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(54) **Mutated acetohydroxyacid synthase genes in brassica**

(57) The invention relates to mutated acetohydroxyacid synthase (AHAS) nucleic acids, expression vectors comprising the mutated nucleic acids, and proteins encoded by the mutated nucleic acids. The invention further relates to plants and seeds, respectively, which comprise

an acetohydroxyacid synthase (AHAS) gene, wherein said gene encodes a mutated AHAS protein. Further provided is a method for producing an herbicide-resistant plant, a method for increasing the herbicide-resistance of a plant, and a method of controlling weeds in a field.

**Figure 2A**

**Bn2 and Bn11 BnAHAS gene I translated**

MAAATSSSPISLTAKPSSKSPLPISRFSLPFSLTPQKDSRLHRPLAISAVLNSPVNVA  
 PPSPEKTDKNKTFVSRYPDEPRKGADILVEALERQGVETVFAYPPGGASMEIHQALTRS  
 STIRNVLP RHEQGGVFAAEGYARSSGKPGIC IATSGPGATNLVSGLADAMLDVPLVAI  
 TGQVPRRMIGTDAFQETPIVEVTRSITKHNYLVMDVDDIPRIVQEAFFLATSGRPGPVL  
 VDVPKDIQQQLAIPNWDQPMRLPGYMSRLPQPPEVSQLGQIVRLI SE SKRPVLYVGGGS  
 LNSSEELGRFVELTGIPVASTLMGLGSYPCNDEL SLQMLGMHGT VYANYAVEHSDLLLA  
 FGVRFD DRTGKLEAFASRAKIVHIDIDS AEIGKNKTPHVSVC GDVKLALQGMNKVLEN  
 RAEELKLD FGVWRSELSEQKQKFP LSFKTFGEAIP PQYAIQILDELTEGKAIISTGVGQ  
 HQMWAAQFYKYRKPRQWLSSSGLGAMGFGLPAAIGASVANPD AIVVDIDGDGSFIMNVQ  
 ELATIRVENLPVKILLNQH LGMVMQWEDRFYKANRAHTYLGDPARENEIFPNMLQFG  
 ACGIPAARVTKKEELREAIQTMLDTPGPYLLDVICPHQEHVLP MIPSGGTFKDVITEGD  
 GR TKY\*

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DOCUMENTS CONSIDERED TO BE RELEVANT			
Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	CLASSIFICATION OF THE APPLICATION (IPC)
A	US 2003/097692 A1 (JANDER GEORG [US] ET AL) 22 May 2003 (2003-05-22) * claims 1-4; sequence 4 *	1-9, 11-15	INV. C12Q1/68 C12N5/04 A01H1/00 C12N9/88 C12N15/82
A	JOSE MARIA BRUNIARD: "Inheritance of imidazolinone resistance, characterization of cross-resistance pattern, and identification of molecular markers in sunflower (Helianthus annuus L.)", DISSERTATION,, 1 September 2001 (2001-09-01), page 89PP, XP009138420, * page 12, line 18 - page 13, line 19; table 20 *	1-9, 11-15	
A	KOLKMAN J M ET AL: "Acetohydroxyacid synthase mutations conferring resistance to imidazolinone or sulfonylurea herbicides in sunflower", THEORETICAL AND APPLIED GENETICS, SPRINGER, BERLIN, DE LNKD- DOI:10.1007/S00122-004-1716-7, vol. 109, no. 6, 1 October 2004 (2004-10-01), pages 1147-1159, XP002417954, ISSN: 0040-5752 * page 1157, line 8 - page 1158, column 2, line 7 * * abstract *	1-9, 11-15	TECHNICAL FIELDS SEARCHED (IPC)  C12N C07K A01H
----- -/--			
<del>The present search report has been drawn up for all claims</del>			
Place of search Munich		Date of completion of the search 8 January 2014	Examiner Obel, Nicolai
CATEGORY OF CITED DOCUMENTS X : particularly relevant if taken alone Y : particularly relevant if combined with another document of the same category A : technological background O : non-written disclosure P : intermediate document		T : theory or principle underlying the invention E : earlier patent document, but published on, or after the filing date D : document cited in the application L : document cited for other reasons ..... & : member of the same patent family, corresponding document	

