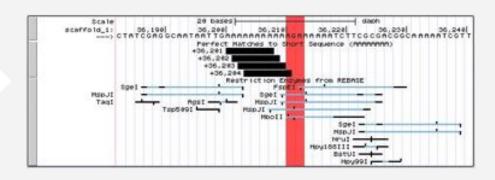
Making an Assembly Track Hub To Visualize New Genomes

```
>Scaffold_1
GTTGTAAATACTCTATTCTACAATAAAACCAA
TCATAGGTTGAATTGGCGTTGAAGTAAAACCAA
TCATAGGTTGAATTGGCGTTGAAGTAAAACAAA
...
>Scaffold_2
AGTTATGACAAACTATAAAAAGTCGGTAGAGACAAAAG
TCGTTCGTGGACGAAGCGACCAAAACTGAGCACAAGAT/
...
>Scaffold_3
CATAAATTCATAAATCAATTCATGAAGAATAATT
TAGAAAATTTCCCAGGAAGTTTGAAGTTGCTA
```

```
hubDirectory
|__hub.txt
|__genomes.txt
|__hg19
|__trackDb.txt
```



1. How Binary Indexed Files are used for Data Visualization

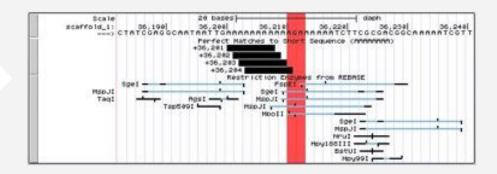
2. Building the Text Files for Track Hubs and additional Tracks

3. Track Hub Resources

Building a 2bit file: binary indexed FASTA

```
PSCOFFOLG_1
GTTGTAAATACTCTATTCTACAATAAAACCAA
TCATAGGTTGAAATTGGCGTTGAAGTAAAACCAA
TCATAGGTTGAATTGGCGTTGAAGTAAAACAAA
...
PSCOFFOLG_2
AGTTATGACAAACTATAAAAAAGTCGGTAGAGACAAAAG
TCGTTCGTGGACGAAGCGACCAAAACTGAGCACAAGATP
...
PSCOFFOLG_3
CATAAATTCATAAATCAATTCATGAAGAATAATT
TAGAAAATTCCCCAGGAAGTTTGAAGTTGCTA
```

```
hubDirectory
|__hub.txt
|__genomes.txt
|__hg19
|__trackDb.txt
```



A BAM file (.bam) is the binary version of a SAM file.

samtools view -S -b sample.sam > sample.bam



PRESLEY_0030:6:5:16900:343200/2 65 falhfafela_cfaddfcaffW_e6fabfcdfcf*ca	chr1	10847 Wa 197221	254 K*V2Ta9	67M95	*	0		CCTA	ACCCTAACCCTAACCC
PRESLEY_8030:6:26:1717:949800/2 65 f_fcdeffhfhaecffdff[ffdcf]ffcff]cb`bb	chrl	10053	254	61M155	*	0		CCTA	ACCCTAACCCTAACCCTAACCC
PRESLEY_8030:5:69:17883:15567#0/2 hgfgghhhhfhhhhhhhhhhhhghghhhhhhqhghhhh	65	chri	10050	255	2573MLS				AACTAACCCTAACCCTAA
PRESLEY_8030:5:75:7248:15014#0/2 hthfeghhethhhitthghhdghcaginhhhitchhfh	65	chrt	10060	255	2573415			0	AACTAACCCTAACCCTAA
PRESLEY_803815110111059613385#8/2 NAMADANANANANANANANANANANANANANANANANANA	65	chri	10050	255	2573915	*			AACTAACCCTAACCCTAA
PRESLEY_8030:7:79:7804:15262#0/2 hithshingflightightighthinhinhinh dhif	65	chrt	10050	255	2573415	+		0	AACTAACCCTAACCCTAA
PRESLEY_8030:7:91:10770:19201#0/2 http://doi.org/10.10770:19201#0/2	65	chri	10050	255	2572925				AACTAACCCTAACCCTAA
PRESLEY_8038:7:101:18643:21267#0/2 Nhifthininghighininghtglinhitgglinhitfaghini	65	chri	10060	255	2573415			0	AACTAACCCTAACCCTAA
brianleeghgwdev -]\$ head -n 40 temp5 PRESLEY 0030:7:02:16832:9421#0/2		chrt	10042	255	20403				CTANCCCTANCCCTANCC
PRESLEY_8030:6:7:18371:16699#0/2	111000				400.000				TAACCCTAACCCTAACCC
Yfffhhgehghhghgghhghhhg_ffffchfehgfff	thatchttt	fl facece	fiffblel	ebT3\^	.geY		1		
PRESLEY_0030:6:5:16900:3432#0/2 65	chet	10847	254	67/495		0	- 0	CCTA	ACCCTAACCCTAACCCTAACCC



