

S2 Fig. HM1 peptide sequence comparison of *hm1* alleles. Sequence comparison of HM1^A with HM1 from the resistant maize inbreds (B73, Va35, W22, and Pr1), maize cultivars (Enano and Pira), HM1 homologs [sorghum (*Sorghum bicolor*), rice (*Oryza sativa*), and barley (*Hordeum vulgare*)], and maize dihydroflavonol 4-reductase (DFR). Among the five amino acid substitutions present in HM1^A (highlighted in red and bold), the L116 residue was a conserved in all *Hm1* alleles and orthologs.

HM1-B73	-----MAEKESNGVRVCVTGGAGFIGSWLVRKLLLEKGYTVHATLRNTGDEAKAGLLRR
HM1A	-----MAEKESNGVRVCVTGGAGFIGSWLVRKLLLEKGYTVHATLRNTGDEAKAGLLRR
HM1-Va35	-----MAEKESNGVRVCVTGGAGFIGSWLVRKLLLEKGYTVHATLRNTGDEAKAGLLRR
HM1-Pr1	-----MAEKESNGVRVCVTGGAGFIGSWLVRKLLLEKGYTVHATLRNTGDEAKAGLLRR
HM1-W22	-----MAEKESNGVRVCVTGGAGFIGSWLVRKLLLEKGYTVHATLRNTGDEAKAGLLRR
HM1-Enano	-----DEAKAGLLRR
HM1-Pira	-----DEAKAGLLRR
HM1-Sorghum	-----MAENESSGVRVCVTGGAGFIGSWLVKKLLEKGYTVHATLRNTGDEEKAGLLRR
HM1-Rice	MAEEGR-SGGVAGDGVRVCVTGGAGFIASWLVKKLLERGCIVHATLRSMGDEEKAGLLRR
HM1-Barley	MAEEGKGTGSNGGRGVRACVTGGAGFIGSWLVRKLLLEAGYTVHATLRSIGDEGKAGLLRG
DFR-Maize	-----MAGGGATVCVTGAGGFIASWVVKLLLSRGTGTVHGTVRHLSDEKT-GHLKR
	S99Y D110Y
HM1-B73	LVPGAA--ERLRLFQADLFDAATFAPAIAGCQFVFLVATPFGLDSAG S QYKSTAEAVV DA
HM1A	LVPGAA--ERLRLFQADLFDAATFAPAIAGCQFVFLVATPFGLDSAG Y QYKSTAEAVV YA
HM1-Va35	LVPGAA--ERLRLFQADLFDAATFAPAIAGCQFVFLVATPFGLDSAG Y QYKSTAEAVV DA
HM1-Pr1	LVPGAA--ERLRLFQADLFDAATFAPAIAGCQFVFLVATPFGLDSAG Y QYKSTAEAVV DA
HM1-W22	LVPGAA--ERLRLFQADLFDAATFAPAIAGCQFVFLVATPFGLDSAG S QYKSTAEAVV DA
HM1-Enano	LVPGAA--ERLRLFQADLFYAATFAPAIAGCQFVFLVATPFGLDSAG S QYKSTAEAVV YA
HM1-Pira	LVPGAA--ERLRLFQADLFDAATFAPAIAGCQFVFLVATPFGLDSAG Y QYKSTAEAVM YA
HM1-Sorghum	LVPGAA--ERLRLFQADLFDAATFAPAIAGCQFVFLVATPFGLQDAG S KYKSTAEAVV DA
HM1-Rice	LVPGAA--ERLRLFQADLFDAATFAPAIAGCQFVFLIATPYGLEASN S KYKNTADA AVDA
HM1-Barley	LVPGGAPPERLVLFQADLFDAATFAPAIAGCHFVFLVANPSAHE-PA S KYKTSAEAA DA
DFR-Maize	LENA---AGNLRIFKADLLDYDAMAAVVGCGVFHVATPVPSEDLT D PELQMLGPAV TG

L116H

HM1-B73	VRAILRQCEESRTVKRVIHTASVAAAASPLLEEE-VSASGVGYRDFIDESCWTSLNVDYPL
HM1A	VRAIHRQCEESRTVKRVIHTASVAAAASPLLEEE-VSASGVGYRDFIDESCWTSLNVDYPL
HM1-Va35	VRAILRQCEESRTVKRVIHTASVAAAASPLLEEE-VSASGVGYRDFIDESCWTSLKVNYP
HM1-Pr1	VRAILRQCEESRTVKRVIHTASVAAAASPLLEEE-VSASGVGYRDFIDESCWTSLNVDYPL
HM1-W22	VHAILRQCEESRTVKRVIHTASVAAAASPLLEEE-VPASGVGYRDFIDESCWTSLNVDYPL
HM1-Enano	VRAILRQCEESRTVKRVIHTASVAAAASPLLEEE-VSASGVGYRDFIDESCWTSLNVDYPL
HM1-Pira	VRAILRQCEESRTVKRVIHTASVAAAASPLLEEE-VSASGVGYRDFIDESCWTSLNVDYPL
HM1-Sorghum	VRAILRQCEESRTVKRVIHTASVSAASPLKEEE-VS-GVIGYREFISESCWTSPDVDYPL
HM1-Rice	VREILRQCAESKTVKRVIHTASISTASPLIDVPGAGVGAAGYRDFIDESCWTPLDVDYPL
HM1-Barley	VRVILRLCAESRTVKRVIHTASVTAASPLTKSSS--AAAVYGDFISESCWTLLDVNYPL
DFR-Maize	TTNVLLKA-ASSANVQRVVVVSSMVAVE-----ISPDKWPEGKVRDERCWSDREFCRSI

