Table S1.RNA used for cDNA library construction.

Variety	RNA concentration (ng/uL)	Total amount of RNA (ug)	RIN	rRNA ratio
Choyang	1,416.2	58.1	8.1	1.0
Daeyang	1,322.2	54.2	7.7	1.0
Darkhorse	1,567.0	64.2	8.2	1.0
Gehl	1,709.4	70.1	8.6	1.1
Gwanghan	1,478.8	60.6	8.2	1.0
Hispeed	899.3	36.9	6.8	1.0
Ilhan	1,489.9	61.1	7.9	1.0
Okhan	1,264.8	51.9	8.4	1.0
Samhan	1,687.1	70.9	8.6	1.2
Swan	1,741.4	71.4	7.8	1.0

RIN: RNA integrity number.

Table S2. Summary of redundant sequences removal for de novo assembly and open reading frames (ORFs) prediction for assembled unigenes.

De novo assembly	Reads	Read Bases (bp)	Min length (bp)	Max length (bp)	Average length (bp)	N50 (bp)	N90 (bp)	BUSCO (embryophyta_odb10)
Raw data	365,685,694	55,218,539,794	151	151	151			
Trimmed data	343,229,248	50,113,234,400	50	151	146			
Trinity (De novo transcriptome assembly)	709,709	617,608,507	177	23,317	870			
CD-HIT-EST (90%) - 1st	440,095	312,460,777	187	23,317	710	1,106	291	
CD-HIT-EST (90%)- 2nd	422,088	298,844,953	187	23,317	708	1,097	291	
CD-HIT-EST (90%) - 3rd	371,180	272,335,823	187	23,317	734	1,157	298	
TGICL	233,449	178,029,002	187	21,849	763	1,334	294	
Coverage Filter (Depth: > 10)	128,244	137,438,033	187	21,849	1,072	1,752	441	C: 82.3% [S: 42.8%, D:3 9.5%], F: 2.7%, M: 15.0%
Predicted partial ORFs	56,483	47,850,738	258	15,213	847	1,089	396	C: 74.1% [S:52.9%, D:21.2%], F: 6.9%, M: 19.0%
Predicted complete ORFs	32,202	32,497,662	258	15,213	1,009	1,287	489	C: 64.8% [S:48.4%, D:16.4%], F: 6.1%, M: 29.1%

C: complete. S: complete single-copy. D: complete duplicate. F: Fragmented. M: missing.

Table S3. Summary of functional annotation of 128,244 assembled unigenes.

Database	Number of annotated unigenes	Percentage (%)
NR	74,288	57.93
GO	47,758	37.24
COG	12,537	9.78
All annotated	74,288	57.93
Not annotated	53,956	42.07
Number of unigenes	128,244	-

Table S4. Summary of genotype-specific SNPs in 10 oat varieties.

Variety	Number of genotype-specific SNPs	Number of SNPs used for development of dCAPS markers
Choyang	11	9
Daeyang	7	6
Darkhorse	0	0
Gehl	147	128
Gwanghan	169	156
Hispeed	0	0
Ilhan	74	67
Okhan	34	29
Samhan	117	105
Swan	77	71
Total	636	571

Table S5. List of 28 oat varieties used in this study.

No.	Variety	Pedigree	Remark[a]
1	Samhan	Early80/Maine PI-590	Transcriptome sequencing
2	Gwanghan	Early80/PA202-210-S	Transcriptome sequencing
3	Darkhorse	Sprinter/73625	Transcriptome sequencing
4	Okhan	Early80/Maine PI-590	Transcriptome sequencing
5	Choyang	Cuauhtemoc/PA202-210-S	Transcriptome sequencing
6	Highspeed	Mex 5 mutant	Transcriptome sequencing
7	Daeyang	FLX446-1-84-Q1/(Selection 3638-2 Purified/LETORIA)	Transcriptome sequencing
8	Swan	Kent/Ballidu	Transcriptome sequencing
9	Ilhan	517A2-121/B-1-2-57	Transcriptome sequencing
10	Gehl	06751//AC Baton*2/05588-297	Transcriptome sequencing
11	Dakyeong	(Osage////Bonda///Hajira//Joanette/Santa Fe)/SWAN	
12	Dajo	(Swan*3/Cuauhtemoc//PA202-210)/Beltsville 61-150	
13	Dahan	Sprinter/73625//PA7507-37	
14	Donghan	Early80/Maine PI-590	
15	Megwiri	Introduced variety	
16	Samjeolgwiri	Introduced variety	
17	Seonyang	Early80/Cuauhtemoc	
18	Suyang	Cuauhtemoc/PA202-210-S	
19	Shinhan	517A2-121/B-1-2-57	
20	Jopung	Early80/PA202-210-S//Walken	
21	Johan	Early80/PA202-210-S	
22	Taehan	PA7836-2093/Illinois 60-3244-1	
23	Punghan	Cuauhtemoc/PA202-210//Beltsville 61-150	
24	Hi-early	517A2-121/B-1-2-57	
25	Joongmo 2005	Early80/PENNCOMP31-3//Early80/PA202-210	
26	Joongmo 2501	Early80/Cuauhtemoc//(Selection 3638-2 Purified/LETORIA)	
27	Joongmo 2507	Mex 5/Samhan	
28	Joongmo 2511	(0-200-10/Southland)/Samhan	