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EUROPEAN SEARCH REPORT

Application Number  
EP 13 17 5389

5

10

15

20

25

30

35

40

45

DOCUMENTS CONSIDERED TO BE RELEVANT			
Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	CLASSIFICATION OF THE APPLICATION (IPC)
A	WHITE A D ET AL: "ISOLATION OF ACETOLACTATET SYNTHASE HOMOLOGS IN COMMON SUNFLOWERS", WEED SCIENCE, WEED SCIENCE SOCIETY OF AMERICA, CHAMPAIGN, IL, US, vol. 51, no. 6, 1 November 2003 (2003-11-01), pages 845-853, XP009058786, ISSN: 0043-1745 * Introduction * * table 4 *	1-9, 11-15	
A,P	WO 2008/071715 A1 (BASF AGROCHEMICAL PRODUCTS B V [NL]; ADVANTA SEEDS BV [NL]; LEON ALBER) 19 June 2008 (2008-06-19) * claims 32-34 *	1-9, 11-15	
E,D	WO 2008/124495 A2 (BASF PLANT SCIENCE GMBH [DE]; MCELVER JOHN A [US]; SINGH BIJAY [US]) 16 October 2008 (2008-10-16) * claim 1r *	1-9, 11-15	
A	WO 2006/060634 A2 (BASF AGROCHEMICAL PRODUCTS B V [NL]; AGRIPRO WHEAT [US]; MOFFATT JOHN) 8 June 2006 (2006-06-08) * the whole document *	1-9, 11-15	
A	WO 2007/005581 A2 (BASF AG [DE]; NIDERA SEMILLAS S A [AR]; SALA CARLOS ALBERTO [AR]; ECHA) 11 January 2007 (2007-01-11) * the whole document *	1-9, 11-15	
A	WO 2005/020673 A1 (BASF AG [DE]; LIVORE ALBERTO B [AR]; PRINA ALBERTO R [AR]; BIRK IWONA) 10 March 2005 (2005-03-10) * the whole document *	1-9, 11-15	TECHNICAL FIELDS SEARCHED (IPC)
----- -/--			
----- -/--			
<del>The present search report has been drawn up for all claims</del>			
Place of search Munich		Date of completion of the search 8 January 2014	Examiner Obel, Nicolai
CATEGORY OF CITED DOCUMENTS X : particularly relevant if taken alone Y : particularly relevant if combined with another document of the same category A : technological background O : non-written disclosure P : intermediate document		T : theory or principle underlying the invention E : earlier patent document, but published on, or after the filing date D : document cited in the application L : document cited for other reasons ..... & : member of the same patent family, corresponding document	

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EP 13 17 5389

5

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30

35

40

45

50

55

DOCUMENTS CONSIDERED TO BE RELEVANT			
Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	CLASSIFICATION OF THE APPLICATION (IPC)
A	WO 01/85970 A2 (UNIV LOUISIANA STATE [US]; CROUGHAN TIMOTHY P [US]) 15 November 2001 (2001-11-15) * the whole document *	1-9, 11-15	
A	WO 2006/024351 A1 (BASF AGROCHEMICAL PRODUCTS B V [NL]; ADVANTA SEEDS BV [NL]; LEON ALBER) 9 March 2006 (2006-03-09) * the whole document *	1-9, 11-15	
X	EP 0 492 113 A2 (AMERICAN CYANAMID CO [US]) 1 July 1992 (1992-07-01) * claims 4,7 *	3,4,7-9, 11-14	
X	WO 2004/062351 A2 (VIRGINIA TECH INTELL PROP [US]; WHALEY CORY M [US]; WILSON HENRY P [US]) 29 July 2004 (2004-07-29) * example 2; sequence 5 *	3-9, 11-15	
Y		1,2	
Y,D	US 6 271 360 B1 (METZ RICHARD A [US] ET AL) 7 August 2001 (2001-08-07) * column 6, lines 34-43; example 6 *	1,2	
A	M.K Tan ET AL: "Characterisation of the acetolactate synthase (ALS) gene of Raphanus raphanistrum L. and the molecular assay of mutations associated with herbicide resistance", Plant Science, 1 January 2002 (2002-01-01), pages 195-205, XP055091823, DOI: 10.1016/S0168-9452(02)00082-1 Retrieved from the Internet: URL:http://www.sciencedirect.com/science/article/pii/S0168945202000821 [retrieved on 2014-01-09] * the whole document *	1-9, 11-15	
----- -/--			
<del>The present search report has been drawn up for all claims</del>			
Place of search Munich		Date of completion of the search 8 January 2014	Examiner Obel, Nicolai
CATEGORY OF CITED DOCUMENTS X : particularly relevant if taken alone Y : particularly relevant if combined with another document of the same category A : technological background O : non-written disclosure P : intermediate document		T : theory or principle underlying the invention E : earlier patent document, but published on, or after the filing date D : document cited in the application L : document cited for other reasons ----- & : member of the same patent family, corresponding document	

