Roadmap) has been worked out considering all recent DoReMi and MELODI workshops and research developments. The 5th and 6th drafts of the SRA were presented at the MELODI Workshops 7-9 October 2014 in Barcelona (6th MELODI WS organized by CREAL) and 7-11 November 2015 in Munich (7th MELODI WS organized by HMGU). Both MELODI Workshops offered great opportunities to tighten the close links between DoReMi members and MELODI and the European projects concerning low dose risk research and radioprotection. For more information, see http://www.melodi-online.eu/sra.html.

WP2 was actively involved in the organisation of the first 5 MELODI workshops. 5th MELODI Workshop organised by SCK-CEN and held October 8-10 2013 in Brussels focused on topics in which most recent and significant breakthrough progress was made (research topics related to radiobiology, dosimetry, epidemiology, radiotherapy, radioecology, emergency planning and other fields of low dose risk research). Parallel sessions helped developing arguments for updating the Strategic Research Agenda and its further implementing into the various low-dose research groups throughout Europe.

It was the first including sessions with contributions originating from the entire field of radiation protection. With nearly 220 attendees, this was the largest workshop since the start of this initiative. Next to low dose research, there were presentations on radioecology, emergency preparedness and dosimetry, and Strategic Research Agendas of MELODI, ALLIANCE, EURADOS and NERIS were presented, indicating the interfacing research areas and/or disciplines between the four different areas of Radiation Protection. This has been paving the way for the set-up and launching of the new European project OPERRA FP7 as well as the first steps for organizing the even larger future European Joint Programme (EJP) and CONCERT (http://www.concert-h2020.eu/) in the framework of Horizon 2020 thus opening the research area from low dose research (DoReMi and MELODI) to radioecology (ALLIANCE), dosimetry (EURADOS), emergency management (NERIS) and, very importantly, medical

aspects. Thus, in recent years the integration of research organisations and the structuring of MELODI have been progressing well. Now MELODI is involved in CONCERT as an independent legal entity.

Very importantly, the DoReMi TRA and statements as well as the MELODI SRA and statements worked on by DoReMi WP2 always included the important complementary issues Education & Training (WP3) and Infrastructures (WP4). The links between RTD work and these latter issues and the support by WP2turned out to be important for the successful enlargement of the DoReMi project and the attraction of new competences and research opportunities within DoReMi and for all MELODI related projects

Dissemination, publications and data management

In addition to RTD activities, the WP2 was also dedicated to dissemination activities. This included mainly the maintenance of the public DoReMi website, the preparation of DoReMi Newsletters, disseminating DoReMi announcements and results of dedicated workshops and conferences as well as support for invited external experts, providing input to DoReMi. An additional internal part of the website was reserved for DoReMi members only facilitating management of scientific and operational issues.

The public DoReMi website available at www.doremi-noe.net served to inform stakeholders and the general public of DoReMi activities and to provide scientific information about low dose research. It provided information on DoReMi activities, the TRA and TRA statements, each work package, infrastructures, training courses, jobs and opportunities and relevant news in the field of radiation protection. In addition, the announcements and supporting materials for the competitive calls for new partners were also published there.

As the scope of the DoReMi website was quite vast, it also provided information on activities organized by various other actors in the field of radiation protection. In recent years, it developed into a central site for information on low dose research in Europe. Moreover, the website provided detailed information on scientific outcomes of the DoReMi project in terms of publications in international peer-reviewed journals, thus increasing general visibility of the DoReMi project. Undoubtedly, the development of the website has been a great success with a steady increase of visitors reaching already in 2014 about 2000 unique visitors per month.

As very useful tools of dissemination a, DoReMi Newsletters were prepared 3-4 times a year. They served widening the scientific community, providing information on various activities in the field of radiation protection, organised by DoReMi, MELODI or other actors. All the issues can be found at http://www.melodi-online.eu/DoReMi/Newsletters.html.

DoReMi Barometer of publications

In order to be able to follow more directly the scientific production of DoReMi, a DoReMi publication "barometer" was set up indicating not only the actual number of peer-reviewed DoReMi publications but also their distribution according to selected key words and WP/ tasks.

By March 2016, there have been altogether 105 peer-reviewed publications disseminating the results of DoReMi, and more are expected to come. In order to illustrate the scientific progress of DoReMi achieved, the peer reviewed DoReMi publications were categorized by the following key words:
1. Cancer
2. Non-cancer
3. Individual sensitivity
4. Radiation quality
5. Tissue sensitivity;
6. Internal emitters (contamination)
7. Epidemiology
8. Modelling
9. Non-targeted effects (bystander)

In addition to key word, the publications have also been categorized according to different tasks in work packages: WP4 Infrastructures, WP5 Shape of dose response, WP6 Individual sensitivities and WP7 Non-cancer effects.

The results are presented in form of a DoReMi Barometer of the DoReMi publications and it is available at http://www.melodi-online.eu/DoReMi/Publications.html.

Looking by the key words, most of the DoReMi publications are related to key word "Cancer" (73 publications out of 105 are related to this key word). In the second place, there are the key word "Non-cancer" and Non-targeted effects (bystander) (42 publications), closely followed by "Tissue sensitivity" (40 publications). For the rest of the key words, the number of publications varies from 10 to 26.

Some analysis on the basis of key wording has also been performed. Most publications are on the impact of immunological and non-targeted processes regarding low dose radiation induced cancers (influence of microenvironment on cancer development or on treatments by radiotherapy). Less publications concern low dose and low dose rate and radiation quality on cancer or non-cancer effects. The number of publications concerning the development of biomarkers has increased favorably. Furthermore, there is clear progress in the mechanisms of low dose radiation induced cancer and non-cancers. Also, there is new information on the use of biomarkers in molecular epidemiology and the use of biomarkers for the detection of individual sensitivity among populations.

In order to be able to conclude on DoReMi outcomes, two main DoReMi WP2 concluding meetings were held before the end of the DoReMi project: a workshop for position papers on consequences on risk assessment and radiation protection 29-30 April, 2015 in Sitges, Spain (see http://www.doremi-noe.net/TRA and Consensus meeting for position papers.html), and a workshop on the implication of science to low dose health risk assessment and radiation protection, including the scientific and societal community and stakeholders (10 December 2015 in Budapest, Hungary) as an annex of the 4th Periodic and Final Meeting of DoReMi. The main scientific and operational outcomes of DoReMi were summarized and are described in some detail in the enlarged Final DoReMi TRA (see http://www.melodi-online.eu/DoReMi/Transitional%20Research%20Agendas.html). It can be seen that the DoReMi project has been performed solid pioneering scientific work to substantially increase our understanding of low dose ionizing radiation-induced health effects to the benefit of an improved future radiation protection.

3.2 WP3 Training and education

This work package has been lead by Andrea Ottolenghi, UNIPV, Italy.

Background

There has been a substantial decline in expertise both in the areas of radiation research and in academic teaching during the past decade throughout Europe, and internationally. Given the current plans to establish new built of NPPs and the increasing application of ionising radiation in medicine, there is an urgent need to maintain competence in radiation risk assessment, including training, and regain expertise in many areas of radiation research.

In 2009 the report of the High Level and Expert Group on European Low Dose Risk Research (HLEG) identified the problem in sustaining expertise in the science areas underlying research into low-dose radiation protection. They saw a need for an action to encourage new students and graduates into the research area, and to develop the depth and range of expertise of the existing research community. The Group proposed that research should be coordinated at the European level, including coordination of the support of E&T. The Network of Excellence (NoE), DoReMi, is the result of their recommendations, and Workpackage 3 puts in place a programme to address their concerns with respect to E&T.

The concept of a NoE is based on 3 different activities:
(i) a Joint Programme of Research (JPR)
(ii) a Joint Programme of Integration (JPI) and
(iii) a Joint Programme for the Spreading of Excellence (JPSE).

E&T plays an important part in each of the activities. Any research programme has a co-dependence on the E&T sector, providing the environment for the development of the students who will become the next generation of research scientists, and at the same time dependent on MSc and PhD students to carry out much of the research work. The integration that is at the heart of forming a network includes integration of universities with research institutes as an essential part. And finally, an essential function of the network is in spreading the excellence and an effective way of disseminating new knowledge is through offering training programmes.

Fundamental to DoReMi is the need to develop the structure of MELODI that will continue after the completion of the programme, to provide a sustainable support for E&T.

Work package 3 was motivated by all of the above drivers, and was formulated in such a way as to be as flexible as possible in order to be responsive to the needs and resources of the low-dose research community. It was envisaged at the time of the proposal that the form taken by the E&T support would evolve in response to dialogue with the stakeholders, and from discovering which actions were the most effective.

Review of the programme

The overall objective of the workpackage was as follows:

To develop a sustainable Integrated Training and Education Network (ITEN) that will be integrated within the DoReMi network, and will provide and coordinate high-level training for research scientists and a career structure that will attract and retain top-level graduates within the research discipline:

- Investigate the needs/demand for training;
- Make an inventory of current courses and educational/research institutes as candidate members of a new integrated virtual network;
- Set up an ITEN that optimally answers the need with available resources;
- Commence new (MSc, PhD) courses with seed funding;
- Develop sustainable funding to take the new ITEN into the future.

Task 3.1: (Months 1 – 12) Preliminary investigative work prior to identify the format of the ITEN
A Training and Education Committee (TEC) was formed from individuals from 6 of the DoReMi partners to carry pout the work of Task 3.1. Correspondence with stakeholders (universities and other research institutes with possible interest in low-dose research) was undertaken by way of a questionnaire. This did not produce useful results. It also became clear that any financial support for E&T activities could only be given to members of the DoReMi Consortium. Since there was no strong enthusiasm for setting up a new legal entity to carry out the work of the ITEN, the picture emerged of the ITEN effectively being formed from the network of DoReMi members, together with a management/guidance role for the TEC. The DoReMi network was programmed to grow significantly (from 12 initially to 36 by the end), so under this scheme a large proportion of institutions with an interest in low-dose research were sure to be included. This is the structure that was decided upon for the ITEN by Task 3.1. With hindsight, this can be seen as a good choice. There was no functionality that was found to be lacking later during the term of the NOE.

Task 3.2: (Months 6-60) Set up a new low-dose risk ITEN following the recommendations from the previous task
Under the structure for the ITEN set up in Task 3.1, an annual programme of E&T was undertaken.
It was recognised that the MSc level was key to attracting students from a science Batchelor's degree into the research area, possibly steering the most capable students into PhD research then career positions. There was a desire to facilitate the formation of a Masters course in radiobiology, to replace the European Masters course that was set up and run by Klaus Trott at UCL, London, from 1992 to 2012. Many of the scientists now working in the field received their introduction to radiobiology during the 20 years that the course ran. At the time of drafting the workpackage it was seen as a possibility that the ITEN could be a stand-alone (possibly virtual) academy able to confer degrees. This is not what happened, and in the event a Masters course in radiobiology was developed by DoReMi partners at the Technical University of Munich and held the first enrolment of students in 2015. More details on the course can be found at:
http://portal.mytum.de/studium/studiengaenge_en/radiation_biology_master

Central to the programme of the ITEN was the annual call for partners to host short (one- to two-week) courses on subjects related to their particular area of expertise. This was very successful. During the 6 years of DoReMi, there were 5 calls, resulting in 38 individual courses, attracting over 500 students, mainly from Europe, but also from non-European countries when space allowed. Each

of the students was able to experience first-hand working in new areas of low dose research. The support from DoReMi meant that courses could be offered free of charge, and so were open to as wide a range of candidates as possible. From the feedback given, the courses provided an excellent opportunity to network with other students of similar interest, to learn from students from other disciplines, and to make contact with the lecturers possibly leading to subsequent project engagements. The courses covered the following range of topics:

- Human radiation genetics
- Molecular radiation carcinogenesis
- Radiation epidemiology and radioecology
- Cellular effects of low doses and low dose-rates with focus on DNA damage and stress response
- Interdisciplinary radiation research focussing on radiation protection
- Modelling radiation effects from initial physical events
- Non-cancer effects of low dose radiation
- Environmental Radiobiology
- Inter-individual responses to low dose ionizing radiation: from damage formation to biomarkers
- Data interpretation and uncertainty analysis

The concept of a fellowship structure to assist young scientists in developing a career in the research area was developed by UniPv and SU, with the setting up of the European Radiation Research Association for Young Scientists (EURAYS - http://eurays.eu/). The goals of EURAYS are:

- To create a network/community of young scientists to promote and facilitate career development within the radiation sciences;
- To support and facilitate scholarly exchange, interdisciplinary communication and establish collaborations, including research training amongst different laboratories across Europe;
- To support and facilitate the participation of members at national and international schools and training courses;
- To encourage the active participation of members at radiation science relevant meetings;
- To establish collaborative links and awareness of European radiation research projects and other societies for young scientists in radiation research;
- To have a link between the young scientist community and the decision making bodies of European Projects.

