



DC-THERA

Dendritic Cells for Novel Immunotherapies

Executive Summary March 2008

Overview of Project Objectives

Dendritic cell (DC) immunobiology has enormous potential for development of new immunotherapies for cancer and infectious disease. Europe possesses a critical mass of leaders in the field who have pioneered many innovative advances and provided initial proof of principle for the approach. DC-THERA, 'Dendritic Cells for Novel Immunotherapies' is a Network of Excellence (NoE) established under the European Commission's Sixth Framework Programme (FP6). Its objectives are to translate genomic, proteomic and bioinformatic information, with knowledge from molecular cell biology and pre-clinical models, into therapeutic endpoints: clinical trials of DC-based therapies for cancer and HIV. Its aims are to promote the integration of the activities of 26 participant groups of scientists and clinicians, and 6 SMEs, across Europe over its five year duration; additionally it has incorporated 39 further groups as Associated Partners of the Network, including several groups from new and future member States – *the Contractors involved are listed at the end of this Executive Summary*. Their collective expertise and resources have been forged into an ambitious Joint Programme of Activities to restructure the field. Towards this end, four thematic S&T Clusters were originally defined, with a fifth for horizontal activities to provide an enhanced infrastructure and promote collaborative working in the Joint Programme of Activities. Following a mid-term review of the Project, 6 new Strategic Priorities (several crossing pre-existing Clusters) have been identified for the remainder of the Contract, and for which a new Cluster (6) has now been created.

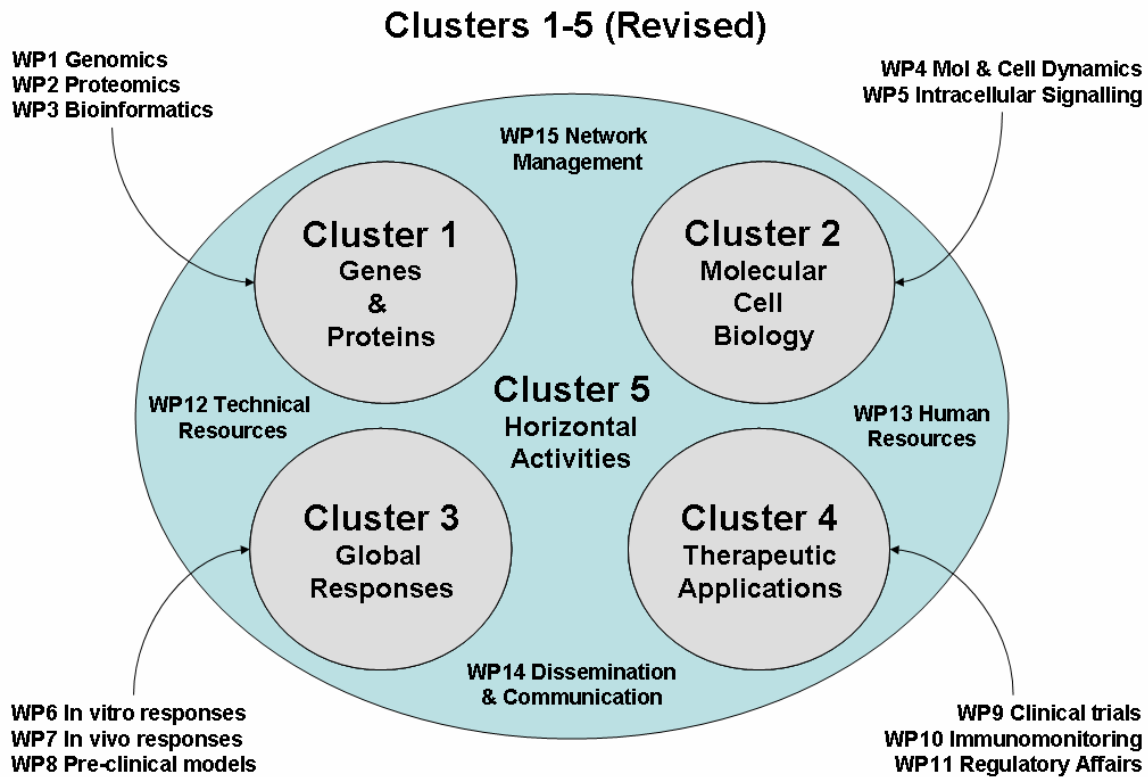
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Clusters 1-5: Progress Report