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DOCUMENTS CONSIDERED TO BE RELEVANT			
Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	CLASSIFICATION OF THE APPLICATION (IPC)
A	EP 0 257 993 A2 (DU PONT [US]) 2 March 1988 (1988-03-02) * the whole document *	1-9, 11-15	
A	US 2007/118920 A1 (LEON ALBERTO JAVIER [AR] ET AL) 24 May 2007 (2007-05-24) * claims 1-7; example 5; table 6 *	1-9, 11-15	
			TECHNICAL FIELDS SEARCHED (IPC)
<del>The present search report has been drawn up for all claims</del>			
Place of search <b>Munich</b>		Date of completion of the search <b>8 January 2014</b>	Examiner <b>Obel, Nicolai</b>
CATEGORY OF CITED DOCUMENTS X : particularly relevant if taken alone Y : particularly relevant if combined with another document of the same category A : technological background O : non-written disclosure P : intermediate document		T : theory or principle underlying the invention E : earlier patent document, but published on, or after the filing date D : document cited in the application L : document cited for other reasons ..... & : member of the same patent family, corresponding document	

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**CLAIMS INCURRING FEES**

The present European patent application comprised at the time of filing claims for which payment was due.

Only part of the claims have been paid within the prescribed time limit. The present European search report has been drawn up for those claims for which no payment was due and for those claims for which claims fees have been paid, namely claim(s):

No claims fees have been paid within the prescribed time limit. The present European search report has been drawn up for those claims for which no payment was due.

**LACK OF UNITY OF INVENTION**

The Search Division considers that the present European patent application does not comply with the requirements of unity of invention and relates to several inventions or groups of inventions, namely:

see sheet B

All further search fees have been paid within the fixed time limit. The present European search report has been drawn up for all claims.

As all searchable claims could be searched without effort justifying an additional fee, the Search Division did not invite payment of any additional fee.

Only part of the further search fees have been paid within the fixed time limit. The present European search report has been drawn up for those parts of the European patent application which relate to the inventions in respect of which search fees have been paid, namely claims:

None of the further search fees have been paid within the fixed time limit. The present European search report has been drawn up for those parts of the European patent application which relate to the invention first mentioned in the claims, namely claims:

see additional sheet(s)

The present supplementary European search report has been drawn up for those parts of the European patent application which relate to the invention first mentioned in the claims (Rule 164 (1) EPC).

**LACK OF UNITY OF INVENTION  
SHEET B**

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The Search Division considers that the present European patent application does not comply with the requirements of unity of invention and relates to several inventions or groups of inventions, namely:

1. claims: 1-9, 11-15(all partially)

Regards variants with a mutations at position 376.

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2. claims: 1-9, 11-15(all partially)

Regards variants with a mutation at position 577.

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3. claims: 1-9, 11-15(all partially)

Regards variants with a mutation at position 653.

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4. claim: 10(partially)

AHAS variants of SEQ ID NO 21.

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5. claim: 10(partially)

AHAS variants of SEQ ID NO 62.

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ON EUROPEAN PATENT APPLICATION NO.

EP 13 17 5389

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This annex lists the patent family members relating to the patent documents cited in the above-mentioned European search report. The members are as contained in the European Patent Office EDP file on The European Patent Office is in no way liable for these particulars which are merely given for the purpose of information.

