



UvA-DARE (Digital Academic Repository)

Identification of reliable reference genes for qRT-PCR studies of the developing mouse mammary gland

van de Moosdijk, A.A.A.; van Amerongen, R.

DOI

[10.1038/srep35595](https://doi.org/10.1038/srep35595)

Publication date

2016

Document Version

Other version

Published in

Scientific Reports

[Link to publication](#)

Citation for published version (APA):

van de Moosdijk, A. A. A., & van Amerongen, R. (2016). Identification of reliable reference genes for qRT-PCR studies of the developing mouse mammary gland. *Scientific Reports*, 6, [35595]. <https://doi.org/10.1038/srep35595>

General rights

It is not permitted to download or to forward/distribute the text or part of it without the consent of the author(s) and/or copyright holder(s), other than for strictly personal, individual use, unless the work is under an open content license (like Creative Commons).

Disclaimer/Complaints regulations

If you believe that digital publication of certain material infringes any of your rights or (privacy) interests, please let the Library know, stating your reasons. In case of a legitimate complaint, the Library will make the material inaccessible and/or remove it from the website. Please Ask the Library: <https://uba.uva.nl/en/contact>, or a letter to: Library of the University of Amsterdam, Secretariat, Singel 425, 1012 WP Amsterdam, The Netherlands. You will be contacted as soon as possible.

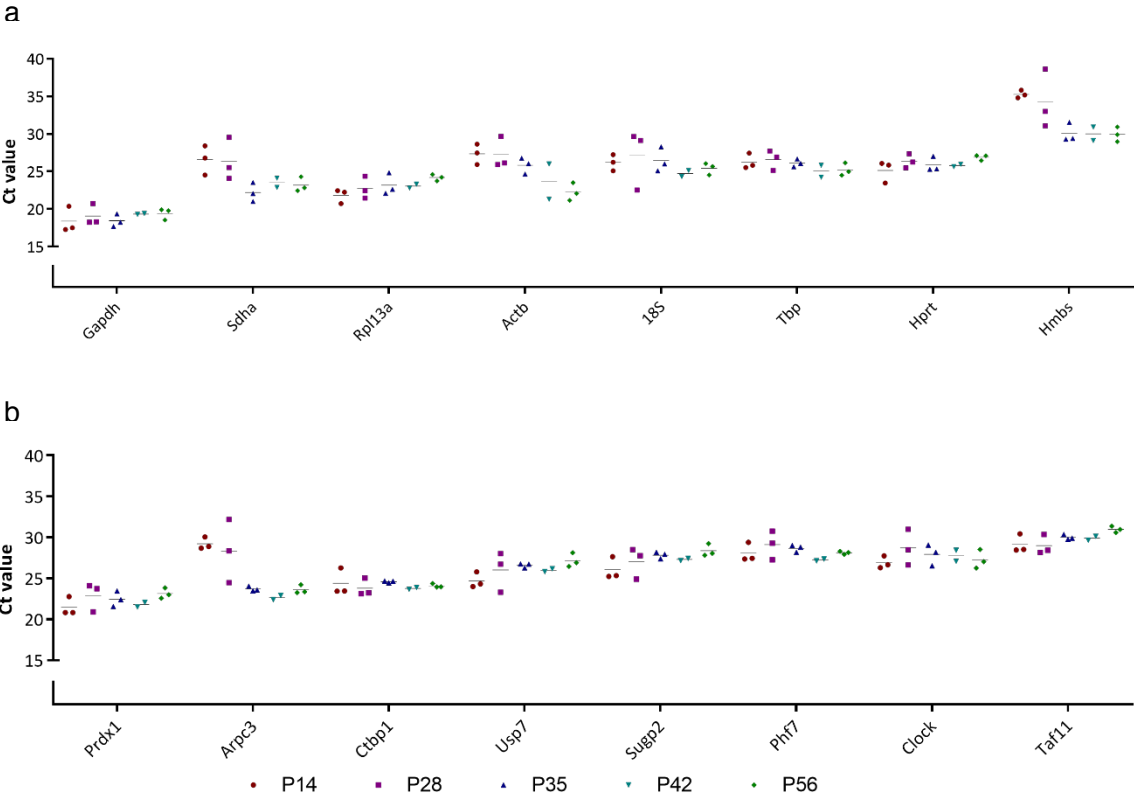
UvA-DARE is a service provided by the library of the University of Amsterdam (<https://dare.uva.nl>)

Identification of reliable reference genes for qRT-PCR studies of the developing mouse mammary gland

Anoeska Agatha Alida van de Moosdijk and Renée van Amerongen

Supplementary Files

- [Supplementary Figure S1 \(this PDF\)](#)
- [Supplementary Figure S2 \(this PDF\)](#)
- [Supplementary Table S1 \(uploaded separately as an .xlsx file\)](#)
- [Supplementary Table S2 \(this PDF\)](#)



Supplementary figure S1. Ct values and reference gene ranking for #3 mammary gland. Scatter plot depicting spread in expression of candidate reference genes in different stages of the #3 mouse mammary gland. a) Traditional reference genes b) Novel candidate reference genes.

