International Agency for Research on Cancer

World Health Organization

Scientific Council **SC/54/7**
Fifty-fourth Session 07/12/2017

Lyon, 31 January–2 February 2018
Auditorium

REQUEST FOR SUPPORT FROM THE GOVERNING COUNCIL SPECIAL FUND

1. The IARC Medium-Term Strategy (2016–2020) highlighted an increasingly multi-disciplinary approach, integrating the latest advances in laboratory-based and epidemiology research. In this context, there is a constant need for acquisition of state-of-the-art scientific instruments and other infrastructure investments essential to support this strategy.

2. Three pieces of equipment have been identified as necessary to support: a) the development of histological activities following recruitment of the new Head of the WHO/IARC Classification of Tumours Group (WCT)[1]; b) to appraise the quality of genomic DNA and RNA isolated in IARC laboratories, and c) to support expanding analytical work in relation to the role of nutrition in cancer etiology.

3. Covering the cost of new equipment on the regular budget has not always been feasible in recent years and obtaining designated funds through competitive grant applications is difficult given the limited number of such opportunities open to the Agency as an international organization.

4. A second part of this request concerns replenishment of the biobank of the European Prospective Investigation into Cancer and Nutrition study (EPIC), housed in the IARC Biobank since the establishment of the cohort in the 1990's. The EPIC biobank is a key resource supporting the Agency's mission on the identification of cancer risk factors and biomarkers of early detection. The large productivity of research performed in EPIC has resulted in the depletion of serum and blood samples from cancer cases. Replenishment of this resource is critical for future research activities in this major cohort.

5. Therefore, the Director would like to request the Governing Council, at its 60th session in May 2018, to provide an allocation of €500 000 from the Governing Council Special Fund (GCSF) for the replenishment of the EPIC Biobank and the purchase of the following equipment:

a) An automated immunostainer

b) An automated device for nucleic acid quality control

c) An automated system for plasma phospholipid fatty acid profiling

6. The annual maintenance costs of the requested equipment will be covered by the regular budget as well as by collaborative programmes through grant applications.

7. The proposed investment is first submitted to the Scientific Council for its consideration.

[1] See IARC Organizational structure for list of acronyms: http://www.iarc.fr/en/research-groups/org_chart.pdf

1) Scientific Equipment

a) Automated immunostainer

8. Immunohistochemisty allows the recognition of molecules, mainly proteins, within cancer biopsies using standard histological methods. It is often required for cancer diagnosis, grading, staging and for predictive oncology. As such, immunohistochemistry is a key method within all histopathology laboratories, required for most cancer research projects involving biopsy materials. Data from the IARC histopathology laboratory covering the period 2013–2016 indicates that the need for this technology is growing. There is anecdotal information to suggest that immunohistochemistry for some research groups is being performed outside the Agency to assure quality of results, due largely to the manual processing used at IARC compared to the more robust automated systems used elsewhere.

9. Automated immunostainers are cost-saving in terms of reagents and staff time. They can perform deparaffinization, antigen retrieval, immunostaining, and cover-slipping. They also provide slides which have been prepared exactly the same between runs, allowing digital imaging and image analysis of the results to provide quantitative rather than qualitative data, as so far obtained at IARC. The IARC histopathology laboratory stained a total of 1171 sections in 2016 by immunohistochemistry, and in the first nine months of 2017, 700 sections were stained for 12 IARC research groups. In addition, immunohistochemistry staining for some major projects such as ESTAMPA has been performed outside IARC. This automated immunostainer is needed today to complement the digital scanner purchased in 2012. On the basis of quality, capacity and running costs there is a strong rationale for automating immunohistochemistry at the Agency.

10. The equipment will be situated in the Histology Laboratory and used by trained histology staff from WCT to provide immunohistochemistry in several large scale projects.