08-01-2014

10

15

20

25

30

35

40

45

50

55

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
US 2003097692 A1	22-05-2003	NONE	
-----			
WO 2008071715 A1	19-06-2008	AR 064298 A1	25-03-2009
		AU 2007331535 A1	19-06-2008
		BR PI0720338 A2	24-12-2013
		CA 2672227 A1	19-06-2008
		CN 101657090 A	24-02-2010
		EA 200900768 A1	26-02-2010
		EP 2091319 A1	26-08-2009
		JP 2010512157 A	22-04-2010
		US 2012023601 A1	26-01-2012
		UY 30781 A1	03-07-2008
		WO 2008071715 A1	19-06-2008
-----			
WO 2008124495 A2	16-10-2008	AR 065938 A1	15-07-2009
		AU 2008237319 A1	16-10-2008
		CA 2682349 A1	16-10-2008
		CN 101668419 A	10-03-2010
		CO 6220904 A2	19-11-2010
		CR 11088 A	19-05-2010
		EA 200901337 A1	30-06-2010
		EA 201370037 A1	30-07-2013
		EC SP099709 A	30-04-2010
		EP 2134159 A2	23-12-2009
		EP 2561749 A1	27-02-2013
		EP 2561750 A1	27-02-2013
		EP 2570024 A1	20-03-2013
		EP 2574233 A1	03-04-2013
		JP 2010523123 A	15-07-2010
		KR 20100016161 A	12-02-2010
		MA 31334 B1	01-04-2010
		NZ 579899 A	31-08-2012
		NZ 598210 A	30-08-2013
		NZ 598211 A	30-08-2013
		NZ 600923 A	25-10-2013
		US 2010287641 A1	11-11-2010
		UY 30997 A1	31-10-2008
		WO 2008124495 A2	16-10-2008
-----			
WO 2006060634 A2	08-06-2006	AR 051690 A1	31-01-2007
		AR 078072 A2	12-10-2011
		AU 2005311754 A1	08-06-2006
		BR PI0518764 A2	09-12-2008
		CA 2588900 A1	08-06-2006
		CN 101443450 A	27-05-2009
		EP 1824981 A2	29-08-2007

EPO FORM P0459

For more details about this annex : see Official Journal of the European Patent Office, No. 12/82



ANNEX TO THE EUROPEAN SEARCH REPORT  
ON EUROPEAN PATENT APPLICATION NO.

EP 13 17 5389

5

This annex lists the patent family members relating to the patent documents cited in the above-mentioned European search report. The members are as contained in the European Patent Office EDP file on  
The European Patent Office is in no way liable for these particulars which are merely given for the purpose of information.

08-01-2014

10

15

20

25

30

35

40

45

50

55

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
		JP 2008521443 A	26-06-2008
		US 2009029860 A1	29-01-2009
		WO 2006060634 A2	08-06-2006
-----			
WO 2007005581	A2 11-01-2007	AR 058429 A1	06-02-2008
		AT 541939 T	15-02-2012
		AU 2006265984 A1	11-01-2007
		BR PI0612384 A2	27-01-2009
		CA 2613087 A1	11-01-2007
		CN 101287836 A	15-10-2008
		CN 103484531 A	01-01-2014
		EA 200800174 A1	29-08-2008
		EA 201301103 A1	28-02-2014
		EP 1902136 A2	26-03-2008
		EP 2400022 A1	28-12-2011
		EP 2532750 A1	12-12-2012
		ES 2380867 T3	21-05-2012
		HR P20120294 T1	31-05-2012
		JP 5265356 B2	14-08-2013
		JP 2009504137 A	05-02-2009
		JP 2013121349 A	20-06-2013
		PT 1902136 E	13-04-2012
		SI 1902136 T1	31-05-2012
		TR 200709185 T2	21-02-2008
		TR 200907590 T2	21-06-2010
		US 2011209232 A1	25-08-2011
		WO 2007005581 A2	11-01-2007
		ZA 200800947 A	30-06-2010
-----			
WO 2005020673	A1 10-03-2005	AR 047107 A1	11-01-2006
		BR PI0413917 A	24-10-2006
		CO 5700668 A2	30-11-2006
		EP 1659855 A1	31-05-2006
		EP 2294913 A1	16-03-2011
		ES 2379553 T3	27-04-2012
		US 2007028318 A1	01-02-2007
		US 2012172224 A1	05-07-2012
		UY 28495 A1	31-03-2005
		WO 2005020673 A1	10-03-2005
-----			
WO 0185970	A2 15-11-2001	AU 6135801 A	20-11-2001
		AU 2001261358 B2	13-07-2006
		CA 2445398 A1	15-11-2001
		EP 1280928 A2	05-02-2003
		US 2003217381 A1	20-11-2003
		US 2005198705 A1	08-09-2005

EPO FORM P0459

For more details about this annex : see Official Journal of the European Patent Office, No. 12/82

ANNEX TO THE EUROPEAN SEARCH REPORT  
ON EUROPEAN PATENT APPLICATION NO.