The resulting binary file sample.bam (with an additional accompanied index file sample.bam.bai) can have data more easily extracted and can also be viewed in Genome Browsers.

faToTwoBit input.fasta output.2bit



>scaffold_1
GTTGTAAATACTCTATTCTACAATAAAACCAAAAGATAACTCTTTATCAG
TCATAGGTTGAATTGGCGTTGAAGTAAAACCAAAAGATAACTGCTCAAAAGG
...
>scaffold_2
AGTTATGACAAACTATAAAAAGTCGGTAGAGACAAAAGCGAAAAGGATCT
TCGTTCGTGGACGAAGCGACCAAAACTGAGCACAAGATAAATCCCGAATA
...
>scaffold_3
CATAAAATTCATAAAATCAATTCATGAAGAATAATTTTAGAAAAATGGTTCA
TAGAAAATTCCCAGGAAGTTTGAAGTTGCTATAATGATTATTTCTCTTG
...
>scaffold_4

TGCTTAAGTTGGTCATCACTGACTGAGTGAGCCCAAACTGGAATAGCTG1

...

The resulting indexed binary file output.2bit can have data more easily extracted and can also be viewed in the UCSC Genome Browser.

Itilities are downloadable tools available on Linux and UNIX platforms.



Itilities are downloadable tools available on Linux and UNIX platforms.



Itilities are downloadable tools available on Linux and UNIX platforms.

```
faFilter
farilters
faFrag
                         09-Jan-2018 15:55
falloise
                         09-Jan-2018 15:55
[a0neRecord
                         09-Jan-2018 15:55
faPolyASizon
                         09-Jan-2018 15:55
faRandomize
                         09-Jan-2018 15:55
faRc
                         09-Jan-2018 15:55
faSize
                         09-Jan-2018 15:55
faSomeRecords
                         09-Jan-2018 15:55
faSplit
faToFastq
Ea/To/Tab
                         09-Jan-2018 15:56
                         09-Jan-2018 15:56
INTrang
                         09-Jan-2018 15:56
                         09-Jan-2018 15:55
fastg7oFa
featureBits
```

faToTwoBit input.fasta output.2bit



Scaffold_1
GTTGTAAATACTCTATTCTACAATAAAACCAAAAGATAACTCTTTATCAG
TCATAGGTTGAATTGGCGTTGAAGTAAAACCAAAAGATAACTCTTTATCAG
TCATAGGTTGAATTGGCGTTGAAGTAAAACCAAAAAATACTGCTCAAAAGG
...
Scaffold_2
AGTTATGACAAACTATAAAAAGTCGGTAGAGACAAAAGCGAAAAGGATCT
TCGTTCGTGGACGAAGCGACCAAAACTGAGCACAAGATAAATCCCGAATA
...
Scaffold_3
CATAAATTCATAAATCAATTCATGAAGAATAATTTTAGAAAAATGGTTCA
TAGAAAATTTCCCAGGAAGTTTGAAGTTGCTATAATGATTATTTCTCTTG
...
Scaffold_4
ATACTGCCCATGCCTCATCTACTCCCTTCTTAACCTTTTCATCAACAACT
TGCTTAAGTTGGTCATCACTGACTGAGTGAGCCCAAACTGGAATAGCTGT

...