[a]The oat variety used for transcriptome sequencing in this study.

Table S6. Characteristics of 30 dCAPS markers and the genetic diversity detected in 28 oat varieties.

Marker name	SNP (Reference/alternate)	Physical location of SNP (Chromosome number, bp)	Forward (5′ to 3′)	Reverse (5′to 3′)	Product size (bp)	Restriction enzyme	Na	MAF	Gene diversity	Het.	PIC
CY-6	T/**C**[a]	Chr. 5A (403,652,401)[c]	AGTTATTTGCTCGGAGCGTCTA	AGTGCTTATGAACGTTTCAGGT	323	AccI	2	0.929	0.133	0	0.124
GH-16-3	C/**A**	Chr. 6C (111,635,848)	GAGGCAACACGCCAACATTCTA	CCTTACCAACAACCTCTGCTAC	302	DdeI	2	0.857	0.245	0	0.215
GH-23-3	G/**C**	Chr. 5D (489,338,915)	CCCCCGTCGTCCCGATGAAGAT	TTTACACACCGCTAACTCACAG	383	BglII	2	0.857	0.245	0	0.215
GH-3	A/**G**	Chr. 6C (109,063,073)	GAACTCAGCGGCCTCTACGATC	TCCTGGCGGTCTAATTTATCTA	318	PvuI	2	0.963	0.071	0	0.069
GH-56	G/**T**	Chr. 6C (111,635,964)	TGTGAACAGCGTGCTAAAAAGC	AGATGGCATAAAGCAGGTAAGA	377	HindIII	2	0.857	0.245	0	0.215
GH-63	T/**C**	Chr. 5D (26,700,645)	CCTCCCCACCACCACTTGTGCG	TTCTAGACTTCCATGCCACTTT	236	HinP1I	2	0.964	0.069	0	0.067
GH-65	T/**C**	Chr. 5D (489,338,827)	CTCCTGCTCCATGCTGTTGGAC	TTTACACACCGCTAACTCACAG	294	AvaII	2	0.857	0.245	0	0.215
GH-99	T/**C**	Chr. 6A (337,540,571)	AAAATTTCTATCGATCGGTCGA	GAGAAGACGATGCTTCGAGAT	340	AccI	2	0.964	0.069	0	0.067
GHA-11	A/**G**	Chr. 5C (322,852,756)	GAGTTGAAGCCGGTGCCCAATT	GGTTGAATCCCTCTCTCTTTCT	395	MunI	2	0.857	0.245	0	0.215
GHA-24	A/**C**	Chr. 3C (569,220,248)	GTAACAGACACAACTCTGCCGG	TATACAAACCACGCACGTAGAG	308	NaeI	2	0.929	0.133	0	0.124
GHA-43-1	G/**C**	Chr. 7C (26,768,619)	GCAACGCGGATCCGAGGGTCTA	CCTACCTAGTCGAATTTCCCTC	244	AccI	2	0.857	0.245	0	0.215
GHA-64	T/**C**	Chr. 5C (563,230,234)	TGTGTGTGAGATGATGCTCGGC	TCAGGCTCACTGTTTACAAATG	269	EagI	2	0.786	0.337	0	0.280
GHA-72	A/**G**	Chr. 5C (322,852,508)	GAGTTGAAGCCGGTGCCCAGCT	GGTTGAATCCCTCTCTCTTTCT	395	MspA1I	2	0.857	0.245	0	0.215
GHA-85	A/**G**	Chr. 7C (67,946,455)	CTCCAAAGCTAGAAAACACCCG	ATGGAAATGATGGGCTAGAAA	252	HpaII	2	0.929	0.133	0	0.124
GHA-101	**T**/C	Chr. 3C (87,054,816)	GTTGCTTCCTAAGAATCAGGCC	ATCCAACCTCTGTTGTGTCTTC	355	StuI	2	0.893	0.191	0	0.173
GHA-102	**T**/C	Chr. 3C (566,615,192)	CGCGCTTGGTGATATCAAGATC	AACTACAGAACCTGATGGCAAC	281	BglII	2	0.929	0.133	0	0.124
