**Registration of 7S β -conglycinin α' and 11S glycinin A3 null
food grade soybean Germplasm, HS-161**

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Registration of 7S β -conglycinin α' and 11S glycinin A₃ null food grade soybean Germplasm, HS-161

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Abstract:

HS-161 is a 7S β -conglycinin α' and 11S glycinin A₃ null food grade soybean line (*Glycine max*. L.) with high protein concentration, yellow hilum, and fair food processing quality. It is adapted to areas of southwest Ontario with 3100 or more crop heat units and has a relative maturity of 2.4 (MG 2.4).

Key words: *Glycine max* (L.) Merr., Soybean, 7S β -conglycinin, 11S glycinin

The 7S β -conglycinin α' and 11S glycinin A₃ null food grade soybean germplasm, HS-161 (Reg No: CN 119052), was developed by Agriculture and Agri-Food Canada (AAFC), Harrow Research and Development Centre (HRDC) at Harrow, Ontario for use in soy food production and released in 2016. It lacks the 7S β -conglycinin α' and the 11S glycinin A₃ subunits, which affect soy food quality, such as tofu texture (Poysa et al. 2005). HS-161 is adapted to areas of southwest Ontario with 3100 or more crop heat units and has a relative maturity (MG) of 2.4 and a protein concentration of 46.2%.

HS-161 has a pedigree of Harovinton*2/IWATE-3//Harovinton*2/IWATE-3/3/X937-1-3-1-3/CL97032 with the last cross that was made in 2009. IWATE-3, kindly provided by Dr. N. Kaizuma, Iwate University, Morioka, Iwate, Japan, lacks the 7S α' subunit of β -conglycinin and all the 11S subunits of glycinin (Zarkadas et al. 2007). Harovinton (Buzzell et al. 1991) and X937-1-3-1-3 (Poysa et al. unpublished) are food grade soybean cultivars developed at AAFC-HRDC, Harrow, Ontario. CL97032 (S25-D3) is a large seeded high protein food grade soybean cultivar developed by Syngenta (D. McClure, Syngenta Canada, personal communication).

The F₁ seeds were sown in the greenhouse at Harrow, Ontario in October 2009. The F₂ seeds were grown in the field at Harrow with single plants selected based on maturity, lodging resistance, seed size, and pot numbers. The selected F₂ plants were threshed individually and the seeds were sent to a winter nursery in Puerto Rico for two generations of advancement by modified single seed descent in 2010. The F₅ seeds were grown in a segregation nursery and selected for maturity, lodging resistance, plant vigor and yield potential at Harrow in 2011. The selected plants were threshed individually and additional visual selections were made in the lab for seed size, shape, and hilum color; seed coat integrity; and uniformity of seed size and shape. The seeds from 330 selected plants were grown in an F₆ protein profile (F₆PP12) nursery at Harrow in 2012 for another round of single plant selection based on total seed protein concentration, plant vigor, lodging and maturity. A subset of 126 lines with acceptable ratings for the criteria described above were analyzed for glycinin and β -conglycinin protein profile by one dimensional polyacrylamide gel electrophoresis (Poysa et al. 2005). The protein profile selection resulted in 15 lines with unique protein profiles. These F₇ seeds were grown in a single plant row yield trial (F₇ CHMIS13) in 2013 using a modified augmented type-2 design (Lin and Voldeng, 1989) at Harrow. The line, SQ09-0040E-M-1, was selected for a protein profile (PP), which lacked the 7S β -conglycinin α' and the 11S glycinin A₃ subunits (Figure 1). The F₈ seeds of SQ09-0040E-M-1 were grown in an observation nursery for seed increase at Harrow in 2014. The F₉ seeds of the SQ09-0040E-M-1 line were evaluated for agronomic performance and seed quality in five rows, three replication yield trials (BYM15) at six locations (Harrow, Chatham, Holiday Beach, Merlin, Ridgetown, and Woodslee, ON) in 2015. The F₁₀ breeder yield (BYM15) seeds were confirmed for its PP purity (Figure 1) and deposited with the Plant Gene Resources of Canada in May 2016 and given Reg No. CN 119052.

In the BYM15 yield trial, with OAC Kent as the check cultivar, HS-161 yielded about 3906 kg ha⁻¹, which was about 11 % lower than the check. HS-161 matured in 127 days, 3 days later than the check. HS-161 had a plant height of 79 cm with a lodging score of 1.6 (with 1.0 being erect and 5.0 lodged flat), and 22.5 g 100 seeds⁻¹, which was 0.3 g 100 seeds⁻¹ lighter than the check. The seed protein concentration of HS-161 on a dry matter basis measured with NIR (near infrared spectroscopy) was 46.2%, which was 4% higher than the check. HS-161 accumulated normal amounts of total protein despite lacking two major protein subunits, consistent with previous reports (Poysa et al. 2005). HS-161 has an indeterminate growth habit, purple flowers, and grey pubescence. The seeds are dull yellow with yellow hilum.

