

Supplementary Table 1. Overview of 82 secondary metabolite gene clusters of FvRN1, FvW15A1, and three reference strains

Chr. no. (No. of clusters)	BGC ^a	Metabolite	Signature ^b	Cluster size (kb)	<i>F. vorosii</i> ^c				<i>F. asiaticum</i> ^c
					FvRN1	FvW15A1	CBS 119177	CBS 119178	KCTC 16664
Chr. 1 (26)	C01			26.6	+	+	ψ	ψ	+
	C77	Apicidin	NPS31	50.6	–	–	–	–	+
	C02	Gramillin	NPS8	73.8	+	+	+	+	+
	C03			12.9	+	+	+	+	+
	C04			7.5	+	+	+	+	+
	C05		STC2	22.7	+	+	+	+	+
	C06		NPS16, NPS19	38.9	+	+	+	+	+
	C07		TPS, NPS-like	52.6	+	+	+	+	+
	C08		TPS, PKS11	37.0	+	+	+	+	+
	C09			22.7	+	+	+	+	+
	C10			10.7	+	+	+	+	+
	C78	W-493	NPS32, PKS40	82.7	+	+	+	+	+
	C11		NPS-like	10.1	+	+	+	+	+
	C12			55.6	+	+	+	+	+
	C70	Fusahexin	NPS4	22.9	+	+	+	+	+
	C13	Aurofusarin	PKS12	27.4	+	+	+	+	+
	C14			80.9	+	+	+	+	+
	C15	Zearalenone	NPS15, PKS4, PKS13	36.7	+	+	+	+	+
	C58		TPS	32.9	+	+	+	+	+
	C59	Culmorin	STC9	37.9	+	+	+	+	+
	C60	Fusarielin	NPS-like, PKS9	22.1	+	+	+	+	+
	C71		NPS3	40.7	+	+	+	+	+
	C61		NPS18, PKS1	49.4	+	+	+	+	+
	C62		NPS-like, PKS-like, TPS-like	30.3	–	–	–	–	+
	C89		PKS-like		–	+	–	+	–
	C87		PKS-like	29.2	+	+	–	+	–
Chr. 2 (29)	C51			78.1	+	+	+	+	+
	C50			10.2	+	+	+	+	+
	C49	Butenolide		17.9	+	+	+	+	+
	C48	Tricinonoic acid	STC5, NPS-like	41.6	+	+	+	+	+
	C76		PKS52	33.0	+	+	+	+	+
	C47	Fusaristatin A	NPS7, PKS6	28.9	+	+	+	+	+
	C46		NPS-like	11.4	+	+	+	+	+
	C79		T3PKS	41.5	+	+	+	+	+
	C45			11.4	+	+	+	+	+
	C44		PKS7	45.1	+	+	+	+	+
	C43			6.3	+	+	+	+	+
	C30			11.9	+	+	+	+	+
	C29		NPS-like	33.2	+	+	+	+	+
	C72		NPS11		+	+	+	+	+

(Continued)

Supplementary Table 1. Continued

Chr. no. (No. of clusters)	BGC ^a	Metabolite	Signature ^b	Cluster size (kb)	<i>F. vorosii</i> ^c				<i>F. asiaticum</i> ^c
					FvRN1	FvW15A1	CBS 119177	CBS 119178	KCTC 16664
	C28	Carotenoids	DTC1	9.2	+	+	+	+	+
	C27		NPS-like	8.8	+	+	+	+	+
	C26	Gibepyrone	PKS8	42.9	+	+	+	+	+
	C25			16.2	+	+	+	+	+
	C24	Fusagramineol	STC4	20.3	+	+	+	+	+
	C23	Trichothecene	TPS	18.9	+	+	+	+	+
	C22		NPS-like	29.4	+	+	+	+	+
	C21	Triacetylfusa- rinine	NPS6	17.3	+	+	+	+	+
	C20			19.4	+	+	+	+	+
	C19			45.1	+	+	+	+	+
	C18	Orcinol	PKS14	55.7	+	+	+	+	+
	C17			21.5	+	+	+	+	+
	C16		PKS15, TPS	23.0	+	+	+	+	+
	C80		CDPS	20.8	+	+	+	+	+
	C67			20.3	–	–	–	–	ψ
Chr. 3 (11)	C32			12.8	ψ	ψ	–	ψ	ψ
	C33	Ferricrocin	NPS2	43.3	+	+	+	+	+
	C34		PKS5	14.5	+	+	+	+	+
	C35		NPS-like	32.2	+	+	+	+	+
	C65		TPS	29.4	+	+	+	+	+
	C64	Fusaotaxin	NPS5, NPS9	54.1	+	+	+	+	+
	C63	Malonichrome	NPS1	24.3	+	+	+	+	+
	C83		TPS	18.1	+	+	+	+	+
	C73		NPS12		+	+	+	+	+
	C69	Koraiol	STC6	21.7	+	+	+	+	+
	C66	Chrysogine	NPS14	33.1	+	+	+	+	+
Chr. 4 (16)	C36		TPS	16.5	+	+	+	+	+
	C37		NPS, NPS-like	39.4	+	+	+	+	+
	C38		NPS10	41.4	+	+	+	+	+
	C39			18.6	+	+	+	+	+
	C68		PKS-like		+	+	+	+	+
	C74		NPS14		+	+	+	+	+
	C40			39.8	+	+	+	+	+
	C85		NPS-like	32.4	+	+	+	+	+
	C41		STC1	6.7	+	+	+	+	+
	C42	Fusarin	PKS10	33.0	+	+	+	+	+
	C57			14.3	+	+	+	+	+
	C56			19.8	+	+	+	+	+
	C55			35.8	+	+	+	+	+
	C54			18.5	+	+	+	+	+