Host **output.2bit** at location that accepts byte-range requests (like CyVerse).

asmBly.2bit files exist for download via **Genome**Data link for all assemblies on the UCSC Browser



asmBly.2bit files exist for download via **Genome**Data link for all assemblies on the UCSC Browser

Sequence and Annotation Downloads

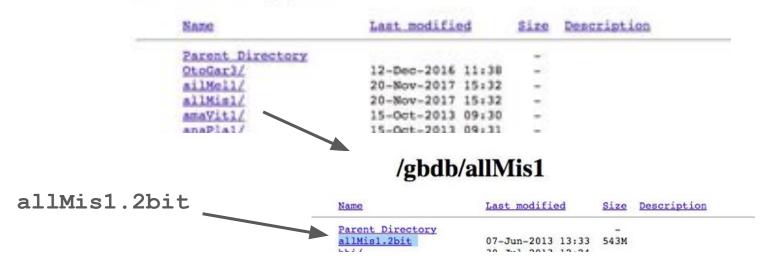
This page contains links to sequence and annotation data d

.....

Other downloads

Access the gbdb fileserver

Index of /gbdb



http://hgdownload.soe.ucsc.edu/gbdb/allMis1/allMis1.2bit

Extracting a specific window location of data from a BAM and 2bit file

```
samtools view http://location of/file.bam
                                                                     "chr1:1499900-1500055" >
output.sam
                                                PRESLEY 0030:6:5:16900:3432#0/2 65
                                                                                                      CCTAACCCTAACCCTAACCC
                                                falhfafela cfaddfcaffW edfabfcdfcf^cace^c\d\aaWaJYZZZ\\K^VZTaB
                                                PRESLEY 0030:6:26:1717:9490#0/2 65
                                                                      chr1
                                                                                                      CCTAACCCTAACCCTAACCC
                                                AACTAACCCTAACCCTAA
                                                PRESLEY 0030:5:75:7248:15014#0/2
                                                                                                          AACTAACCCTAACCCTAA
                                                hfhfgghhafhhhhffhghhdghcagfhhhhfchhfhcfhcqcedfaff hdfeeeheacc[Rchhh]egbd[bb
                                                PRESLEY 0030:5:101:10596:3305#0/2
                                                                      65
                                                                           chr1 10060 255
                                                                                                          AACTAACCCTAACCCTAA
```

```
twoBitToFa -seq=chr1 -start=1499900 -end=1500055
```

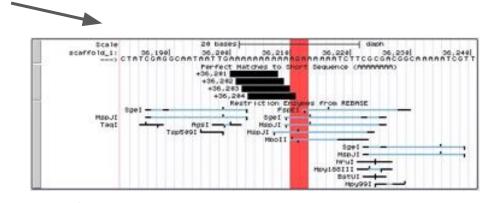
twoBitPath http://yourGenome/your.2bit output.fa



>chr1:1499900-1500055
GCTACCATCACCCAAAAAGCTGAGGAGTTTGAATTCACTTCAGCACAACT
ATCATTAATTAATTTTTGAACCTCTGAGCCTGGAAGAGAAAACAGGTTTG
GTTCAACATGAAGAATACTGTGATTTGACCCGTGACAGAGCTTTCTGTTA

Assembly Track Hubs

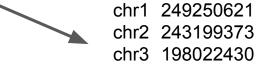
bigDataUrl http://location_of/file.bam



twoBitPath http://yourGenome/your.2bit

Extracting data from a the 2bit for later use: your2bit.chrom.sizes

twoBitInfo your.2bit your2bit.chrom.sizes



..

chr16 90354753 chr17 81195210

. . . .

