Assembly of *Norana najaformis*

Table 1: Basic statistics of *Norana najaformis* assemblies, presenting the strategies that were used for each assembly (purging haplotigs, assembly polishing, scaffolding), assembly size, number of contigs, N50, number of BUSCO single complete features and BUSCO duplicate complete features. The tool purge_dups v1.0.1 is designated as PD.

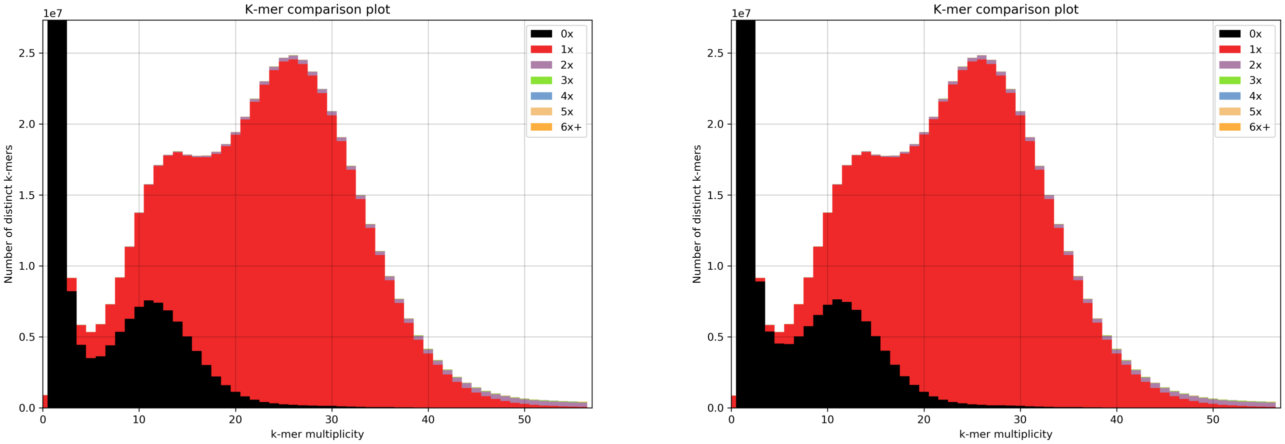
Assembler	**Purging**	**Scaffolding**	**Gap filling**	**Polishing**	**Assembly size**	**# fragments**	**N50**	**Gaps**	**BUSCO**	
									single	**dup.**
Flye v2.8	-	-	-	-	822 Mb	12,176	521 kb	78	80.3%	15.6%
	PD	-	-	-	659 Mb	7,049	788 kb	252	91.8%	4.3%
	PD x2	-	-	-	611 Mb	1,860	860 kb	240	92.8%	3.2%
Flye v2.9	-	-	-	-	859 Mb	8,585	242 kb	0	63.5%	30.3%
Flye v2.9 --keep-haplotypes	-	-	-	-	876 Mb	16,893	105 kb	0	58.9%	33.3%
HiCanu v2.1	-	-	-	-	1.2 Gb	11,992	326 kb	0	25.7%	68.8%
	PD	-	-	-	654 Mb	2,488	1.1 Mb	428	92.0%	3.5%
	PD x2	-	-	-	629 Mb	1,540	1.1 Mb	391	91.8%	3.5%
	PD x2	instaGRAAL	-	-	630 Mb	490	35.6 Mb	1,299	92.0%	3.0%
	PD x2	instaGRAAL	TGS-GapCloser	-	630 Mb	490	35.6 Mb	1,032	92.1%	3.0%
	PD x2	instaGRAAL	TGS-GapCloser	HyPo	631 Mb	490	35.6 Mb	910	92.0%	3.0%
hifiasm v0.12 -l 2 p_ctg	-	-	-	-	660 Mb	1,057	2.6 Mb	0	90.4%	4.5%
	PD	-	-	-	628 Mb	711	2.6 Mb	64	91.8%	3.1%
hifiasm v0.12 -l 0 p_utg	-	-	-	-	1.1 Gb	8,500	213 kb	0	86.3%	8.7%
hifiasm v0.15 Hi-C -l 2 p_ctg	-	-	-	-	668 Mb	1,199	2.4 Mb	0	89.6%	5.1%
hifiasm v0.15 Hi-C -l 0 hap1 p_ctg	-	-	-	-	659 Mb	1,735	1.4 Mb	0	88.3%	5.9%
hifiasm v0.15 Hi-C -l 0 hap2 p_ctg	-	-	-	-	644 Mb	1,302	1.3 Mb	0	88.6%	6.2%
hifiasm v0.16 Hi-C -l 2 p_ctg	-	-	-	-	669 Mb	1,174	2.4 Mb	0	89.2%	5.6%
	-	instaGRAAL	-	-	669 Mb	1,842	36.9 Mb	702	91.0%	4.3%
	-	instaGRAAL	TGS-GapCloser	-	669 Mb	1,842	36.9 Mb	644	90.9%	4.4%
	-	instaGRAAL	TGS-GapCloser	HyPo	670 Mb	1,842	36.9 Mb	626	90.9%	4.5%
hifiasm v0.16 Hi-C -l 2 hap1 p_ctg	-	-	-	-	626 Mb	1,369	1.5 Mb	0	90.8%	3.9%
hifiasm v0.16 Hi-C -l 2 hap2 p_ctg	-	-	-	-	618 Mb	1,117	1.3 Mb	0	92.7%	3.0%
mdBG v0.1.0	-	-	-	-	1.2 Gb	1,010	1.6 Mb	0	54.8%	18.2%
NextDenovo v2.4	-	-	-	-	624 Mb	1,066	2.1 Mb	0	91.7%	4.4%
	PD	-	-	-	606 Mb	868	2.2 Mb	77	93.1%	3.0%
	PD	instaGRAAL	-	-	606 Mb	82	35.0 Mb	860	93.4%	2.6%
	PD	instaGRAAL	TGS-GapCloser	-	606 Mb	82	35.0 Mb	748	93.2%	2.8%
	PD	instaGRAAL	TGS-GapCloser	HyPo	613 Mb	82	35.3 Mb	632	93.3%	2.7%
Raven v1.5 --weaken	-	-	-	-	587 Mb	3,736	294 kb	0	91.0%	3.4%
wtdbg2 v2.5 -x ccs -g 600m	-	-	-	-	588 Mb	1,925	1.2 Mb	0	89.4%	3.5%
wtdbg2 v2.5 -x ccs -g 1200m	-	-	-	-	588 Mb	1,930	1.1 Mb	0	89.5%	3.5%

Three assemblies are designated then as:

- Nn1: HiCanu v2.1 + purge_dups x2 + instaGRAAL + TGS-GapCloser + HyPo
- Nn2: hifiasm v0.16 + instaGRAAL + TGS-GapCloser + HyPo
- Nn3: NextDenovo v2.4 + purge_dups + instaGRAAL + TGS-GapCloser + HyPo

Table 2: Final statistics of the 17 main scaffolds. rRNA sequences were detected with BARRNAP.