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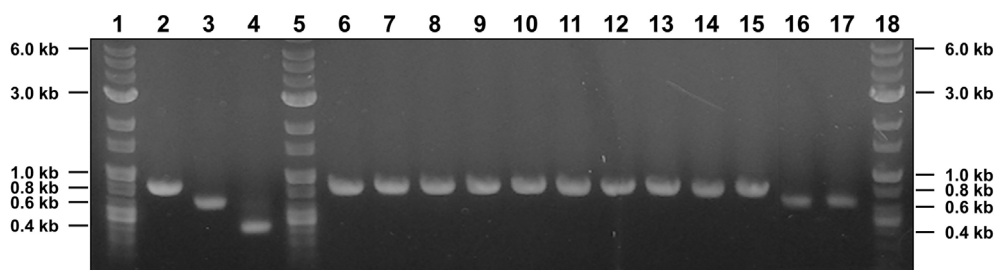
Supplementary Table 1. Continued

Chr. no. (No. of clusters)	BGC ^a	Metabolite	Signature ^b	Cluster size (kb)	<i>F. vorosii</i> ^c				<i>F. asiaticum</i> ^c
					FvRN1	FvW15A1	CBS 119177	CBS 119178	KCTC 16664
	C53	Bostryocoidins	PKS3	36.4	+	+	+	+	+
	C52			69.1	+	+	+	+	+
			Total		78	79	76	79	80

^aBGC: Biosynthetic Gene Cluster; Cluster designations C01 to C76 were previously described (Sieber et al., 2014; Adpressa et al., 2019; Westphal et al., 2021), C77 to C88 are newly designated in our previous study (Jeong et al., 2023), and C89 is newly designated in this study.

^bKey enzymes were identified using antiSMASH v.6.0.1 and manually curated (Adpressa et al., 2019; Hansen et al., 2015). NPS, non-ribosomal peptide synthetase; PKS, polyketide synthase; T3PKS, type III polyketide synthase; CDPS, cyclodipeptide synthase; TPS, terpene synthase; STC6, stanniocalcin 6.

^c+ , genome of each strain includes this secondary metabolite gene cluster; – , genome of each strain does not include this secondary metabolite gene cluster; ψ , genome of each strain includes the cluster in which the key enzyme was pseudogenized or 1/3 of the clusters were deleted.



Supplementary Fig. 1. Agarose gel electrophoresis for amplified *TRII2* gene of Fv strains. Amplified *TRII2* gene of *Fusarium vorosii* (Fv) strains were separated using 1% agarose gel electrophoresis at 100 V for 25 min. The expected PCR products size for nivalenol (NIV), 15-acetyl-deoxynivalenol (15ADON), and 3-acetyl-deoxynivalenol (3ADON) type were about 840 bp, 670 bp, and 410 bp, respectively. Lanes 1, 5, and 18, 1 kb DNA ladder; lane 2, *F. asiaticum* KCTC 16664, positive control for NIV type; lane 3, *F. graminearum* KCTC 16659, positive control for 15ADON type; lane 4, *F. graminearum* KCTC 16661, positive control for 3ADON type; lane 6, *F. vorosii* WN031; lane 7, *F. vorosii* WN037; lane 8, *F. vorosii* WN038; lane 9, *F. vorosii* CN008; lane 10, *F. vorosii* CN009; lane 11, *F. vorosii* CN010; lane 12, *F. vorosii* CN011; lane 13, *F. vorosii* WN036; lane 14, *F. vorosii* RN1; lane 15, *F. vorosii* RN030; lane 16, *F. vorosii* W15A1; lane 17, *F. vorosii* B15A6. Lanes 6-15 are NIV type, and lanes 16 and 17 are 15ADON type.

References

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