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RESEARCH ARTICLE

Allelic variants of *EFL3* and their association with early flowering traits in chickpea (*Cicer arietinum* L.)

Alok Das*, P.S. Shanmugavadivel, Biswajit Mondal[1], P. S. Basu[2] and G. P. Dixit

Abstract

Allele mining of the *ELF3* gene, associated with flowering time in chickpeas, was conducted using whole genome resequencing data from 254 chickpea genotypes within the chickpea reference set, utilizing the GATK tool. A total of 671 genetic variants were identified within the *ELF3* genic region, encompassing not only its genic region but also its 1 kb promoter region and intergenic regions. Among these, biallelic SNPs were predominant (641), followed by multi-allelic InDels (19), multi-allelic SNPs (9) and least of biallelic Indels (2). Out of these 641 biallelic SNPs, 44 SNPs were located across 4 exons of *ELF3* gene [exon 2 (16), exon 5 (10), exon1 (9) and exon 4 (9)] while exon 3 displayed no genetic variants. Remarkably, the distinctive 11 bp deletion within the first exon of ICC96029's *Elf3* was not detected in this analysis. Instead, a missense mutation within *ELF3* was identified within the studied chickpea reference set. Utilizing a candidate gene-based association mapping approach, 20 variants (comprising 18 biallelic SNPs and 2 InDels) were employed based on their presence in at least 95% of genotypes. Employing a general linear model (GLM) approach with three years of phenotypic data, a total of 4 significant marker-trait associations (MTAs) were identified. Specifically, the variants SNP_021164.1_36025048 (G/A) and SNP_021164.1_36021869 (A/C) exhibited associations with the FLD/s trait. Additionally, the SNP locus SNP_021164.1_36011429 (C/T), located within exon 5 of ELF3a, and SNP_021164.1_36013862 (C/A) located within intron 2, displayed associations with the pod_D/S trait. These allelic variants, particularly the missense mutation, carry significant importance due to their potential impact on the interaction of *ELF3* with interacting proteins that eventually lead to variations in flowering time within the chickpea population.

Keywords: EFL3, Early flowering, Association mapping, Molecular markers, chickpea.

Introduction

Chickpea is an important grain legume to nutritional security and sustainable agriculture. Chickpea is traditionally grown as a low-input crop in areas predominantly relying on conserved soil moisture where the crop often experiences terminal drought stress (Gaur et al. 2008). The variability in climate conditions has impacted production drastically, and terminal drought coupled with elevated temperature regimes has become a major constraint in many chickpea-growing areas. Early flowering has been identified as an important trait for increasing and stabilizing chickpea productivity.

Four major loci *viz.*, *Efl1* to *Efl4*, with recessive alleles controlling early flowering time in chickpeas, have been reported earlier (Or et al. 1999; Kumar and van Rheenen 2000; Hegde 2010; Gaur et al. 2015). Of these, the *Efl1* genetic locus was further validated in ICCV96029 for flowering time and photoperiod sensitivity trait following the QTL mapping approach and identified *Arabidopsis* ortholog circadian gene *ELF3* as a candidate in one of the major QTL identified on chromosome 5, for early flowering in short-day conditions (Ridge et al. 2017). ELF3 is a versatile protein with dual functions. Firstly, it serves as a component of the

Division of Plant Biotechnology, ICAR-Indian Institute of Pulses Research, Kanpur 208 024, India.

[1]Division of Crop Improvement, ICAR-Indian Institute of Pulses Research, Kanpur 208 024, India.

[2]Division of Basic Sciences, ICAR-Indian Institute of Pulses Research, Kanpur 208 024, India.

***Corresponding Author:** Alok Das, Division of Plant Biotechnology, ICAR-Indian Institute of Pulses Research, Kanpur 208 024, India, E-Mail: alok.das@icar.gov.in

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evening complex (EC), which is formed by the interaction of the ELF3, ELF4, and LUX proteins), contributing to the regulation of the plant circadian oscillator. EC functions as a transcriptional repressor complex and a core component of the plant circadian clock**.** Secondly, ELF3 plays a role in modulating phyB signaling to control light input and gating in the oscillator (Kolmos et al. 2011).