Task 3.3: (Months 24 - 72) Develop sustainable funding in collaboration with WP2

The objective of DoReMi Workpackage 2 was to create a sustainable integrated approach to low-dose research in Europe through the MELODI platform. Accordingly, Task 3.3 made use of the structure of MELODI to set up a body to continue supporting and setting priorities for E&T beyond the end of DoReMi, and in 2014 a Working Group for Education and Training was set up under the rules of MELODI.

The membership of the WG was:
University of Pavia, Italy: Andrea Ottolenghi (Chair) and Vere Smyth (Secretary)
Helmholtz-München, Germany: Michael Atkinson (Vice Chair)
SCK•CEN, Belgium: Michèle Coeck
URV, Spain: Victoria Linares

PHE, UK: John Moody
NRIRR, Hungary: Geza Safrany
BfS, Germany: Annemarie Schmitt-Hannig
NCRPP, Bulgaria: Nina Chobanova
IST/ITN, Portugal: Pedro Vaz
Stockholm University, Sweden: Andrzej Wojcik

The primary purpose the E&T WG was to:

- Ensure the continuing existence of a body that would oversee the support of E&T in the Research Area beyond the term of DoReMi;
- Set out the plan of how E&T should be developed and managed in the context of the overall aims of the low-dose research area;
- Ensure E&T is integrated into the MELODI SRA;
- Ensure E&T support within new H2020 calls.

The work plan included drafting a section on E&T for the MELODI Strategic Research Agenda (SRA), and production of a stand-alone document setting out a strategic agenda for E&T in support of the aims of MELODI. The policy element key to sustainability was the putting of the case that E&T must be an integral part of all RTD project work: that RTD relies heavily on the contribution of work carried out by PhD students, and in turn new research provides a vital educational environment for students. These policies were instrumental in gaining significant support for E&T in the European Joint Programme for the Integration of Radiation Protection Research, 'CONCERT', funded under the Euratom programme Horizon 2020 (http://www.concert-h2020.eu). Drafting of the CONCERT workpackage dedicated to E&T was led by UniPv, with assistance from members of the MELODI E&T WG. The work package tasks will enable a continuation and extension of the initiatives first developed in DoReMi.

When Task 3.3 was initially drafted, it was hoped that the financial support for E&T through MELODI would be come from the MELODI partners, or from national funding bodies. However, unlike the provision of training for operational radiation protection, where there is generally a regulatory requirement and industry accepts the need to pay, E&T actions to attract new students into the research area would be unproductive if they were offered on a cost-recovery basis. Supporting the competence and expertise in an essential research area must be seen as a public good. Therefore the focus of this task was maintained on the Commission, through the Euratom programmes.

Task 3.4: (Months 24 – 72) Oversee management of ITEN during the period of transition to sustainable funding

The EJP CONCERT began in June of 2015, just as the last series of DoReMi courses was finishing. So the mechanism was set up to transfer the funding support previously provided by DoReMi to the new programme. This has now been achieved, and support for short courses, will continue for a further 5 years under CONCERT. The annual call engages a similar format to that used successfully in DoReMi, now incorporating the interests of the other partner platforms ALLIANCE, NERIS, EURADOS, and a new medical RP platform. The E&T workpackage will also make provision for student travel grants, integration of E&T into research projects, outreach to other parties, and targeted support of individual selected students.

Task 3.5: (Months 1-72) Funding training activities
The E&T activities carried out under Task 3.2 (setting up and managing the ITEN) included the series of short courses already described. In addition, support was provided for travel grants to present work at conferences or undergo training at another institution, and for student projects. The activities were directed by the TEC and the administration and financial arrangements were executed by the DoReMi Coordinator (STUK).

3.3 WP4 Infrastructures

This work package has been lead by Laure Sabatier, CEA, France.

Infrastructures are essential in order to fulfil low dose risk research objectives. Many types of facilities are required ranging from radiation facilities like large accelerators to data bases, human cohorts, tissue banks and platforms for sample analysis. The WP4 objectives are:

- To describe available facilities;
- To identify the needs for existing facilities and for new ones with WP5,6,7;
- To define with WP2 the infrastructures to be implemented within this NoE and those implemented with MELODI support in order to set-up sustainable funding;
- To facilitate access to infrastructures in collaboration with WP3;
- To launch calls for infrastructure accesses in collaboration with WP1,2,5,6,7;
- To give DoReMi participants access to the NMBU (previously named UMB, Norway) irradiation facility;
- To integrate the STORE platform into DoReMi and implement it as a pointer to existing datasets and biomaterial and/or a long-term data storage;
- To give DoReMi participants access to the SNAKE microbeam facility.
- To implement a Laboratory infrastructure for retrospective radon and thoron dosimetry

Task 4.1 Survey of existing facilities for low dose risk research
The survey was completed concerning the different types of infrastructures and D/M4.1 Review of infrastructures in DoReMi was submitted during month 23 (November, 2011). The website was updated to include the new DoReMi partners. The infrastructure lists established during the development of this tool will be re-used by the EJP CONCERT project to develop new communication tools that will further improve access to infrastructures for the radioprotection community.

Task 4.2 Characterization of infrastructure needs and roadmap of implementation
In the roadmap document (D4.3), we provide a review of the status of current and planned infrastructures for radiobiology research, including irradiation facilities, exposure cohorts, databases, ESFRI platforms and analytical platforms. We also review the process and criteria for selecting qualifying infrastructures, and discuss strategies for improving access to infrastructures.

- Promote the accessibility and use of available infrastructures among researchers *(through dissemination activities such as Education& Training, Website information and workshops)*
- Promote efforts for harmonization and integrative research through the use of available infrastructure *(through dissemination activities such as Education& Training, Website information and workshops)*
- Identify as yet missing infrastructures that need to be developed

- Improve reproductibility by supporting infrastructurues that meet essential quality criteria (example: 'Omics')
- Improve sustainability of rare but essential infrastructures (example: internal contamination acility for radon etc.)
- Improving access by focusing on infrastructures and facilities with recommended criteria
- Rationalizing the use of existing infrastructures and available financial resources
- Attract and train scientists to use the recommended infrastructures

Task 4.3 Implementation of DoReMi support activities for shared infrastructures
To assess the barriers to infrastructure access with the goal of further improving access to infrastructures, a recent survey of DoReMi scientists was performed. The results clearly highlight the need for targeted funding to access research infrastructures. DoReMi scientists have a high level of interest in using a variety of external research infrastructures and facilities including irradiation facilitates, omic's and imaging platforms, and archived data and samples from human and animal studies, but only if targeted funds are available. The survey also showed that lack of targeted funding is considered to be the greatest barrier to infrastructure access, followed by logistical issues, and the need for adequate on-site staff and laboratories.

Access to research infrastructures has been the major theme in DoReMi, The major means to support infrastructure access has been an extensive survey on available infrastructures, identification of gaps, and then financing access to specific infrastructures by providing funds for their upgrade and/or use, notably for low dose gamma irradiation and microbeam facilities as well as a retrospective radon dosimetry platform. A major workshop was held in 2015 to prepare the way for the CONCERT-EJP via OPERRA.

Task 4.4 Development and implementation of access to Infrastructure
An ad hoc approach to implementing infrastructure access has been taken following the identification of gaps and infrastructures at risk. This has consisted of the 3 external calls, each of which has brought in new irradiation infrastructures by providing beam time as well as the use of flexibility funding to aid in the integration of STORE into DoReMi as a step towards its sustainability. The tool developed (D4.4) was a 3-tier helpdesk to assist researchers to identify and access suitable research infrastructures for their projects. The helpdesk offered both passive and interactive means of communication, allowing researchers to consult a website or to interact directly with dedicated staff by telephoning the helpdesk or contacting a dedicated expert onsite at an infrastructure facility.

Task 4.5 - Open Access to the UMB low dose irradiation facility (FIGARO)
The FIGARO low dose/low dose rate gamma irradiation facility has successfully completed the pilot mouse irradiation experiment and is now being used to support two DoReMi projects for use of the Figaro facility that have been funded through internal DoReMi calls: OSTINATO (WP7) and CLOGICAT (WP5). These two projects have utilised the available time for DoReMi partners, and hence the project has been successful in stimulating access to facilities and collaboration between DoReMi partners.

Task 4.6 - Dose/Dose-rate Radiation Effects in Brain Cancer Risk (DDRE-BrainCancer)
The first results are now available from the low dose/low dose rate gamma irradiation study on brain cancer risk. Irradiations and necropsies for all mice enrolled in the tumour induction study (n=532) have been completed. Sample collection for molecular analyses (n=376) has also been completed as planned.

The results from histopathological analysis shows a trend towards a lower incidence of brain tumors in irradiated mice compared to controls. These findings suggest that radiation exposure at low doses elicits radio-protective responses in neural cells that may both counter the harmful effects of radiation and act to inhibit spontaneous cancer not related to radiation exposure, especially at very low dose rates. Furthermore, by accessing an overseas facility for mouse irradiation, Task 4.6 provides proof of principle for the added value offered by shared use of key infrastructures on a national and international scale.

Task 4.7 Low dose/dose rate gamma irradiation facility for in vitro and in vivo biological systems
Construction of the facility at ISS permits to perform the dosimetry tests and the first biological test.

Construction of the low dose radiation facility at SU has been performed permitting the licensing by the radiation authority and installation in the animal facility : calibration (dosimetry) of the radiation facility, training of the personal, approved ethical permission for mice exposure and performing the first animal exposures.

Task 4.8 - Integration of STORE into DoReMi as a trustable and viable database and/or pointer to biobanks and ascertain sustainability
During the STORE project, a considerable effort has been made and resources invested in the creation of a database (RBStore) to house data from existing, current, and future experiments as well as to serve as a pointer to archived biological material. The STORE project ended in October 2012, and the database is being transferred to DoReMi. While the database itself has been completed, it still contains a very limited amount of data. Investigators are invited to enter data from their experiments.
In order to promote the sustainability of RBStore and to make it a useful research tool, it is imperative that the radiobiology community uses it to store and analyse data as well as to find and request biological samples. To this end, data available from thousands of animal at CEA, SCK-CEN and HMGU were provided for RBStore.

Work is now ongoing with SUBI for collecting and collating information on data and biomaterial and its preparation for inclusion into STORE, and a contract has been negotiated and with the Kazakh Research Institute on Radiation Medicine and Ecology, Semey, to collect and collate information on data an biomaterial from a 3-generation study where the parents were exposed because of nuclear bomb testing.

Task 4.9 - Provision of ion microbeam irradiation facility SNAKE (MicroRAD)
The SNAKE microbeam facility has successfully established and commenced two collaborative projects with DoReMi partners. The first project is with IC to study the recruitment of the chromatin re-modelling factor ALC1 to sites of ion damage, while the second is with USAAR to visualize the recruitment of DNA damage response factors (pKu70, 53BP1) at gamma-H2AX-labelled regions by TEM (transmission electron microscope) following high LET irradiation of samples.

Task 4.10. Laboratory infrastructure for retrospective radon and thoron dosimetry
The laboratory infrastructure for retrospective radon and thoron dosimetry has been implemented, based on the use of stored indoors CDs/DVDs as retrospective radon and thoron detectors. The calibration equipment includes certified ^{222}Rn and ^{220}Rn sources, reference radon and thoron monitors, hermetic box and accessories.The survey design is oriented towards evaluation of dating

accuracy, collection efficiency and identification of potential bias/confounders. Making measurements in no less than 200 buildings, using different approaches for disk collection has been performed. .

It will be important to continue to work towards shaping and implementing policies which will positively impact on infrastructure access

3.4 WP5 Shape of dose-response

This WP has been lead by Simon Bouffler, DH-PHE (formerly HPA), UK.

The two over-arching objectives of WP5 were:

- To improve knowledge of low dose/dose rate radiation cancer risk in humans;
- To improve low dose/dose-rate risk projection models based on knowledge of the processes that drive carcinogenesis.

WP5 was structured into ten tasks, expanding the original five. Workshops were run that aimed to explore opportunities to further develop DoReMi and related research activities. Most tasks have considered processes that relate to carcinogenesis rather than human population studies of cancer risk, this has been necessary due to the limitations imposed by the available funds.

Task 5.1 has been examining dose and dose-rate response relationships of cancer-related processes including gene expression responses and senescence. With regard to senescence there were phase shifts in responses observed for the proteome response related to senescence that are best described as on/off effects at different doses/ dose rates. In addition, a priming dose effects was observed at the proteome level, suggesting a form of adaptive response may be acting. However it was also found that the stage of cellular differentiation influences the radiation induced proteome response. Work on gene expression developed a highly sensitive *in vivo* reporter system for p53-dependant transcription response based on luciferase expression driven by the p21 promotor; this shows genetic background dependence and time-dependence. Plasmid integration has been observed to increase in the dose range 5-200 mGy and fall above about 500mGy, which may relate to the upregulation of the variant end-joining pathway of DNA double strand break repair.