Estimated cost: €120 000

b) Automated device for nucleic acid quality control

11. Sample quality control is crucial at multiple stages of next-generation sequencing (NGS) applications, e.g. when analysing FFPE DNA or preparing NGS libraries, or when assessing genomic DNA quality for biobanking. For NGS applications, a Bioanalyzer is currently used by IARC scientists to assess DNA and RNA quality, which is based on a proven technology and requires small amounts of sample. Inherent limitations, however, include its relatively low throughput (manual operation, 12 samples maximum/run) and the inability to assess the quality of genomic (high-molecular weight) DNA. This low throughput has become incompatible with several large-scale ongoing projects.

12. The proposed device for nucleic acid quality control is envisioned to allow quality control of up to 96 samples per run, including NGS libraries, genomic DNA and RNA, but is also scalable allowing the flexibility to run smaller numbers of samples without wasting consumables. Another advantage over the Bioanalyzer will be the degree of automation, which will significantly reduce work-intensive steps of gel preparation and sample loading. The proposed piece of equipment uses gel matrices pre-packaged on a ready-to-use chip, which are loaded and run automatically as part of the program.

13. In light of the upsurge in NGS-based projects over the last few years at IARC, the in-house NGS capacity expansion (acquisition of the NextSeq 500 sequencing system in 2017) and the continuous requirement for quality control of large numbers of DNA samples by the Biobank (~2500/year in LSB Group), the proposed high-throughput device for nucleic acid quality control will complement multiple lines of research at the Agency. Research projects include experimental and epidemiological studies, performed with external collaborators or as cross-cutting projects among IARC groups. Specific applications include NGS library preparation (MMB, EGE, GCS, ICB), genomic DNA and RNA quality control (MMB, EGE, GCS, LSB, ICB), genotyping (GCS) or multiplex PCR fragment analysis (multiple groups).

14. The equipment will be under the responsibility of MMB but accessible to different laboratory research groups at IARC, operated as a shared resource accompanied by training for individual users.

Estimated cost: €50 000

c) Automated system for plasma phospholipid fatty acid profiling

15. The link between dietary fat and fatty acid metabolism with cancer development is a long-standing interest within the Section of Nutrition and Metabolism (NME). Over the last four years, 9000 plasma phospholipids have been profiled for fatty acid composition within three large epidemiological studies on cancer risk. In the past five years the Agency has already been successful in obtaining grant funding for large-scale epidemiological studies which require 14 600 samples to be profiled for fatty acid levels. In addition, new opportunities are arising to measure fatty acids in tissue samples, notably breast tumour biospecimens and to determine short-chain fatty acids in plasma and stool samples.

16. The Biomarkers Group (BMA) has established a platform with two gas chromatographs capable of providing quantitative measurements of 60 phospholipid fatty acids in a single analytical run in plasma samples as well as tissue sections. However analytical throughput is limited by manual sample preparation which includes lipid extraction, purification of the phospholipid fraction, derivatization of fatty acids and extraction of the methylated fatty acids.

17. Currently two technicians working together analyse 250 samples per month. This limited throughput presents analytical challenges given the large number of samples planned to be measured in the coming years. The acquisition of a robot dedicated to the extraction and derivatization of fatty acids is requested for the fully automated preparation of fatty acids.

18. An automated method was previously validated on 860 batches and a total of 20 000 samples over a period of 20 months (Wang LY *et al*, 2013, *Genome Med.* **5**:39). The proposed system, in use at the Medical Research Council (Cambridge, UK), would allow the analysis by two technicians of about 7000 samples/year with two chromatographs, versus 2500 samples/year with the current manual procedures. This method also showed an improved precision when compared to the manual method for most of the fatty acids analysed. The acquisition of this automated system will then result in significant saving of time and cost per sample and improvement of the quality of the data.

Estimated cost: €115 000

2) Replenishment of the EPIC biobank

19. EPIC is a longitudinal cohort comprising ~520 000 participants recruited across ten European countries from 1992–1999. The cohort was originally funded by the European Commission as well as national funds from the participating countries and central support from IARC. The cost of establishing the EPIC cohort is estimated to be in the region of €150 million, with continuing substantial investment since. More than 8.4 million person-years have now accrued across the cohort and as of the last round of follow up in 2015, more than 62 000 EPIC participants had received a cancer diagnosis. This large number of incident cases included 15 637 cases of breast cancer, 7328 prostate cancers, 6738 colorectal cancers and 4298 lung cancers. In addition to the large numbers of commonly occurring cancers, the combination of size and follow-up time of the cohort has meant that even for less common and rare tumours, the number of incident cases is sizeable including, for example, 1246 cases of kidney cancer, 1397 pancreatic cancers, 1724 ovarian cancers and 544 gliomas.