In its first year, the Network made considerable progress in beginning to implement its infrastructural measures and the majority of these were consolidated during the second. Over the current period (2007) further progress was made towards meeting the Project's S&T objectives and, by the mid-point of the Project (mid-2007), the last of its planned horizontal activities has been implemented. This has now resulted in fulfilment of essentially all of the Project's anticipated Deliverables, and the majority of its Milestones, by the end of the current period (end 2007). Overall, the Network's research activities have continued to make very significant contributions to the state-of-the-art in the field of DC immunobiology and translational medicine, and its horizontal activities have led to further integration of its Partners and increased involvement of its Associated Partners. In addition, an internal mid-term review of the Project, together with an opportunity to reconsider the two previous reviews of it, have now led to the identification of several new Strategic Priorities (below) that will provide a new focus for the Network's activities, further integrate the different areas being covered by the Project, and provide new opportunities for collaborative working by its participants. Finally the pre-existing Clusters (1-5) of the Project were restructured to reflect more accurately the many different activities being undertaken by the Network's participants; these are summarised diagrammatically below, following which some of the achievements of the current period are highlighted.



Cluster 1, Genes and Proteins, comprises three Work Packages (WPs): Genomics (WP1), Proteomics (WP2), and Bioinformatics (WP3). This jointly executed research project aims to study both gene and protein expression (the transcriptome and proteome respectively) in DC subsets from different sources and/or activated by a variety of stimuli, as well as developing the bioinformatic tools to enable interrogation of the datasets that are obtained in order to link them with cell functions. In **WP1**, collaborations between several Partners have now resulted in additional transcriptomes for mouse DC activated with different stimuli (or treated with kinase inhibitors; see WP5), rat DC subsets from pseudo-afferent lymph, and human DC; related work on macrophages has also been initiated. In **WP2**, collaborations between Partners and two newly-incorporated Associated Partners have additionally led to preliminary mass spectrometric analysis of the mouse DC plasma membrane proteome, as well as proteomic analysis of clinical grade DC before and after maturation. Finally, work undertaken in **WP3** has led to the development of a platform for automated transcriptional data analysis, and has continued to underpin efforts to create a database of transcriptomic signatures that, in Q4 2007, was made accessible to DC-THERA participants via the Genopolis (IT) website.

Cluster 2, Molecular Cell Biology, comprises two WPs: Molecular & Cellular Dynamics (WP4) and Intracellular Signalling (WP5); it originally included a third (WP6) but this has now been incorporated into Cluster 3 (below). In **WP5** an array of state-of-the-art imaging techniques (including two-photon and confocal microscopy, and MRI) has enabled the visualisation of interactions between DC and CD8 T cells in intact lymph nodes, and between CTL and tumour cells in vivo; the acquisition of the very first images of NK cells in lymph nodes; and both the tracking of clinical-grade DC into lymph nodes of melanoma patients, and migration of mouse DC transfected with E- and L-selectin directly from the blood into nodes. Techniques such as multi-parametric flow cytometry have also yielded further insights into the characterisation of helper T cell subsets, and enabled the analysis of translation activation in T cells activated by DC. In addition, methods have been refined in order to introduce new genes into DC (e.g. via lentiviral constructs) or to silence them (e.g. use of siRNA and shRNA approaches). Meanwhile, work in **WP6** has led to some real insights into signalling pathways of DC, including the application of kinase inhibitors to uncouple cytokine secretion from costimulatory molecule induction; the first definition of a signalling pathway from a C-type lectin receptors of DC (Dectin-1); and the importance of the STAT-1 pathway in the response of DC to Th1-inducing stimuli.

Cluster 3, Global Responses, now comprises three WPs: In Vitro Responses (WP6), In Vivo Responses (WP7), and Pre-Clinical Studies (WP8); the former has been incorporated from Cluster 2 due to the increasing overlap with work being undertaken in the latter two. In **WP6**, in vitro studies have led to a deeper understanding of stimuli that: modulate DC responses (including their antigen- and cross-presentation pathways); polarise CD4 T cell responses (Th1, Th2, Th17 and Treg); and induce CTL responses. It also includes studies of the cross-talk between DC and the specialised subset of NKT cells, and between DC and NK cells. These studies are extended to the in vivo setting in **WP7**, which also includes the development of techniques to enhance the targeting and

delivery of antigens to DC (e.g. via DC-SIGN, Fc receptors and Toll-like receptors). The aim of **WP8** is then continue to apply some of these approaches to pre-clinical models of infection (e.g. viral infections such as HIV in the SCID-hu model), malignancies (including a spontaneous transgenic model of breast cancer) and inflammatory diseases (including an autoimmune colitis model) – and ultimately to the clinic (below).