Task 5.2 has been investigating the contributions that non-targeted and systemic effects make to carcinogenesis following low dose exposures. A workshop helped guide this task and reviews on the role of 3D tissue models (Acheva et al 2014) and non-targeted effects in general (Campa et al 2013) have been published. Investigation of an *in vitro* adaptive response in primary human fibroblasts has indicated that the cytokine IL-6 and TGF-β do not play a role in adaptive responses as assessed by γ-H2AX foci formation (Dieriks et al 2011). While DoReMi has not placed substantial efforts on adaptive responses, they remain difficult to define and their impact on disease following low dose exposure is not clear. Most effort and productivity in Task 5.2 concerns effects on immune system and inflammation. Many papers have been published (13-17, 19, 21, 26, 41-43, 46, 47, 55) on modulation of inflammation. *In vitro* studies suggest modest reduction of inflammatory cytokine (IL-1β, TNF-α) production following 0.5-0.7 Gy exposure (Rödel et al 2012). Effects of low dose (0.1-0.5 Gy) irradiation but not 2 Gy irradiation on mouse macrophages increased chemotaxis but reduced migration (Wunderlich et al 2014). This was paralleled by increased anti-inflammatory cytokine profiles at exposures below 0.5 Gy. The impact of low and intermediate doses of radiation on attenuation of inflammation was however found to be dependent on the basal/individual

radiosensitivity on the basis of mouse strain dependent differences. These studies indicate that cytokine expression and response may be non-linear in dose-responsiveness. Linking these observations to disease manifestation following low dose exposure will be important. In this context the finding that low doses of radiation did not impact on outgrowth of tumours, but slightly modulated the infiltration of immune cells in existing tumours at later time points after radiation exposure, may be of relevance.

Further work on immune system T-cell receptor gene rearrangement was included towards the end of DoReMi. This work found that changes in the murine TCR repertoire are greater following acute low dose (0.1 Gy) compared to intermediate (1 Gy) dose exposure *in vivo*. No evidence was obtained suggestive of genetic instability (detected by illegitimate TCR rearrangements) following acute (0.1 Gy, 1 Gy) or chronic (0.2 Gy, 1 Gy, 3 wks) low and intermediate dose exposures *in vivo.*

Task 5.3 examined pre-neoplastic changes particularly in relation to acute myeloid leukaemia in the mouse though some potentially relevant studies on thyroid have been reported (Abou-El-Ardat et al 2011, 2012, Gerard et al 2012). In terms of monitoring disease progression over time substantial progress has been made in developing fluorescence reported gene systems to monitor critical gene loss events (Olme et al 2013, a,b). These continue to be developed to incorporate longer wavelength fluorochromes that are potentially more valuable for *in vivo* monitoring. Much effort has also been placed on integrated transcriptomic and proteomic analysis of mouse acute myeloid leukaemias with papers now submitted (Badie et al). This analysis highlighted the need to use primary *in vivo* passaged AML material to identify AML-associated alterations rather than derived cell lines which carried as many alterations associated with adaptation to culture as realted to AML. A 17 member signature that allowed AMLS to be distinguished from normal cells and primary material to be distinguished from cell cultures was developed, many of the members of this signature have known role in human leukaemogenesis, confirming the value of the mouse model in relation to the human disease. C-Myc was found to have a key role, paralleling the human situation.

A productive workshop was held on stem cells and DNA damage (Raj and Bouffler, 2012) that was influential in the development of an application for a full project (RISK-IR) that received EC funding in 2012.

Modelling approaches are of great importance to combine data from a range of sources and *Task 5.4* worked in this area. Lung cancer risk modeling comparisons of mechanistic and empirical models has had mixed findings (Eidemüller et al 2012). A model to describe the phenomenon of inter cellular induction of apoptosis has been developed (Kundrat et al 2011). Modelling of colon cancer risk in the Japanese A-bomb survivor cohort Colorectal cancer (CRC) in the Japanese A-bomb survivor Life span study cohort suggested that of the two known pathways contributing to CRC, microsatellite instability (MSI) and chromosomal instability (CIN), the latter predominated in radiation associated cases. Analysis of non-tumour thyroid tissue derived from biopsies from the UkrAm cohort identified some cases of BRAF mutation and RET/PTC rearrangements, moreover gene expression analysis suggested that MAPK pathway activation may act as an early indicator of papillary thyroid tumorignesis.

Task 5.5 concerned cancer risk associated with internal radiation exposure. A scoping workshop provided an important evaluation of priorities and helped build consensus (Laurier et al 2012). This effectively paved the way for *Task 5.8*, CURE – a concerted approach to evaluation of occupational uranium exposure risk. This piece of work confirmed the feasibility of pooled epidemiological analysis and of a molecular epidemiology approach to assess Uranium-specific risk. A full protocol was

prepared that includes consideration of harmonized dosimetric approaches, biomarkers, standardized operating procedures for biobanking, logistics etc. conducting the study is dependent on funding availability. An evaluation of one uranium miller cohort has been published (Kreuzer et al, 2014) indicating uranium exposure was not associated with increased death although organ doses were low.

Other work in *Task 5.5* concerned dose reconstruction for Czech and UK radiation worker cohorts. The Czech uranium miner study found that leukaemia and skin cancer risk followed a linear dose-response and the results have allowed an improved compensation scheme for uranium miners, The UK study converted some 300,000 paper records relating to UKAEA worker internal dosimetry into electronic format; databases now hold over 1 million sample records relating to internal dose. These will be available for future studies of internal exposure risk.

Task 5.6 considered radiation track structures and initial events in relation to the impact radiation quality on risk. Several publications have focused on the role and modeling of DNA fragmentation (Alloni et al 2011, 2012, 2013). In particular certain ions in the 125-225 keV μm^{-1} range have been found to produce large number of very small DNA fragments (Alloni et al 2013). Modelling of DNA repair kinetics following multiple fractionated exposures has been successfully implemented (Mariotti et al 2013). More recent investigation of potential damage to mitochondrial DNA in the 0.2-2 Gy range suggested that it is unlikely that direct damage to mitochondrial DNA will play a role in effects at these dose levels.

Investigation of the induction of chromothripsis by low dose radiation exposure (*Task 5.7)*, led to the development of a model system to determine the impact of radiation exposure on the frequency with which micronuclei are taken up and integrated in the genome of recipient cells, and the subsequent fate of re-integrated chromosomes. A potential role of TP53 status on uptake and integration of micronuclei has been observed. This work will help contribute to determining the importance of this novel genome destabilization/rearrangement pathway in low dose effects.

The role of microvesicles (MV) in mediating non-targeted effects was investigated in *Task 5.9*. MVs were extracted from *in vivo* irradiated mice (0.1 – 2 Gy) and characterized in terms of contents, effects and distribution on re-injection into hosts Western blotting confirmed the presence of TSG101 protein, a typical MV protein marker, in MV from unirradiated and irradiated mice. Radiation was found to increase MV release (but not size) independent of dose. No simple correlation between chromosomal instability and MV concentration has yet been observed. miRNA profiling will further define the cargos that MVs carry under different irradiation conditions and so contribute to identifying and defining their biological role, and potentially role in radiogenic disease.

Task 5.10 was a quantitative intestinal tumourigenesis study following low dose rate exposure using the FIGARO facility, a number of short term endpoints relating to genotoxicity were included also. Exposure to 1.7 or 3.1 Gy of low-dose rate (2.1 mGy h^{-1}) γ-radiation was found to lead to a 3-fold increase in MN formation in immature and mature red blood cells ($p<0.001$); However no increase in Pig-a mutation in blood cells, or DNA lesions was observed and the Apc^{min} mutation status did not influence the genotoxicity read-outs. Full pathological and quantitative analysis of intestinal tumours remains to be completed, with publication expected in the 2106 calendar year.

Overall WP5 has made substantial progress in exploring dose-responses for early responses to radiation. In common with many others, the challenge remains relating these early responses to

cancer and ensuring adequate experimental models of radiation carcinogenesis are available. A combination of experimental, epidemiological and modeling approaches continues to be required to develop a better understanding of cancer risk at low doses and dose-rates.

3.5 WP6 Individual sensitivities

This work package has been lead by Michael J. Atkinson, HMGU, Germany.

Objectives of the workpackage

Our overarching objective has been to provide a scientific basis for decision-making on the inclusion of individual sensitivity as a modifier of risk at low doses. Current risk models use LNT to extrapolate from high to low doses. By convention this assumes an equal risk distribution amongst all members of an exposed population. Consequently, the risk assessments delivered by the models can only represent the average risk across the population. Despite inbuilt safety margins the present state-of-the-art knowledge does not allow us to assume that exposed individuals with a greater natural predisposition due to age, gender, genetic background or interactions of these variables with lifestyle and/or environment are effectively protected. Consequently in the 11 tasks of WP6 we have striven to understand how, and to what extent, individual susceptibility influences the carcinogenic effects of low doses and low dose rates.

Major achievements of the DoReMi WP6 are:

Task 6.1 Identification and evaluation of research strategies for conducting future molecular epidemiological studies
Epidemiological studies that incorporate individual susceptibility into their risk assessments have not been performed. Molecular epidemiology offers a potential solution but has not yet been taken up by the radiation protection community. We held an international workshop to bring together epidemiological and biological experts in order to evaluate options for initiating molecular epidemiological research in low dose radiation risk. As a consequence of this workshop a position paper was produced and published (Pernot et al 2012). A second workshop to assess progress and take-up was recently held under the auspices of task 6.6 (see below).

Task 6.2 Identification of genetic modifiers of individual cancer susceptibility and their mechanisms of action
In order to evaluate the contribution of individual susceptibility to risk it is necessary to identify which factors operate, and to understand their mechanism of action. To do this task 6.2 explored a range of model systems:

We developed methodology for irradiating neonatal mice with reproducible doses of I-131, simulating exposure scenarios relevant to the Chernobyl accident. Using this method we investigated genetic susceptibility to radio-iodine-induced thyroid cancer. We demonstrated that genetic factors indeed strongly influence susceptibility of mouse strains to the development of both adenoma and carcinoma (Dalke et al 2012). A possible association between iodine deficiency (environmental factor) and the (genetic) risk of developing thyroid cancer was studied. The radiation responses of both thyroid and breast cancer cells were shown to be dependent upon cells being iodine deficient or iodine sufficient (Gérard et al Thyroid 2012).

We identified the Rb1 gene as a strong modifier of sensitivity of strain differences in radiation-induced cancer in a radiogenic mouse osteosarcoma model. This study established that the cell cycle regulating tumour suppressor Rb1 regulates the length of telomeres by a non-canonical mechanism. The result of the telomere attrition caused by inheritance of allelic variants of Rb1 was shown to be increased genomic instability after irradiation. This is the first mechanistic understanding of how susceptibility influences carcinogenic risk (Rosemann et al 2014, Gonzales-Vasconcellos et al 2013, Rümenapp et al 2012, Gonzales-Vasconcellos et al 2011).

As telomere length varies dramatically between individuals, and steadily decreases with age, this makes it a promising biomarker of individual susceptibility. We have evaluated mean telomere length and changes in telomere length following irradiation as potential biomarkers. This study (Sabatier et al) has provided preliminary evidence from a small cohort of healthy donor suggesting that in peripheral blood leucocytes the combination of initial telomere length and the subsequent change in telomere length following 2Gy of gamma irradiation is a possible predictor of individual radiosensitivity.

The tumour suppressor gene p53 encodes a protein essential for the early phase of the DNA damage response. Rare mutations in p53 predispose to radiation-induced cancer, but the effects of common allelic variants present in the population are unknown. The response to radiation, including post-radiation changes in TP53 mRNA processing, varied across a panel of human cell lines harbouring different TP53 genotypes. A number of p53 gene polymorphisms outside the coding region were identified in these cells, and their possible contribution to individual sensitivity to radiation examined. We have shown that polymorphisms in the 3' flanking region impact p53 pre-mRNA processing and after exposure to low doses of IR result in differences in the p53 transcripts being found between individuals (Sagne et al. 2013, Sagne, et al. 2014, Perriaud et al 2014).