Interestingly, the *ELF3* gene in ICCV96029 carries an 11 bp mutation in the first exon as *Elf3* that negatively affects the function of this gene. This deletion was predicted to cause a premature stop codon, leading to a truncated or non-functional *Elf3* protein in ICC96029. The 11-bp deletion associated with early flowering in chickpeas is considered to have arisen recently, as this mutation is not widely distributed (Ridge et al. 2017). Despite the knowledge about 11 bp deletion associated with flowering time in chickpea cultivar ICCV96029, a substantial amount of natural variation exists in mini core lines of chickpea and their association with flowering time remains unexplored. The main objective of the study was to document the allelic variants of the *ELF3* gene in the chickpea reference set and associate variations in the locus with flowering time and pod-filling traits following the candidate gene-based association approach.

Materials and methods

A set of 254 chickpea reference genotypes were raised in the light textured sandy loam soil of New Research Farm of ICAR-Indian Institute of Pulses Research, Kanpur (Coordinates: 26.449923°N 80.331874°E) in alpha design with three replications. About 50 mm of irrigation was applied the next day to ensure complete emergence. Need based insecticide sprays against pod borer (*Helicoverpa armigera* H.) were provided, and the plots were kept weed free by manual weeding. Pre-emergence weedicide pendimethalin at 3 mL/L was applied immediately after sowing the crop. Manual weeding was followed thereafter at regular intervals. By regular observation, the date when first flower appeared and similarly when 50% or more of the plants in a plot flowered was recorded as first and 50% flowering time of the plot, respectively and the dates when first pod initiated in a plot were recorded as 1st pod for each accession. During *rabi* seasons of three consecutive years *viz*., 2018-19, 2019-20 and 2020-21 (Supplementary Table S1). The *ELF3a* gene sequence (LOC101489432) of CDC Frontier genotype comprised of 5 kb gene sequence (Ca5, NC_021164.1: 36011384..36016600) along with 1 kb upstream promoter region and intergenic region between LOC101489432 and LOC101489846 were retrieved from CDC Frontier genome assembly and used as a reference sequence for downstream analysis to call variants.

The whole genome resequencing data of 254 chickpea genotypes were retrieved from the National Center for Biotechnology Information (nih.gov) (NCBI) database. The SRA files obtained were subjected to read filtering using the Trimmomatic tool (Bolger et al. 2014). This filtering process was aimed to remove various types of unwanted sequences, including adapter contamination, low-quality bases, and reads shorter than 70 bases. The filtered reads of the 254 genotypes were aligned individually to the *ELF3a* gene sequence (LOC101489432) of the CDC Frontier genotype using the BWA tool (Li and Durbin 2009). The resulting alignment files, in the ".bam" format, for each genotype were then processed to identify and mark duplicate reads using the markduplicates functions of the PICARD tool (https://broadinstitute.github.io/picard/). The resulting files were used for genetic variant calling using a series of functions from the genome analysis tool kit (GATK) tool, namely, HaplotypeCaller, CombineGVCFs, and GenotypeGVCFs with a standard minimum confidence threshold of 20 (Poplin et al. 2017). The resulting ".vcf" file was hard filtered to select good quality variants with minimum quality by depth of < 2.0, Fisher Strand value of >60, Strand Odds Ratio of > 3.0, RMS Mapping Quality of < 40, Mapping Quality Rank Sum Test of < -12.5 and Read Position Rank SumTest of < -8. The resulting filtered genetic variants obtained from the previous step were subjected to annotation and prediction of their functional effects using the SnpEff tool (Cingolani et al. 2012). TASSEL v2.0.1 (Bradbury et al. 2007) was employed to evaluate marker-trait associations (MTA) for the traits using genotypic data and 3 years of phenotypic data following a general linear model (GLM). In the analysis, significant marker-trait associations (MTAs) were declared based on a significance threshold of a p-value ≤ 0.05 with the relative magnitude of variation explained by the marker, which is represented as R^2.