chr21_gl000210_random 27682 chrUn_gl000231 27386 chrUn_gl000229 19913 chrM 16571

Using your2bit.chrom.sizes to make an artificial Cytoband

Track (banding options gneg, gpos25,... programmatic steps could be done manually) for

later: yourCytoBandIdeo.bed (BED4+1)



chr1 249250621 chr2 243199373 chr3 198022430

chr16 90354753 chr17 81195210

chrUn_gl000231 27386 chrUn_gl000229 19913 chrM 16571 chr1 0 249250621 chr1 gneg chr2 0 243199373 chr2 gneg chr3 0 198022430 chr3 gneg

chr16 0 90354753 chr16 gneg chr17 0 81195210 chr17 gneg

chrUn_gl000231 0 27386 chrUn_gl000231 gneg chrUn_gl000229 0 19913 chrUn_gl000229 gneg chrM 0 16571 chrM gneg

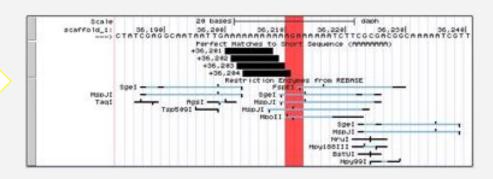
Turning yourCytoBandldeo.bed text file into a binary indexed version: yourCytoBandldeo.bigBed

bedToBigBed -type=bed4 yourCytoBandIdeo.bed -as=cytoBand.as your2bit.chrom.sizes yourCytoBandIdeo.bigBed table cytoBandIdeo chr1 0 249250621 chr1 gneg "cytoBandIdeo describes...." chr2 0 243199373 chr2 gneg Host file.bigBed chr3 0 198022430 chr3 gneg string chrom; "Reference.... " at location that uint chromStart; "Start..." chr16 0 90354753 chr16 gneg accepts uint chromEnd; "End..." chr17 0 81195210 chr17 gneg "Name of item" string name; byte-range chrUn_gl000231 0 27386 chrUn_gl000231 gnegstring gieStain; "Giemsa stain..". requests (like chrUn gl000229 0 19913 chrUn gl000229 gneg) chrM 0 16571 chrM gneg CyVerse).

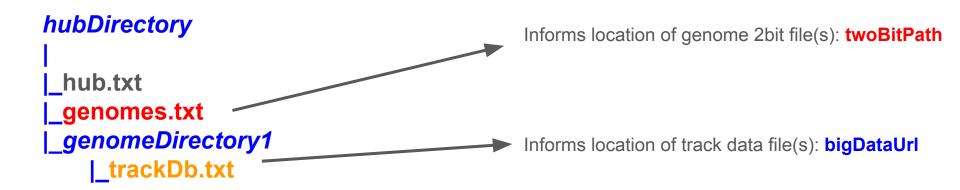
Building the Hub Text (.txt) Files

>Scaffold_1
GTTGTAAATACTCTATTCTACAATAAAACCAA
TCATAGGTTGAATTGGCGTTGAAGTAAAACCAA
TCATAGGTTGAATTGGCGTTGAAGTAAAACAAA
>Scaffold_2
AGTTATGACAAACTATAAAAAGTCGGTAGAGACAAAAGA
TCGTTCGTGGACGAAGCGACCAAAACTGAGCACAAGAT
>Scaffold_3
CATAAATTCATAAATCAATTCATGAAGAATAATT
TAGAAAATTTCCCAGGAAGTTTGAAGTTGCTA

hubDirectory
|_hub.txt
|_genomes.txt
|_hg19
|_trackDb.txt



Track Hubs are text files (hub.txt, genomes.txt, trackDb.txt) that describe and point to the location of binary indexed data files (twoBitPath, bigDataUrl)



Track Hubs are text files (hub.txt, genomes.txt, trackDb.txt) that describe and point to the location of binary indexed data files (twoBitPath, bigDataUrl)

hub.txt

hub MyHubsNameWithoutSpaces
shortLabel My Hub's Name
longLabel Longer label about my hub.
email myEmail@address
genomesFile http://location_of/genomes.txt

genomes.txt

genome yourGenome
trackDb http://location_of/trackDb.txt
twoBitPath http://location_of/output.2bit

trackDb.txt

track cytoBandIdeo
type bigBed
longLabel Chromosome ideogram with cytogenetic bands
shortLabel cytoBandIdeo
bigDataUrl http://location_of/yourCytoBandIdeo.bigBed

track bam1
type bam
shortLabel BAM example
longLabel A BAM file for my genome data
visibility dense
bigDataUrl http://location_of/file.bam

Each new track gets a new stanza

bigDataUrl lines point to location of binary indexed data

More trackDb parameters can be defined too (color, ect.)