Cluster 4, Therapeutic Applications, also includes three WPs: Clinical Trials (WP9), Immunomonitoring (WP10), and Regulatory Affairs (WP11). In **WP9**, the current period saw the completion of 10 clinical trials of DC-based and other therapies for cancer by Network participants, while a further 15 are still in progress or are planned for the next phase of the Project, including at least 2 for therapy of HIV-infected individuals. This is a truly impressive joint effort that reflects the fact that DC-THERA includes many of the European leaders in the field. Equally impressive is the wide variety of different approaches that is available across the Network for immunomonitoring of patient responses in **WP10**, including assays to assess responses of different subsets of CD4 T cells, including Treg; CTL; and NK cells. In the current period the participants of this area of the Project have also made recommendations (and have developed standardised SOPs) for routine (tetramer, ELISPOT) monitoring, as well as more specialised T cell assays (MLPC assays; analyses of DTH infiltrating T cells; gene expression in individual T cells; etc). The latter work now substantially overlaps with the original objectives of **WP11** which was (and is) to lead to a durable re-shaping of European clinical practice. This WP also included the initiation of dialogue with European Regulatory Authorities (now dealt with in WP14 below) but has now been re-focussed towards ensuring that all current and future clinical trial protocols (and the associated SOPs) are designed in accordance with National and European Regulatory Guidelines.

As an NoE, DC-THERA wishes to enhance not only the integration of joint research activities, as exemplified by the four S&T clusters above, but also the infrastructure underpinning the fields of DC immunobiology, vaccinology and immunotherapy in Europe. **Cluster 5**, for Horizontal Activities, has been constructed towards the latter goal and it comprises four WPs that have been partly restructured: Technical Resources (WP12), Human Resources (WP13), Dissemination & Communication (WP14) and Network Management (WP15).

WP12 is designed to enhance the technical capabilities of the Network, and to remove bottlenecks to progress, by establishing core technological platforms (initially for functional genomics, advanced imaging, and cellular therapeutics) and promoting the development of shared tools and protocols. In the current period, for example, the genomics platform has provided ‘services’ to other participants that have enabled collaborative transcriptomic profiling studies to be undertaken (see WP1), while a new proteomics platform has likewise facilitated collaborative proteomic studies to be initiated (see WP2). **WP13** is designed to enhance the human resources of the Network by focusing on the development of education and training activities, and the sharing and dissemination of expertise and resources. Activities within this area of the Project have previously included the creation of thirteen new collaborative PhD studentships financed for a total of 37 person years, and an annual Graduate School, and the incorporation of

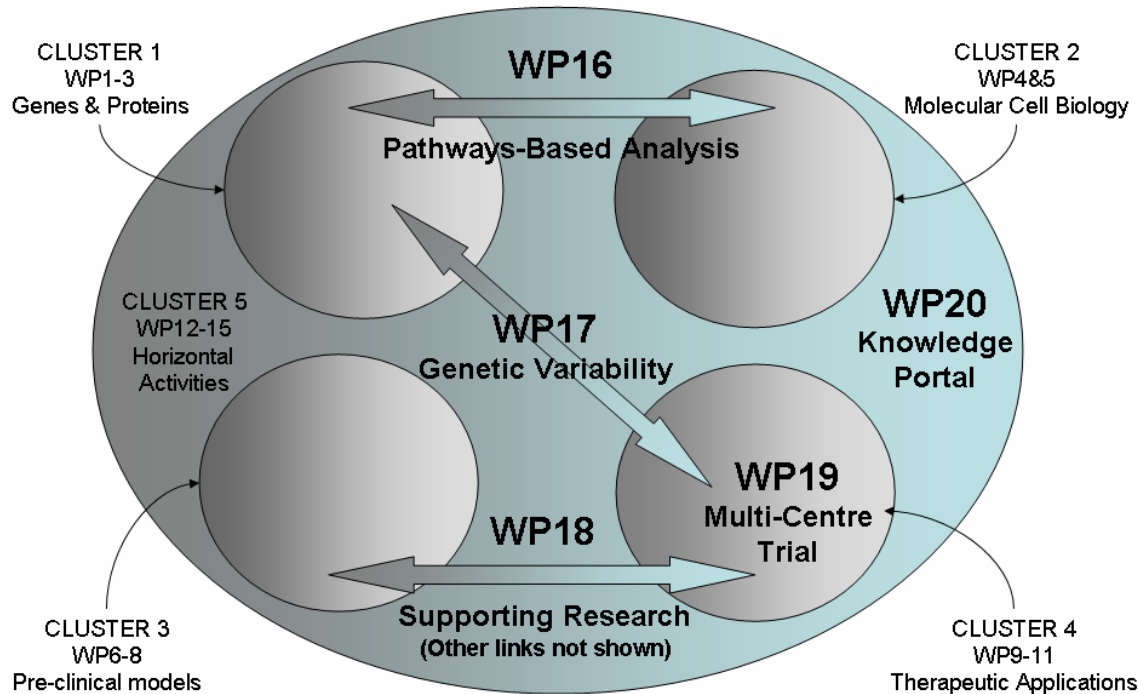
Associated Partners. Over the current period a Visiting Scholars Scheme has also been initiated (initially with 6 appointees, some between Associated Partners) and other activities have continued (including delivery of intensive training courses for GMP production of clinical grade DC).

