

**External RNA Control
Consortium (ERCC) Workshop**

Product Description Section

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The ERCC proposes to develop two “products”

To be used as tools to monitor microarray assay performance and to facilitate comparisons across platforms.

- 1.) A set of characterized clones and associated data for producing *in vitro* transcripts for use as external RNA standards.
- 2.) A set of standardized protocols and formulations for the expected [commercial] development of transcript pools as spiked-in standards.

Benchmark controls for determination of sensitivity, dynamic range, non-specific background and reproducibility

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ERCC working group validation

Clones and protocol testing

- **Stages of production**
- **Quantification**
- **Quality control assessment**
- **Cross-reactivity screening**
- **Testing on different expression platforms**

An emphasis will be placed on defining formulations for standardized pools

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Clone selection

A set of 100 clones

- Unique random sequences
- *Bacillus subtilis*
- *Arabidopsis thaliana* sequences

Tested for cross-reactivity

- *H. sapiens*
- *M. muscularis*
- *R. norvegicus*
- *S. cerevisiae*
- *A. thaliana*
- *E. coli*
- *C. elegans*
- *D. melanogaster*

Clones will have a GC content of 40-60%, be void of repetitive elements, with marginal cross reactivity defined as having no more than 20 contiguous bases of identity and overall homology less than 70%.

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Clone information

- **Sequence selection**
- **Clone construction**
- **Quality control**
- **Characterization**
- **Nucleic acid sequences**
- **Recommended handling and storage**
- **Stability data**
- **Recommended protocols for use**
- **Quantitative information on nucleic acids**
- **Observed performance characteristics**
- **Standardized vector with defined poly-A tail length and T7 promoter**

***All relevant information will be published
in the open, archival literature.***

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Two functional classes of candidate controls

Fixed Length Transcripts (700-800 nt) for:

- quantitative assessment of sensitivity
- dynamic range
- transcript abundance
- relative difference in abundance (ratio)

Variable Length Transcripts (500-2000 nt) for:

- Monitoring fidelity of RNA isolation
- labeling methods

The ERCC will propose a set of DNA sequences for designing capture probes that will serve as negative controls

The ERCC project phases

Specific Aim 1:

- **Will address the definition, assembly and initial production of the candidate clones and transcripts**

Specific Aim 2:

- **Will address testing the clones on various platforms and in different configurations**

The overall aim is to define a set of validated transcripts and recommendations, for use on various platforms which enables subsequent commercial development and distribution as standardized pooled transcripts for research applications.