In tests conducted at Harrow, HS-161 had fair soymilk and tofu processing qualities (Table 2), being inferior to Harovinton, the standard soybean cultivar used for soy food quality evaluation in Canada. The A₃ subunit has significant effects on tofu firmness (Poysa et al. 2005). The softer tofu produced from HS-161 is in agreement with the previous report. However, the softer tofu may be a favorable trait for silken tofu production.

Requests for seed should be made to Plant Gene Resources of Canada, 107 Science Place, Saskatoon, SK, Canada S7N 0X2 indicating Accession No: CN 119052.

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Cultivar			Seed					Plant	
	Yield	Maturity	Weight	Protein ^a	Oil ^a	Sugar ^a	Carb ^{ac}	Height	Lodging
	(t/ha)	(d)	(g/100 sd)	%	%	%	%	(cm)	(1-5) ^b
HS-161	3.9 ^a	126.9 ^a	22.5 ^a	46.2 ^a	20.1 ^a	10.9 ^a	15.9 ^a	79.1 ^a	1.6 ^a
OAC Kent	4.4 ^b	124.3 ^a	22.8 ^a	42.2 ^b	22.3 ^b	11.6 ^b	16.7 ^b	74.6 ^a	1.7 ^a
SE ^d	0.3	2.7	1.6	1.1	0.5	0.2	0.4	4.7	0.2
No. Sites	6	6	6	6	6	6	6	6	4

Table 1. Summary of agronomic performance and seed quality of HS-161 and the check cultivar OAC Kent, in Southwest Ontario, in 2015. The trial was conducted by AAFC at Chatham, Harrow, Holiday Beach, Merlin, Ridgetown and Woodslee, Ontario, Canada.

^aWhole seed, dry matter basis, by near infrared spectroscopy

^b1 = no lodging, 5 = complete lodging.

^cCarb is total hydrolysable carbohydrates

^dStandard errors between corresponding means of the trials at $P \leq 0.05$

a, *b* in columns, means followed by different letters differ significantly

Table 2. Soymilk and tofu yield and quality of HS-161 and Harovinton. Seed was sampled from the field test grown at Harrow, Ontario, Canada by AAFC in 2015

	HS-161	Harovinton	LSD _a
Water Uptake ^b	2.48	2.55	0.02
Soymilk yield (l kg ⁻¹ beans)	7.17	7.31	0.06
Solids concentration of soymilk (%)	9.63	9.59	0.23
Tofu (GDL) ^c yield (kg kg ⁻¹ beans)	6.86	7.03	0.07
Tofu (GDL) ^c compression hardness (N) ^f	0.56	1.05	0.10
Tofu (GDL) ^c compression firmness (N/mm) ^g	0.13	0.22	0.01
Tofu (GDL) ^c springiness ^h	0.47	0.69	0.10
Tofu (CaSO ₄) ^d yield (kg kg ⁻¹ beans)	6.81	7.02	0.08
Tofu (CaSO ₄) ^d compression hardness (N) ^f	0.43	0.68	0.08
Tofu (CaSO ₄) ^d compression firmness (N/mm) ^g	0.13	0.22	0.01
Tofu (CaSO ₄) ^d Springiness ^h	0.41	0.42	0.02
Tofu (MgCl ₂) ^e yield (kg kg ⁻¹ beans)	6.90	6.20	0.15
Tofu (MgCl ₂) ^e compression hardness (N) ^f	0.62	0.97	0.07
Tofu (MgCl ₂) ^e compression firmness (N/mm) ^g	0.15	0.18	0.03
Tofu (MgCl ₂) ^e Springiness ^h	0.46	0.64	0.10

^a LSD_{0.05} between HS-161 and the check, Harovinton.

^b (weight of hydrated beans) / (initial weight of beans), soybeans were soaked for 22 h at 13°C

^c Tofu made with Glucono-delta-lactone coagulant

^d Tofu made with Calcium sulphate dihydrate coagulant

^e Tofu made with Magnesium chloride coagulant

^f Hardness in Newtons

^g Firmness in Newtons per mm

^h The height that the sample springs back after the first compression divided by the full distance of the first compression, dimensionless

Figure Legend

Figure 1. 1D polyacrylamide gel illustrating the protein variant line and the two parent lines. From left to right: Protein molecular weight ladder; IWATE-3, the parent lacking β -conglycinin α' and all glycinin subunits; HS-161, selected as SQ009-0040E-M-1 and lack β -conglycinin α' and glycinin A₃ subunits; Harovinton, a parent with all the protein subunits present. Note: very low molecular weight proteins including the glycinin A₅ subunit are run to end of the gel in this protocol.

254x190mm (96 x 96 DPI)