WP14 has now been restructured to focus on dissemination and communication issues. This includes maintenance of the Network's website; continuing development of links with other Networks, including joint meetings; and dialogue with the European Medicines Agency, EMEA (initiated in former WP11). Over the current period DC-THERA has continued to collaborate closely with the CIMT 'CANCERIMMUNOTHERAPY' Integrated Project (e.g. see WP18 below); has opened dialogue with the 'EXPERTISSUES' Network of Excellence in the field of Tissue Engineering and Regenerative Medicine (a joint satellite meeting is planned for the next phase); and has held a joint meeting with the European Macrophage and Dendritic Cell Society, EMDS (a further meeting is planned in the next phase). In addition, DC-THERA initiated dialogue with the EMEA Q1 2007, provided (in association with CIMT) a written response on its draft guidelines for cell-based medicinal products early Q3 2007, and was subsequently represented at its last meeting Q4 2007. Finally, **WP15** addresses management issues per se, including the organising of Cluster meetings (3 to date) which have been most successful, and the revision of the JPA and associated reallocation of the budget for the next period. It was also responsible for the internal mid-term review which has now lead to the identification of new Strategic Priorities (below) for the remainder of the Contract.

Cluster 6: Strategic Re-Alignment of the Project

The new **Cluster 6** comprises 5 new WPs: Pathways-Based Analysis (WP16), Genetic Variability (WP17), Supporting Research (WP18), Clinical Trial (WP19) and the Knowledge Portal (WP20). These WPs, and their relationship to pre-existing (but partially restructured) Clusters 1-5 are summarised diagrammatically below.

Cluster 6 Strategic Re-alignment of the Project



WP16 will draw upon the vast wealth of (publicly available, and Network-generated) data relating to genes and proteins of DC much of which has not yet been ‘mined’; incorporate the data obtained from a ‘Golden Reference Experiment’ (WP17); and provide an entirely new and powerful instrument to begin to understand the genomic and proteomic pathways that underlie the function of the cells. WP16 links Clusters 1 & 2. The goal of **WP17** is, for the first time, to obtain sufficient genomic information from retrospective studies of a large number of stored human DC samples to understand the genetic basis of inter-donor variability in responses to DC maturation stimuli. WP17 links Cluster 1 (e.g. genomics) with Cluster 4 (e.g. clinical trials and immunomonitoring) and will also provide a dataset for WP16. The objective of **WP18** is to conduct arguably the first well-standardised, two-armed clinical trial in 3 countries using consensus SOPs. This trial will assess immune responses of melanoma patients. WP18 is assigned to Cluster 6 as it transcends the work that will be performed concurrently in Cluster 4 by individual Partners. The purpose of **WP19** is to fund small, short-term research projects that underpin any and all of the other initiatives in Cluster 6, and which will also specifically target the ‘interface’ between pre-clinical and clinical work; a substantial portion of the budget for this initiative has been ring-fenced for Associated Partners. WP19 thus explicitly links Clusters 3 & 4, but potentially also underpin all other Clusters. Finally, the objective of **WP20** is to develop a database of the existing expertise and resources of all Partners and Associated Partners within the Network, the DC-THERA Knowledge

Portal. Once established, this initiative – which potentially integrates all WPs of the Project – may be opened to all European scientists and clinicians working in the field.

Summary

Over the current period, excellent progress has continued to be made in all areas covered by the Project's four original S&T Clusters (1-4) as well as its horizontal activities (Cluster 5). Following an internal mid-term review, several new Strategic Priorities, now addressed in Cluster 6, were additionally identified for the remainder of the Contract. At the end of the current period, the JPA was completely revised (including some restructuring of Clusters 1-5 and re-definition of associated Deliverables & Milestones) and the budget was reallocated to enable the implementation of the new Strategic Priorities whilst still maintaining support for all activities that are currently underway in other areas of the Project. Finally, at this stage of the Project, serious consideration is now being given to different mechanisms that could be put in place to ensure the future sustainability of the DC-THERA Network beyond the end of the Contract.

Principal Partners

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