Júpiter-INIA: A new oat variety with improved β-glucan and protein contents

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ABSTRACT

Júpiter-INIA is a new oat (*Avena sativa* L.) cultivar selected in preliminary field trials for higher β-glucan and protein content, resistance to disease and lodging, and similar yield than the other cultivated oat varieties. The objective of the present research was to compare agronomic, industrial performance and groat nutritional quality of 'Júpiter-INIA' with the widespread commercial 'Supernova-INIA' in southern Chile. Field trials were conducted at four different locations during four seasons in a randomized complete block design with three replicates. 'Júpiter-INIA' had about 20% more β-glucan (4.25 *vs*. 3.31 g 100 g^{-1}, $P < 0.0001$) and protein (16.14 *vs*. 12.78 g 100 g^{-1}, $P < 0.0001$) contents than 'Supernova-INIA'. The overall plant height, lodging, and disease reaction to crown rust were lower in 'Júpiter-INIA' than in 'Supernova-INIA'. Grain yield, 1000-grain weight, and hectoliter weight, were lower in 'Júpiter-INIA' than in 'Supernova-INIA'; however, its mean values were within the range for oat cultivars. Dehulled-grain yield and stained grains were similar to 'Supernova-INIA'. This results point out that 'Júpiter-INIA' groat had superior nutritional quality than 'Supernova-INIA', and thus it could be an important contribution to be included in the development of healthy oats-containing food products.

Key words: *Avena sativa*, β-glucan, lodging, protein.

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INTRODUCTION

Oat (*Avena sativa* L.) whole grain consumption exerts beneficial effects on human health, contributing to reduce rates of oxidative stress, chronic age-related diseases and various forms of cancer (Gangopadhyay et al., 2015); which is relevant since cardiovascular disease and cancer cause 48% and 20% of global mortality, respectively (McDonald and Pickart, 2011). Due to its health benefits, oats-based food products such as breads, biscuits, cookies, probiotic drinks, breakfast cereals, flakes and infant food, are increasingly gaining consideration (Rasane et al., 2015).

The health promoting effects of oat grains are due to their fiber, essential amino acids and fatty acids, as well as their antioxidants (Butt et al., 2008; Cavazos and Gonzalez de Mejia, 2013; Gangopadhyay et al., 2015). The consumption of at least 3 g oat soluble fiber, (1-3, 1-4) β-D-glucan (from now on, β-glucan), lowers serum and low-density lipoprotein (LDL) cholesterol by 0.30 and 0.25 mmol L^{-1}, respectively (US Food and Drug Administration; FDA, 1997; Whitehead et al., 2014); also of blood glucose (Shen et al., 2011). In addition, amino acid profile of oat proteins matches human requirements better than other cereals (Robbins et al., 1971; Bewley et al., 2013), containing bioactive peptides with "antihypertensive" effect (Cavazos and Gonzalez de Mejia, 2013; Gangopadhyay et al., 2015).

Oat groat contain between 1.44% and 6.5% β-glucan, mainly depending on the genotype, and in some cases on the environment and seeding date (Peterson et al., 2005; Fan et al., 2009; Güler, 2011; Newell et al., 2012). On the other hand, the protein content ranges between 9% and 24.5% (Youngs, 1972; Pomeranz et al., 1973; Mirmoghtadaie et al., 2009), depending on the genotype and environment, with an important effect of the N fertilization (Peterson et al., 2005; Fan et al., 2009; Güler, 2011). The genetic regulation of these traits in oat is poorly understood (Orr and Molnar, 2008). Thus, seven quantitative trait *loci* (QTLs) regions has been found explaining up to 20% of β-glucan content in oat groat (Kianian et al., 2000); whereas, 17 QTLs have been found explaining up to 42% of the protein phenotypic variation, in a particular population (Zhu et al., 2004). Several PCR-based sequence-characterized amplified region (SCAR) markers linked to β-glucan and protein QTLs have been developed, however, due to mapping populations are rather wide crosses these QTLs may not represent the *loci* and alleles most important in modern elite germplasms (Orr and Molnar, 2008).

