

EMMAservice project summary



Project acronym: EMMAservice
Funding scheme (FP7): Integrating activities
EU financial contribution: €8 million
Total budget: €10.3 million
EU project officer: Brigitte Sambain
Duration: 48 months
Start date: 1 January 2009
Completion date: 31 December 2012

Partners:

Medical Research Council (UK)
Centre National de la Recherche Scientifique (FR)
Karolinska Institutet (SE)
Fundação Calouste Gulbenkian (PT)
Helmholtz Zentrum München Deutsches Forschungszentrum für Gesundheit und Umwelt GMBH (DE)
Europäisches Laboratorium Für Molekularbiologie (DE)
Centre Européen de Recherche en Biologie et Médecine (FR)
Genome Research LTD (UK)
Consejo Superior de Investigaciones Científicas (ES)

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Project webpage: www.emmanet.org

EMMAservice project objectives

The mouse shows great similarities in development, physiology and biochemistry to humans, which makes it a key model for research into human disease. The major challenges for mouse functional genomics in the 21st century are to:

- 1) Develop a series of mutant alleles for every gene in the mouse genome
- 2) Determine the phenotypic consequences of each mutation
- 3) Identify mouse models for the complete disease spectrum in humans

To exploit this emerging resource, mouse models must be preserved and made available to the European biomedical research community. Building on EMMA's previous achievements as the primary mouse repository in Europe, EMMAservice aims to meet the future challenges presented by archiving and disseminating mouse models in the ERA as follows:

- Archiving of 1224 new mouse mutant lines in support of individual depositors and also European mouse genetics programs
- Support of eligible customers with free of charge Transnational Access for up to 20% of requested mouse resources
- Technology development will underpin the archiving and distribution efforts by advancing current sperm freezing technology
- Training courses will promote the shipment of frozen germ plasm rather than live mice
- EMMA informatics will support user services by setting new standards for user friendly accession of EMMA services, extensive data curation and cross referencing with other mouse database resources
- Outreach efforts to attract users will be widened and addressed at the translational research community

EMMA service project – activities and results

A first key objective of the EMMA service networking activities (**WP2**) was to effectively **integrate three new partners into EMMA** leading to a significantly increased archiving and distribution capacity and to the representation of a new member state (Spain) in the network. **Outreach efforts** focused on attracting new EMMA users by presenting the EMMA activities at 16 major mouse genetics meetings. In addition, EMMA was represented on project meetings of collaborating consortia such as EUCOMM and EUCOMMtools, EUMODIC, IMPC and CREATE. Furthermore, EMMA was widely presented as part of the intensive promotion of the INFRAFRONTIER project. Overall, EMMA PR activities covered a total of 115 presentations at scientific and science policy meetings. Promotion activities covered in addition publications, advertisements and the use of Email lists. The existing know-how and cryopreservation expertise was transferred to the community by organizing a total of 16 training courses for a total of 104 students.

A key objective of the EMMA service project was the **archiving of mouse mutant lines** contributed from the wider community and in support of large European mouse production projects such as EUCOMM (**WP4**). The work package milestone for the project was the archiving of a total of 1224 mouse mutant lines. During the 48 months of the EMMA service project a total of 1224 mouse mutant lines were submitted to the EMMA repository out of which 1094 are archived and publicly available. Thus, with regard to submitted mouse mutants the envisaged project milestone was fully achieved. A total of 67% of the submitted lines account for community contributed lines and the remainder for lines originating from EU funded projects such as EUCOMM and EUMODIC. Of the submitted lines 793 were archived as embryos and 398 were archived as sperm. Some are archived as both formats. The mean archiving time for embryo freezing was 3.8 months. Most submitted lines were targeted mutations, mainly KO mutations due to the contributions from the EUCOMM mutagenesis project. This class accounts for 901 of the submitted lines, transgenic lines account for 176 lines, followed by chemically induced mutations, spontaneous mutations and gene traps. Strain donations were made across Europe with major contributions from UK, France and Germany. Mouse mutant lines were donated by a total of 250 different depositors.