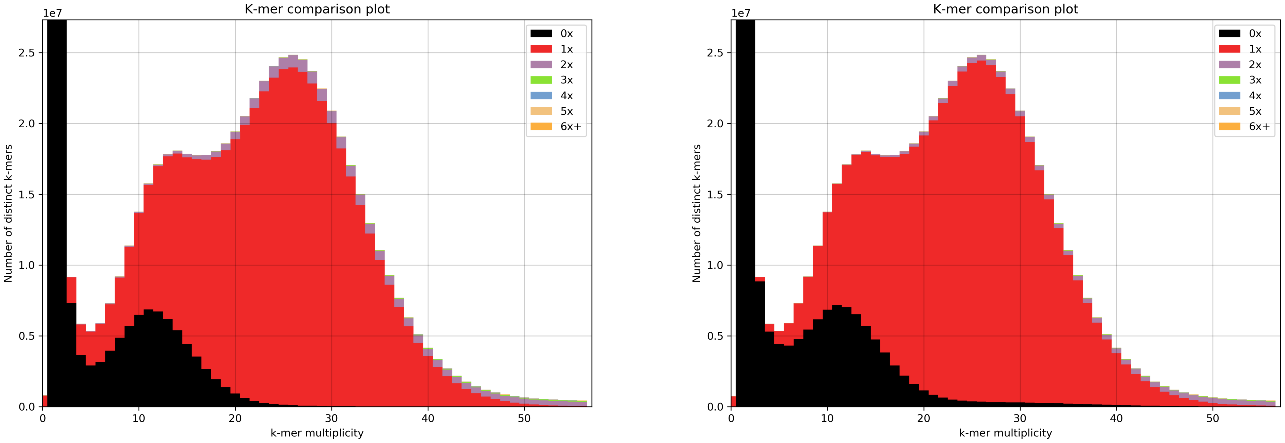
Assembly	**Assembly size**	**Scaffold sizes**	**HiFi mapping**	**BUSCO**		**rRNA hits**	
				single	**dup.**	**#**	**size**
Nn1	612 Mb	19.6-64.3 Mb	99.33%	93.0%	2.8%	253	44.3 kb
Nn2	613 Mb	19.9-64.0 Mb	98.93%	92.1%	2.9%	97	17.5 kb
Nn3	608 Mb	19.4-63.9 Mb	99.33%	93.6%	2.6%	70	19.6 kb



(a) All scaffolds.

(b) Main scaffolds.

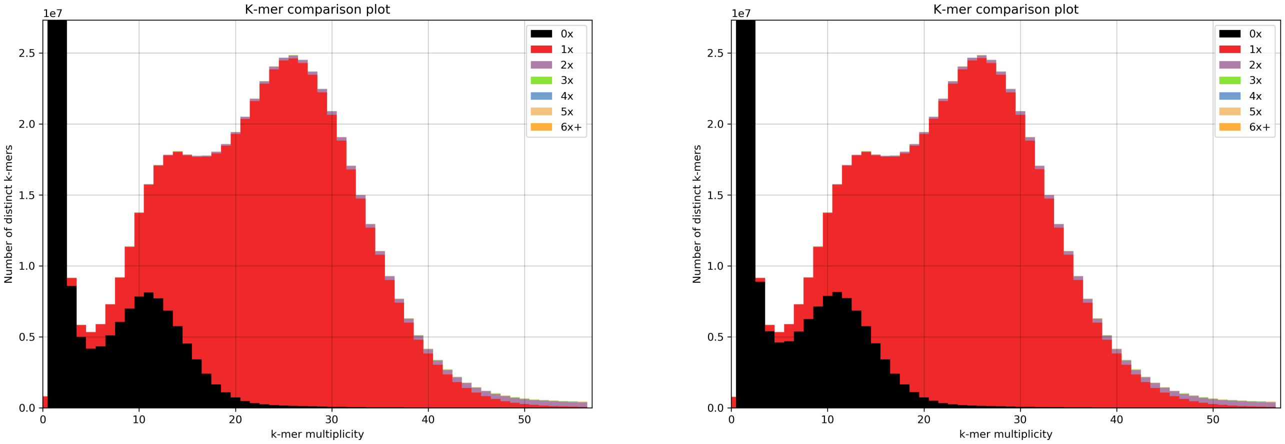
Figure 1: KAT plot of Nn1.



(a) All scaffolds.

(b) Main scaffolds.

Figure 2: KAT plot of Nn2.



(a) All scaffolds.

(b) Main scaffolds.

Figure 3: KAT plot of Nn3.