(A)

Ranking Order (Better--Good--Average)								
Method	1	2	3	4	5	6	7	8
Delta CT	Tbp	Rpl13a	Hprt	18S	Gapdh	Sdha	Actb	Hmbs
BestKeeper	Rpl13a	Tbp	Hprt	Gapdh	18S	Sdha	Actb	Hmbs
Normfinder	Tbp	Rpl13a	Hprt	18S	Sdha	Gapdh	Actb	Hmbs
GeNorm qBase	Rpl13a	Hprt	Gapdh	Tbp	18S	Sdha	Actb	Hmbs
Recommended comprehensive ranking	Rpl13a	Tbp	Hprt	Gapdh	18S	Sdha	Actb	Hmbs

(B)

Ranking Order (Better--Good--Average)								
Method	1	2	3	4	5	6	7	8
Delta CT	Prdx1	Sugp2	Ctbp1	Phf7	Usp7	Taf11	Clock	Arpc3
BestKeeper	Prdx1	Ctbp1	Phf7	Sugp2	Taf11	Usp7	Clock	Arpc3
Normfinder	Prdx1	Phf7	Ctbp1	Sugp2	Clock	Usp7	Taf11	Arpc3
GeNorm qBase	Phf7	Prdx1	Ctbp1	Sugp2	Usp7	Taf11	Clock	Arpc3
Recommended comprehensive ranking	Prdx1	Phf7	Ctbp1	Sugp2	Usp7	Taf11	Clock	Arpc3

Supplementary Figure S2.

Comprehensive ranking of the old/traditional (A) and novel candidate reference genes (B) using RefFinder (available at <http://fulxie.0fees.us/?type=reference>). A combined comprehensive ranking of both the traditional and novel candidate genes is presented in Table 1.

Gene name	Accession Number	Primer sequence (5' – 3')	Amplicon length	PCR Efficiency %
Reference genes				
<i>18S</i>	NR_003278	F: CCGCCGCCATGTCTCTAGT R: CTTTCCTCAACACCACATGAGC	150 bp	90
<i>Arpc3</i>	NM_019824	F: TGAAGCGGACAGGACATTGAT R: AGTTGGTGATTCTAGCGTGT	122 bp	92
<i>Actb</i>	NM_007393	F: GAGCTGCCTGACGGCCAGGT R: TACTCCTGCTTGCTGATCCA	365 bp	72
<i>Clock</i>	NM_001289826	F: TTTACAGGCGTTGTTGATTGGA R: ACGCAAGGCCGTCTTCTG	92 bp	97
<i>Ctbp1</i>	NM_001198859	F: GTGCCCTGATGTACCATACCA R: GCCAATTCGGACGATGATTCTA	83 bp	92
<i>Gapdh</i>	NM_001289726	F: CTGGTGCTGCCAAGGCT R: CTGCTTCACCACCTTCTTGATGTC ATCATA	161 bp	99
<i>Hmbs</i>	NM_013551	F: GCCCATGTGCCTTCAGTC R: TCTTTGAGCCGTTTTCTCCGC	151 bp	101
<i>Hprt</i>	NM_013556	F: TGTTGTTGGATATGCCCTTG R: TTGCGCTCATCTTAGGCTTT	108 bp	97
<i>Phf7</i>	NM_027949	F: TGTTGCTTCTATGCCTTCAA R: GGCAGTCTGCTAGACAGGATAAG	114 bp	88
<i>Prdx1</i>	NM_011034	F: AATGCAAAAATTGGGTATCCTGC R: CGTGGGACACACAAAAGTAAAGT	150 bp	96
<i>Rpl13a</i>	NM_009438	F: CCCTCCACCCTATGACAAGA R: GCCCCAGGTAAGCAAACCTT	95 bp	101
<i>Sdha</i>	NM_023281	F: GTTGCTGTGTGGCTGATCG R: GCACAGTGCAATGACACCAC	149 bp	89
<i>Sugp2</i>	NM_001168290	F: TGACCAGAGGCCGATCTCTAA R: GGAAGTAATACGACTGGCAACAG	158 bp	105
<i>Tbp</i>	NM_013684	F: GTCATTTTCTCCGAGTGCC R: GCTGTTGTTCTGGTCCATGAT	151 bp	94
<i>Taf11</i>	NM_026836	F: GATGGAGACGCGGACTTGAAA R: GCAGGAGTAAGTAAGGACGAGTT	113 bp	99
<i>Usp7</i>	NM_001003918	F: CCACAAGGAAAACGACTGGG R: GTAACACGTTGCTCCCTGATT	196 bp	88
Target genes				
<i>Wnt4</i>	NM_009523	F: ACTGGACTCCCTCCCTGTCT R: TGCCCTTGCTACTGCAAA	109 bp	97

Supplementary table S2. Overview of all genes and primer sequences used in this study. The length of the amplicon and the efficiency of the primers is also listed.