Results and discussion

The early flowering trait in most crops, including chickpeas, is a complex trait influenced by a combination of genetic and environmental factors. According to Fornara et al. (2010), there are six major pathways *viz*., photoperiod pathway, ambient temperature pathway, vernalization pathway, autonomous pathway, aging pathway, and gibberellins pathway that coordinate to regulate flowering in plants. Early flowering in chickpeas can indeed confer several advantages, particularly in regions characterized by short growing seasons or crops prone to drought or heat stress. This is especially relevant in northern India, where chickpea cultivation faces challenges such as terminal heat and drought stress during the early completion of winter, exacerbated by changing climatic conditions. Early maturing chickpea varieties can align their reproductive phase with more favorable environmental conditions, resulting in improved yield and overall crop performance. Hence, there is a pressing need for breeding efforts and research studies that focus on understanding the underlying mechanisms and identifying genetic markers associated with early

flowering to develop improved chickpea varieties with desirable traits for achieving food security in the current scenario.

ELF3 gene sequence and genetic variants

The *ELF3* gene is located on the complementary strand of DNA and spans a region of 5216 base pairs (bp) on chromosome 5 (NC_021164.1). It consists of five exons and four introns, with its genomic position ranging from 36011384 to 36016600 on the chromosome. This gene is flanked by an uncharacterized protein-coding gene on the left end (LOC101489099: 36007947..36011130) and zinc finger CCCH domain-containing protein 20-like coding gene on the right side (LOC101489846: 36029820..36031169). In the gene location of *ELF3*, including its 1kb upstream and intergenic regions between *ELF3* and LOC101489846 loci, a total of 671 genetic variants were identified. Of these, biallelic SNPs were predominant (641), followed by multi-allelic InDels (19), multi-allelic SNPs (9) and least of biallelic Indels (2) (Table 1). Of these 641 biallelic SNPs, 533 SNPs were present in intergenic region between *ELF3* and LOC101489846 followed by 33 SNPs in 1kb upstream region of *ELF3*, 44 SNPs in 4 exons [exon 2 (16), exon 5 (10), exon1 (9) and exon 4 (9)], five SNPs in downstream of a gene, 26 SNPs in 4 introns of *ELF3* and exon 3 does not have any genetic variants (Table 1). Of these 44 SNPs present in exons, 14 SNPs present in exon 2 (9) and exon 4 (5) cause missense mutations. The majority of these biallelic SNPs consisted of transitions (427), outnumbering transversions (214). Moreover, there were fewer purine-to-purine transitions (209) compared to pyrimidine-to-pyrimidine transitions (216). Among transversion, G to T (35) were predominant followed by T to G (33) (Fig. 1).

In addition to these biallelic SNPs, there were 9 multi-allelic SNPs identified, along with 2 biallelic single base pair InDels located in the intergenic region between ELF3 and LOC101489846. Furthermore, 19 multi-allelic InDels

Table 1. Genetic variants documented in *EFL3* genic region

S. No.	Gene Structure/Genetic Variant	Biallelic SNP	Multi-allelic SNP	Biallelic InDels	Multi-allelic Indels	Biallelic SNP impact (Missense)
1	Exon 1 with 5′ UTR	9	0	0	1	-
2	Intron 1	2	0	0	0	-
3	Exon 2	16	0	0	0	9
4	Intron 2	18	0	0	0	-
5	Exon 3	0	0	0	0	0
6	Intron 3	2	0	0	0	-
7	Exon 4	9	0	0	0	5
8	Intron 4	4	0	0	0	-
9	Exon 5 with 3′ UTR	10	0	0	0	-
10	1 kb upstream	33	1	0	1	0
11	down steam of a gene	5	1	0	0	0
12	Intergenic between *EFL3* and LOC101489846	533	7	2	17	-
Total (671)		641	9	2	19	14