Task 6.3 Evaluation of strategies for integrating mathematical modelling and molecular epidemiology to provide insight into the extent individual variability influences risk at the population and personal level
Although the LNT concept and two-stage clonal expansion models of radiation action are widely used a number of alternative approaches need to be considered to incorporate new knowledge on the contribution of individual sensitivity. A workshop was held in 2011 (together with task 5.4) to consider tools and processes needed to implement individual sensitivity into the existing models. One of the main conclusions of the workshop was that the pathways and processes influenced by susceptibility are too complex to be accommodated by simple models. As a consequence the workshop urged the adoption of systems biology approaches to modelling. A report of the workshop was delivered, an internal call for DoReMi was released resulting in the inclusion of Task 5.4. Moreover, the input into the SRA and TRA has led to the EURATOM project EpiRadBio and to studies in DoReMi on the modelling of the MAP kinase pathway as a start to systems analysis. (Abouelaradat et al 2012)

Task 6.4 Studies on the effects of modifying genes on low dose exposures
In conjunction with WP4 (Infrastructures) we developed an external call to identify international facilities suited to studies of the effects of chronic low dose exposure. This work was subsequently performed under WP4 and WP5 by ENEA in partnership with IES Rokkasho, Japan. These experiments, using the Patched mouse as a model system, to determine the effects of low dose rate are included in the report of WP4 (Infrastructures).

Task 6.5 Studies on the contributions of genetic and epigenetic effects towards sensitivity
The role of epigenetic changes in the cellular response to irradiation, in particular the connection between radiation, DNA repair and epigenetic changes, was the subject of a workshop "Contribution of epigenetic mechanisms that influence susceptibility to radiation induced cancer" held .in Stockholm on 24-26 April 2013, organised by SU. From this workshop we were encouraged to study microRNA expression as an epigenetic component of the radiation response, with the assumption that individual differences may be recognisable. Six hours after exposure of TPC-1 cells to low (62.5 mGy), moderate (0.5 Gy), and high (4 Gy) doses of X-rays a cluster of radiation responsive miRNAs were round to be regulated. Of these a small number were altered in a dose specific manner, including let-7g, whose mRNA target p21 was found to be regulated in transcript profiling microarray data (Abouelaradat et al 2012).

Task 6.6 Implementation of the DoReMi strategy for a large-scale molecular epidemiological study to quantify genetic contribution to individual susceptibility
Following on from the workshop organized under task 6.1 we held the second workshop of epidemiologists and biologists within DoReMi to review the results of biomarkers studies to date and draw lessons from the pilot molecular epidemiological studies conducted. During the course of the meeting organized and chaired by CREAL we reviewed biomarkers that were being developed and field tested in molecular epidemiological studies investigating the impact of low dose radiation exposure.

Task 6.7 Planning the future expansion of research portfolio
This task was designed to identify areas of research developing in adjacent scientific disciplines that would be of relevance for future low dose risk research involving individual susceptibilities. The first meeting was held in Stockholm in October 2011 on the subject of "Radiation and Systems Biology". This was instrumental in sustaining the nascent activities of the International Conference on Systems Radiation Biology, which in 2016 will hold its 8th meeting. The workshop also served as the initiating template for SOPRANO project in the OPERRA programme.

The second exploratory workshop, in conjunction with task 6.5, took place in 2013 in Stockholm. The chosen subject area was "Radiation Epigenetics", with a focus on supporting planning for future epigenetic research activities. Here the task 6.11 resulted from this workshop.

The final workshop was on "Mitochondria and Radiation" and was held in Munich in 2015, where the key role of the mitochondria in radiation responses was highlighted. A number of key questions for future research were identified. These included: Is the response of the mitochondria to radiation due to intrinsic changes or external factors? Are metabolic changes due to increased ROS generation, or are they due to the counterbalance of increased ROS removal depleting energy, calcium or both. Are the lipids targeted by oxidation acting as signal molecules? How are radiation-induced changes to mitochondrial function able to persist over several months? Are individual differences in mitochondrial response to radiation relevant for non-cancer and cancer diseases?

Task 6.8 Predicting individual radiation sensitivity with Raman microspectroscopy
Our search for biomarkers has focussed upon the relatively new technology of Raman spectroscopy to identify cellular components associating with an increased sensitivity to radiation. In a pilot approach a panel of human lymphocyte samples were assembled and individual radiation sensitivity assessed in

vitro before acquisition of the Raman spectra. Comparison of the Raman spectra and G2 radiosensitivity showed a very good correlation. A support vector machine classification model delivered 94% sensitivity and 90% specificity. Investigation of spectral features associated with treatment progression following hormone therapy and radiotherapy showed that the effects of treatment stage were observable spectrally with different spectral features identified for hormone therapy and radiotherapy. In a first test on clinical material cancer patient blood samples were examined after radiotherapy for toxicity using Raman spectroscopy and classification models demonstrated that spectral discrimination between lymphocytes from patients displaying no/minimal toxicity and severe toxicity immediately after radiotherapy was possible. This is the first study to demonstrate the capability of Raman spectroscopy to assess individual radiosensitivity in lymphocytes of healthy controls and prostate cancer patients (Maguire et al 2015).

Task 6.9 Integrating radiation biomarker into epidemiology of post-Chernobyl thyroid cancer from Belarus

The dramatic increase in incidence of thyroid cancer in young people in those regions most contaminated by fallout from Chernobyl has provided much information about radiation induced thyroid cancer. Previous studies among thyroid cancer patients exposed to fallout from the Chernobyl accident in Ukraine identified CLIP2 overexpression as a possible biomarker of radiation induced thyroid cancer. We have conducted a study to validate CLIP2 as a biomarker in an independent cohort of post-Chernobyl childhood thyroid cancer patients from Belarus. We have studied CLIP2 associations with dose and age at exposure and have evaluated modifying effects of iodine deficiency and other factors. We have found that 84% of the PTC tissue samples (53 out of 63 cases) were CLIP2 positive in this independent cohort of cases of thyroid cancer that. This is numerically similar to the frequency previously reported for both UkrAm and Genrisk-T cases (Selmansberger et al., 2015). If all available subjects are considered, including those who subsequently declined to participate in the interviews, then 83% of tissue samples (75 out of 90 cases) were CLIP2 positive. The scope for interpretation of the value of CLIP2 was limited due to a number of circumstances, inn particular uncertainties in dose estimates and the small number of cases with full information. In spite of these limitations it appeared that the proportion of CLIP2 positive was greater amongst those older ATA and ATO, among males and, particularly, in areas of greater iodine deficiency. An association between radiation exposure and CLIP2 status could not be adequately evaluated in this study.

Task 6.10 Characterization of DNA lesions in the nuclear ultrastructure of differentiated and tissue-specific stem cells after protracted low-dose radiation

The biological impact of low doses of ionizing radiation on human health and the genetic factors influencing whole organism radiosensitivity at low doses are unclear. Using mouse strains that varied in genetic DNA repair capacity (C57BL/6, $ATM^{+/+}$, $ATM^{+/-}$, $ATM^{-/-}$, SCID) we have analysed DNA damage in differentiated cell populations and tissue-specific stem cells of healthy tissues after repeated low doses of radiation.

In all analysed tissues, the gradual accumulation of DNA damage with increasing doses of fractionated radiation was observed. No verifiable threshold-dose was detected, even in repair-proficient organisms (C57BL/6, $ATM^{+/+}$). The number of radiation-induced foci varied significantly between the different cell populations, suggesting differing vulnerability to ionizing radiation. During repeated low-dose radiation, cortical neurons in brain tissues of all mouse strains had a significant increase of persisting foci with cumulative doses, with most pronounced accumulation of large-sized foci in repair-deficient mice. Electron microscopic analysis revealed that persisting foci in repair-proficient

neurons reflect chromatin alterations in heterochromatin, but no persistently unrepaired DSBs. Repair-deficient SCID neurons, by contrast, showed high numbers of unrepaired DSBs in eu- and heterochromatin, emphasizing the fundamental role of DNA-PKcs in DSB re-joining, independent of chromatin status. In repair-deficient $ATM^{-/-}$ neurons, large persisting damage foci reflect multiple unrepaired DSBs concentrated at the boundary of heterochromatin due to disturbed KAP1-phosphorylation.Thus, multiple unrepaired DSBs account for large-sized foci in repair-deficient neurons, thus counting foci alone may underestimate extent and complexity of persistent DNA damage. Collectively, repeated low-dose radiation leads to the accumulation of persisting DNA damage foci in differentiated stem cell populations and tissue-specific stem cells, and thus may adversely affect healthy tissues and increase the risk of carcinogenesis. These findings suggest that even very low doses of DNA-damaging radiation increase the health risks of individuals, particularly of those with compromised DNA repair capacity (Grewenig et al 2015), (Schanz et al 2014) (Flockerzi et al 2014).

Task 6.11 Mechanism of low dose response to ionizing radiation and its significance in radiation protection (RADSENS)
The non-coding transcriptome was identified in the workshop on radiation epigenetics as a potentially significant source of biomarkers of individual radiation responses. In this task we have studied the value of microRNAs in clinical samples from radiation sensitive and radiation non-sensitive individuals. We examined epigenetic pathways in human leucocytes that are up- or down-regulated in response to doses in the mGy range. Here we identified 31 miRNAs that were differentially regulated between the groups of sensitive and non-sensitive donors in non-irradiated samples. Several other miRNAs that differ in their dose response between the sensitive versus non-sensitive group were also identified. The regulatory pathways influenced by the regulated microRNAs may indicate mechanisms of sensitivity. Based on the databases TarBase, TargetScan and miRecords interactions between proteins and those miRNAs differentially expressed in normo-sensitive and sensitive patients were identified by the IPA software. These interactions were further verified for seed sequence matches by manually searching in microrna.org and miRBase databases. A pathway analysis was done for the predicted targets and the five most significant pathways

3.6 WP7 Non-cancer effects

This work package has been lead by Jean-René Jourdain, IRSN, France.

Recognizing that the epidemiological evidence for the non-cancer effects of low doses is still suggestive rather than persuasive, the multi targeted biological effects observed in chronically exposed experimental models with internal emitters are quite puzzling, and no convincing mechanistic explanations are available that can account for the findings observed, the overarching strategic objective of WP7 was to implement a long-term, integrated approach involving several disciplines, namely, epidemiology, radiobiology, immunology and toxicology, for the purpose of risk evaluation for radiation-induced non-cancer effects.

Major achievements of the DoReMi WP7 are:

In *Task 7.1 – Structuring the research effort on non-cancer effects according to the HLEG roadmap: organisation of consultation/exploratory meetings and funding integrative RTD projects*, two exploratory workshops were organized with the view of editing recommendations on the most promising research lines to better understand the vascular damages and lens opacities that may result

from exposures to low doses of ionizing radiation. This task proved that even though the organization of exploratory workshops required considerable resources, bringing together basic scientists and clinicians with complementary skills to work together for a week on specific issues relating to a particular area of research was a productive way to identify unresolved scientific issues and reach a consensus between experts who usually do not collaborate extensively.

In *Task 7.2 – Preparation of a pilot study to conduct molecular epidemiology studies in vascular radiation damage,* as a result of think-tank meetings of epidemiologists and biologists, a detailed review on types of biomarkers and cohorts of persons exposed to low dose was performed and strategies to conduct molecular epidemiology studies in vascular damage were determined (Kreuzer M et al. 2015).

In *Task 7.3 – Feasibility study towards a systems biology approach of radiation response of the endothelium,* the results obtained demonstrate for the first time that acute low doses of X-rays induce DNA damage and apoptosis in endothelial cells. Moreover, a non-linear dose-response relationship for DNA damage has been shown, and the findings clearly indicate that cellular effects seen below 0.5 Gy, may have an impact on the long term current radiation protection system that assumes a threshold dose for non-cancer effects. The work performed within task 7.3 also confirms that an early stress response is observed after one week of exposure of HUVEC to 4.1 mGy/h, which is replaced by a more inflammation-related expression profile after three weeks and onwards. This early stress response may trigger the radiation-induced premature senescence previously observed in HUVEC irradiated with 4.1 mGy/h (Yentrapalli R et al. 2013). Furthermore, even exposures at lower dose-rate (2.4 mGy/h) significantly inhibit endothelial cell growth and induce premature senescence. Protein expression and metabolic pathway analysis revealed that the p21-mediated pathway and an altered PI3K/Akt/mTOR and Rho GDI signaling inhibition are triggered in premature senescence induced by low-dose rate exposure (Yentrapalli R et al. 2013). X-ray doses as low as 50 mGy induce DNA damage in primary endothelial cells without significant growth arrest; however, apoptosis is observed after a dose of 0.5 Gy (Rombouts C et al. 2013). A dedicated transcriptomic analysis points to the involvement of insulin-like growth factor binding protein 5 (IGFBP5) signaling in radiation-induced premature senescence (Rombouts C. et al. 2014). Finally, this task demonstrated that chronic gamma-irradiation induces a dose-rate dependent pro-inflammatory response and associated loss of function in human umbilical vein endothelial cells (Ebrahimian T. et al. 2015). The pathways and proteins involved in endothelial-low dose responses open the way to the development of biomarkers of radiation-induced cardiovascular diseases.