Track Hubs are text files (hub.txt, genomes.txt, trackDb.txt) that describe and point to the location of binary indexed data files (twoBitPath, bigDataUrl)

twoBitPath http://location of/output.2bit

genomes.txt shares where to find the 2bit (and what to call new genome)

genome yourGenome

trackDb http://location_of/trackDb.txt

scientificName Your Genome description Feb. 2017 Assembly organism Your organism

defaultPos chr1:1000000-2000000

groups groups.txt

htmlPath http://yourGenome/description.html twoBitPath http://location_of/output.2bit Genomic Window of Sequence viewed at UCSC

Three tracks are generated from the 2bit on the fly:

- Base Position
- Restriction Enzymes
- Short Match Track (30 bases)

Track Hubs are text files (hub.txt, genomes.txt, trackDb.txt) that describe and point to the location of binary indexed data files (twoBitPath, bigDataUrl)

groups groups.txt



genomes.txt shares where to groups.txt to label the track groups

genome yourGenome

trackDb http://location_of/trackDb.txt

scientificName Your Genome description Feb. 2017 Assembly

organism Your organism

defaultPos chr1:1000000-2000000

groups groups.txt

htmlPath http://yourGenome/description.html

twoBitPath http://location_of/output.2bit



Very small file labels and priority ordering

name map label Mapping priority 1 defaultIsClosed 0

name genes label Genes priority 2 defaultIsClosed 0

Viewing the 2bit at UCSC

Browser requests AGCTs for only the genomic window currently viewing



Viewing the 2bit at UCSC

Browser requests *AGCTs* for only the genomic window currently viewing



QuickStart Guide to Assembly Hubs

http://genome.ucsc.edu/goldenPath/help/hubQuickStartAssembly.html

Quick Start Guide to Assembly Hubs

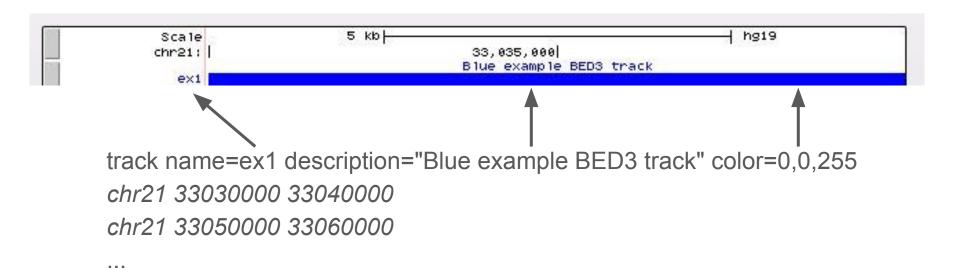
Assembly Hubs allow researchers to create Track Data Hubs on assemblies that are not in the UCSC Browser. By including the underlying reference sequence in UCSC twoBit format, as well as data tracks, researchers can browse and annotate any genome. For more information please refer to the Assembly Hub Wiki. Below is also a section about starting GBiB Assembly Hubs.

STEP 1: In a publicly-accessible directory, copy this Arabidopsis thaliana plant assembly hub, which includes an araTha1.2bit file, using the following wget command:

```
wget -r --no-parent --reject "index.html*" -nH --cut-dirs=3
http://genome.ucsc.edu/goldenPath/help/examples/hubExamples/hubAssembly/plantAraThal/
```

Adding Annotations

simple as adding text-based custom tracks



Adding tracks inside Track Hubs

create binary indexed versions of text files (bedToBigBed)

bedToBigBed in.bed chrom.sizes out.bb



The resulting binary file **out.bb** (bigBed) can be hosted online:

chr1 33050000 33060000 chr1 33050000 33060000

...