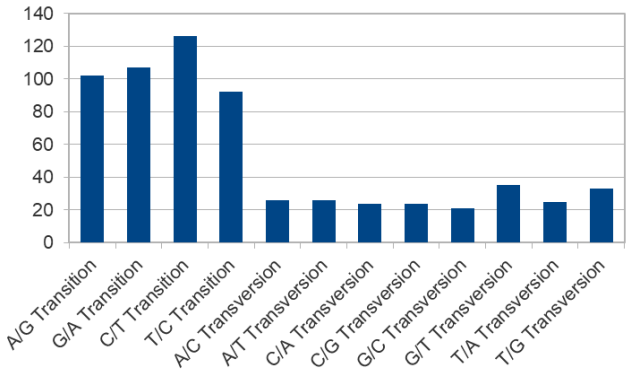

Fig. 1. Transitions and transversions in *EFL3* gene

Table 2. Markers information used in association analysis

S. No.	*ELF3* gene structure	No. of biallelic SNPs	No. of biallelic InDels
1	Exon 5	1	0
2	Intron 2	1	0
3	1 kb upstream	1	0
4	Intergenic	15	2
Total (20)		18	2

Table 3. Association of flowering time and days to pod setting traits using GLM approach

S.No.	Trait	Locus	Type	Variant location on Gene Structure	Y1			Y2			Y3		
					F value	P value	R^2 Marker	F value	P value	R^2 Marker	F value	P value	R^2 Marker
1	FLD/S	SNP_021164.1_36025048	G/A	Intergenic	2.7884	0.0412	0.0324				2.7131	0.0455	0.0315
2	FLD/S	SNP_021164.1_36021869	A/C	Intergenic	2.7282	0.0446	0.0317				3.2641	0.022	0.0377
3	pod_D/S	SNP_021164.1_36011429	C/T	Exon 5				3.7448	0.0117	0.0432			
4	pod_D/S	SNP_021164.1_36013862	C/A	Intron 2				3.3615	0.0194	0.0391			

were identified, with the majority of them (17) found in the intergenic region between *ELF3* and LOC101489846, followed by each one in 1 kb promoter region and exon1. Among these multi-allelic InDels, variations ranged from 1-base insertions to a maximum of 60-base pair deletions and 46-base pair insertions. In the current analysis, we did not detect the presence of this unique 11-bp deletion in the first exon of *ELF3* among the 254 chickpea accessions from the chickpea reference collection and this further validates it as a very rare variant in the genotype included in the collection. In addition to the unique 11 bp deletion identified in ICC96029, a total of 671 mutations, including SNPs and InDels, were discovered from the analysis of 254 genotypes of chickpeas. These mutations were found in various regions, including exons, introns, 1 kb upstream regions, and intergenic regions between the *ELF3a* and LOC101489846 loci. However, it is worth noting that while most of the identified mutations did not have a detrimental effect on the function of the *ELF3a* gene, there were a few missense mutations that could potentially alter the protein's structure and function by introducing amino acid changes that could eventually impact the protein's activity or interaction with other protein molecules. There are many reports on the impact of a missense mutation in genes determining flowering time in various crops. A missense mutation in a large subunit of ribonucleotide reductase confers temperature-gated tassel formation in maize (Xie et al. 2020). A natural variation in the highly conserved serine/threonine kinase domain of *EL1* in H143 of heading-date gene namely *Early flowering1* enables *japonica* rice to flower early under long-day (LD) conditions at high latitudes (Kwon et al. 2014). Missense mutation of C to T in exon 4 of one copy of VRN-A1 in wheat affects heading date variation (Xue et al. 2023).

ELF3 interaction with other protein partners

ELF3 directly interacts with several proteins, including protein time for coffee-like isoform x1 (LOC101496754), transcription factor PIF4-like isoform X1 (LOC101488979), phytochrome a-like isoform x1 (LOC101496082; LOC101506511), serine-rich adhesin for platelets-like (LOC101510087), transcription factor PCL1-like/two-component response regulator ARR2-like isoform X1(LUX), and protein EARLY FLOWERING 4-like (LOC101498412) (Fig. 2). Han et al. (2019) employed a genome editing technique to create an early flowering genotype in soybeans targeting $E1$ gene controlling soybean flowering. They identified two novel types of mutations, 11 and 40 bp deletion at E1 coding region, respectively. Frameshift mutations produced premature translation termination codons and truncated E1 proteins, causing obvious early flowering under long-day conditions. In our study, we observed missense mutations in *ELF3* that may have a negative effect on the interaction with LUX and ELF4 and further formation of effective EVENING COMPLEX. These