In *Task 7.4 – Pilot epidemiological study of lens opacities among a cohort of interventional radiologists and cardiologists,* a study (O'CLOC) including 106 interventional radiologists versus 99 unexposed was performed. For all participants, information regarding the risk factors for lens opacities was collected; for exposed people, information regarding the description of type and frequency of IC procedures and the radiation protection material used was also collected. In all participants, a slit lamp examination was performed to determine when diagnosed the LOCS III classification of the detected lens opacity. The O'CLOC study showed that the interventional radiologists have significant higher risk of developing a posterior subcapsular lens opacity compared to the unexposed group. This result wasn't found for nuclear and cortical lens opacities.

In *Subtask 7.4.1- Lens opacities: Methodology implementation (ELDO),* a method has been developed to estimate cumulative eye lens doses for past practices based on personal dose equivalent values, H_p (10), determined above the lead apron at the collar, chest and waist levels (Farah J et al. 2013). Using

anthropolymorphic phantoms it could be shown that eye lens doses correlate best with H_p (10) measured on the left side of the phantom at the level of the collar. In spite of a relatively high spread of the correlative dose estimates when using chest-left whole body dose measurements, the method appears to be very useful for first order of magnitude retrospective eye dose assessments in individuals exposed to high exposure levels. For estimating cumulative eye lens doses for past practices, the evolution of the X-Ray systems and procedures and their effects on eye lens dose have to be taken into account. Thus, health professionals should benefit from wearing specific eye dosemeters for accurate dose assessments. Both methodologies increase the precision of epidemiological studies on cataract development in accidentally or professionally exposed individuals.

In *Task 7.5 – Pilot study of external irradiation versus internal contamination effects on neurogenesis,* time-dependent analysis of gene expression following irradiation of 14 HIV neuron cell cultures revealed modulation of different pathways at different time points after irradiation. An up-regulation of p53-dependent genes 6h after irradiation was observed, which was attenuated at 14h and 24h after irradiation indicating the induction of cellular stress, which is known as the classical early response following oxidative damage such as radiation. Nevertheless no significant apoptosis was induced at 24h after irradiation indicating rather a cell cycle arrest and DNA damage repair. The down-regulation of genes involved in synaptogenesis, extracellular structure biogenesis and cytoskeleton organization at 14h after irradiation correlates with the reduction in neurite length and branching of neurites observed at the morphological level 24h after irradiation. Modulation of those specific genes 14h after exposure appears to induce morphological changes in neuron cells 10h later (24h after irradiation). These morphological changes are significant to irradiation doses as low as 0.1 Gy. Finally no dose-dependent gene modulation was observed 24h after exposure indicating that the transcriptional changes related to the neurite outgrowth delay are transient and persist for less than 24h. Also, new results (Samari N et al. 2013) obtained on the neuronal maturation and synapse establishment using moderate doses of X-rays (0.2- 0.6 Gy) revealed a new non-conventional radiation-induced cell death pathway, involving excitotoxicity of immature neuronal cells mediated by the activation of N-methyl D-aspartate receptors, that is independent of direct radiation-induced DNA damage. This apoptotic pathway involves the activation of calpain, a proteolytic enzyme thought to play an important role in neurodegenerative diseases (Parkinson's disease etc.). Interestingly, a certain link between calpain and caspase-3 activation has been established as well showing a cross-talk between different cell death pathways involved in radiation-induced neuronal excitotoxicity in the developing brain. In spite of this link, the calpain dependent apoptotic pathways appears to be specifically involved in the elimination of immature neuronal cells after ionizing irradiation, and thus may explain the high radiation sensitivity of maturing neurons during fetal development.

This task included also the study of the effects on neurogenesis of internal contamination with depleted uranium (DU). Two different contamination protocols have been implemented, the first one during the first 13 days of gestation (E13) and the second protocol covers the 21 days of gestation and 21 days after birth (PND21).

At PND 21, cognitive tests showed that exploratory and locomotors activities were not significantly affected by DU at 40 or 120 mg/l at PND 21 in comparison with control group. At PND 21, the spontaneous alternation doesn't differ significantly between DU groups (40 and 120 mg/l) and control rats, indicating no significant effect on memory. The significant decrease of the number of visits and the time spent in the open arms of the elevated plus-maze in DU at 120 mg/l compared to control suggested no effect an anxiety-like behaviour. The depressive-like behaviour, measured by the

immobility time, was also not affected by exposure to uranium in our experimental conditions.

Cell proliferation in neurogenic regions was assessed by immunohistochemistry for BrdU after in utero and postnatal exposure to low dose of radionuclide internal contamination. The results showed an active cell proliferation in both control and contaminated animals at E13 in the telencephalic region of E13 embryos and in the different regions of the hippocampal formation of PND21 pups.

At E13, BrdU staining was very intense and largely distributed in the developing brain. Histological analyses restricted to the telencephalic region revealed a slight disturbance of BrdU incorporation in the animal group contaminated with DU at 120mg/mL. It was also decided to quantify the number of BrdU positive cells in the dentate gyrus (DG) of the PND21 animals. Although the total number of BrdU positive cells tends to decrease in the DG from animals contaminated at 120mg/L compared with control group, this difference is not statistically significant.

Neural differentiation in telencephalic region at E13 was assessed by double immunohistochemistry for nestin (stem cell marker) and doublecortin (neuroblast marker). No major difference has been observed in the distribution of nestin and doublecortin staining in contaminated embryos compared with control embryos.

Neurosphere cultures made from neurogenic zones of E13 embryos and PND21 pups all give rise to primary and secondary spheres whether cells originate from control or contaminated animals. These results showed that contamination with DU at 40 or 120mg/L does not seem to affect the capacity of stem cells to form clones at both stages. No difference was seen in the number of primary or secondary neurospheres in cell cultures originating from control or contaminated E13 embryos. A slight decrease in the number of secondary neurospheres was observed in cultures made from PND21 contaminated pups compared to the controls. This experiment would need to be replicated to ascertain if there is a statistical difference.

Differentiation tests using specific markers for neurons, oligodendrocytes and astrocytes showed that whatever the origin of the cells is (E13 or PND21), stem cells retain their multipotency in the contaminated group. Finally, examination of Golgi preparations revealed no major difference of dendritic morphology in the principal areas of the hippocampus (CA1, CA3 and the dentate gyrus) in contaminated rats (40 and 120 mg/l) compared with non-contaminated rats (Legrand M et al. 2015).

In *Task 7.6 - Study on contribution of low dose X-radiation in induction of anti-inflammation,* an analysis of the anti-inflammatory effects of low dose ionizing radiation, after exposures of activated peritoneal macrophages from Balb/c mice to 0.5 or 0.7 Gy of X-rays showed a reduced release of inflammatory cytokines such as IL-1b and TNF-a, whereas this was not the case in the more radioresistant C57/BL6 mice (Frischholz et al. 2013). After differentiation of human monocytic cells (THP-1) into macrophages low dose X-rays decreased secretion of IL-1β has been observed in a discontinous manner with dose, most pronounced between 0.5 and 0.7 Gy. The decrease was accompanied by a reduced translocation of ReIA (part of the NF-kB complex) into the nucleus, and also the serine/threonine proteinase kinase B (Akt) and the mitogen-activated protein (MAP) kinase p38 were reduced. This shows that low doses of X-rays induce an anti-inflammatory phenotype of activated macrophages (Lödermann B et al. 2012; Rödel F. et al. 2012).

This task also included the aim to elucidate the mechanisms underlying the clinically documented low-dose X-irradiation (0.5- 1 Gy) anti-inflammatory effects on benign diseases and chronic degenerative disorders such as painful shoulder and elbow syndrome and calcaneodynia (inferior heel pain syndrome). Several clinical trials were performed to optimize single dose treatments in terms of clinical efficacy and radiation protection (Ott OJ. 2012, a, b; Ott OJ. et al. 2013, a, b; Ott OJ. et al. 2014, b, c). The results obtained in these prospective randomized trials show that a single dose 0.5 Gy is somewhat equally effective as a single dose of 1 Gy (or 3Gy). Following these trials, for treatments of benign painful elbow syndrome a single dose of 0.5 Gy and total dose of 3 Gy are recommended (Ott OJ et al. 2012, a; Ott OJ. et al. 2014, b), and for treatments of benign painful shoulder syndrome (Ott OJ et al. 2012, b; Ott OJ et al. 2014, c), achyllodynia and calcaéneodynia (inferior heel pain) single doses of 0.5 Gy (and 1 Gy) and total doses of 3-6 Gy are regarded as dose standard. A review of recent data reveals that low dose-radiation therapy (LD-RT) modulates immunological processes including leukocyte/endothelial (EC) cell adhesion, cytokine expression, apoptosis induction and the metabolism of mononuclear (PBMC) and polymorphonuclear cells (PMN) with maximum effect in the range between 0.3 and 0.7 Gy (Rödel F et al. 2012). For example, a single dose between 0.5 and 0.7 Gy has been shown to induce the expression of X-chromosome linked inhibitor of apoptosis (XIAP) and TGF-b 1, reduce the expression of E and L selectin from EC and PBMC, IL-1 or chemokine CCL20 secretion from macrophages and PMN. In most cases discontinous or biphasicv dose responses similar to non-targeted (NT) and bystander responses are observed (Rödel F. 2012, Rödel F et al. 2013). Interestingly, such exposures may also foster innate and adaptive responses against tumors (Rubner Y et al. 2012).

This task showed also that low and intermediate doses of ionising radiation reduce the expression of surface MHCII molecules on activated macrophages. When these macrophages get in contact with T cells they slightly reduce their proliferation starting at a radiation dose of 0.1Gy. By this, irradiated macrophages induce bystander effects in T cells. Supernatants (SN) of activated and irradiated macrophages only slightly impact on dendritic cells (DC): a decreased surface expression of CD40 on DC was observed after contact with SN of macrophages that had been irradiated with 0.01Gy, 0.05Gy, 0.1Gy, 0.3Gy, 0.5Gy, 1.0Gy or 2.0Gy. However no consecutive impact on T cell proliferation was detectable. This suggests that only mild bystander effects on DC had been induced by irradiated macrophages. A reduced adhesion of leukocytes (PBMC and PMN) to endothelial cells is considered to display a pivotal mechanism in the anti-inflammatory properties of low and intermediate dose irradiation. By applying activators of the redox sensitive transcription factor Nrf2 and ROS scavengers, reduced adhesion events, most pronounced after a 0.5 Gy exposure, were significantly abrogated indicating a correlation between a non-linear expression of major factors of the cellular anti-oxidative defense, ROS production and immune modulatory properties of ionizing radiation.

In *Task 7.7 – Low dose Gene Expression signature and its impact on Cardiovascular disease (LoGiC),* repetitive exposure to CT radiations did not lead to noticeable changes in aortic or ventricular dimensions, showing that radiation did not have a major impact on these parameters. Further analysis will have to address more subtle molecular and cellular changes that may result from radiation exposure.

In *Task 7.8 – Study on contribution of low dose X-radiation in induction of cataractogenesis and influencing genetic and cell communication factors (LDR-OPTI-GEN),* the results showed that lens epithelial cells express the telomerase enzyme and that the activity of this enzyme is reduced after irradiation in a dose-dependent manner. Furthermore, it was found that these cells show an increase

in telomere length with age, a trend that gets reversed after exposure of cells to doses above 0.02 Gy. It has been found so far that telomerase activity is present in HLE cells, and that telomere length increases both dose dependently and after each passage. HLE cells have effective repair ability especially at lower doses of 0.02Gy and 0.2Gy.

In *Task 7.9 – Low and moderate dose radiation effects on brain microvascular pericytes: epigenetic mechanisms and functional consequences (PERIRAD),* it was shown that DNA damage of primary pericytes, as evaluated by the gamma-H2AX assay, was dose dependent in the dose range of 0.1-2 Gy and persisted even 24 hours after irradiation. In the dose range of 0.01-0.1 Gy, it was also shown the persistence of damage up to 168 hours after irradiation and that these late damages were not dose-dependent any more, with low doses showing almost similar levels of DNA damage than high doses. The results obtained in this task could also prove in pericyte-endothelial cocultures that DNA damage levels were higher in pericytes compared to endothelial cells. Overall data suggest that although in terms of viability pericytes are radioresistant cells, due to persistent, unrepaired DNA damage they are prone to genetic instability.

Overall data from other experiments suggest that pericytes have important roles in brain inflammation both in acute (septic shock-like) and chronic, autoimmune-type inflammation and radiation exposure can modulate their response to inflammatory stimuli. It was also can concluded that radiation induced epigenetic changes (namely DNA methylation and differential expression of miRNA) are present in pericytes although not at a very pronounced level. Changes do not seem to be dose-dependent (Persa E et al. 2015).

