

BBF RFC 45: Cloning Standard for Mammalian BioBrick Parts and Devices

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1. Purpose

To introduce a common cloning standard for BioBrick parts that find application in mammalian cells.

2. Relation to other BBF RFCs

Comments upon and extends RFC-12; Replaces RFC-10, RFC-20, RFC-21, RFC-25 for use in mammalian cells.

3. Copyright Notice

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4. Definition

RFC-12 (Tom Knight's Bbb proposal) SHOULD be applied to Parts and Devices for use in mammalian cells.

5. Rationale

The existence of multiple cloning standards presents a problem to biological engineers. Considering the weaknesses of the original BioBrick standard (does not allow for protein fusions), it is not surprising that a multitude of cloning standards has been proposed. As current discussion shows, each of the proposed alternatives has several disadvantages [1], but still, each standard is applied, for most registry parts are in the original BioBrick standard and compatibility is desired. RFC-12 is recognized as the most advanced standard with no disadvantages other than incompatibility with existing standards [1].

On the other hand, the registry contains a very small number of mammalian parts and devices. By introducing RFC 41-43, we hope to set a process in motion that will alter this fact. In mammalian systems, the ability to perform protein fusions is a central requirement. For example, eukaryotic cells are compartmentalized and protein targeting can only be achieved by fusing proteins to targeting sequences.

6. Promoter structure

In eukaryotic cells, promoters can be subdivided into a core and a proximal promoter[2]. We suggest that core and proximal promoter MAY be separated by a HindIII site. This allows for rapid swapping of core promoters (alters strength [3]) and proximal promoters (alters regulation).

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References

[1]

http://openwetware.org/wiki/The_BioBricks_Foundation:Standards/Technical/Formats

[2] Heintzman ND, Ren B. The gateway to transcription: identifying, characterizing and understanding promoters in the eukaryotic genome. Cellular and Molecular Life Science 64, 386-400 (2007).

[3]

http://2009.igem.org/Team:Heidelberg/Project_Measurement#Different_core_promoters_result_in_different_expression_strength