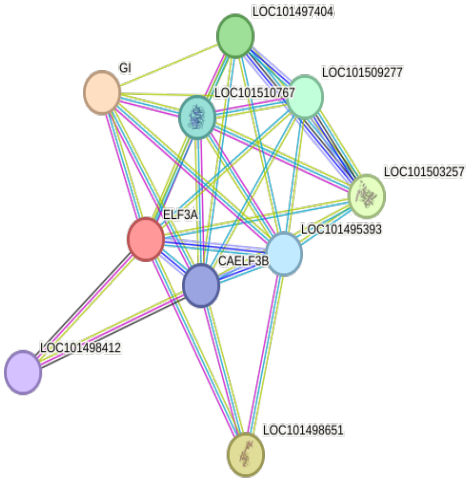

Fig. 2. Interaction of EFL3 with other proteins

missense mutations could potentially contribute to early flowering in chickpeas by derepression of PIF4 from EC and subsequent activation of PIF4 target genes. By creating recessive alleles of *elf3* that disrupt the interaction with LUX and ELF4, it is possible to alter the regulatory dynamics of the plant circadian clock, resulting in accelerated flowering in chickpeas.

Association mapping

For candidate gene-based association mapping, only the biallelic SNPs and InDels that were called in at least 95% of genotypes were considered for analysis. A total of 20 variants, consisting of 18 SNPs and 2 InDels, that were present in at least 241 of the genotypes studied were selected for analysis (Table 2). These 18 SNPs were present in exon 5 (1), intron 2 (1), 1kb upstream of the gene (1) and intergenic region (15). Both the InDels were present in the intergenic region. Of these selected 18 SNPs, 11 were of transition type, and 7 were of transversion type. Association with these selected 20 variants and 3 years of phenotypic data identified 4 MTAs for FLD/s (2) and pod_D/S (2) following the general linear model (GLM) approach (Table 3). Of which, SNP_021164.1_36025048 (G/A) and SNP_021164.1_36021869 (A/C) were associated with FLD/s trait in two years data but not associated with year 2 data. The SNP loci SNP_021164.1_36011429 (C/T) present in exon5 of *ELF3a* and SNP_021164.1_36013862 (C/A) present in intron 2 were associated with pod_D/S trait in year 2 data only.

From the association mapping study, we could identify two MTAs linked with the FLD trait in Y1 and Y3 data and their linked markers located upstream of the *ELF3a* gene. It is possible that these markers could fall in the effector/suppressor/insulator region. Unfortunately, none of the missense mutations identified in this study were included in the association analysis as their allele frequencies did not meet the threshold of >0.95. This is attributed to inadequate sequence coverage in the studied genotypes. Although the linked markers associated with Pod D/S explain approximately 4% of a variation for the considered trait, the MTAs were statistically highly significant. Candidate gene-based association studies in plants have contributed significantly to our understanding of the genetic basis of various traits such as kernel composition and starch production in maize (Wilson et al. 2004), starch gelatinization temperature in rice (Bao et al. 2006), flowering time and water-soluble carbohydrate content in *Lolium perenne* (Skøt et al. 2007), flowering time in *Arabidopsis* (Ehrenreich et al. 2009), flowering time and plant height in soybean (Han et al. 2021), drought tolerance in European beech (Cuervo-Alarcon et al. 2021), and girth growth in rubber trees (Bhusudsawang et al. 2021).

The development of high-yielding early-maturing chickpea cultivars could be highly beneficial for chickpea farmers, particularly in mitigating yield losses caused by adverse terminal heat and drought conditions. Additionally, the saved time due to early flowering would provide farmers with the opportunity to grow subsequent crops, enhancing agricultural efficiency and potentially increasing overall yield. These advancements in manipulating flowering time have the potential to be valuable tools for chickpea farming and contribute to improved resilience in the face of environmental challenges.