In recent years, oat production in Chile has been increasingly derived from high yielding cultivars that mainly target high

industrial quality for milling (Beratto et al., 1994; Beratto, 2006), and which are cultivated using high input of agrochemicals. Today, concepts such as nutritional quality and environmentally friendly agriculture, have gained importance. Chilean oat varieties, are in the lower end of the protein range, containing up to 11.5% (Beratto, 2006), whereas β-glucan content has not been reported so far. Thus, varieties exceeding 12% protein would be a contribution to increase plant protein intake by the human population, while varieties containing minimum 4% β-glucan are required to reach the established β-glucan recommended dose (FDA, 1997). Consequently, the Instituto de Investigaciones Agropecuarias (INIA), Chile, released 'Júpiter-INIA' (pedigree: T99/1021-8t-3t) due to its higher groat nutritional quality, lodging and disease resistance. Thus, the aim of the present research was to describe 'Júpiter-INIA' and compare its agronomic performance, industrial and groat nutritional quality with regard to the widespread commercial 'Supernova-INIA', in different locations and growing seasons of southern Chile. In addition, the characterization of β-glucan QTL regions and their utility in MAS breeding, also of the relevance of 'Júpiter-INIA' for food, are discussed.

MATERIALS AND METHODS

Experimental design and data analysis

'Júpiter-INIA' and 'Supernova-INIA' were evaluated under a randomized complete blocks design with three replicates per treatment. Each replicate was represented by one plot of 3 m long by 1 m wide. Field experiment was replicated in four locations of Southern Chile (Table 1), during four growing seasons from 2011-2012 to 2014-2015. Each combination of location and sowing date was defined as an "environment".

A mixed model was applied using the genotypes as a fixed factor, and environment and year as random factors. The model was fitted using restricted maximum likelihood. Non-significant interactions (restricted maximum likelihood [REML] variance ratio < 1) were eliminated and the reduced model re-fitted. Data were analyzed using JMP 11.0.0 (SAS Institute, Cary, North Carolina, USA).

Plant material and planting

Seeds of 'Júpiter-INIA' and 'Supernova-INIA' were provided by the Oat Breeding Program of INIA. Plots were hand-planted using 120 kg seed ha^{-1} and 0.2 m between rows at different dates depending on the location (Table 2). Nutrients (Vitra S.A., Rancagua, Chile) doses chosen for each environment depended on the soil analysis, fluctuating between 60 to 90 kg P_2O_5 ha^{-1} applied as monocalcium phosphate at seeding; 120 to 150 kg N ha^{-1} using magnesium calcium ammonium nitrate 27% applied 20% at seeding, 40% at early tillering (Zadoks Stage Z-21), and 40% during full tillering (Z-27) (Zadoks et al., 1974). In addition, 50 kg K_2O ha^{-1}, 40 kg MgO ha^{-1} and 50 kg S ha^{-1}, were applied using potassium/magnesium sulfate mixed with monocalcium phosphate. Weeds were controlled using a post-emergence herbicide mixture of 10 g ha^{-1} metsulfuron methyl (2-[4-methoxy-6-methyl-1,3,5-triazin-2-ylcarbamoylsulfamoyl]benzoic acid; Anasac Chile S.A., Santiago, Chile), 0.5 L ha^{-1} MCPA-dimethylammonium (A.H. Marks and Company Ltd., West Yorkshire, UK) and 170 g ha^{-1} dicamba-sodium (Syngenta Crop Protection AG, Basle, Switzerland), diluted in 200 L ha^{-1} water. Crops were grown without application of fungicides, insecticides, or growth regulators and were harvested with a combine harvester machine.

Evaluation of agronomic traits

Grain yield was determined from a cleaned seeds sample from each plot, after correction to 12% grain moisture. Days to heading were recorded when the first panicle spikelet had emerged in 50% of the plants. Plant height (cm) was measured at maturity adding stem and panicle length (cm). Plant lodging was recorded immediately before harvest as a visual estimation of the percentage of plot area presenting lodging, as well as by a 1 to 5 scale, where 1 is absence and 5 is complete inclination of the plants.

Table 2. Field trial sowing and heading dates at southern Chile.

Location	Sowing date	Heading date	
		Júpiter-INIA	Supernova-INIA
Chillán	2 Sept 2011	30 Nov 2011	25 Nov 2011
	21 Aug 2012	28 Nov 2012	22 Nov 2012
Traiguén	24 May 2011	10 Nov 2011	6 Nov 2011
	24 May 2012	16 Nov 2012	14 Nov 2012
	20 June 2014	10 Nov 2014	6 Nov 2014
Vilcún (1st date)	23 June 2011	20 Nov 2011	21 Nov 2011
	26 June 2012	22 Nov 2012	20 Nov 2012
	25 June 2013	20 Nov 2013	19 Nov 2013
	4 July 2014	24 Nov 2014	21 Nov 2014
Vilcún (2nd date)	20 Aug 2011	24 Nov 2011	26 Nov 2011
	7 Aug 2012	25 Nov 2012	2 Dec 2012
	26 July 2013	27 Nov 2013	24 Nov 2013
	14 Aug 2014	1 Dec 2014	29 Nov 2014
Purranque	6 Oct 2011	24 Dec 2011	15 Dec 2011
	25 Aug 2013	12 Dec 2013	2 Dec 2013
	11 Sept 2014	14 Dec 2014	28 Nov 2014

Table 1. Field trial locations at southern Chile.

