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Genome wide analysis of NAC gene family 'sequences' in sugarcane and its comparative phylogenetic relationship with rice, sorghum, maize and Arabidopsis for prediction of stress associated NAC genes

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ABSTRACT

A total of 85 NAC genes of sugarcane (ScNAC) were retrieved from GRASSIUS (grass regulatory information server). An overview of this gene family is presented including conserved domains, phylogenies, comparative analysis of NAC genes of sugarcane with its closest relative sorghum and with other monocot species. Among the Poaceae family, the NAC genes from sugarcane showed high sequence identity with most of the NAC genes of *Sorghum bicolor*. A highly conserved two proline residues, a glycine, phenyl alanine and leucine residues are present in N-terminal domain. Conserved amino acid residues and phylogeny helps us to classify the ScNAC gene family into two major groups (Group I and II) and five subgroups (A–E). The analysis of phylogenetic tree of NAC protein sequences of sugarcane with sorghum, rice, maize and Arabidopsis reveals distinct clades with several orthologs and paralogs. A total of 30 pairs of paralogous NAC genes were identified in sugarcane. Based on the orthology, putative stress associated NAC genes were predicted in sugarcane. These stress associated NAC genes of sugarcane and their orthologs from other species were clustered in the phylogenetic tree and shared common motifs, revealing the possibility of functional similarities within this subgroup.

1. Introduction

NAC gene family is one of the largest and most important transcription factors in plants. The term NAC has been derived from three genes sharing common domain (Nac domain). These genes are; NAM (No apical meristem), ATAF 1/2 (Arabidopsis transcription activator factor 1/2) and CUC2 (cup shaped cotyledon) (Souer et al., 1996).

NAC transcription factors have been implicated in plant growth and development, including flowering (Kim et al., 2007; Sablowski and Meyerowitz, 1998), cell division (Kim et al., 2006), lateral root development (Xie et al., 2000), leaf senescence (Guo and Gan, 2006; Yang et al., 2011; Podzimska-Sroka et al., 2015), secondary cell wall biosynthesis (Zhong et al., 2007), response to pathogen infection (Xie et al., 1999; Saga et al., 2012) and adaption to the abiotic stress (Puranik et al., 2012; Wu et al., 2012; Nakashima et al., 2007; Tran et al., 2004; Shahnejat-Bushehri et al., 2012). Based on their response to abiotic stimuli, a set of NAC genes have been designated as Stress associated NAC (SNACs). These genes were used to develop transgenic plants. A transgenic rice with SNAC1 (ONAC045), OsNAC5 and OsNAC6 genes showed improved tolerance to drought and salt stresses (Hu et al., 2006; Zheng et al., 2009; Song et al., 2011); OsNAC10 and ONAC022 over-expressing rice plants showed improved drought tolerance (Jeong et al., 2010; Hong et al., 2016); NAC gene from finger millet (EcNAC1) conferred abiotic stress tolerance in tobacco (Ramegowda et al., 2012).

Considering their significance in plant growth and development, whole genome search was done to identify NAC genes in many plants like Arabidopsis (117 NAC genes), rice (151), grape (79), citrus (26), poplar (163) soybean and tobacco (152), foxtail millet (147) and cotton (145) (Rushton et al., 2008; Hu et al., 2010; Nuruzzaman et al., 2010;

Abbreviation: NAC, Nam Ataf Cuc2; TF, transcription factor; NAM, no apical meristem; ATAf, Arabidopsis transcription activator factor; CUC, cup shaped cotyledon; SNAC, stress associated NAC; Sc, sugar cane; Os, *Oryza sativa*; At, *Arabidopsis thaliana*; Zm, *Zea mays*; Sb, *Sorghum bicolor*.

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Table 1
List of NAC genes of sugarcane retrieved from Grassius Grass Gene Regulatory Information Server (http://grassius.org/family.html?KEYWORDS=NAC&SPECIES=4).