20. A major resource of EPIC is the centralized biorepository that houses blood-based biological samples from over 350 000 subjects from the EPIC cohort in eight of the ten EPIC countries, with an identical set of samples stored locally in each of the participating centres. EPIC has been extremely productive in developing cancer specific studies on risk biomarkers and also biomarkers of early detection that have used biological samples from this central biorepository, typically serum or plasma.

21. A consequence of the scientific productivity of the EPIC network is the depletion of serum and plasma samples from a proportion of incident cancer cases at the IARC Biobank. This is particularly so for frequently studied cancers (e.g. colorectal, pancreas, lung) and for cancer cases diagnosed within the first five years of follow-up, which are of particular interest for studies of early detection of cancer. This situation presents problems for future studies that will require access to biological samples for specific cancer studies particularly when many large scale analytical tools are only now becoming available.

22. To ensure that EPIC continues to lead and contribute to cutting edge research that requires biological samples, including biological markers of early detection or cancer risk, we propose to undertake a replenishment of the IARC EPIC biobank for serum and plasma samples for all incident cancer cases and matched controls across EPIC. This will ensure that at least three plasma samples and two serum samples are available for each EPIC participant. This number will mirror exactly half of what was originally stored at both IARC and the local EPIC centres (i.e. six straws of plasma and four straws of serum). One straw (0.5 ml) can provide sufficient sample for multiple studies, particularly for newer technologies (e.g. metabolomics, proteomics) that require very small amounts of material (<100 µl).

23. The principle to replenish the EPIC biorepository was agreed at the recent EPIC Steering Committee meeting that took place in London in September 2017. The local EPIC centres, where additional serum and plasma samples are housed, have agreed to participate in the replenishment of the central IARC biobank. It was also agreed at the EPIC Steering Committee level that there will be strict governance and oversight on how the biological samples are used for future projects and, in particular, samples from cancer cases that were diagnosed within five years of blood draw will be preserved for early detection studies. Rules on the future access to biospecimens in EPIC will be developed in the coming months.

24. The proposed replenishment of the EPIC Biobank will parallel ongoing efforts supported by internal NME resources to substantially enhance the EPIC database at IARC. This entails centralization of data collected by the local EPIC centres during the follow-up period and includes updated information on anthropometry, lifestyle (smoking, alcohol, physical activity), diet and co-morbidities (diabetes, cardiovascular disease, neurodegenerative disorders) as well as all existing genetic (GWAS) and biomarker data. It is anticipated that the upgraded database along with the replenishment of samples will ensure that EPIC remains a resource for cutting-edge cancer research in the years to come.

25. Funding is required to identify and move the approximately 22 000 straws for 5656 cancer cases and 5656 matched controls from the regional centres to IARC. The estimated budget for replenishment of the EPIC biorepository is €215 000: €95 000 to the EPIC partners to draw the straws and ship them to IARC, and €120 000 for new in-house sample storage equipment and for IARC personnel to receive and document all samples. These costs are an approximation; if the proposed budget should prove overestimated, the unused funds will be returned to the GCSF. It is estimated that this work will be undertaken over an 18-month period.

Requested budget

	Approximate price (€)	**Annual maintenance costs (€)**
a) Automated immunostainer	120 000	4000
b) Automated device for nucleic acid quality control	50 000	3000
c) Automated system for plasma phospholipid fatty acid profiling	115 000	Nil
Replenishment of the EPIC biobank		Nil
- Retrieval and shipment from EPIC Centres	95 000	
- Liquid nitrogen tank	30 000	
- Other materials and reagents	30 000	
- Staff costs – Biobank	60 000	
Total	215 000	
Grand total	**500 000**	