Region	Homogeneous area	Commune	Province	GPS coordinates
Biobío	Intermedium depression	Chillán	Ñuble	36°31' S, 71°54' W
La Araucanía	Interior unirrigated soil	Traiguén	Malleco	38°10' S, 70°00' W
La Araucanía	Unirrigated valley	Vilcún	Cautín	38°41' S, 72°42' W
Los Ríos	Unirrigated valley	Purranque	Osorno	40°52' S, 73°12' W

Protein and β-glucan contents

Samples of 50 g dehulled grain were ground in a laboratory mill with a 0.5 mm sieve. The moisture content of each sample was estimated in 20 g flour using an electronic moisture meter (MLB 50-3N, Kern & Sohn GmbH, Balingen, Germany). Determinations of β-glucan and protein contents were conducted in 250 and 100 mg flour subsamples, respectively. The content of β-glucan was determined by a Mixed Linkage beta-glucan Assay kit (Megazyme, Wicklow, Ireland), which correspond to the AOAC Method 995.16. Crude protein content was determined with a TruSpec Micro (LECO Corporation, St. Joseph, Michigan, USA), which correspond to the AOAC Method 992.15. A factor of 5.83 was used to convert total N to crude protein (FAO, 2003). Crude protein and β-glucan contents were expressed on a DM basis.

Grain industrial and physical quality traits

These quality traits were estimated on a sample taken from each experimental plot at 100% purity and 12% moisture. Weight of 1000-grains was determined by hand according to International Seed Testing Association (International Seed Testing Association; ISTA, 2016). Test weight was determined using a 0.25-L hectoliter weight balance (Nr 8884, Dalle Molle, Caxias do Sul, Brazil), using the Bauart 1938 conversion table. Dehulled-grain yield was determined in a 100 g grain sample using an impact dehuller machine, and weighing the resulting dehulled grains. Then, a manual counting of stained grains was conducted in representative samples of 50 g dehulled-grains, considering those exhibiting over 5% of stained epidermis.

Reaction to diseases

Diseases scores were recorded when the first node of the stem became visible, at the end of flowering, and during dough grain, using different scales notes. Powdery mildew (*Blumeria graminis* f. sp. *avenae*) was recorded using the double-digit X/Y modified scale of Saari and Prescott (1975), while crown rust (*Puccinia coronata* var. *avenae* f. sp. *avenae*) and stem rust (*Puccinia graminis* f. sp. *avenae*) were evaluated with the binomial system of Cobbs as modified by Peterson et al. (1948). Scores of Halo blight (*Pseudomona syringae* pv. *coronafaciens*) were a visual estimation of the affected leaf area expressed as percentage of damage, whereas *Barley yellow dwarf virus* (BYDV) was recorded as the estimation of percentage of affected plants in the plot. The highest scores in each season per experimental replicate were used for the comparison between 'Júpiter-INIA' and 'Supernova-INIA'. The final note of disease reaction was considered the highest note observed across the four growing seasons, locations, and sowing dates.

Morphological and genetic characterization

The morphological characterization was conducted during 2014-2015 using 24 oat morphological descriptors and the methodology described by International Union for the Protection of New Varieties of Plants (UPOV, 1994). Grain color was determined by comparing with a Munsell Plant Tissues Color Charts (Munsell Color, Grand Rapids, Michigan, USA).

For genetic characterization, in order to detect genetic factors explaining the β-glucan composition of the oat grains, QTLs were characterized in 'Júpiter-INIA', according to Orr and Molnar (2008). 'Marion' and 'Kanota' (Orr and Molnar, 2008) were used as positive and negative controls. The dwarfing gene DW6 was also identify as described by Tanhuanpää et al. (2006). For this gene, the oat line OT207 and 'Curt' were used as positive and negative controls (Milach et al., 2002), respectively. Seeds of control lines were kindly provided by Dr. Stephen A. Harrison, Quaker International Oat Nursery curator.