chr21 33030000 33040000 chr21 33050000 33060000

. . .

chr22 33030000 33040000 chr22 33050000 33060000

...

chr1 248956422

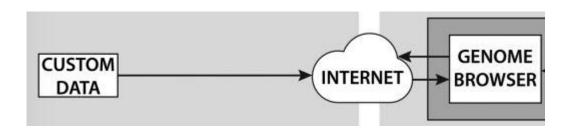
. . .

chr21 46709983 chr22 50818468

. . .

bigDataUrl=http://path.lab.edu/to/out.bb

The serving of files requires byte-range requests, which allows only portions of file to transfer. Some locations, like Dropbox, prevent such requests as people can then watch videos from their servers.



bigDataUrl http://path.lab.edu/to/out.bb

The trackDb.txt shares descriptions and where to find binary indexed tracks

track ex1bb type bigBed shortLabel ex1 longLabel Blue example BED3 track color 0,0,255 bigDataUrl http://path.lab.edu/to/out.bb

track track2
bigDataUrl http://path.lab.edu/to/out2.bb

View Data Tracks at UCSC

A wide variety of track binary indexed formats exist:

- BAM, CRAM, VCF
- bigGenePred, bigBarChart
- bigPsl, bigChain, bigMaf,
- bigNarrowPeak, halSnake
- bigBed, bigWig

Additional Track Types In Hubs Example

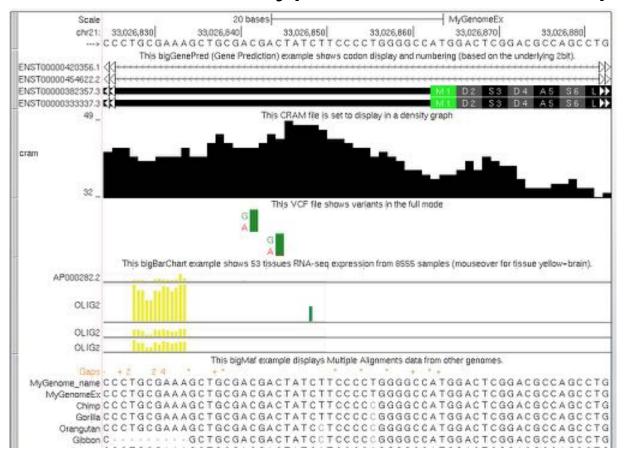
bigGenePred

CRAM

VCF

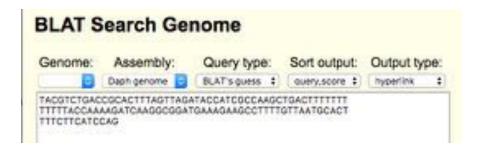
bigBarChart

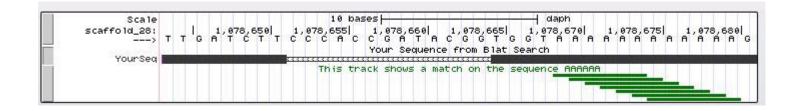
bigMaf



Additional Tools Available on Assembly Hubs

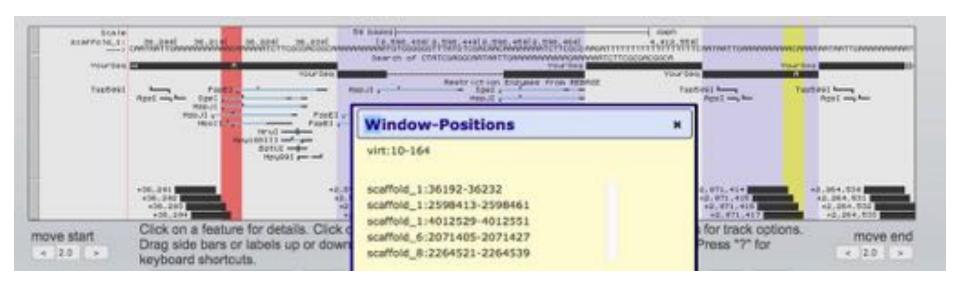
BLAT search sequence or amino-acid matched