Supplementary material

Supplementary Table S1 is provided, which can be accessed at www.isgpb.org

Authors' contribution

Conceptualization of research (AD, GPD); Designing of the experiments (AD, PSS, BM, PSB); Contribution of experimental materials (AD, PSB); Execution of field/lab experiments and data collection (AD, PSB, PSS); Analysis of data and interpretation (PSB, PSS); Preparation of manuscript (AD, PSS, PSB, GPD).

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Supplementary Table S1

Accession No	FL D/S	pod D/S
ICC 14815	82	96
ICC 12537	80	91
ICC 10755	81	102
ICC 4363	83	97
ICC 12851	87	105
ICC 3776	86	101
ICC 16654	83	96
ICC 9862	81	96
ICC 13719	86	102
ICC 6579	84	99
ICC 6874	81	99
ICC 8350	84	100
ICC 6294	86	101
ICC 10018	84	99
IG 10569	82	101
ICC 8718	86	101
ICC 2969	87	99
ICC 2242	93	101
ICC 15868	85	99
ICC 8058	84	107
ICC 2679	88	105
ICC 4841	87	110
ICC 15518	85	112
ICC 791	100	111
ICC 13764	86	105
ICC 9137	84	108
ICC 1398	85	103
ICC 5434	84	105
ICC 14778	85	100
ICC 11498	86	101
ICC 506	85	100
ICC 1710	89	101
ICC 4463	80	100
ICC 13461	89	102
ICC 10701	89	100
ICC 5135	85	101
ICC 7255	85	99
ICC 14098	80	95
ICC 12928	98	105
ICC 11121	85	99
ICC 13187	86	97
ICC 8621	80	97
ICC 1205	85	100
ICC 7150	82	102
ICC 9434	95	105
ICC 13892	85	101
ICC 3230	89	101
ICC 11664	93	108
ICC 5639	85	102
ICC 4567	85	101
ICC 67	87	99
ICC 12155	79	97
ICC 10341	85	101
ICC 3391	84	100
ICC 8607	81	99
ICC 1915	100	116
ICC 15762	87	100
ICC 15294	85	105
ICC 15610	95	103
IG 6047	87	103
ICC 1083	80	97
ICC 16261	87	105
ICC 10685	87	103
ICC 4991	87	103
IG 7087	100	112
ICC 2507	80	100
ICC 3421	83	101
ICC 7554	85	102
ICC 6279	86	103
IG 69438	87	105
ICC 13219	84	97
ICC 7305	83	93
ICC 7571	78	94
ICC 2580	82	94
ICC 12947	85	100
ICC 15612	80	95
ICC 10393	78	98
ICC 8740	81	99
ICC 1098	87	100
ICC 95	86	101
ICC 2629	87	103

ICC 8151	83	102	ICC 9872	84	102
ICC 5845	97	107	ICC 3776	80	99
ICC 5337	99	110	ICC 15510	83	102
ICC 10939	93	101	ICC 12028	84	105
ICC 1230	88	103	ICC 4093	80	99
ICC 11279	86	100	ICC 4814	81	99
IG 6055	83	101	ICC 10399	85	101
ICC 14199	81	99	ICC 12492	86	108
ICC 7272	80	101	ICC 1397	88	105
ICC 7296	85	100	ICC 15785	85	106
ICC 3512	83	102	ICC 6816	82	101
ICC 5221	84	101	ICC 456	81	100
ICC 14051	83	100	ICC 12824	83	101
ICC 9590	81	99	ICC 16915	81	101
ICC 3410	80	97	ICC 8318	78	92
ICC 6293	85	101	ICC 1510	85	104
ICC 11764	88	105	ICC 8521	90	105
ICC 11198	87	101	IG 72109	85	101
ICC 4593	98	105	ICC 2263	93	105
ICC 15333	87	101	ICC 7323	83	100
ICC 16269	81	100	ICC 4495	80	99
ICC 1923	100	112	IG 6343	86	100
ICC 12299	90	101	ICC 10885	80	97
IG 72070	89	103	ICC 14831	82	100
IG 10500	81	95	ICC 15248	85	105
ICC 9002	85	99	IG 5909	84	102
ICC 8200	84	97	ICC 10466	84	100
ICC 6263	81	100	ICC 9895	81	101
ICC 13628	85	101	ICC 3325	82	101
ICC 762	98	113	ICC 4853	78	100
ICC 14669	80	99	ICC 6802	99	109
ICC 12379	83	101	ICC 15567	80	101
ICC 7867	84	102	ICC 12866	84	99
ICC 15802	81	97	ICC 4872	78	99
ICC 16524	85	102	ICC 15888	77	96
ICC 4418	82	101	ICC 867	80	98
ICC 2929	85	100	ICC 1715	97	107
ICC 13441	93	104	ICC 1180	85	102
ICC 6571	87	107	ICC 8384	82	99
ICC 2210	98	111	ICC 14799	87	102
ICC 9848	85	100	ICC 6875	85	104