In *Task 7.10 – Influence of a chronic LD and LDR exposure onto the development of Parkinson symptoms in genetically predisposed Pitx3-EYL/EYL Ogg1-/- mouse mutant (OSTINATO),* a significant impairment of motor coordination (Beam walk) in Pitx3Eyl/+;Ogg1+/- mice was observed after 0.2 Gy chronic neonatal LDR irradiation. This effect was less pronounced (reduction below significance threshold) in Pitx3Eyl/+;Ogg1-/- and at the same genotype but after pre-natal instead of post-natal irradiation. No effect onto mouse motor coordination was seen at the high dose (1 Gy cumulative dose). A reduction by 30% in the number of DA-neurons in SNC, but not in VTA was seen, but the effect has not reached significance yet. Additional samples are under investigation to test if the difference can be validated. At the moment it seems as if at the next time-point of follow up (12 months) no impairment of motor coordination was seen any more. Measurements of ROS-induced markers and miRNA-profiling are on-going.

In *Task 7.11 Epidemiological pilot study on radiation-induced cataract in interventional cardiology: validation of methodology explored in Sub-Task 7.4.1 (EVAMET),* a pilot epidemiological study of lens opacities within a group of 89 Polish interventional cardiologists was performed (Domienik J et al. 2016, in press). Regarding the analysis of data from occupational questionnaire the results show clearly which kind of data are already available and which kind of data are still lacking to evaluate as precisely as possible cumulative eye lens doses. A reasonable data concerning the frequency of various procedures, access type, geometry of X-ray tube and the use of radiation protective devices were obtained. In view of the above, in order to provide the credible estimation of cumulative eye lens doses extra information is needed. It concerns the information, still lacking in the literature, about the dose per procedure for less common, but possibly contributing significantly to the eye lens dose, procedures and not included in the previous studies analyzing the cumulated eye lens doses. Also the correction factors for the use of various protective devices (lead glasses and ceiling lead glass) and

different geometries (X-ray tube configuration, the use of bi-plane systems) are important and necessary when the direct doses on eyes are not at our disposal. All information presented above allows to plan an extra measurements which help, first, in assessing the eye lens doses for new scenarios (new procedures, geometries, etc. for which a little amount of data or no data at all are available) and, then, in estimating the cumulative eye lens dose.

This task aimed also to validate_the two approaches developed in ELDO project for retrospective assessment of cumulative eye dose lens and evaluation of corresponding uncertainties. To achieve this objective, the thorough measurement program was set up in routine practice with recruited interventional cardiologists. Every physician was asked to wear two dosimeters during each procedure he/she has been performing for at least two months (one measurement period): one near the eye lens which is closer to the X-ray tube and another one on the chest also on the side which is closer to the X-ray tube.

Approach based on the whole body measurements: in total the data (Hp(10) and Hp(3) doses) from 33 measurement periods were collected. For individual data set the equivalent eye lens doses Hp(3) were calculated based on the reading from Hp(10) dosimeter and ELDO coefficients specific for the position of Hp(3) and Hp(10) dosimeter during the measurements. The results were then compared with the Hp(3) dose measured in the same time period. The relative differences (RF) calculated as the difference between the measured and calculated Hp(3) values changed from +50% up to -190% of the measured dose. In 70% measurements the calculated dose overestimated the measured one.

Approach based on the questionnaire and the dose data for single procedure: validation of the second method based on the data from the questionnaire was performed for 25 physicians who worn the EYE-D dosimeter. The cumulative annual eye lens dose was calculated on the basis of occupational data from the questionnaire (concerning the mean number of procedures performed annually, the percentage of procedures performed with the use of ceiling suspended shield, position of the X-ray tube (above or below) and in the case of coronary interventions the access type (radial or femoral)) and the input data. The latter included the doses per single procedure of different type collected by NIOM during ORAMED project and later and various correction factors derived in parallel project launched in Poland. In the first attempt the mean doses per procedure were used. In the next step the calculated doses were compared with the measured ones extrapolated to the same time period (one year). The maximum relative difference (the difference between the measured and calculated dose) were about -550% of the measured dose which means that in most extreme case the calculated dose overestimates the measured one about 6.5 times.

In *Task 7.12 – Effect of low doses of low-LET radiation on impaired vascular endothelium (ELDORENDO),* the objectives were to evaluate the effects of low doses of X-rays on the structure and function of vascular endothelium in mice before and during the development of hyperglycemia, presuming that endothelial effects of low doses of low-LET radiation will afford protection against atherothrombotic complications of diabetes. The results obtained in the ELDoREndo project suggest that:

- db/db mice display hyperglycemia associated with alterations in lipid profile (elevated total cholesterol and triglycerides), inflammation (elevated TNF-α, decreased IL-4), endothelial dysfunction (impaired NO-dependent function, elevated endothelin-1, reduced thrombomodulin).
- WBI at the tested low doses of X-rays does not induce significant and consistent effects of any of the measured parameters in db/db mice.

- If there is any beneficial effect of low doses of low-LET radiation, it can be related only to a total of 0.05 Gy and to the effects on early diabetes that showed some tendency for improvement that was, however, not consistent and in most cases not significant.

To confirm or exclude the possible beneficial effects of low doses of low-LET radiation on endothelial dysfunction in diabetes, further studies need to be conducted with a) considerably larger experimental groups (at least 25 animals per group) and b) different exposure schemes (including chronic irradiation for e.g., 4-8 weeks) performed at the facilities now available at some of the DoReMi partners' institutions.

In *Task 7.13 – Low-dose ionizing radiation-induced cataracts in the mouse: in vivo and in vitro studies (RadCat),* the objective of this task was to establish the dose-response curve of radiation-induced cataracts resulting from low-dose exposure (from 0 to 0.5 Gy) and to quantify incidence and mechanisms of cataract formation in a mouse model following a single low dose exposure. Also, taking advantage of the established dose-response curve, this task aimed to investigate for the lifetime of the mice the transparency of the eye lens by the most sensitive method (i.e. Scheimpflug imaging), to perform in a cohort of healthy heterozygous mutant mice suffering from a novel mutation in the *Ercc2* gene (also known as *Xpd*) cytogenetic analyses including telomere length determination in bone marrow samples from irradiated and non-irradiated samples, to evaluate the induction of chromosomal aberrations in a cohort of wild-type mice, and to perform micronuclei analysis in ex-vivo human lens epithelial cells put into culture.

Mice were sacrificed at the scheduled time points (4 hrs, 24 hrs, 12 months, 18 months & 24 months post-irradiation). The eyes were embedded in plastic for histological and immunohistochemical analysis. Individual bone marrow samples from the different time-points were distributed to two partners involved in this task for chromosomal analysis and telomere length measurement.

Even at the highest dose of 0.5 Gy no differences in lens density were observed among the groups (irradiated vs. non-irradiated, females vs. males or wild types vs heterozygous mutants). In contrast, the retinae of the irradiated (0.5 Gy) mutant mice were statistically significantly thinner than the non-irradiated controls; this effect was not seen in the wild-type mice.

To date the results of the chromosomal analysis has showed that a dose response was observed in the majority of samples (wild type and mutant mice) at 4 hrs following 0.5Gy irradiation. Moreover, the mutants (male & female) irradiated by 0.063 Gy demonstrated a higher number of chromosomal aberrations than expected from a linear dose-response relationship. However by 24 hrs, damage levels had returned to near background levels in all groups with exception of female wild type. In contrast, at both 12 & 18 month time-points, the frequency of aberrations was again higher in all groups compared to the untreated control counterparts with exception of female wild type, with highest levels observed following 0.5 Gy irradiation. Analysis of the available 24-month samples is in progress. A more detailed statistical analysis will be performed when all samples have been analysed.

As of the average telomere length at 4 hours after radiation, it was showed that the male samples have shorter telomeres compared to female samples. It can be seen that the wildtype samples have longer telomeres. The same trend in telomere length as seen in 4 hours after irradiation can be seen in samples 24 hours after irradiation. Female samples have longer telomeres compared to male samples and wild-type samples have longer telomeres compared to mutant samples. Also, the samples showed

that telomere length decreases after 24 hours of irradiation in both female and male samples. When comparing the mutant and wild-type samples a decrease in telomere length can be seen in samples 24 hours after irradiation.

3.7 WP1 Project management

This work package has been led by Sisko Salomaa, STUK, Finland.

During the course of the project, the coordinator has performed various overall legal, contractual and administrative managerial duties, including being responsible of overall financial management and management of the flexibility funding, establishment and implementation of the Quality Assurance procedures, organising the periodic meetings as well as meetings of the General Assembly, Management Board and External Advisory Board, measuring the success of integration by using performance indicators, and preparing the contingency plans.

As part of Task 1.1 Overall legal, contractual and administrative management and Task 1.2 Overall financial management, the Coordinator has taken care of the day-to-day management of the project. This has included the preparation of the Consortium Agreement (original and two updates), scientific and financial reporting towards EC and also internally within project, administering the Community's financial contribution and processing payments to partners, administering the inclusion of new partners and the legal changes of existing partners as well as the preparation of altogether 8 amendments for the grant agreements. In addition, the Coordinator has continuously supported the partners in various ways, responding to their daily questions and assisting them in legal and administrative matters.

In order to support the project management, the Coordinator established the quality assurance (QA) procedures (Task 1.3). The function of the QA Handbook was to clarify and summarise the various legal rules as well as administrative and financial procedures established in the Grant Agreement and in the Consortium Agreement from more practical point of view, making them easier for the partners to follow and understand. The QA Handbook included descriptions of organisational structure of the project management, decision-making procedures, publication procedures, meeting arrangements and responsibilities, reporting guidelines, description of how non-compliance with the project procedures would be handled in practise as well as ethical guidance. During the course of the project, the QA handbook was updated regularly and new items added, when considered necessary.

Part of the managerial activities has also been the organisation of the periodic meetings and activities of the General Assembly (GA), Management Board (MB) and External Advisory Board (EAB) (Task 1.4). In the beginning of the project, there was the kick-off meeting and then there have been four periodic meetings scheduled in connection with the periodic reporting (app. every 18 months). The periodic meetings have gathered together the entire consortium, with the opportunity to share the results of the work and to interact with other fellow scientists. These events have also included the meetings of the General Assembly, where all partners were represented, and allowing the discussion of issues that affect the entire project. This has been more strategic tool meanwhile the Management Board (MB) has been the principal decision-making body, convening for meetings or teleconferences several times per year and making decisions also by e-mail. In addition, DoReMi has been supported by the External Advisory Board EAB, consisting of five prominent scientists external to the consortium member organisations. The EAB followed the progress of the project and gave impartial view on

project's scientific issues. A major duty of the EAB was to evaluate the proposals of the three competitive calls and internal calls organised during the course of the project.

To measure the success of integration, DoReMi established performance indicators (Task 1.5). The performance indicators characterized the quality and success of the project and provided a systematic approach to follow the success of the project and to support managerial decisions. The success factors and performance indicators derived from them were organised according to the Balanced Score Card structure. To be successful, all four main areas of Effectiveness, Processes and Structures, Development and Work Capacity and Resources and Financing had to be in balance. The performance indicators were analysed in connection with the periodic reporting. At the mid-term of the project, the DoReMi MB organised a self-review and assessment meeting discussing in details the results of the analysis based on the performance indicators, and various corrective measures to develop the project were taken.

The most demanding part of the coordination work has surely been the management of the large flexibility funding budget (Task 1.6), with the allocation of more than 5,6 million euros. Most of the flexibility budget was used in competitive calls for new partners. Altogether three calls were organised and as a result, 24 new partners joined the consortium and the DoReMi work plan expanded to cover new fields. The first and second call were open to organisations all over the world and while most or the applicants were from Europe, one of the new partners selected was from Japan so this further expanded the scope of DoReMi. On the other hand, the 3rd competitive call was directed only to organisations from new EU member states, in order to strongly support their further integration.

The flexibility budget also allowed the organisation of three internal calls directed to existing DoReMi partners, in order to again expand the DoReMi work plan with topical issues identified in the TRA and TRA Statements, and to enhance the collaboration of the partners. In addition, there was always the possibility for the partners to apply for ad hoc funding, in case there was a need to extend an existing task or even establish a new one, bearing in mind the overall objectives of the task/work package/project as a whole. DoReMi also provided travel grants in order to support the dissemination of the results and attending training. Specific travel grants were directed to young scientists. Finally, DoReMi also established student projects where students from EU-based universities could be hired by the partners. The work plans of the students were to be relevant to low dose risk research and DoReMi objectives.

DoReMi started in year 2010 with 12 original partners, and it was clear from the beginning of the project that consortium will grow with new partners and the work plan will be widely expanded. Due to the size of the project and various updates to the work plan, it was considered important to make sure that potential problems are detected and tackled in advance. Therefore, a contingency plan was prepared as an early managerial deliverable and it was since updated on a regular basis (Task 1.7). The contingency plans identified the following items for continuous follow-up: recruitment issues, use of person-months, access to infrastructures, status of deliverables and milestones, status of Transitional Research Agenda (TRA), violations of the good scientific practise and evolution of the consortium: competitive calls. The contingency plan also handled any other major issues identified as potentially endangering the progress of the project, and sought solutions in case of possible problems.