Additional Tools Available on Assembly Hubs

- BLAT search sequence or amino-acid match
- Multi-region: simultaneously view non-adjacent regions across chromosomes
- Ability to annotate variants in added VCFs against added bigGenePred tracks



Assembly Track Hub Summary:

Remotely Hosted Data

Binary indexed files: Text files: hub.txt,

twoBitPath,

bigDataUrl

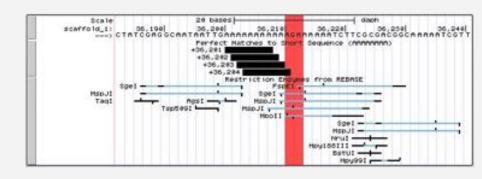
>SCATTOLG_1
GTTGTAAATACTCTATTCTACAATAAAACCAA
TCATAGGTTGAATTGGCGTTGAAGTAAAACCAA
...
>SCATTOLG_2
AGTTATGACAAACTATAAAAAGTCGGTAGAGACAAAAGA
TCGTTCGTGGACGAAGCGACCAAAACTGAGCACAAGAT
>SCATTOLG_3
CATAAATTCATAAAATCAATTCATGAAGAATAATT
TAGAAAATTTCCCAGGAAGTTTGAAGTTGCTAY

hubDirectory
|_hub.txt
|_genomes.txt
|_hg19
|_trackDb.txt

genomes.txt,

trackDb.txt

Visualizable at UCSC



Examples of Hubs and Resources

Genbank Assembly Hubs

http://genome-test.soe.ucsc.edu/gbdb/hubs/genbank/

hubDirectory |_hub.txt |_genomes.txt

trackDb.txt

Ensembl Track Hub Registry

https://trackhubregistry.org/

UCSC Public Hubs

http://genome.ucsc.edu/cgi-bin/hgHubConnect

G-OnRamp

http://gonramp.wustl.edu/

CyVerse Discovery Environment byte-range supported data hosting https://de.cyverse.org/de/

Existing Working Assembly Hub Templates

Genbank Assembly Hubs

http://genome-test.soe.ucsc.edu/gbdb/hubs/genbank/

94 94 8,548,615 19 0,643,657 45 4,577,423 14	2,835,004	30,159	4-
mblies: tal contig ount 4 v 94 8,548,615 19 0,643,657 49 4,577,423	2,835,004 93,684,015,605	contig size	assembly size
mblies: tal contig ount 4 v 94 8,548,615 19 0,643,657 49 4,577,423	2,835,004 93,684,015,605	contig size	assembly size
94 8,548,615 19,0643,657 4,577,423	2,835,004 93,684,015,605	contig size	assembly size
94 8,548,615 19 0,643,657 45 4,577,423 14	2,835,004 93,684,015,605	contig size	assembly size
8,548,615 19 0,643,657 49 4,577,423 14	93,684,015,605		945,00
0,643,657 49 4,577,423 14	and the same of th	40 444	
4,577,423 14	00 004 450 500	10,441	1,126,069,85
The second second	98,264,459,566	16,259	2,442,472,84
	45,341,422,954	4,203	540,302,68
3,939,128	16,816,724,183	4,269	49,753,62
2,264,511 17	70,439,035,382	5,282	346,420,80
4,143,097 3	38,677,096,556	9,335	31,833,00
57,569	2,010,246,046	34,918	2,709,22
8,397,216 23	34,147,691,500	27,883	3,991,53
2,571,310 1,29	99,383,526,796	9,801	20,925,39
8 2	,264,511 1 ,143,097 57,569 ,397,216 2 ,571,310 1,20	,264,511 170,439,035,382 ,143,097 38,677,096,556 57,569 2,010,246,046 ,397,216 234,147,691,500	264,511 170,439,035,382 5,282 ,143,097 38,677,096,556 9,335 57,569 2,010,246,046 34,918 ,397,216 234,147,691,500 27,883 ,571,310 1,299,383,526,796 9,801

Existing Working Assembly Hub Templates

Genbank Assembly Hubs

http://genome-test.soe.ucsc.edu/gbdb/hubs/genbank/

Scroll Down to see assemblies... Click common name to load hub.