A further focus of the EMMA service research activities is **technology development (WP5)** which underpins the EMMA cryopreservation service by refining existing EMMA sperm freezing technologies. Focus of WP5 were **1)** a thorough assessment and implementation of new sperm freezing technologies, **2)** setting up laser IVF technology at participating centers, **3)** initiate trainings to establish an ICSI capability at EMMA and **4)** to test for optimum conditions for transportation of unfrozen embryos. The first two reporting periods of WP5 were characterised by variable IVF fertilisation rates associated with our attempts to develop a new and robust sperm freezing procedure. Nevertheless, following discussions with collaborators in Japan, plus further development work in the laboratory we have been able to establish a robust sperm freeze/recovery protocol that works extremely well on sperm harvested from inbred strains of mice. The efficacy of this technique has been demonstrated by several partners within the EMMA network and across the most common genetic backgrounds used in mouse genetics. This protocol has been presented on the EMMA website and is available to the community. Furthermore, we have demonstrated that laser assisted IVF increases fertilisation rates over and above conventional IVF techniques. Laser IVF is firmly established within the EMMA consortium as a robust recovery technique and a protocol has been uploaded on to the EMMA website. However, the need to resort to laser IVF has diminished with the advent of the improved sperm freezing and IVF recovery techniques. ICSI is a technically challenging procedure that requires a considerable investment in time to perfect. Nevertheless, we have made good progress in this area and have been able to generate ICSI derived embryos using sperm and oocytes from several different strains of mice (CD-1, B6D2 and C3H). There is still further potential to refine the efficiency of the ICSI technique; consequently we feel it is too early to publish a protocol on the EMMA website. Nevertheless, we have developed a reliable ICSI protocol at two nodes (MRC and CNR). The transportation of unfrozen embryos will simplify the exchange of mouse stocks between laboratories. In keeping with this idea, we have demonstrated that frozen/thawed 2-cell embryos from several different strains (C3H, C57BL/6 & 129) will resume *in vitro* development after being held at 8°C for 72hrs. We now believe that we have defined the optimum conditions for

transporting unfrozen embryos. As a proof of concept we have used a regular courier service to exchange unfrozen embryos between five EMMA nodes. Subsequent embryo transfer experiments and the production of live born progeny have clearly demonstrated that *in vivo* development is not compromised after long term low temperature storage.

Distribution of mouse mutant resources is supported in the EMMA service project via the **Transnational Access (TA) activity (WP3)**. The TA program facilitates free of charge access to a defined number of requested EMMA mouse resources. In general, the EMMA distribution continues to grow. A total of 550 strain requests were received in 2012 compared to a total of 473 requests for 2011. The total number of strain requests by the end of the EMMA service reporting period was 2814. A first objective of WP3 in the first reporting period was the implementation of the TA system and its integration into the existing EMMA ordering system. Milestone for the project was the provision of 330 free of charge access units. To this effect, a total of 15 TA calls were published on the EMMA website. In response to the calls a total of 339 applications were received out of which at various stages 28 were cancelled. Overall, 311 genuine TA applications were submitted out of which 23 were rejected and 288 accepted. TA applications were submitted from 23 different countries with most applications being from Germany, France and the UK.

Another objective of the EMMA service activities was to support interested customers with the transfer of up to 12 mouse mutant lines into germ-free conditions (**WP7**). The **EMMA germ-free service** offered by the Gulbenkian Institute (IGC, Portugal) is a unique service not provided by other major repositories. The IGC received a total of 18 service requests during the course of the project. By the end of EMMA service a total of 14 service requests were fulfilled, while 2 projects are expected to be finalized in 2013. Furthermore, customers could benefit from the availability of mice already made germ-free and available for distribution. This concerns C3H mice, the seeding and foster colony; C57BL/6, often used as controls for various mutants and C57BL/6 Rag2^{-/-}, a line requested several times by independent users. Cells from germ-free C3H animals were isolated and shipped to customers as well as serum from C3H and C57BL/6J wild type young adults born and raised in germ-free conditions.