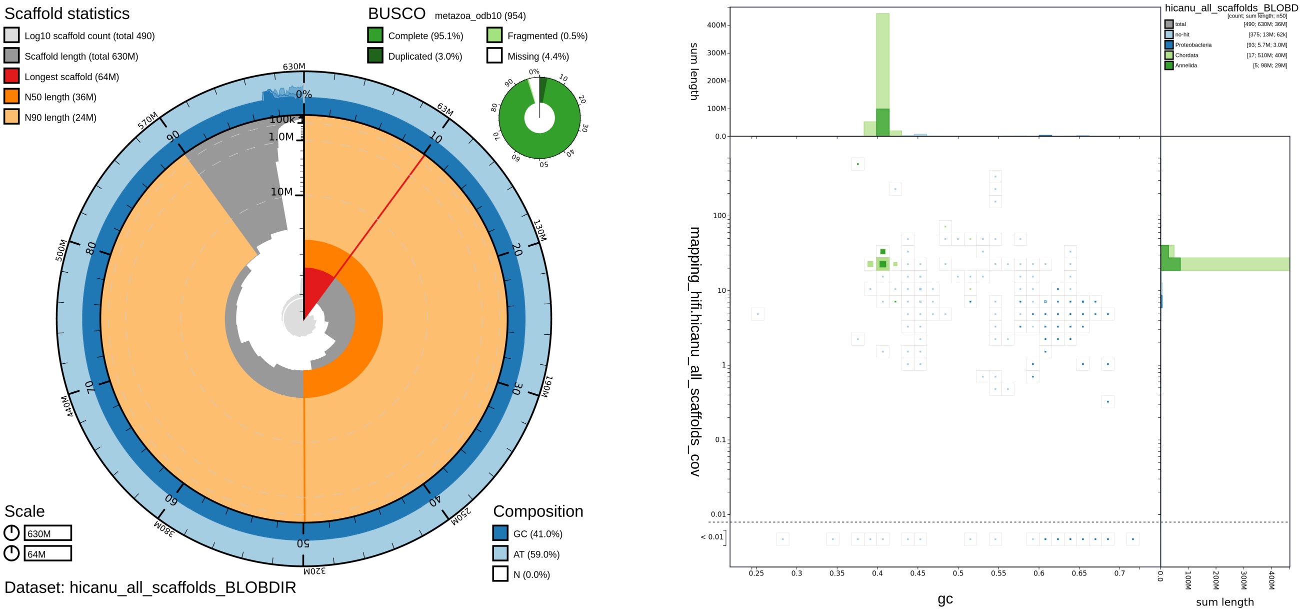


Figure 4: Blobtools of Nn1.

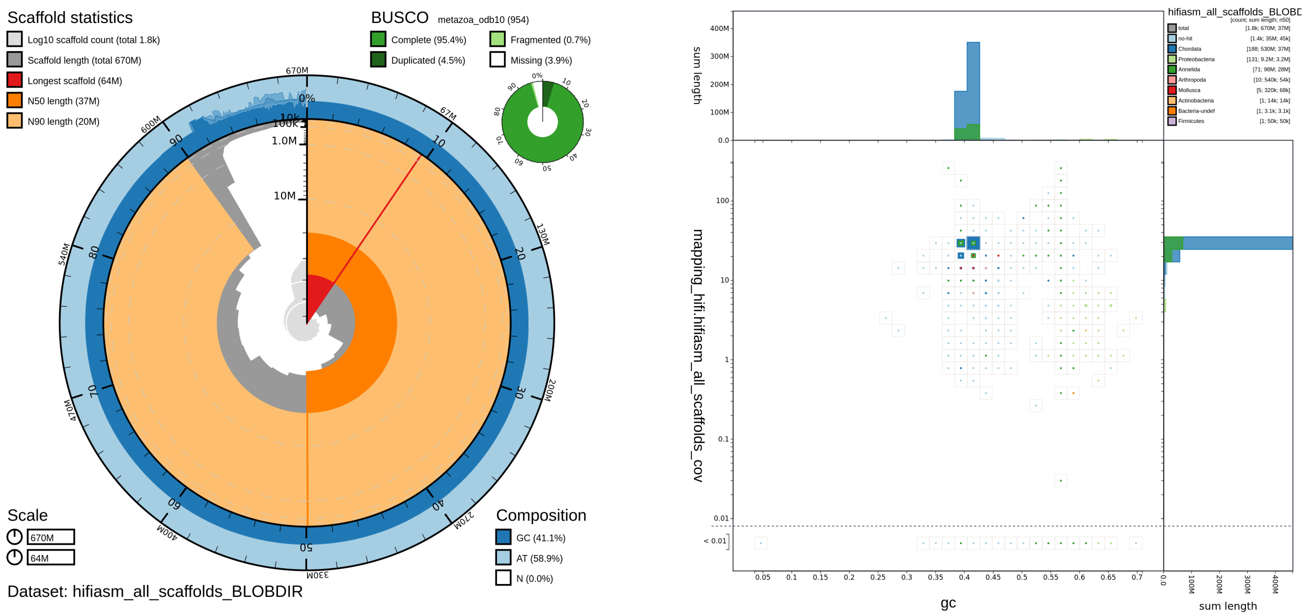


Figure 5: Blobtools of Nn2.

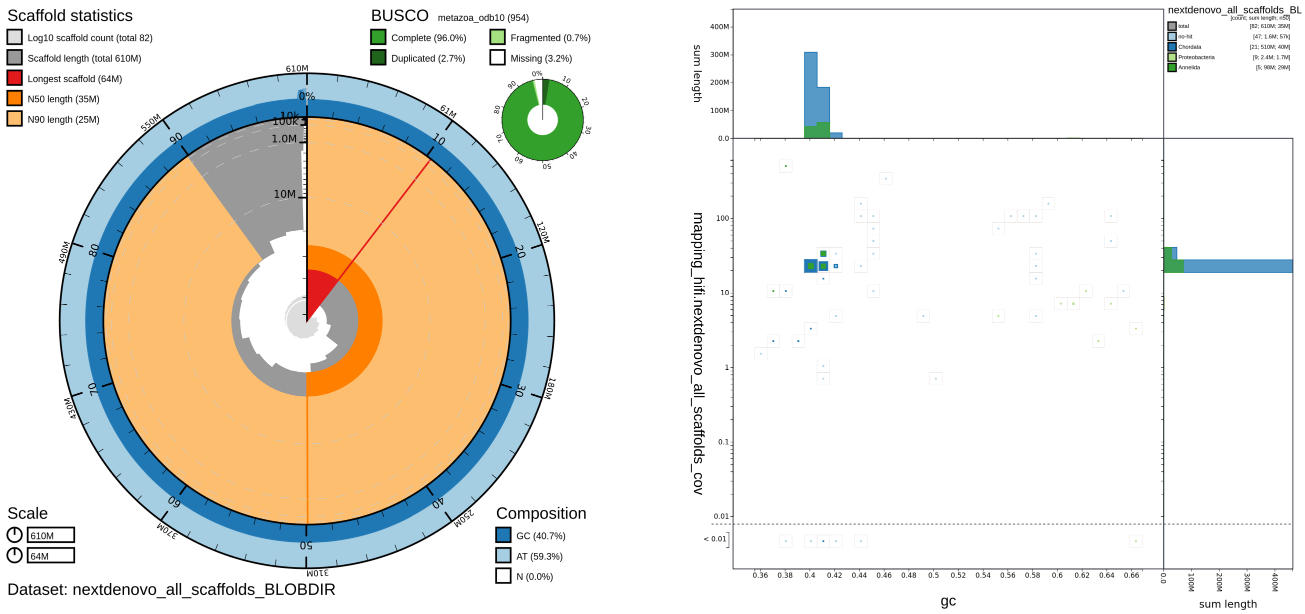


Figure 6: Blobtools of Nn3.