EP 13 17 5389

5

This annex lists the patent family members relating to the patent documents cited in the above-mentioned European search report. The members are as contained in the European Patent Office EDP file on  
The European Patent Office is in no way liable for these particulars which are merely given for the purpose of information.

08-01-2014

10

15

20

25

30

35

40

45

50

55

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
		US 2009025108 A1	22-01-2009
		WO 0185970 A2	15-11-2001
-----			
WO 2006024351 A1	09-03-2006	AR 050088 A1	27-09-2006
		AU 2005279457 A1	09-03-2006
		BR PI0513981 A	20-05-2008
		CA 2576813 A1	09-03-2006
		CN 101031646 A	05-09-2007
		EP 1776457 A1	25-04-2007
		TR 200700491 T2	24-04-2007
		TR 200900517 T2	23-03-2009
		US 2007300321 A1	27-12-2007
		US 2011138503 A1	09-06-2011
		UY 29044 A1	24-02-2006
		WO 2006024351 A1	09-03-2006
-----			
EP 0492113 A2	01-07-1992	AU 9005591 A	02-07-1992
		BR 9105628 A	15-09-1992
		CA 2058349 A1	28-06-1992
		CS 9103958 A3	14-10-1992
		EP 0492113 A2	01-07-1992
		FI 916117 A	28-06-1992
		IE 914542 A1	01-07-1992
		JP H04311392 A	04-11-1992
		NO 915094 A	29-06-1992
		PL 292942 A1	11-01-1993
		PT 99896 A	31-12-1992
		YU 200091 A	10-06-1994
		ZA 9110136 A	28-10-1992
-----			
WO 2004062351 A2	29-07-2004	US 2006130172 A1	15-06-2006
		WO 2004062351 A2	29-07-2004
-----			
US 6271360 B1	07-08-2001	AT 478687 T	15-09-2010
		AU 773242 B2	20-05-2004
		AU 7076700 A	26-03-2001
		AU 2004205139 A1	16-09-2004
		BR 0013590 A	07-05-2002
		CA 2382120 A1	08-03-2001
		CN 1384759 A	11-12-2002
		DK 1210123 T3	20-12-2010
		EP 1210123 A1	05-06-2002
		EP 2266627 A1	29-12-2010
		EP 2324856 A1	25-05-2011
		ES 2358292 T3	09-05-2011
		JP 2003508451 A	04-03-2003

EPO FORM P0459

For more details about this annex : see Official Journal of the European Patent Office, No. 12/82

**ANNEX TO THE EUROPEAN SEARCH REPORT  
ON EUROPEAN PATENT APPLICATION NO.**

EP 13 17 5389

5

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The European Patent Office is in no way liable for these particulars which are merely given for the purpose of information.

08-01-2014

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15

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30

35

40

45

50

55

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
		JP 2012184246 A	27-09-2012
		NZ 517942 A	30-07-2004
		PT 1210123 E	30-11-2010
		US 6271360 B1	07-08-2001
		US 6479292 B1	12-11-2002
		US 2004023392 A1	05-02-2004
		WO 0115740 A1	08-03-2001
		ZA 200201228 A	28-05-2003
-----			
EP 0257993	A2	02-03-1988	
		AT 145648 T	15-12-1996
		AU 619167 B2	23-01-1992
		AU 7638387 A	03-03-1988
		CA 1314506 C	16-03-1993
		CN 87106531 A	06-07-1988
		DE 3751963 D1	09-01-1997
		DE 3751963 T2	28-05-1997
		DK 443187 A	27-02-1988
		EP 0257993 A2	02-03-1988
		EP 0730030 A1	04-09-1996
		ES 2095202 T3	16-02-1997
		IL 83348 A	08-12-1995
		JP 2759135 B2	28-05-1998
		JP S6371184 A	31-03-1988
		NZ 221233 A	28-05-1990
		RU 2099422 C1	20-12-1997
		ZA 8706314 A	26-04-1989
-----			
US 2007118920	A1	24-05-2007	
		AR 056793 A1	24-10-2007
		AU 2006313700 A1	18-05-2007
		BR PI0618382 A2	30-08-2011
		CA 2629075 A1	18-05-2007
		EP 1948801 A2	30-07-2008
		US 2007118920 A1	24-05-2007
		UY 29910 A1	29-06-2007
		WO 2007054555 A2	18-05-2007
-----			

EPC FORM P 0459

For more details about this annex : see Official Journal of the European Patent Office, No. 12/82