RESULTS

'Júpiter-INIA' showed facultative grown habit with a somewhat later cycle from planting to heading than 'Supernova-INIA' (Tables 2 and 3), reaching lower grain yield, plant height, and lodging in comparison to 'Supernova-INIA' (Table 3). There was a significant interaction effect of "environment/year" over days to heading (variance ratio = 9.68), yield (variance ratio = 3.88), plant height (variance ratio = 1.8) and lodging (variance ratio = 4.4) (results not shown).

Values of industrial grain quality were in the typical range for food-oats. Thus, 'Júpiter-INIA' exhibited similar yield of dehulled grains, and slightly lower hectoliter weight and weight of 1000-grain, in comparison to 'Supernova-INIA' (Table 4). The interaction "environment by year" was significant over de-hulled grain yield (variance ratio =

Table 3. Overall effect of the genotype over agronomic traits during four growing seasons and locations.

Traits	Júpiter-INIA	Supernova-INIA	SE	*P*
Grain yield, t ha^{-1}	7.88	8.69	0.41	< 0.0001*
Plant height, cm	111.42	120.77	3.39	< 0.0001*
Lodging (Index 1-5)	1.20	1.93	0.28	< 0.0001*
Lodging area, %	8.73	19.12	6.18	0.0003*
Heading time, d	122.92	116.20	15.66	0.0002*

SE: Standard error.
*$P < 0.05$.

Table 4. Overall effect of genotype over grain quality traits during four seasons and locations.

Traits	Júpiter-INIA	Supernova-INIA	SE	*P*
Industrial and physical quality				
Dehulled-grain yield, g 100 g^{-1}	66.18	66.60	1.76	0.3399
1000-grain weight, g	39.81	44.22	2.57	< 0.0001*
Test weight, kg hL^{-1}	53.85	54.98	0.25	< 0.0001*
Stained grains, nr 50 g^{-1}	141.65	104.04	65.08	0.3845
Nutritive quality				
Protein, g 100 g^{-1} DW	15.83	12.78	0.88	< 0.0001
β-glucan, g 100 g^{-1} DW	4.25	3.31	0.14	< 0.0001*

SE: Standard error. *$P < 0.05$.
DW: Dry weight basis.

1.86), and hectoliter weigh (variance ratio = 2.20) (result not shown). Presence of stained grains caused by weather conditions were on average similar for both genotypes (Table 4); however, 'Júpiter-INIA' had more stained grains in 2012 (interaction genotype by year variance ratio = 4.38).

Dehulled grains of 'Júpiter-INIA' had around 20% higher protein and β-glucan contents than 'Supernova-INIA' (Table 4). The random effect of the environment (variance ratio = 1.79), resulted in different protein values in Purranque (16.60 ± 0.60 g 100 g^{-1} DW, $P < 0.05$), Cañete and Traiguén (15.58 ± 0.67 g 100 g^{-1} DW and 14.72 ± 0.67 g 100 g^{-1} DW, $P < 0.05$), and Vilcún (13.09 ± 0.55 g 100 g^{-1} DW, $P < 0.05$) (results not shown). On the other hand, the genotype effect was stable over β-glucan content, whereas there was not effects of environment, year, or their interactions on this trait (result not shown).

Disease resistance of 'Júpiter-INIA' was greater than 'Supernova-INIA' against crown rust (Table 5). There were no differences with regard to the rest of the diseases evaluated, although 'Júpiter-INIA' had lower final disease reactions scores for almost all diseases except oidium (Table 5).

Both 'Supernova-INIA' and 'Júpiter-INIA' exhibited a semi-erect growth habit and dark green foliage with equilateral panicles showing absent or very weak tendency to be awned (Table 6). Eight out of 24 morphological descriptors differed between 'Júpiter-INIA' and 'Supernova-INIA' stands (Table 6). Some of those characteristics in 'Júpiter-INIA' were its strong frequency of flag leaf recurved, semi-erect attitude of spikelets in panicles, and shorter length of panicles compared with 'Supernova-INIA' (Table 6). Grains of 'Júpiter-INIA' were light yellow with a slight difference between hulled and dehulled grains, determining values of 8/6 - 2.5 Y (darker) and 8/5 - 2.5 Y (lighter) of the color chart, respectively (results not shown). The stem mass of 'Júpiter-INIA' was greater, exhibiting wider main stems in comparison with 'Supernova-INIA' (result not shown).