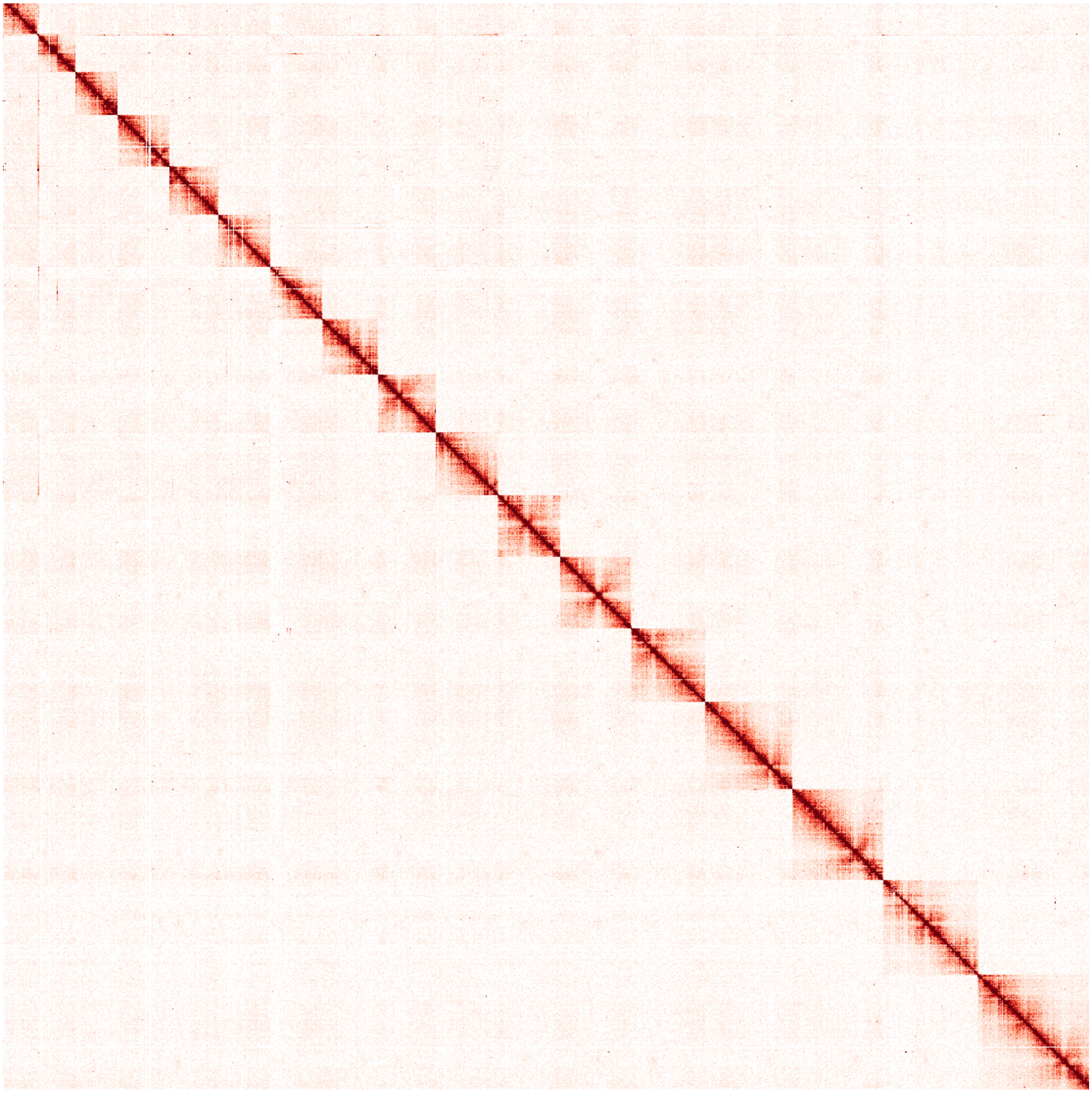


Figure 7: Hi-C contact map of Nn1 (binning = 1000).