GHA-115	T/**A**	Chr. 7C (26,768,697)	TTCTCGCGAATCTCTCCACTCG	CTCTACCTAGAAGCAACGCAAT	323	AvaI	2	0.857	0.245	0	0.215
IH-12-2	**C**/A	Chr. 6C (151,698,993)	GCACCTCCGTCGTGGTAGTATA	TCAACAACAATGGTGTCTATGG	282	AccI	2	0.778	0.346	0	0.286
IH-39	T/**G**	Chr. 3D (424,852,131)	ACAATACACGAGGAGATAACCG	GGAATGGTGATTACAAGTTCAGA	266	HpaII	2	0.893	0.191	0	0.173
OH-6-1[b]	T/**C**	Chr. 7C (2,572,432)	GCTGTAGCTAGCGTGCGAGGTC	GCATCTCTCAACACCATCTCAT	306	AvaII	2	0.821	0.293	0	0.250
SH-4	T/**C**	Chr. 1C (30,457,950)	TATGGTCCATGCGTCACAGGTA	CTTCTGCAAATAGTGAACGACA	255	RsaI	2	0.857	0.245	0	0.215
SH-5-1	T/**C**	Chr. 2C (468,854,520)	GGCAAGAGGTGCACCTGGGAAT	AGTAGGCTCAGATGACGATGAG	298	HinfI	2	0.893	0.191	0	0.173
SH-20-1[b]	A/**T**	Chr. 3D (439,425,986)	GGATTAGTCGTGCATTGTCCGC	CCAAAGTTGTTCACTCTTTGGTA	244	MspA1I	2	0.750	0.375	0	0.305
SH-28-2	**C**/G	Chr. 6C (39,649,991)	GGATGTCGCTGGGTAGCGGTAC	TTCCAACTGAACAATCTTGACA	373	KpnI	2	0.714	0.408	0	0.325
SH-34-4	G/**A**	Chr. 2D (31,590,429)	CTGCCACGTGAGGCCTCGACTA	TACATTGGTGATGATTAGTCGC	283	DdeI	2	0.643	0.459	0	0.354
SW-3-2	G/**A**	Chr. 2C (228,398,434)	CCAGGCCGTGTCATCTATCATG	ACGAGGGAGTACTGTACGTGAT	233	BspHI	2	0.857	0.245	0	0.215
SW-21-2	G/**C**	Chr. 3C (541,837,616)	TGGTTTACAGATGAGTGGCCGG	CCTCAAGAGGAAAGGGTAAAGT	302	NaeI	2	0.786	0.337	0	0.280
SW-23-1	T/**A**	Chr. 2C (180,580,925)	TGCCCCCAGCATGACCTTCATG	CCTGTGTACATACACGTTTGCT	341	BspHI	2	0.857	0.245	0	0.215
SW-27	T/**C**	Chr. 2C (148,179,408)	TTCAGGCGCACGGGAGATGGTC	GTGTTTCACTGAACTGCGAATA	209	AvaII	2	0.857	0.245	0	0.215
SW-45	A/**C**	Chr. 6C (75,155,534)	CCAGCATATACGGTGATGTCAA	CCATCTACCCGAGGTTCGT	224	HincII	2	0.857	0.245	0	0.215

[a] Allele type of SNP with bold character which showed the shorten band generated by restriction enzyme. [b] Genotyping by InDel variation. [c] The chromosomal location of the SNP used for dCAPS marker development was searched using genome sequence data of PepsiCo OT3098 Hexaploid Oat v2 pseudomolecules (2021) through BLASTN hosted by GrainGenes (https://wheat.pw.usda.gov/GG3/). Na: number of alleles. MAF: major allele frequency. Het.: heterozygosity. PIC: polymorphism information content.