CALERIE™ SAMPLE ACCESS AND ANCILLARY STUDY GUIDELINES

Note: *potential applicants should carefully review this guidelines document. Access to the biospecimens must be approved by the SC prior to any submission of a grant application to any funding agency. Failure to follow this guidance may not get access to CALERIE™ samples for their studies as proposed, even if funding is secured.*

An ancillary study involves analysis of the existing data and generation of new data using biological samples (blood, urine, muscle or adipose) collected from CALERIE™ participants and stored in the CALERIE™ Data Repository. A list of all currently planned analyses by investigators using CALERIE™ data and samples is maintained on the CALERIE™ Website in the *Ongoing Projects tab*. All proposals for secondary analysis studies are reviewed, scored and approved by the CALERIE™ Steering Committee (SC) using the procedure outlined below.

A. What proposals will be considered?

Investigators are encouraged to conduct secondary ancillary studies using CALERIE™ dataset and biorepository samples with the stipulation that they be scientifically sound and are consonant with the overall CALERIE™ study goals, stated as "*to test the hypothesis that two years of sustained caloric restriction (CR), involving a reduction in energy intake to 75% of baseline (25% CR), in healthy men aged 21 to 50 and healthy women aged 21 to 47 will result in the same adaptive changes occurring in rodents subjected to CR*."

Investigations proposing the use of CALERIE™ biorepository samples to address scientific questions duplicative of already existing and approved investigations will not be considered. We encourage investigators to check the *Ongoing Projects tab* at the CALERIE™ Web site. Requests for the use of "depleted samples" to addressing questions not furthering the goals of CALERIE™ will not be further considered. Please refer to *Guidelines for Maintenance of the Biorepository* section at the end of this document.

A data dictionary for the CALERIE™ database, and a listing of available samples of serum, plasma, urine, muscle and fat and extracted material (DNA and RNA) from whole blood and PAXgene tubes, is listed at the CALERIE™ web site. Access to the database and samples will be handled through the CALERIE™ web site.

B. The CALERIE™ Steering Committee

The SC is composed of CALERIE™ Investigators with spectrum of expertise relevant to CALERIE™. This may include the following:

- Biology of aging
- Metabolic physiology/energy balance/endocrinology
- Molecular predictors of aging: metabolomics/gene expression/epigenetics
- Behavioral lifestyle medicine

Additional membership consists of the Principal Investigator(s) of the sponsoring CALERIE™ U24 Network Resources Grant and the NIA. Membership quorum consists of a minimum of four voting members.

C. The CALERIE™ External Science Committee (ESC)

The CALERIE™ ESC provides independent expertise and opinion on the use of CALERIE™ Research Network resources that is independent of the CALERIE™ Investigators. It provides independent guidance on the approval of ancillary studies. The ESC is composed of membership representing a broad spectrum of expertise relevant to CALERIE™:

- CR studies in animal models
- Disease risk epidemiology - especially focusing on primordial risk
- Biology of aging
- Metabolic physiology/energy balance/endocrinology
- Molecular predictors of aging: metabolomics/gene expression/epigenetics

Additional membership consists of the two co-Principal Investigators of the sponsoring CALERIE™ Research Network Resources Grant. Membership quorum consists of a minimum of four voting members.

D. Application process and proposal format

Time line for submission of ancillary proposals and review. The CALERIE™ SC reviews proposals on a quarterly basis, on the fourth Friday in January, April, August and October. To be considered at a meeting, investigators are encouraged to submit new proposals at a minimum on the following dates: November 15 for January, February 1 for April, June 1 for August and August 1 for October meeting. This provides time for investigators to get feedback from the SC for revision of proposals in advance of the SC meeting. Proposals failing to meet these dates may not receive an approval letter for inclusion in the application.

An investigator wishing to conduct an ancillary study or perform secondary data analyses will submit a proposal to the chair of the SC who will track the proposal through the approval process. All materials should be submitted as a single pdf.

Please include a Research Proposal with the suggested format:

- General Research Proposal
- Structured Abstract
- Background and Rationale
- Specific Aims
- Methods: Justify the requested sample numbers (e.g., ***sample size calculations***) and amounts (e.g., ***fluid volume, tissue mass***).
- Describe the laboratory performing the assays, the laboratory methods, and quality control with enough detail that reviewers can assess the validity of the methods.
- Literature References

Assistance with proposals, including clarification on obtaining access to the database, guidance on what resources are available, and other items can be obtained at the CALERIE™ Website (http://calerie.duke.edu/)

When the application is complete, the chair will send the proposal to the Committee for review as outlined, below. If additional expertise is required, the SC may seek additional external review.

E. Review process

The following criteria will be used by the CALERIE™ SC to make a decision about whether to approve a proposed study:

- The proposed study addresses an important scientific problem related to the objectives of CALERIE™;

- The aims are achievable and advance scientific knowledge in the topic area
- The study is not a duplication of the ongoing studies/analyses.
- Proposed methods are scientifically sound, feasible and reliably measure the proposed outcomes.
- The requesting investigators are appropriately trained and well suited to carry out the project, and their experience level is appropriate for the proposed work.
- Required equipment is available.
- A funding source has been identified.
- The scientific environment in which the work will be done contributes to the probability of success.
- Potential overlap with other similar projects. If significant overlap exists, projects may be asked to combine resources to preserve and optimally use non-renewable samples.