The various activities of the DoReMi WP1 Network Coordination have ensured the effective administrative and financial management of the project, and supported partners in their scientific

work, providing the structures for efficient and participatory decision-making and flow of information, reporting, financing, knowledge management, fluent day-to-day management and guidance on project activities.

4 The expected final results and their potential impact

Humankind is exposed to low levels of ionising radiation from natural sources. Other low dose exposures can be received from medical and industrial uses of radiation, as well as from high altitude air travel. For the general public natural background and medical sources predominate. As even these low doses can be potentially damaging to health it is important to understand the risks radiation poses at all exposure levels but especially at the low levels that predominate, and where direct evidence of health effects are masked by the high incidence of sporadic diseases. An improved understanding of the risk at low doses is important for the further development of an effective and transparent system of radiation protection.

Although much is known about the quantitative effects of exposure to ionising radiation, considerable uncertainties and divergent views remain about the health effects at low doses. In 2009, the European High Level and Expert Group (HLEG) identified a series of key policy questions to be addressed by a strategic European research agenda. This resulted in the establishment of the MELODI European Research Platform, Multidisciplinary European Low Dose Research Initiative) to sustain the impetus and continue evolution of the research programme via the SRA. DoReMi acted as an operational tool for the sustained development of the MELODI platform during the years 2010-2015, creating sustainable integration of European research on low dose risk and providing answers to key policy questions. The DoReMi joint programme for research focused on the areas identified by the HLEG and MELODI as being the most promising in terms of addressing and resolving the key policy questions. By addressing the scientific basis underlying the system of radiation protection DoReMi, contributed directly to strengthening the credibility of scientific evidence relevant to the development of radiation protection policy. Ultimately DoReMi can be expected to contribute more widely to radiation protection through engagement with International Commission on Radiological Protection and other national and international bodies. The High Level and Expert Group pointed out that many EU member states have lost key competences and are no longer capable of independently retaining their current research activities in radiation sciences, with implications for effectively fulfilling operational and policy needs and obligations. Up-to-date research and education and training activities carried out by DoReMi and MELODI are urgently needed to ensure the European competence in radiation sciences and radiation protection.

In the final DoReMi TRA, the main achievements of DoReMi in low dose health risk research have been stated and the most urgent research needs and priorities in this domain emphasized for the benefit of improved radiation protection. Now, it will rely on the new wider-ranging European initiatives launched within Horizon 2020 to follow up the low dose research lines. The experiences on integration of research gained by MELODI and DoReMi have been exploited when preparing for the Horizon 2020. By now, Strategic Research Agendas have been prepared not only for low dose risk research but also for radioecology, emergency preparedness and dosimetry. Furthermore, an additional strategic research agenda is currently developed for the use of radiation in medicine. There is hope that low dose research can be further extended and consolidated by MELODI within the European Joint Programme (EJP) for the Integration of Radiation Protection Research (CONCERT). Further improvement of Radiation Protection in Europe remains an important goal.

Quantification of biological and health effects (cancer, other diseases and cell damage) associated with exposure to ionizing radiation has been a major issue for the ICRP since its foundation in 1928. While there is plenty of information on the human health for whole body doses above approximately 100 mGy, the effects associated with doses below 100 mGy are still being investigated and debated intensively. The current radiological protection approach, proposed by ICRP for workers and the public, is largely based on risks obtained from high-dose and high-dose-rate studies, such as Japanese Life Span Study on atomic bomb survivors. The risk coefficients obtained from these studies can be reduced by the dose and dose rate effectiveness factor (DDREF) to account for the assumed lower effectiveness of low dose and low dose rate exposures. The 2007 ICRP Recommendations continue to propose a value of 2 for DDREF, while other international organisations suggest either application of different values or abandonment of the factor.

To complement the traditional line of radiation research addressing DNA damage and response that typically follow the linear dose response pattern, DoReMi has focused its investigations on various non-linear cellular and tissue responses and underlying mechanisms. DoReMi results call for some caution in applying linear extrapolation from high dose results to estimates of low dose risk as nonlinearities in cell/ tissue responses exist. Cellular responses to low doses are complex. DoReMi has shown that they include a mixture of simple cellular transcriptional, proteomic and metabolic responses and more complex regulatory responses integrated across tissues (including stress responses, damage responses, and immunological responses and other non-targeted effects). Neither the simple nor complex responses can be assumed to follow linear dose response kinetics. In fact, cells may respond differently to high and low doses and to dose-rate changes in the manner and scale by which different response pathways and distinct regulatory pathways are activated.

Dose limits applied in radiation protection have been set to protect an “average person”, based on studies of risks (mostly cancer) seen in large populations such as the A-bomb survivors in Japan. For cancer induction, it is well established that there are differences in radiation sensitivity between individuals and population subgroups, depending on their gender, age, genetic make-up lifestyle such as smoking, and exposures to other agents. In general, however, even though these differences are recognized, they are not specifically accounted for in the setting of dose limits for planning purposes in radiation protection practice apart from very few situations. At present, there is insufficient information to establish how large these various differences in sensitivity may be between individuals or between groups of individuals and their consequent influence on risk estimates at low dose. Differences in radiation sensitivity between individuals, or groups, raise the ethical and policy question as to whether some individuals, or groups, are adequately protected by the present system and regulations. In order to address these policy questions, it is necessary to obtain better scientific information on the extent of variations in the sensitivity of the population, both in the sizes of the variations and also in the proportions of the population that are affected. Therefore, research is needed to identify the factors that affect individual sensitivity to radiation risk and to obtain realistic estimates of how large the differences may be in extreme cases and also on the spread of sensitivities in average population groups.

DoReMi has contributed to the knowledge basis on individual susceptibility. Research in DoReMi reveals that quite a number of specific genetic and epigenetic factors are involved in the observed high variability between individuals (patients). A genetic basis for differences in individual sensitivity to cancer induced by radiation has been demonstrated. This unequivocally establishes that genetic

differences between exposed individuals contribute to differences in risk.

With the view of better understanding non-cancer radiation-induced effects, the DoReMi project has permitted the implementation of a number of pilot experimental and epidemiological studies addressing vascular effects, lens opacities, neurological and cognitive defects, and anti-inflammatory reactions. These studies have offered the opportunity to establish strong, close and productive collaborations amongst European teams and have opened new routes for future research activities. The results obtained so far have shown clearly that studying the non-cancer effects resulting from exposure to ionizing radiation is highly relevant, even at doses below 100 mGy. As an achievement of the project DoReMi, a new dynamic research on non-cancer effects is in place. DoReMi has stimulated new teams, including scientists from new Member States, to take interest in this topic that was up to present an issue for a small number of research organizations only. While the study of non-cancer effects of exposure to low doses of ionizing radiation has long been considered too risky because that cannot lead to high-level publications due to the potential lack of significant results, research conducted on this topic in the context of DoReMi have shown that they could allow scientists to publish their results, recognizing that even the results showing no notable effects deserve to be published. Indeed, beyond the scientific value of the results is the benefit that these studies are likely to provide for the future development of doctrines in radiation protection by considering, not only cancerous diseases, but also non-cancer effects, with the final objective of a better evaluation of the radiation-induced detriment and a better understanding by the public of future new recommendations governing radiation protection.

By early 2016, there have been 105 publications by DoReMi consortium reporting the scientific progress in low dose risk research. These publications will be available for the international scientific community and risk assessment bodies, contributing to the knowledge base and reducing uncertainties related to the estimation of low dose risk. A long-term research agenda has now been established for the European research community to continue on the work on lines of research that are essential for the resolution of the uncertainties related to low dose risk. More than 30 European research organisations and funding bodies have now expressed their commitment in continued efforts to work along the lines of the MELODI Strategic Research Agenda and this vision is supported by coordination of research infrastructures and education and training activities.

Annex I: Evolution of the consortium and expansion of the work plan

It was recognized already in the beginning of the project, that the scientific challenges related to the low dose risk research are substantial and require a multidisciplinary approach and new competencies and capacities. Even though the DoReMi Annex I (Description of Work) included some research work to be started in the beginning of the project, it was already foreseen that research needs will be more clearly identified and justified only after the initial pilot and feasibility studies and surveys have been carried out. Therefore it was planned to launch competitive calls for proposals to deal with the emerging needs for research. A large proportion of the flexibility budget was reserved for this purpose.

The enlargement of the consortium was expected to benefit the consortium, as the additional competencies provided by new beneficiaries supplement the initial field of know-how. It would also benefit the wider research community, offering open and transparent process for additional beneficiaries to join the project and take part in the actual research.

The DoReMi consortium started its work in January 2010 with 12 original partners. The first ten new partners joined after the first competitive call, starting their work in the beginning of the 2nd project period in July 2011, and another ten new partners joined after the 2nd competitive call, starting their work in the beginning of the 3rd project period in January 2013. The third competitive call, directed to new member states, brought in four more new partners, starting their work in the beginning of fourth and last project period, in July 2014. In addition, the partial transfer of rights and obligations from partner 5 Health Protection Agency HPA to Department of Health, Public Health England DH-PHE gave DH-PHE the partner number 37. From initial 12 partners, the consortium has grown threefold.

The DoReMi work plan has been amended several times, mainly via three competitive calls for new partners, as well as via three internal calls, providing opportunities to existing partners. Also many tasks that started in the beginning of the project have been extended via internal ad hoc mechanism that has allowed the DoReMi programme to develop further and to respond to current and topical needs.

The four tables below show the enlargement of the WP's:

Table 1: WP4 Infrastructures programme enlargement

Task	Work	Starting
4.1	Survey of existing facilities for low dose risk research	2010
4.2	Characterization of infrastructure needs and roadmap of implementation	2010
4.3	Implementation of DoReMi support activities for shared infrastructures	2010
4.4	Development and implementation of access to Infrastructure	2010
4.5	**Open Access to the UMB low dose irradiation facility (FIGARO)**	**2011**
4.6	**Dose/Dose-rate Radiation Effects in Brain Cancer Risk (DDRE-**	**2011**

	BrainCancer)	
4.7	**Low dose/dose rate gamma irradiation facility for in vitro biological systems (LIBIS)**	**2012**
4.8	**Integration of STORE into DoReMi as a trustable and viable database and/or pointer to biobanks and ascertain sustainability**	**2012**
4.9	**Provision of ion microbeam irradiation facility SNAKE (MicroRAD)**	**2013**
4.10	**Laboratory infrastructure for retrospective radon and thoron dosimetry (RETRODOS)**	**2014**

Table 2: WP5 Shape of dose response program enlargement

Task	Work	Starting
5.1	Phase – shifts in responses and processes at high/low doses and dose rates	2010
5.1.1	**Low dose Gene Expression signature (LoGiC)**	**2011**
5.2	Assessing the relative contribution of targeted (DNA), non-targeted and systemic processes to radiation carcinogenesis	2010
5.2.1	**Modulation of Inflammation by low and moderate dose Ionising Radiation (ModInIR)**	**2011**
5.3	The dynamics of pre-neoplastic change and clonal development	2010
5.4	Mathematical models to link experimental findings and epidemiological data	2010
5.5	Assessing the risk from internal exposures	2010
5.5.1	**Internal Emitters in Uranium Miners (INTEMITUM)**	**2013**
5.5.2	**Assembly of internal radiation dose for UKAEA and AWE epidemiology cohorts (AIRDoseUK)**	**2013**
5.6	**Track structures and initial events: an integrated approach to assess the issue of radiation quality dependence (INITIUM)**	**2012**
5.7	**Induction and facilitation of chromothripsis by low dose ionizing radiation (In-FaCT-IR)**	**2013**
5.8	**Concerted Action for an Integrated (biology-dosimetry-epidemiology) Research project on Occupational Uranium Exposure (CURE)**	**2013**
5.9	**Low dose radiation-induced non-targeter effects in vivo: the role of microvesicles in signal transduction (Rad-Mvivo)**	**2014**
5.10	**Effects of Chronic LOw-dose Gamma Irradiation on GAstrointestinal Tumorigenesis (CLOGICAT)**	**2014**