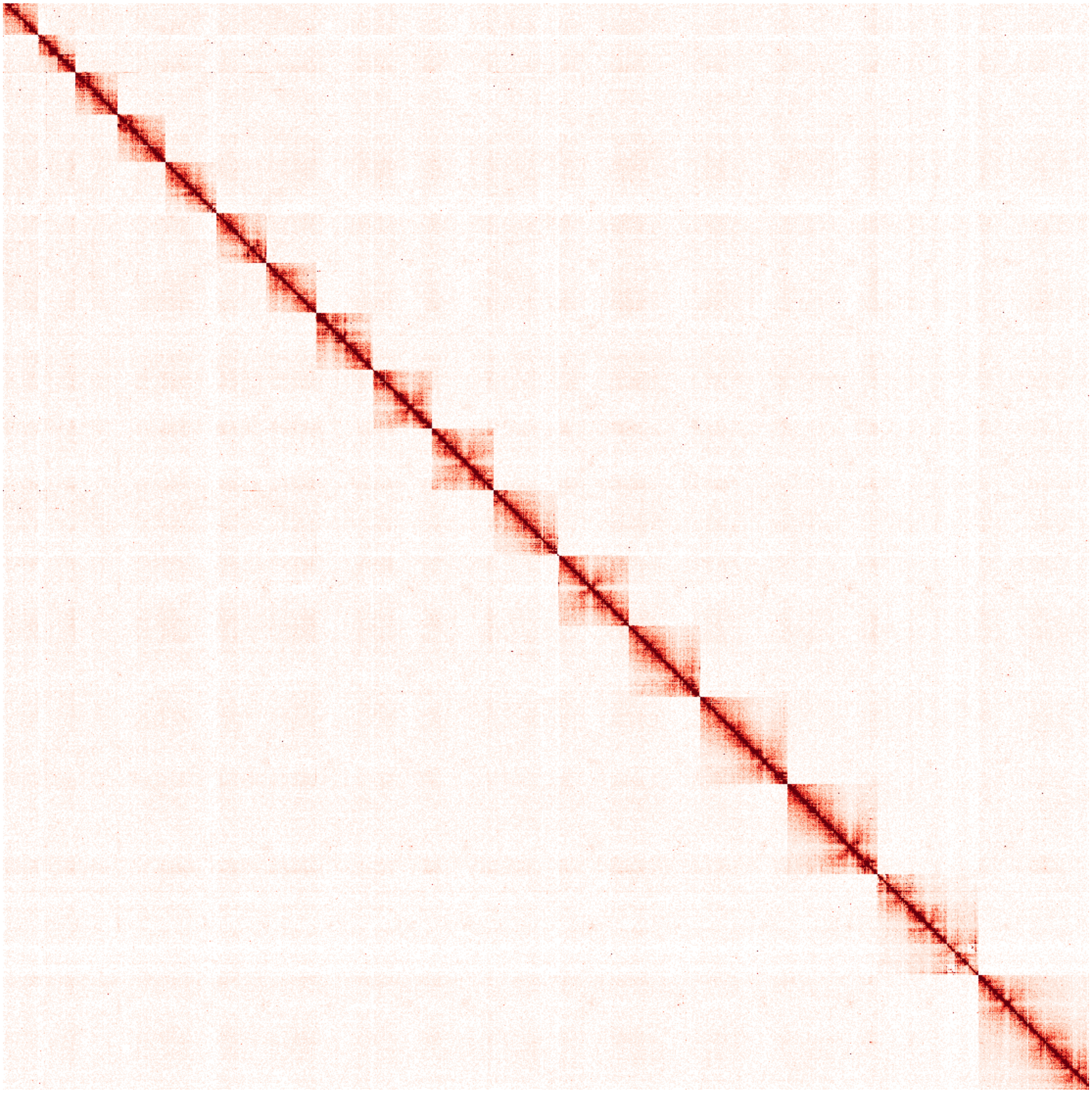


Figure 8: Hi-C contact map of Nn2 (binning = 1000).

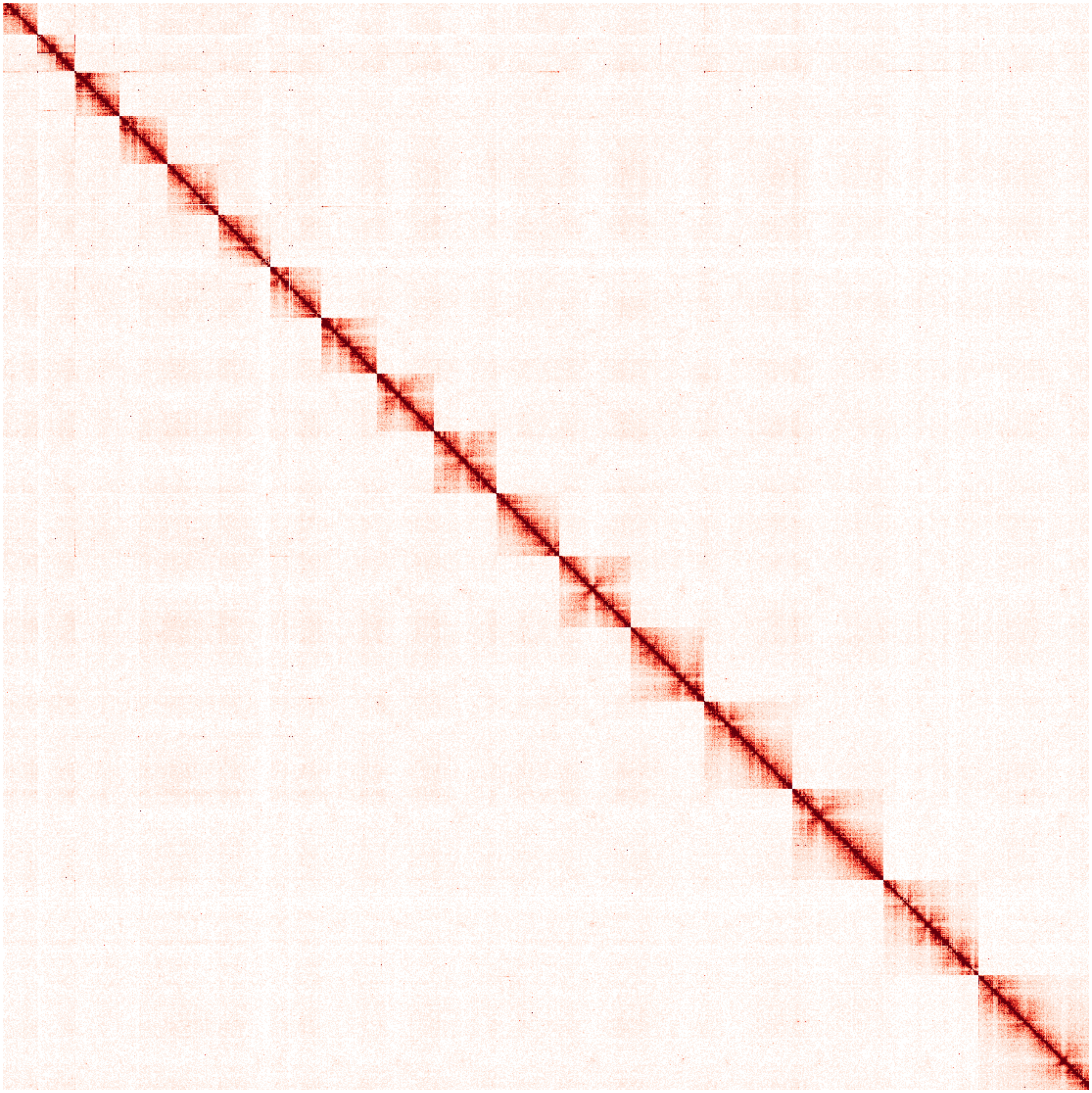


Figure 9: Hi-C contact map of Nn3 (binning = 1000).

Assembly of *Carpetania matritensis*

Table 1: Basic statistics of *Carpetania matritensis* assemblies, presenting the strategies that were used for each assembly (purging haplotigs, assembly polishing, scaffolding), assembly size, number of contigs, N50, number of BUSCO single complete features and BUSCO duplicate complete features. The tool purge_dups v1.0.1 is designated as PD. Polishing was performed either before scaffolding (BS) or after scaffolding (AS).