'Júpiter-INIA' shares the same β-glucan content QTLs of 'Marion' and 'Ogle', while 'Supernova-INIA' lacked these QTLs (Table 7). In addition, 'Júpiter-INIA' has the DW6 gene, while 'Supernova-INIA' lacked this gene (Figure 1, Table 7).

Table 5. Overall effect of the genotype over disease reactions and final reaction score during four growing seasons and locations.

Diseases	Júpiter-INIA	Supernova-INIA	SE	*P*
Crown rust, %	0.51 (30 MR)	5.18 (80 S)	2.31	0.0008*
Stem rust, %	0.00 (0 R)	0.37 (20 S)	0.27	0.3189
Bacteriosis, %	3.91 (3 MS)	6.77 (4 S)	4.21	0.2085
Powdery mildew, %	9.79 (8/60 MS)	14.02 (8/60 S)	8.51	0.0846
BYDV, %	2.52 (30 MS)	1.52 (30 MS)	2.11	0.3006

SE: Standard error. *$P < 0.05$.
BYDV: *Barley yellow dwarf virus*.
Means are indicated at the first line whereas the final disease-reaction score in parenthesis on second line.
R: Reaction of resistance, MR: moderately resistant, MS: moderately susceptible, and S: sensible.

DISCUSSION

The oats breeding program of INIA Chile created 'Júpiter-INIA' during the three last decades, integrating diverse germplasms (Figure 2), being the first released cultivar with food quality. Despite that 'Supernova-INIA' is the most yielding cultivar in Chile, occupying near to 80% of cropping area, it is not reaching a good performance in term of nutritional groat quality oriented to healthy food. For these reasons, 'Júpiter-INIA' would allow diversifying the oat crop at southern Chile not only by its nutritional attributes, but also by its better resistance to lodging and diseases. Crop diversification would contribute with a greater ability to suppress pest outbreaks, as well as by buffering crop production from the effects of greater climate variability and extreme events (Lin, 2011).

The creation of 'Júpiter-INIA' (pure line AVE 11.04) started in 1990 with a bi-parental cross between pure lines AVE 15.96 (Pedigree: Nehuén-INIA/Zeta) and AVE 8.95 (Pedigree: T90 1009 -3t-1t-2t-3t), with subsequent mass and pedigree selection until F_6 at southern Chile environment (Figure 2). Part of the original germplasm were old Chilean cultivars such as 'Pony-Baer' and 'Nehuén-INIA', along with Canadian (OT 222 × 212) and Brazilian (C7512/SR cpx) lines, both within the collaborative effort of the Quaker International Oat Nursery. The pure line AVE 11.04 was obtained during 2004; however, its groat nutritional value was discovered during 2010, when the genetic breeding program made efforts in the characterization of its germplasm in terms of groat β-glucan and protein.

'Júpiter-INIA' resulted having near 10% lower overall yield than 'Supernova-INIA' (Table 3), and slightly lower industrial quality (Table 4); it was, however, within ranges satisfying the requirements of the industry and producers. The best performance of 'Júpiter INIA' was reached in early-sowed trials during years without water restriction (results not shown). For this reason, 'Júpiter-INIA' is recommended for earlier sowing dates in southern Chile. It is important to point out that due to its longer cycle (Table 3); it is highly probably that yield was negatively affected in this research. Probably there might be no difference in yield compared to 'Supernova-INIA', when the planting date will be adjusted considering its flowering time (Table 2).

The most important feature of 'Júpiter-INIA' is its groat nutritional quality, having about 20% more β-glucan and protein than 'Supernova-INIA' (Table 4). The stable effect of the genotype across all environments and years, suggests a strong genetic effect on protein and β-glucan contents. Similar results have been obtained in other studies for β-glucan (Kianian et al., 2000; Cervantes-Martinez et al., 2001; Redaelli et al., 2013), and protein (De Koeyer et al., 2004) contents. The effect of environment over β-glucan

Table 6. UPOV morphological descriptors of 'Júpiter-INIA' and 'Supernova-INIA'.