WP6 coordinates the **EMMA informatics** activities such as database development and integration with other resources, web interface development and database curation. Key objectives for the EMMA service project were **1)** maintenance and updating of the existing EMMA database including extensive curation of strain nomenclature, **2)** further development of the internal and external websites to improve the user-friendliness for customers and internal users and **3)** to facilitate integration of the EMMA database with other appropriate strain, genomic and phenotypic resources. The objectives of **task 1** were met during the reporting periods, with considerable development of the database schema and the database population mechanisms as well as via extensive data curation. During the project a total of 1942 strains had their gene/allele and strain names/symbols manually reviewed and curated. Concerning **task 2** we achieved the deliverable i.e. releasing the Spring internal interfaces, new searching and strain description pages and a new statistics/reporting package. The reporting package is important to control in more detail the progress of the EMMA archiving activities to underpin further process improvements. **Task 3** was achieved on time and in fact the BioMart interface was delivered more than a year ahead of the delivery date. The BioMart is a widely used informatics distribution resource and allows that EMMA data are accessible by multiple resources. Furthermore, links were set up to phenotype data of EMMA stocked mouse resources available at the Europhenome database or at the Sanger Mouse Portal. Finally, information available from MGI on human disease associations to mouse models were integrated into EMMA, now allowing the searching of EMMA mice by associated diseases. New data sources that will become available such as high-throughput phenotype data will be integrated with EMMA to improve the usefulness of the resource to the whole scientific community, either by bringing the data into the EMMA database itself or through the ability of the BioMart interface to perform distributed and integrated querying. All tasks of the informatics work package were managed and prioritized via an online informatics task manager (JIRA).

Project management (WP1) underpins all EMMAservice activities. Its specific objectives for the EMMAservice project were **1)** to ensure achievement of project results, **2)** to provide decision making and quality control, **3)** to support the implementation of changes in the activities and in the consortium if needed and **4)** to provide timely and efficient contractual, financial and administrative coordination of the project where needed. To this effect foremost a consortium agreement was concluded. Furthermore, a total of eight general project meetings were held. The documentation of all meetings is available via a newly developed specific internal website. Also the external website was extensively updated and redesigned with the EMMA informatics team. Management activities covered further the appointment of new Advisory Board and Evaluation Committee members as well as the integration of four additional partners into the EMMA network. During the course of the project Austria, Finland, Czech Republic, Greece, Israel and Netherlands were approved as new member states of the EMMA network. Finally, the network was and is continuously developed further by numerous collaborations with other European consortia. A key milestone achieved during the second period was the organization and successful completion of the midterm review meeting. The reviewer provided valuable advice that was considered and turned into specific project objectives in a grant application submitted to the EC in response to the publication of call 10 of the Capacities Specific Program in July 2011. The successful application resulted in the INFRAFRONTIER-I3 project that will underpin the continuation of the EMMA operation.

Expected project impact

The EMMAservice project contributed so far a large number of new mouse models to the EMMA repository. The projected further substantial quantitative and qualitative growth and development of the EMMA repository in the coming years will be essential to support the transition to a sustainable research infrastructure. EMMAservice and complementary efforts led to a repository holding nearly 4000 mouse mutant lines in 2013. For many of these lines a comprehensive first line phenotypic screen will be undertaken and the data will be made freely available. This comprehensive and unique physical and data resource will support biomedical research in Europe by offering the opportunity to decipher molecular disease mechanisms and aiding the development of therapeutic strategies. Thus, the EMMA network will play a critical role in ensuring Europe maintains its leading role in the development of resources and knowledge of medically relevant mouse models by providing a user driven, sustainable platform offering access to unparalleled services and physical as well as data resources.