S191N

HM1-B73	RSAHFDKYILSKLRSEQELLSYNGGESPAFEVVTLPLGLVAGDTVLRAPETVESAVAPV
HM1A	RSAHFDKYILSKLRSEQELLYNNGGESPAFEVVTLPLGLVAGDTVLRAPETVESAVAPV
HM1-Va35	RSAHFDKYILSKLRSEQELLSYNGGESPAFEVVTLPLGLVAGDTVLRAPETVESAVAPV
HM1-Pr1	RSAHFDKYILSKLRSEQELLSYNGGESPAFEVVTLPLGLVAGDTVLRAPETVESAVAPV
HM1-W22	RSAHFDKYILSKLQSEQELLSYNGGESPAFEVVTLPLGLVAGDTVLRAPETVESAVAPV
HM1-Enano	RSAHFDKYILSKLRSEQELLYNNGGESPAFEVVTLPLGLVAGDTVLRAPETVESAVAPV
HM1-Pira	RSAHFDKYILSKLRSEQELLYNNGSESPAFEVVTLPLGLVAGDTVLRAPETVESAVAPV
HM1-Sorghum	RSAHFDKYILSKVQSEQELLSYNGGESPAFEVVTLALGLVAGDTVLRVPETVESAVSPV
HM1-Rice	RSAHFDKYVLSKMMSEKELLYGNDGEGRAFEVVTLPCGLVAGDTVLRAPETLENVAVSPV
HM1-Barley	RSVHFDKYIESKVLSEKELLSYNDGENPAFEVVTLPLGLVGGNTVLGYLPETMESVAVPV
DFR-Maize	E----SWYPVAKIISEEAALAYG--QQTGLDVVTINPGLVFGPMLQPTVNTTIQFLI---

L240P

HM1-B73	SRSEPCFGLLRILQQLLGSPLVHVDDVCDALVFCMERRPSVAGRFLCAAAYPTIHDVVA
HM1A	SRSEPCFGLPRILQQLLGSPLVHVDDVCDALVFCMERRPSVAGRFLCAAAYPTIHDVVA
HM1-Va35	SRSEPYFGLPRILQQLLGSPLVHVDDVCDALVFCMERRRSVAGRFLCAAAYPTIHDVVA
HM1-Pr1	SRSEPCFGLPRILQQLLGSPLVHVDDVCDALVFCMERRPSVAGRFLCAAAYPTIHDVVA
HM1-W22	SRSEPYFGLLRILQQLLGSPLVHVDDVCDALVFCMERRPSVAGRFLCAAAYPTIHDVVA
HM1-Enano	SRSEPYFGLLRILQQLLGSPLVHVDDVCDALVFCMERRPSVAGRFLCAA-----
HM1-Pira	SRSEPCFGLPRILQQLLGSPLVHVDDVCDALVFCMERRPSVAGRFLCAA-----
HM1-Sorghum	SRNEAYFGLPRILQQLLGSPLVHVDDVCDALIFCMERRHSIAGRFLCAAAYPTIHDVVG
HM1-Rice	SRNEPSFAFLRLLQRLVGSVPLVHADDVCDALVFCMDQ-PSLAGRFLCSAAYPTIHDIVE
HM1-Barley	TKQEPFMLPRILQRLVGSVPLVHVDDVCAALIFCMEQ-PALSGRFLCAAAYPTIHDILD
DFR-Maize	-----YFLKGGPDPVKNKLWHIVDVRDVADAMLLLYEV-PEATGRHICAPHVISARDLLD

HM1-B73	HYASKFPHLDILKET-EAVATV---RPARDRLGELGFKYKYGMEEILDSSVACAARLGSL
HM1A	HYASKFPHLDILKET-EAVATV---RPARDRLGELGFKYKYGMEEILDSSVACAARLGSL
HM1-Va35	HYASKFPHLDILKET-EAVATV---RPARDRLGELGFKYKYGMEEILDSSVACAARLGSL
HM1-Pr1	HYASKFPHLDILKET-EAVATV---RPARDRLGELGFKYKYGMEEILDSSVACAARLGSL
HM1-W22	HYASKFPHLDILKETTEAVATV---RPARDRLGELGFKYKYGMEEILDSSVACAARLGSL
HM1-Enano	-----
HM1-Pira	-----
HM1-Sorghum	HYARKFPHLDILKET-EAVARV---QPDGDR LGELGFVYKYGIEEILDSSVACAARLGCL
HM1-Rice	HFAAKYPHLDVLKEPEREVARV---QPAADKLGELGFRYKYGMEEILDGSVGCAARLGYI
HM1-Barley	HYGSKYPHLDLLRET-DEVARV---QPDKNKLGELGFYKYGLKEILDESIDCAVRLGSL
DFR-Maize	LLKSMYPDYPCIANE-SILDRDHPAPMTSDKLLKIGWSCRPL-LEETIVDTVECCLRAGFL

HM1-B73	DASKLGLQKG-----
HM1A	DASKLGLQKG-----
HM1-Va35	DASKLGLQKG-----
HM1-Pr1	DASKLGLQKA-----
HM1-W22	DGSKLGLQKG-----
HM1-Enano	-----
HM1-Pira	-----
HM1-Sorghum	DATKLSMQKG-----
HM1-Rice	DAAKLRPQEG-----
HM1-Barley	DASKLIVQQG-----
DFR-Maize	DDVGGETSCRFPPLLNQI