Sl. No	Gene name	Gene ID	Length (in bp)
1	ScNAC01	PTSo00907.1	565
2	ScNAC02	PTSo00903.1	675
3	ScNAC03	PTSo00884.1	591
4	ScNAC04	PTSo00908.1	875
5	ScNAC05	PTSo00901.1	628
6	ScNAC06	PTSo00889.1	1079
7	ScNAC07	PTSo00892.1	977
8	ScNAC08	PTSo00894.1	667
9	ScNAC09	PTSo00886.1	677
10	ScNAC10	PTSo00895.1	1193
11	ScNAC11	PTSo00905.1	847
12	ScNAC12	PTSo00891.1	672
13	ScNAC13	PTSo00904.1	561
14	ScNAC14	PTSo00898.1	767
15	ScNAC15	PTSo00902.1	645
16	ScNAC16	PTSo00887.1	697
17	ScNAC17	PTSo00888.1	612
18	ScNAC18	PTSo00897.1	657
19	ScNAC19	PTSo00899.1	612
20	ScNAC20	PTSo00909.1	498
21	ScNAC21	PTSo00906.1	871
22	ScNAC22	PTSo00896.1	1834
23	ScNAC23	PTSo00900.1	697
24	ScNAC24	PTSo00890.1	995
25	ScNAC25	PTSo00893.1	795
26	ScNAC26	PTSo00885.1	620
27	ScNAC27	SCCCLR2003E10.g	1262
28	ScNAC28	SCCCFL2002D10.g	451
29	ScNAC29	SCAGRT2039C10.g	1259
30	ScNAC30	SCCCCL3140E02.g	1495
31	ScNAC32	SCJLRT2049C12.b	756
32	ScNAC33	SCQGRT3042B03.g	667
33	ScNAC34	SCCCLR2003C07.g	1332
34	ScNAC35	SCCCCL5072D12.g	706
35	ScNAC36	SCCCCL3001D02.g	1774
36	ScNAC37	SCMCLB2082D05.g	605
37	ScNAC38	SCCCCL4007D12.g	1559
38	ScNAC39	SCMCSB1109C11.g	880
39	ScNAC40	SCJLRT1015C08.g	610
40	ScNAC41	SCMCRT2106G06.g	600
41	ScNAC42	SCEZLB1010H05.g	2834
42	ScNAC43	SCEQRT1028D09.g	628
43	ScNAC44	SCCCCL6005A08.g	807
44	ScNAC45	SCACLR2007C06.g	642
45	ScNAC46	SCJFRZ2014D06.g	1329
46	ScNAC47	SCRLCL6031C08.g	2578
47	ScNAC48	SCBGLB2012G03.g	691
48	ScNAC49	SCRLCL6032B05.g	481
49	ScNAC51	SCRUAD1132D09.g	913
50	ScNAC52	SCBGLR1118D05.g	2282
51	ScNAC53	SCCCRZ1004G12.g	1160
52	ScNAC54	SCCCCL4009H05.g	556
53	ScNAC55	SCJLRT1018B06.g	581
54	ScNAC56	SCCCFL4092A07.g	719
55	ScNAC57	SCEZRT2023G12.g	729
56	ScNAC58	SCEQRT1024G05.g	2525
57	ScNAC59	SCEZHR1086A06.g	674
58	ScNAC60	SCVPLB1019G11.g	710
59	ScNAC61	SCJFRT2054C09.g	628
60	ScNAC62	SCEQRT2030D12.g	605
61	ScNAC63	SCVPLR2027G07.g	516
62	ScNAC64	SCQGAM2026C10.g	797
63	ScNAC65	SCSGLV1011C04.g	616
64	ScNAC67	SCRLST3165E07.g	612
65	ScNAC68	SCEQRT1024G08.g	890
66	ScNAC69	SCQSRT2036F07.g	1221
67	ScNAC70	SCQGAM2028E09.g	648
68	ScNAC71	SCJLHR1027E08.g	597
69	ScNAC72	SCMCCL6052E02.g	410
70	ScNAC73	SCMCLR1122B12.g	601
71	ScNAC74	SCCCLR1C04H01.g	1372
72	ScNAC75	SCCCSB1002A01.g	974
73	ScNAC77	SCEPRT2048G05.g	1260
74	ScNAC78	SCEZRT2019E05.g	615
75	ScNAC79	SCBGFL3094F02.g	826
76	ScNAC80	SCCCST3005F09.g	1068
77	ScNAC81	SCEQRT1027H04.g	517
78	ScNAC82	SCCCLB1024F12.g	716
79	ScNAC83	SCCCCL4014A04.g	1380
80	ScNAC84	SCEPLB1042D02.g	2439
81	ScNAC85	SCBGLR1002H04.g	724
82	ScNAC86	SCEQRT1031E06.g	1247
83	ScNAC87	SCEPAM1021E09.g	503
84	ScNAC88	SCJFRT1009A08.g	456
85	ScNAC89	SCCCLR1070C08.g	1558