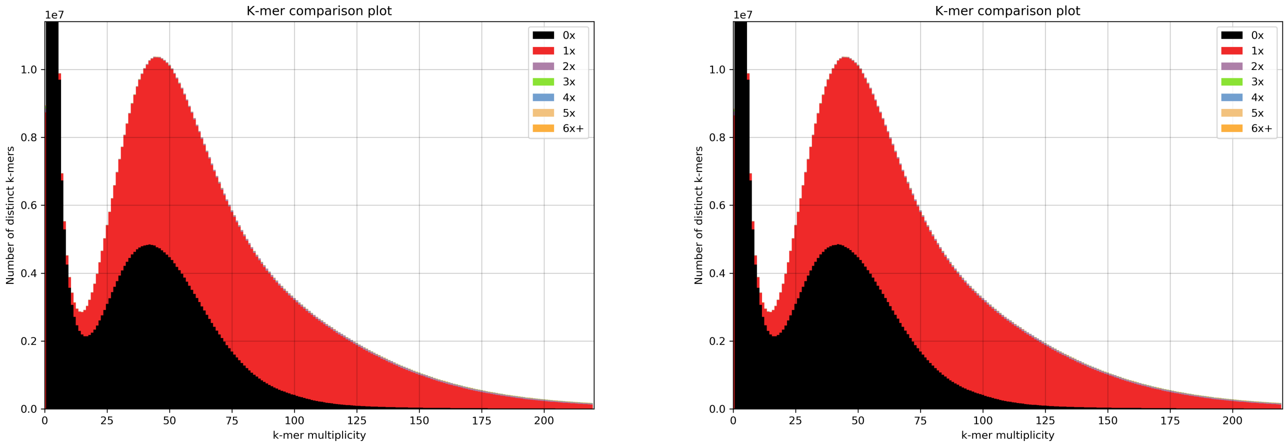
Assembler	**Reads correction**	**Purging**	**Polishing**	**Scaffolding**	**Gap filling**	**Assembly size**	**# contigs**	**N50**	**Gaps**	**BUSCO**	
										single	**dup.**
Canu v1.9	Canu	-	-	-	-	1.2 Gb	8,998	250 kb	0	26.4%	69.5%
	Canu	PD	-	-	-	602 Mb	2,885	403 kb	1,151	90.6%	3.8%
	Canu	PD x2	-	-	-	594 Mb	2,745	407 kb	1,092	90.9%	3.7%
Flye v2.8	-	-	-	-	-	792 Mb	16,565	266 kb	91	78.9%	16.5%
	-	PD	-	-	-	624 Mb	6,162	392 kb	190	91.4%	4.2%
	Canu	-	-	-	-	659 Mb	8,728	546 kb	39	90.7%	5.5%
	Canu	PD	-	-	-	632 Mb	6,064	573 kb	39	90.4%	5.7%
	MECAT2	-	-	-	-	662 Mb	9,107	534 kb	38	89.5%	6.6%
	MECAT2	PD	-	-	-	584 Mb	3,303	614 kb	100	92.1%	3.5%
MECAT2	MECAT2	-	-	-	-	765 Mb	5,896	325 kb	0	75.9%	8.0%
	MECAT2	PD	-	-	-	590 Mb	2,999	416 kb	435	79.8%	1.2%
NextDenovo v2.4	NextDenovo	-	-	-	-	604 Mb	1,309	906 kb	0	92.1%	3.2%
	NextDenovo	PD	-	-	-	588 Mb	1,168	933 kb	136	92.6%	2.7%
	NextDenovo	PD	HyPo (BS)	-	-	587 Mb	1,168	932 kb	114	92.2%	3.4%
	NextDenovo	PD	-	instaGRAAL	-	588 Mb	59	36.0 Mb	1,349	92.2%	2.6%
	NextDenovo	PD	-	instaGRAAL	TGS-GapCloser	590 Mb	59	36.1 Mb	609	92.0%	2.6%
	NextDenovo	PD	HyPo (AS)	instaGRAAL	TGS-GapCloser	589 Mb	59	36.1 Mb	519	92.9%	3.2%
Raven v1.5	-	-	-	-	-	658 Mb	2,620	568 kb	0	88.9%	5.0%
	-	PD	-	-	-	628 Mb	2,214	597 kb	468	90.3%	3.0%
	-	PD x2	-	-	-	621 Mb	2,144	600 kb	573	90.0%	3.0%
Shasta v0.7.0	-	-	-	-	-	740 Mb	17,768	85 kb	0	75.2%	10.7%
wtdbg2 v2.5 -g 600m	-	-	-	-	-	585 Mb	2,351	1.6 Mb	0	90.5%	2.4%
	Canu	-	-	-	-	564 Mb	2,265	943 kb	0	92.0%	2.5%
wtdbg2 v2.5 -g 1200m	-	-	-	-	-	575 Mb	1,762	2.2 Mb	0	89.6%	2.5%
	-	-	HyPo (BS)	-	-	576 Mb	1,762	2.2 Mb	0	92.7%	2.6%
	-	-	-	instaGRAAL	-	575 Mb	600	33.2 Mb	1,607	90.3%	2.3%
	-	-	-	instaGRAAL	TGS-GapCloser	578 Mb	600	33.3 Mb	1,032	90.1%	2.3%
	-	-	HyPo (AS)	instaGRAAL	TGS-GapCloser	578 Mb	600	33.3 Mb	1,024	92.7%	2.7%
	MECAT2	-	-	-	-	571 Mb	2,235	1.3 Mb	0	91.4%	2.8%
	Ratatosk	-	-	-	-	566 Mb	2,750	1.5 Mb	0	90.5%	2.6%

Two assemblies are then designated as:

- Cm1: NextDenovo v2.4 + purge_dups + instaGRAAL + TGS-GapCloser + HyPo
- Cm2: wtdbg2 v2.5 + instaGRAAL + TGS-GapCloser + HyPo

Table 2: Final statistics of the 17 main scaffolds. rRNA sequences were detected with BARRNAP.