After discussion, the members of the SC will reach consensus and/or bring the proposal to a vote.

Notes / special situations:

A letter will be provided by the SC Chair to the study investigator upon approval of the completed funding application. The investigators will be required to sign the data/sample use agreement (Appendix 1) prior to receiving access to CALERIE™ data and / or biospecimens.

F. IRB approval

IRB approval is not required to submit a proposal to the CALERIE™ SC. However, access to samples and data will require approval of the local IRB of the proposing investigator.

G. Funding

All studies must be approved by the SC before funding applications are submitted to the funding agencies. Proposals for funding must include coverage of all the costs, including administration, data access costs, and sample access and mailing costs. The cost of sample access and mailing can be obtained through consultation with the Biorepository Director at the University of Vermont (contact information will be available at the CALERIE™ Web page). An ancillary study investigator may not enter into any verbal or written agreement or contract with industry or private individuals that will provide funding for anything related to CALERIE™ without prior review and written approval from the CALERIE™ Steering Committee.

H. Changes after approval

The proposing investigator is required to provide annual (semi-annual) reports to the SC informing the committee about the study progress. Studies supported by the National Institutes of Health (NIH) may submit their annual progress reports to the CALERIE™ SC at the deadlines required by the funding Institute. Changes in study design between submissions of revised proposals must be considered by the SC before a revised letter is provided. Once a proposal is approved, samples will be reserved in the investigator's name for up to three years in order for the investigator to receive final funding (that is, the approval letter is good for up to three years from the date on the support letter.

I. Data

Data collected by the ancillary study and any relevant documentation should be provided to the CALERIE™ Research Network in a timely manner for integration into the main data repository. The

ancillary study PI will be given the first and exclusive opportunity to analyze, present and publish data collected under the auspices of the ancillary study. Within 12 months of publication of ancillary study results or at the time ancillary study funding ends (whichever occurs first), the ancillary study data should be made available for additional uses by other investigators.

J. Data analysis, publications and presentations

All the publications, presentations and abstracts must acknowledge the CALERIE™ study (U24-AG047121).

GUIDELINES FOR MAINTENANCE OF THE BIOREPOSITORY

The CALERIE™ biorepository serves several purposes:

1. Provide access to biological samples for research directed at the effects of calorie restriction in humans.
2. Generate molecular data to curate the CALERIE™ data repository such that the CALERIE™ data repository can serve as a platform for the greater scientific community to use systems biology to study the integrated responses to calorie restriction in humans

Stewardship of CALERIE™ biologic samples will be the responsibility of the CALERIE™ Steering Committee (SC) until such time as the committee yields such responsibility to another entity.

The CALERIE™ Steering Committee — with guidance from an External Science Committee (ESC) — will provide access to samples according to the directives as set forth in the CALERIE™ Sample Access and Secondary Analysis Studies Guidelines Document to which this document is appended.

Proposals will be considered in the context of a biorepository impact statement generated by the proposing investigator in contact with CALERIE™ Biorepository personnel.

A running record of the contents of the CALERIE™ Biorepository will be maintained on the CALERIE™ Web site for use by CALERIE™ investigators. It will be updated on a quarterly basis.

The Biorepository contents will be reviewed by the CALERIE™ Steering Committee regularly and when necessary, but no less frequently than on a yearly basis.

Smart stewardship of samples will be the responsibility of the Steering Committee, according to the following table.

Sample Type	Depleted Short-term, Limited, Restricted Use	Critical Long-term Storage. Use under Exceptional Circumstances	Comments
Muscle	50 mg	10 mg	
Muscle RNA	5 mcg	2 mcg	
Fat	100 mg	25 mg	
Fat RNA	5 mcg	2 mcg	
Plasma	2 mL	500 µL	
Blood RNA	5 mcg	2 mcg	
Blood DNA	10 mcg	2 mcg	

Some limited mounted samples are available for histochemistry. Investigators seeking access to histochemistry samples should confer with the Steering Committee before submitting their proposals.

Note, these guidelines apply to each time point for a given participant. For example, if baseline, and 24-month samples are plentiful but 12-month samples are not for a participant, an investigator may only be allowed access to the baseline and 24-month samples.

CALERIE™ samples are considered non-renewable resources.

Depleted and Critical specimens shall be used only for studies that have direct relevance to the CALERIE™ primary hypotheses.

Samples qualified as **Depleted** will require consent of a majority of the SC and consent of a majority of a quorum of the ESC to provide access to the samples by study investigators.

Samples qualified as **Critical** will require unanimous consent of the SC and the ESC to provide access to the samples by study investigators.

All data generated by use of CALERIE™ human tissues will be return to the CALERIE™ SC to be added to the data repository at the earliest convenience of the investigator, but no later than six months after generation. An embargo period for publication may be provided to the investigator for publication of results prior to public release of the data.