Puranik et al., 2013; Shang et al., 2013). Even though we have extensive literature on NAC genes in many crops, surprisingly there are no reports on NAC genes of sugarcane crop. In this report, we present the results of phylogenetic characterization of sugarcane NAC genes, their conserved motifs, comparative analysis of NAC genes from sugarcane and its closest relatives. Furthermore, in this manuscript we emphasize on NAC genes that are implicated in plant abiotic stresses.

Table 2
Comparative analysis of sugarcane NAC with sorghum NAC proteins.

Sl. no	Sugarcane NAC genes	Sorghum NAC gene showing maximum similarity	Max identity (in %)	e-value	Bit Score	Query coverage (in %)
1	ScNAC79	SbNAC31	30.0	2.00E − 10	53.9	86
2	ScNAC11	SbNAC87	43	7.00E − 27	97.8	83
3	ScNAC72	SbNAC94	50	7.00E − 08	44.7	55
4	ScNAC18	SbNAC88	53	8.00E − 57	182	82
5	ScNAC64	SbNAC117	54	3.00E − 46	158	59
6	ScNAC78	SbNAC34	67	3.00E − 77	232	97
7	ScNAC46	SbNAC99	74	7.00E − 156	444	92
8	ScNAC17	SbNAC57	83	8.00E − 86	256	99
9	ScNAC30	SbNAC34	84	9.00E − 146	416	68
10	ScNAC35	SbNAC53	87	3.00E-124	354	98
11	ScNAC42	SbNAC92	88	0	1102	88
12	ScNAC16	SbNAC41	89	1.00E − 54	174	85
13	ScNAC86	SbNAC91	88	5.00E − 148	423	86
14	ScNAC47	SbNAC15	88	0	1239	93
15	ScNAC28	SbNAC36	88	4.00E − 55	175	87
16	ScNAC59	SbNAC17	89	3.00E − 142	402	99
17	ScNAC74	SbNAC102	90	5.00E − 149	420	76
18	ScNAC21	SbNAC92	90	9.00E − 138	400	99
19	ScNAC04	SbNAC15	90	7.00E − 120	353	95
20	ScNAC01	SbNAC65	91	5.00E − 74	223	100
21	ScNAC34	SbNAC47	92	0	545	80
22	ScNAC61	SbNAC108	92	4.00E − 97	278	100
23	ScNAC58	SbNAC111	92	0	844	79
24	ScNAC82	SbNAC56	92	4.00E − 137	387	99
25	ScNAC70	SbNAC57	93	1.00E − 137	390	99
26	ScNAC03	SbNAC118	93	1.00E − 120	348	98
27	ScNAC57	SbNAC20	93	4.00E − 133	376	83
28	ScNAC83	SbNAC20	94	0	563	82
29	ScNAC56	SbNAC34	94	2.00E − 139	393	88
30	ScNAC43	SbNAC111	94	2.00E − 120	347	100
31	ScNAC22	SbNAC111	94	0	653	89
32	ScNAC05	SbNAC111	95	8.00E − 126	360	100
33	ScNAC84	SbNAC96	96	0	1242	94
34	ScNAC39	SbNAC118	95	0	531	95
35	ScNAC38	SbNAC49	95	0	566	88
36	ScNAC27	SbNAC4	97	0	628	85
37	ScNAC29	SbNAC63	98	0	745	96
38	ScNAC85	SbNAC4	98	7.00E − 128	363	77
39	ScNAC23	SbNAC111	98	2.00E − 125	359	99
40	ScNAC09	SbNAC41	99	2.00E − 113	331	88
41	ScNAC25	SbNAC41	100	6.00E − 99	292	100