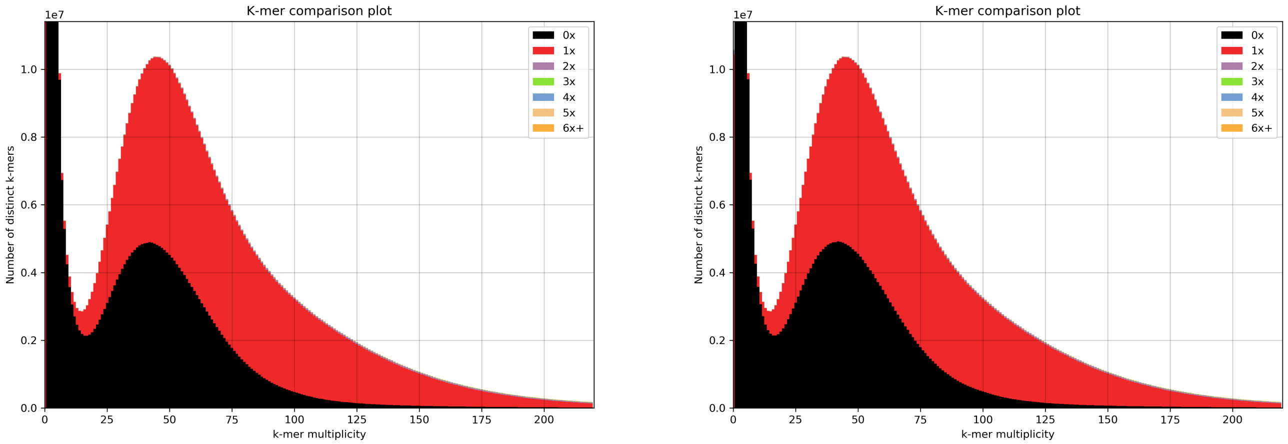
Assembly	**Assembly size**	**Scaffold sizes**	**HiFi mapping**	**BUSCO**		**rRNA hits**	
				single	**dup.**	#	**size**
Cm1	588 Mb	17.5-75.1 Mb	90.02%	92.8%	3.2%	32	32.6 kb
Cm2	573 Mb	17.0-65.4 Mb	90.13%	92.1%	2.7%	21	21.6 kb



(a) All scaffolds.

(b) Main scaffolds.

Figure 1: KAT plot of Cm1.



(a) All scaffolds.

(b) Main scaffolds.

Figure 2: KAT plot of Cm2.

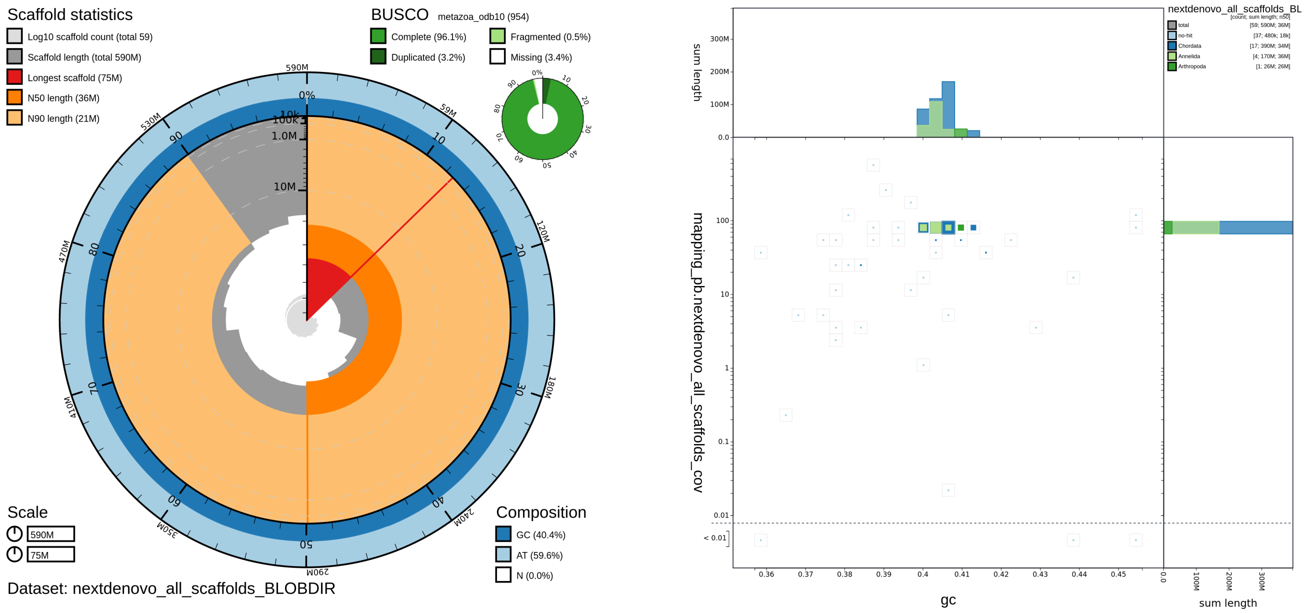


Figure 3: Blobtools of Cm1.

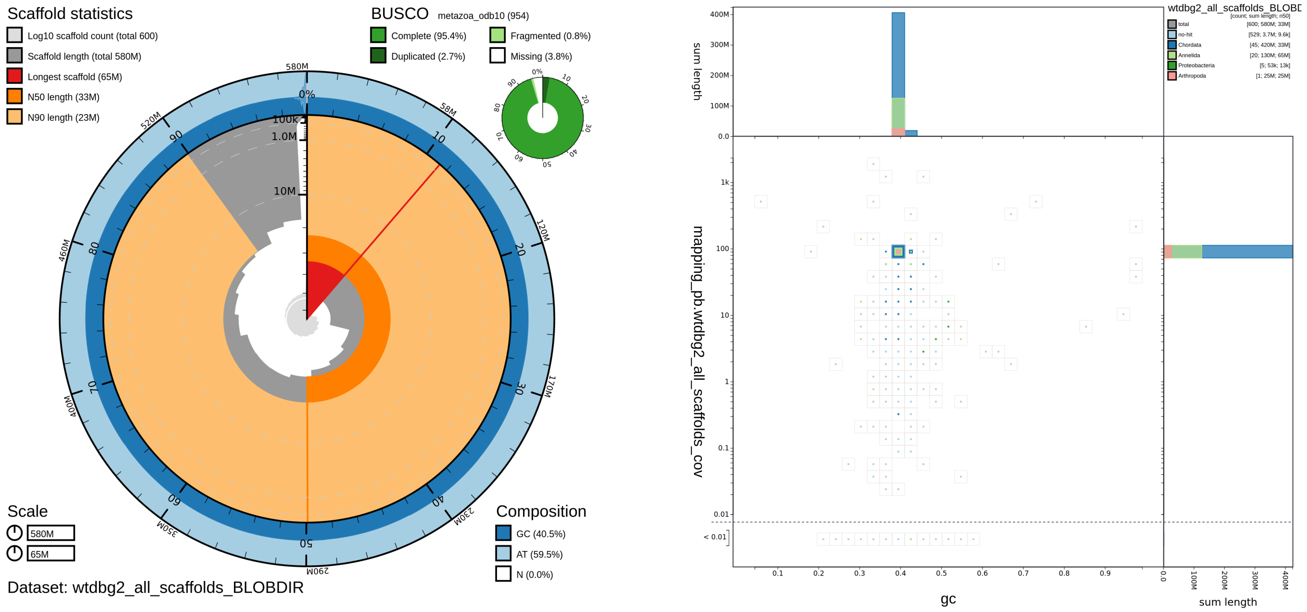


Figure 4: Blobtools of Cm2.