Table 3: WP6 Individual sensitivities program enlargement

Task	Work	Starting
6.1	Molecular epidemiological studies to address the role of individual genetic variation in determining susceptibility to low doses	2010
6.2	Identification of genetic modifiers of individual cancer susceptibility and their mechanisms of action	2010
6.3	Modelling of the effects on risk prediction models due to changes in biological processes influenced by genetic variability	2010
6.4	The effect of genetic modifiers on carcinogenesis following low dose rate exposure	2010
6.5	Contribution of genetic and epigenetic mechanisms that indirectly influence susceptibility to radiation-induced cancer	2010
6.6	Implementation of the DoReMi strategy for a large scale molecular epidemiological study to quantify genetic contribution to individual susceptibility	2010
6.7	Planning expansion of research portfolio	2010
6.8	**Predicting individual radiation sensitivity with Raman microspectroscopy (PRISM)**	**2011**
6.9	**Integrating radiation biomarker into epidemiology of post-Chernobyl thyroid cancer from Belarus (INT-Thyr)**	**2012**
6.10	**Characterization of DNA lesions in the nuclear ultrastructure of differentiated and tissue-specific stem cells after protracted low-dose radiation (Zif-TEM)**	**2013**
6.11	**Mechanism of low dose response to ionizing radiation and its significance in radiation protection (RADSENS)**	**2013**

Table 4: WP7 Non-cancer effects program enlargement

Task	Work	Starting
7.1	Structuring the research effort on non-cancer effects according to the HLEG roadmap: organisation of consultation/exploratory meetings and funding integrative RTD projects	2010
7.2	Preparation of a pilot study to conduct molecular epidemiology studies in vascular radiation damage	2010
7.3	Feasibility study towards a systems biology approach of radiation response of the endothelium	2010
7.4	Pilot epidemiological study of lens opacities among a cohort of interventional radiologists and cardiologists	2010

7.4.1	**Lens opacities: Methodology implementation (ELDO)**	**2012**
7.5	Pilot study of external irradiation versus internal contamination effects on neurogenesis	2010
7.6	**Study on contribution of low dose X-radiation in induction of anti-inflammation**	**2011**
7.7	**Low dose Gene Expression signature and its impact on Cardiovascular disease (LoGiC)**	**2011**
7.8	**Study on contribution of low dose X-radiation in induction of cataractogenesis and influencing genetic and cell communication factors (LDR-OPTI-GEN)**	**2013**
7.9	**Low and moderate dose radiation effects on brain microvascular pericytes: epigenetic mechanisms and functional consequences (PERIRAD)**	**2013**
7.10	**Influence of a chronic LD and LDR exposure onto the development of Parkinson symptoms in genetically predisposed Pitx3-EYL/EYL Ogg1-/- mouse mutant (OSTINATO)**	**2013**
7.11	**Epidemiological pilot study on radiation-induced cataract in interventional cardiology (EVAMET)**	**2014**
7.12	**Effect of low doses of low-LET radiation on impaired vascular endothelium (ELDORENDO)**	**2014**
7.13	**Low-dose ionizing radiation-induced cataracts in the mouse: invivo and invitro studies (RadCat)**	**2014**

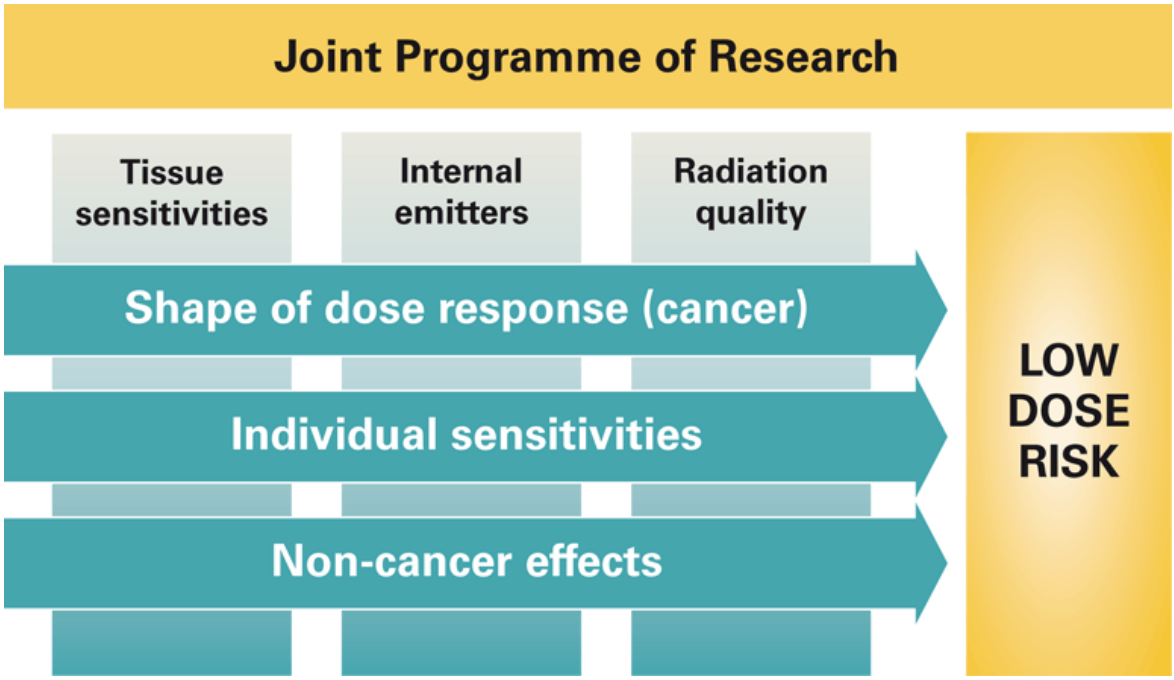
Annex II: List of partners

Beneficiary no.	Beneficiary name	Beneficiary short name	Country	Date enter project	Date exit project
1 (Coordinator)	Radiation and Nuclear Safety Authority	STUK	Finland	1	72
2	Institut de Radioprotection et de Sûreté Nucléaire	IRSN	France	1	72
3	Helmholz Zentrum München	HMGU	Germany	1	72
4	Commissariat à l'Energie Atomique	CEA	France	1	72
5	Health Protection Agency	HPA	UK	1	72
6	University of Pavia	UNIPV	Italy	1	72
7	Istituto Superiore di Sanitá	ISS	Italy	1	72
8	Belgian Nuclear Research Centre	SCK-CEN	Belgium	1	72
9	Bundesamt für Strahlenschutz	BfS	Germany	1	72
10	University of Stockholm	SU	Sweden	1	72
11	Centre for Research in Environmental Epidemiology	CREAL	Spain	1	72
12	Institut Curie	IC	France	1	72
Third party	The Institut national de la santé et de la recherche médicale	Inserm	France	1	72
13	Universitaetsklinikum Erlangen	UKER	Germany	19	72
14	Johann Wolfgang Goethe Universitaet, Frankfurt am Main	GUF	Germany	19	72
15	Universitaet Rostock	UROS	Germany	19	72
16	Norges miljo- og biovitenskapliga universitetet	NMBU	Norway	19	72
17	Norwegian Radiation Protection Authority	NRPA	Norway	19	72
18	Nasjonalt Folkehelseinstitutt	NIPH	Norway	19	72
19	Agenzia Nazionale per le Nuove Tecnologie, l'Energia e lo Sviluppo Economico Sostenibile	ENEA	Italy	19	72
20	Institute for Environmental Sciences	IES	Japan	19	72
21	Dublin Institute of Technology	DIT	Ireland	19	72
22	Erasmus Universitair Medisch Centrum Rotterdam	Erasmus MC	Netherland	19	72
23	Oxford Brookes University	OBU	UK	37	72

24	Brunel University	UBRUN	UK	37	72
25	National Public Health Center - National Research Directorate for Radiobiology and Radiohygiene	NRIRR	Hungary	37	72
Universal transfer of rights and obligations from “Frédéric Joliot Curie” National Research Institute for Radiobiology and Radiohygiene to National Public Health Center - National Research Directorate for Radiobiology and Radiohygiene validated in August 2015 (month 68).					
26	National Radiation Protection Institute	SURO	Czech Republic	37	72
27	NUVIA Limited	NUVIA	UK	37	72
28	Atomic Weapons Establishment	AWE	UK	37	72
29	Universitaet des Saarlandes	USAAR	Germany	37	72
30	Leiden University Medical Center	LUMC	Netherlands	37	72
31	Universität der Bundeswehr München	UBWM	Germany	37	72
32	Ludwig-Maximilians- Universität München	LMU	Germany	37	72
33	Sofia University “St. Kliment Ohridski”	SUN	Bulgaria	55	72
34	Nofer Institute of Occupational Medicine	NIOM	Poland	55	72
35	Military Institute of Hygiene and Epidemiology	MIHE	Poland	55	72
36	Jagiellonian University	UJ	Poland	55	72
37	Department of Health	DH-PHE	UK	40	72
(Partial transfer of rights and obligations from beneficiary 5 HPA to Department of Health, Public Health England)					

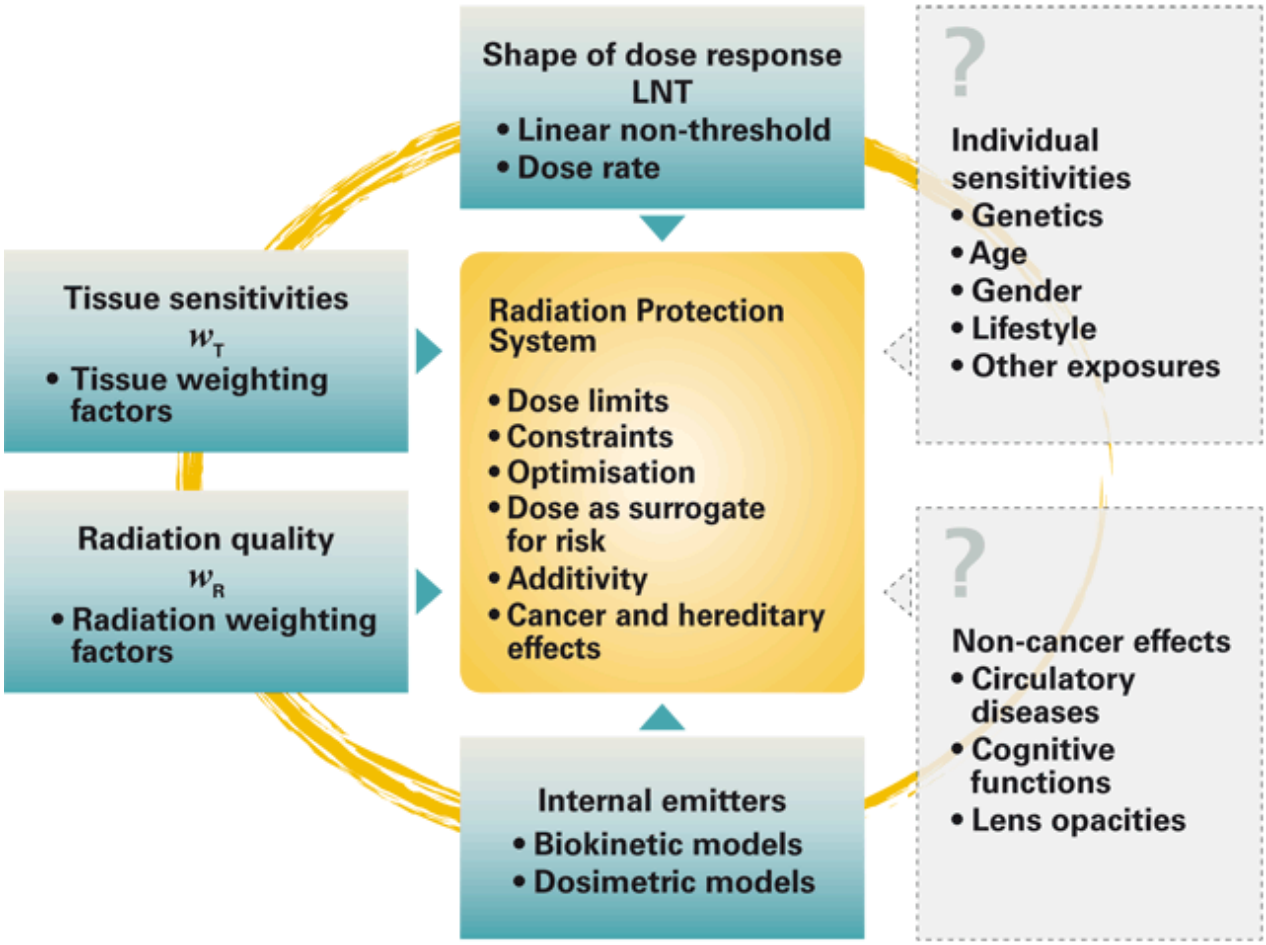
Annex III : Illustrative pictures

Picture 1: Joint programme of research



Picture 2: How robust is the system of radiation protection and risk assessment?

The current system of radiation protection makes judgements in several important areas: the four blue boxes indicate judgements that fall directly within the system of protection against the low dose radiation effects as recommended by ICRP, whereas the boxes on the right identify issues that are, at present, included only to a minor degree.



Picture 3: DoReMi work package structure