Stage and characteristics	Scale	Note	
		Júpiter-INIA	Supernova-INIA
Stage 25-29: Main shot and 5 to 9 tillers			
1 Plant: growth habit	Erect (1), semi-erect (3), intermediate (5), semi-postrate (7), postrate (9)	3	3
2 Lowest leaves: hairiness of sheaths	Absent or very weak (1), weak (3), medium (5), strong (7), very strong (9)	1	7
Stage 40-45: botting			
3 Leaf blade: hairiness of margins of leaf below flag leaf	Absent or very weak (1), weak (3), medium (5), strong (7), very strong (9)	1	1
Stage 47-51: Flag leaf sheath opening to first spikelet just visible			
4 Plant: frequency of plants with recurved flag leaves	Absent or very weak (1), weak (3), medium (5), strong (7), very strong (9)	9	1
Stage 50-52: First spikelet just visible to 1/4 of inflorescence emerged			
5 Time of panicle emergence (first spike let visible on 50% of panicles)	Very early (1), early (3), medium (5), late (7), very late (9)	5	5
Stage 60-65: Beginning of anthesis to anthesis half-way			
6 Stem: hairiness of uppermost node	Absent (1), present (9)	1	1
7 Stem: intensity of hairiness of uppermost node	Very weak (1), weak (3), medium (5), strong (7), very strong (9)	5	3
Stage 70-75: Milk development to medium milk			
8 Panicle: orientation of branches	Unilateral (1), sub-unilateral (2), equilateral (3)	3	3
9 Panicle: attitude of branches	Erect (1), semi-erect (3), horizontal (5), drooping (7), strongly droopling (9)	3	7
10 Panicle: attitude of spikelets	Erect (1), pendulous (2)	1	2
Stage 65-69: Anthesis half-way to complete			
11 Glumes: glaucosity	Absent or very weak (1), weak (3), medium (5), strong (7), very strong (9)	1	1
Stage 65-69: Milk development to medium milk			
12 Glumes: length	Short (3), medium (5), long (7)	3	5
13 Primary grain: glaucosity of lemma	Absent (1), present (9)	1	1
14 Primary grain: intensity of glaucosity of lemma	Very weak (1), weak (3), medium (5), strong (7), very strong (9)	1	1
Stage 80-85: Dough development to soft dough			
15 Plant: length (stem and panicle)	Very short (1), short (3), medium (5), long (7), very long (9)	5	5
16 Panicle: length	Very short (1), short (3), medium (5), long (7), very long (9)	3	5
Stage 92: Ripening (Caryopsis hard)			
17 Grain: husk	Absent (1), present (9)	9	9
18 Primary grain: tendency to be awned	Absent or very weak (1), weak (3), medium (5), strong (7), very strong 9)	1	1
19 Primary grain: length of lemma	Very short (1), short (3), medium (5), long (7), very long (9)	5	5
20 Grain: color of lemma	White (1), yellow (2), brown (3), grey (4), black (5)	2	2
21 Primary grain: hairiness of back of lemma	Absent (1), present (9)	1	1
22 Primary grain: hairiness of base	Absent or very weak (1), weak (3), medium (5), strong (7), very strong (9)	1	1
23 Primary grain: length of basal hairs	Short (3), medium (5), long (7)	3	3
24 Primary grain: length of rachilla	Short (3), medium (5), long (7)	3	5

UPOV: International Union for the Protection of New Varieties of Plants.

content is unclear. Lower water availability positively affected β-glucan in Peterson (1991); however, the opposite occurred in Doehlert et al. (2001). In some studies, N fertilization incremented the β-glucan and protein contents (Fan et al., 2009; Güler, 2011); whereas in others only there was a positive effect in protein content (Saastamoinen et al., 2004; Weightman et al., 2004). In this work, only there was effect of the environment in the protein content (result not shown).

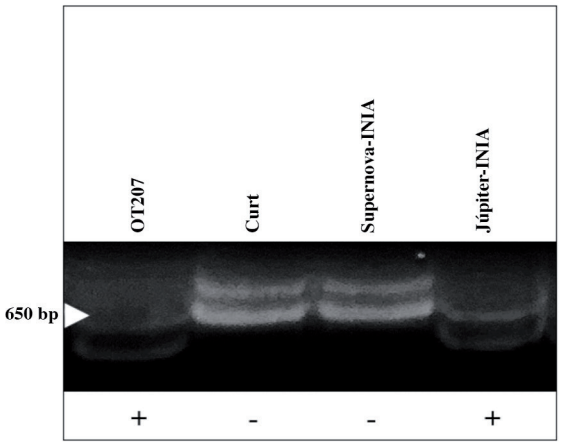
The effect of environment and its interaction with the genotype were important sources of variability in β-glucan content in Redaelli et al. (2013); contrary to the results obtained in this work. This might be due to the locations in Redaelli et al. (2013), were five countries with very

Table 7. Genetic characterization of β-glucan content quality trait *loci* (QTLs) and a dwarfing gene.