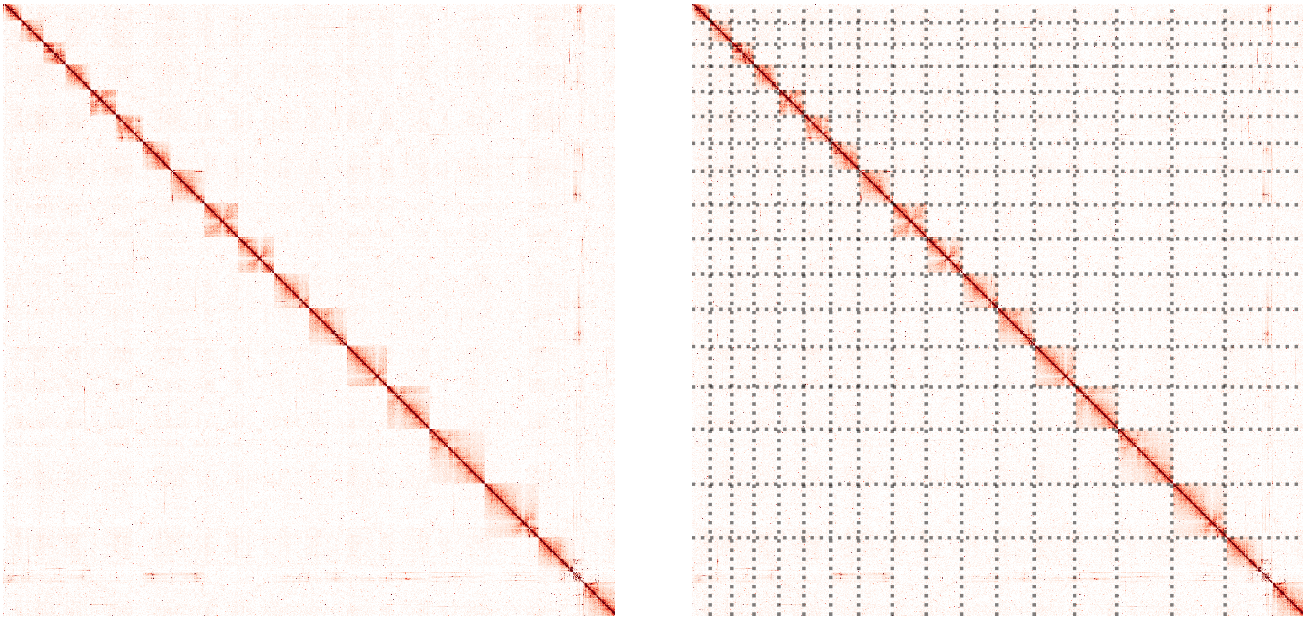


Figure 5: Hi-C contact map of Cm1 (binning = 1000).

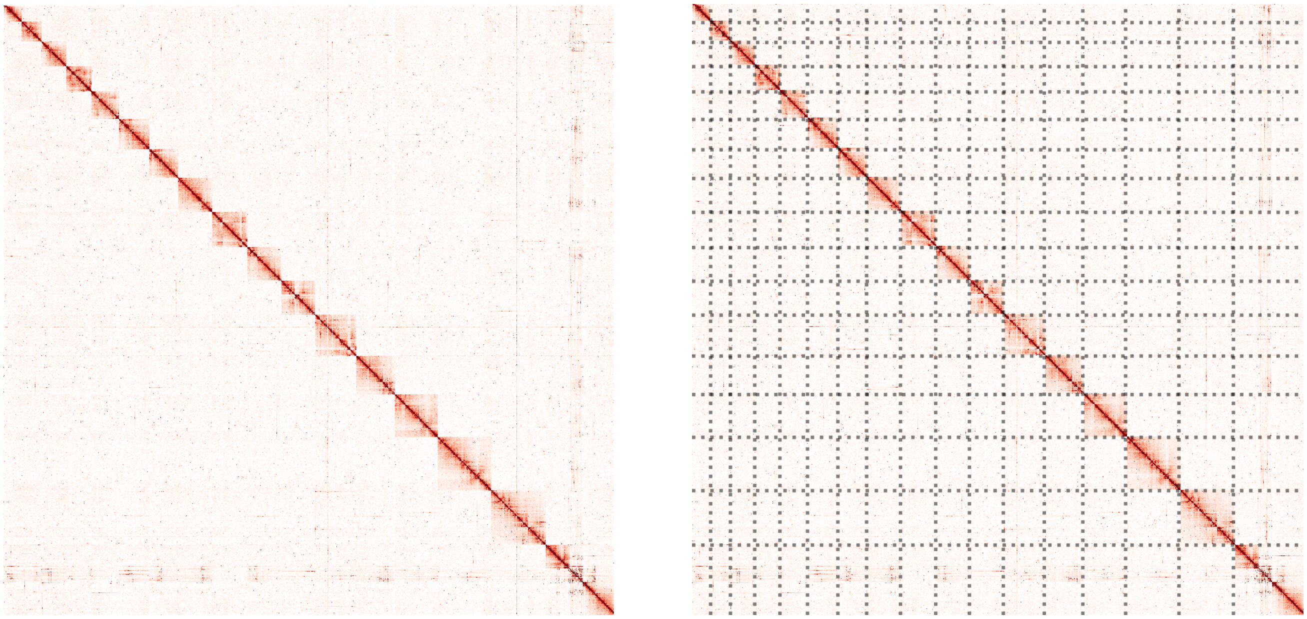


Figure 6: Hi-C contact map of Cm2 (binning = 1000).