ICC 9402	86	105	ICC 13124	80	102
ICC 8752	85	109	ICC 7052	82	100
ICC 2737	85	105	ICC 15606	81	99
ICC 2990	86	101	ICC 11627	88	105
ICC 13523	80	102	ICC 5613	80	101
ICC 11584	95	101	ICC 5912	85	103
ICC 5504	87	100	ICC 1161	97	112
ICC 637	80	99	ICC 9636	90	105
ICC 12654	78	94	ICC 13283	80	102
ICC 13816	85	102	ICC 7668	80	95
ICC 6877	85	102	ICC 11303	81	97
IG 73458	85	100	ICC 7326	85	95
ICC 9586	91	105	ICC 8195	98	106
ICC 4958	85	103	ICC 7308	83	98
ICC 15614	80	99	ICC 3582	86	105
IG 6905	88	102	ICC 15618	80	99
ICC 10945	82	96	IG 6067	82	101
ICC 13863	77	96	ICC 8855	79	99
ICC 7184	85	101	ICC 4639	95	105
IG 6044	78	99	ICC 9643	85	99
ICC 9418	86	102	ICC 12307	86	101
ICC 12037	80	97	IG 10309	84	91
ICC 1882	82	102	ICC 3631	83	100
ICC 2277	83	99	ICC 9702	87	103
ICC 9942	80	95	ICC 1356	80	99
ICC 7441	81	97	ICC 5878	88	102
ICC 8522	83	98	ICC 8318	85	99
ICC 4918	81	99	ICC 16374	80	102
ICC 15264	84	102	ICC 16796	80	101
ICC 708	81	104	ICC 11284	85	99
ICC 3218	85	101	ICC 6306	85	105
ICC 14077	89	102	ICC 5383	80	101
ICC 3362	89	101	ICC 2720	95	103
ICC 3239	83	102	ICC 1422	80	100
ICC 10673	89	107	ICC 11879	81	101
ICC 1194	88	112	ICC 13524	81	100
ICC 16207	87	111	ICC 2065	95	105
ICC 6811	88	103	ICC 15406	83	103
ICC 1052	85	105	ICC 283	81	104
ICC 12916	90	101	ICC 13077	81	102
ICC 4533	81	99	ICC 07110	81	99

ICC 11944	85	100	ICC 1413	85	103
ICC 12726	82	101	ICC 9755	81	101
ICC 8950	85	101	ICC 1164	85	101
ICC 14595	81	104	ICC 15697	80	100
ICC 3892	83	105	ICC 13357	80	96
ICC 1392	86	105	ICC 4657	85	102
ICC 7315	85	107	ICC 440	85	102
ICC 16903	83	99	ICC 4182	79	99
ICC 7819	88	103	ICC 9712	85	101
ICC 2884	83	98	ICC 16487	99	112
ICC 2072	90	100	ICC 3761	81	99
ICC 8515	89	101	ICC 11378	88	99
ICC 8261	86	102	ICC 15435	80	102
IG 10419	75	91	IG 6154	100	112
ICC 13599	85	101	ICC 5879	80	100
ICC 6537	91	104	ICC 12324	80	101
ICC 12328	86	99	ICCV 92944	80	95
ICC 7413	80	94			