Marker	Gene/QTL	Trait	Position[1] (cM)	Fragment size (bp)	Júpiter-INIA	Supernova-INIA
SCARs						
Ubc 254s	KO3+38 QTL	β-glucan	2	350	+	-
Ubc109as	KO14 QTL	β-glucan	11	1500	+	-
SNPs						
REMAP	DW6 gene	Dwarfism	5	650*	+	-

[1]Position of the marker with regard the gene or QTL.
*650 bp allele containing the three-nucleotide mutation feature of DW6 gene (Figure 1).
+: Presence; - : absence of the respective allele.

Figure 1. Banding pattern of DW6 gene in 'Júpiter-INIA' and 'Supernova-INIA'.



Native sequencing polyacrilamide gel separating the 650 bp SNP-REMAP fragments based on a three nucleotide sequence mutation feature of DW6 gene. OT207: DW6 positive control, Curt: DW6 negative control.
+: Presence, -: absence of DW6 allele.

contrasting environments, and cultivars were created in some of these countries. In contrast, 'Supernova-INIA' was not selected by β-glucan but introduced from New Zealand, whereas 'Júpiter-INIA' was subjected to selection by β-glucan at the end of the breeding period, considering data from different locations.

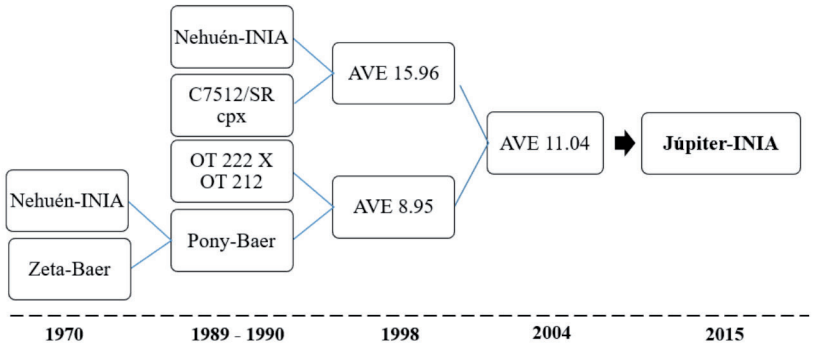
Within the genetic factors regulating β-glucan, seven QTLs were found in a 'Kanota' 'Ogle' population (KO), explaining 29.4% of the phenotypic variance; QTLs located on 3 and 11 linkage groups, explained 12.5% and 5.3% of the variation in β-glucan, respectively. In addition, four QTLs were found in a 'Kanota' × 'Marion' (KM) population, explaining 21.8% of the phenotypic variance in β-glucan; QTLs located on 11 and 14 linkage groups, explained 5.5% and 7.6% of variation in β-glucan (Kianian et al., 2000). On the other hand, seventeen QTLs have been found explaining 29% to 42% of the protein phenotypic variance in oat (De Koeyer et al., 2004).

Several SCARs PCR-based markers has been developed for detecting β-glucan QTLs, some of them are linked to QTL regions for protein or other traits (Orr and Molnar, 2008). Thus, the better β-glucan content of 'Júpiter-INIA' might be explained in part because of it contains KO3 and KO14 QTLs regions (Table 7, Kianian et al., 2000; Orr and Molnar, 2008). The PCR markers used in this work, are based on QTLs detected in rather broad genetic populations, KO and KM being winter × spring crosses, and TM naked × hulled crosses (Orr and Molnar, 2008). These factors could explain why 'Júpiter-INIA' together with other lines and cultivars, failed to amplify some of these markers (results not shown). The validation of KO3 and KO14QTLs in populations at southern Chile environment, will allow determining if they have a significant effect over β-glucan content. The high cost of measuring groat chemical compositions in breeding lines makes it attractive to consider strategies based on marker assisted selection (MAS) (Orr and Molnar, 2008).

Associations among heading date and β-glucan QTLs have been found on different oat genomic regions (Kianian et al., 2000; De Koeyer, 2004). The common location of these QTLs could indicate a pleiotropic effect caused when later maturing lines fail to complete the optimum grain-filling period, which results in shrunken kernels with less endosperm and enhanced relative β-glucan content (De Koeyer et al., 2004). This associations observed in different oat populations are coincident with the phenotype of 'Júpiter-INIA', which exhibit longer period to heading, higher groat β-glucan and protein, in combination with a lower 1000-grain weight and test weight (Tables 3 and 4).

The lower plant height was a positive feature of 'Júpiter-INIA', which is explained in part by the presence of the *Dw6* dwarfing gene (Figure 1, Table 7), and thus it has good association with low lodging (Table 3) (Milach et al., 2002; Tanhuanpää et al., 2006). The *Dw6* produces a shortening in the length of the three uppermost internodes, particularly the peduncle, resulting in panicles that failed to emerge fully from the leaf sheath (Milach et al., 2002). Panicles of 'Júpiter-INIA', however, fully emerged from the leaf sheath (results not shown). This observation suggests the presence of panicle exertion genes in 'Júpiter-INIA', which partially

Figure 2. Time line of 'Júpiter-INIA' creation.



The motherly and paternal lines, AVE 15.96 and AVE 8.95, were created in 1989 and 1990. Parental lines were subsequently crossed in 1998 originating the F_1 progeny. Cycles of mass selection followed from F_2 to F_4, and pedigree selection to F_6, giving rise to the breeder line AVE 11.04. During the following years, AVE 11.04 was evaluated in field and laboratory trials, being selected for its higher protein and β-glucan content, and being registered as 'Júpiter-INIA' during 2015.

reverse the phenotypic effects of *Dw6* by elongating the peduncle (Farnham et al., 1990). Other dwarfism features of 'Júpiter-INIA' were the more condensed stem internodes and wider main stems (subjective observations), in comparison to 'Supernova-INIA', which would confer extra strength to the stem with lower vulnerability to lodging (Milach et al., 2002; Tanhuanpää et al., 2006). These observations are in agreement with Konga et al. (2013), where morphological features such us increased stem width, width of mechanical tissue layer, and stem density, have been the best indicators of improved lodging resistance in wheat. The stem attributes of 'Júpiter-INIA' will allows the non-application of growth regulators. This is a positive contribution either in reducing the production cost or the claim of non-hormone application within the plant growth in the field.

The greater tolerance of 'Júpiter-INIA' against crown rust, together with its numerically lower disease-reaction scores to the most prevalent diseases in southern Chile (Table 5), are desirable features to obtain healthy crops without application of fungicides. Crown rust is the most damaging disease of cultivated oat while genetic resistance is the more effective, economical, and environmentally friendly means of controlling the disease (Jackson et al., 2010; Montilla-Bascón et al., 2015). Rust-specific resistance genes have been found in *Avena sativa* and *A. sterilis*, while several resistance QTLs with minor and major effects have been found in oat germplasms (Jackson et al., 2010). The resistance genes and QTLs in 'Júpiter-INIA' are still unknown. In addition, races occurring in natural populations of crown rust at southern Chile, are unknown; as well as the effectiveness of resistance genes and QTLs to control them, doing difficult their application in breeding programs. 'Júpiter-INIA' is similar to 'Supernova-INIA' in the response to BYDV. This virus, which is transmitted by aphids, is considered the most important viral disease of oats, and currently none of the oat cultivars is highly resistant (Stewart and McDougall, 2014). For this reason, it is important to consider in future cultivars the search of genetic resistance sources to this virus.

A daily dose of β-glucan of ≥ 3 g is recommended to reduce total and LDL cholesterol by 5%. A decrease between 1% to 2% of total cholesterol may reduce coronary mortality in 4% to 5% (FDA, 1997). Consequently, a serving of 80 g 'Júpiter-INIA' grains would contribute with the total β-glucan recommended dose. On the other hand, a safe intake of milk or egg protein in adult humans is about 0.57 g per day and per kilogram of body weight (Whole Health Organization; WHO, 2007). Thus, 80 g 'Júpiter-INIA' would contribute with 50% of the recommended daily intakes of plant protein, considering adults of 65 kg, 72% digestibility of oat protein, and a consumption 50% total protein from plant sources according to WHO (2007). Consequently, the development of 'Júpiter-INIA' contributes to increase the quality and diversity of oat-based food products for a healthy diet.

CONCLUSIONS

'Júpiter-INIA' is a new oat cultivar exhibiting a 20% increase in protein and β-glucan content in comparison to current commercial varieties, being the first oat cultivar released in Chile within the international quality standards for healthy food products.

'Júpiter-INIA' could plays an important role, in relation to a more environmentally friendly agriculture; due to there is no need of chemical application to control diseases and lodging.

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