The "Taxon ID" link will go to the Entrez taxonomy for that ID.

The "common name" link will go to the UCSC genome browser for that assembly.

The "biosample" link will go to the Entrez biosample for that ID.

The "accession" link will go to the Entrez assembly for that ID.

The "assembly" link will go to the NCBI Genbank FTP source directory.

	Taxon ID a=	date a~	common name +	scientific name av	biosample	count av	genome size x-	NS0 size a-	GC percent	unknown bases count/percent	gene count bases percent **	accession A+
1	9785	15 Jul 2009	African bush elephons	Loxodonta africana	SAMN02953622	2,352	3,196,738,035	46,401,353	% 39.76	78,195,493 % 2.45		GCA_000001905
1	9646	15 Dec 2009	Aluropoda melanoleuca	Alluropoda melanoleuca	SAMN00008160	81,466	2,299,492,210	1,281,781	% 40.62	54,196,184 % 2.36	52,206 473,410,926 % 20.59	GCA_000004335.
1	37293	16 Mar 2015	Actus nancymeas	Actus nancymeae	SAMN03121886	29,222	2,926,565,220	8,280,397	% 35.21	391,667,293 % 13.38	% 0.00	GCA_000952055.
1	9555	05 Jun 2012	baboon	Papio anubis	SAMN02981400	63,250	2,948,380,710	139,646,187	% 40.20	55,130,419 % 1.87		GCA_000264685
1	118797	31 Jul 2013	beij	Lipotes vexitifer	SAMN02953859	30,712	2,429,195,737	2,419,148	% 40.75	32,195,939 % 1.33	% 0.00	GCA_000442215.

Existing Working Assembly Hub Templates

Genbank Assembly Hubs

http://genome-test.soe.ucsc.edu/gbdb/hubs/genbank/

1. Right-click and copy and paste a link to explore the hub's architecture.



2. Visit just the hub.ncbi.txt and then the genomes.ncbi.txt and then look at a specific GCA...trackDb.ncbi.txt

Ensembl Track Hub Registry

https://trackhubregistry.org/

The Track Hub Registry

A global centralised collection of publicly accessible track hubs

The goal of the Track Hub Registry is to allow third parties to advertise track hubs, and to make it easier for researchers around the world to discover and use track hubs containing different types of genomic research data.

mouse epigenomics

Q

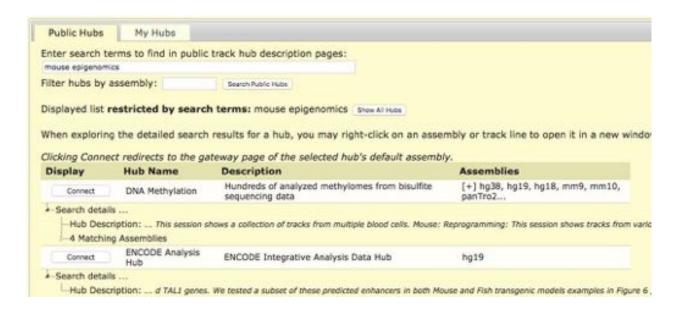
Ensembl Track Hub Registry -Link out from Gramene

https://trackhubregistry.org/



UCSC Public Hubs moderated collection of track hubs

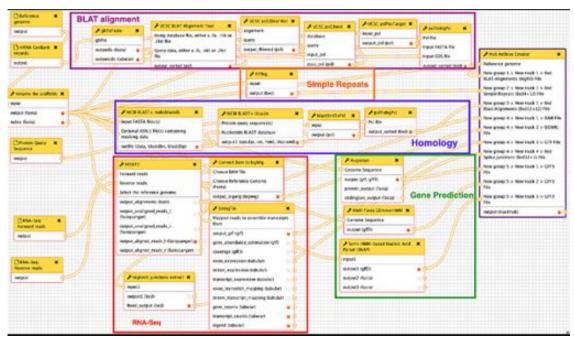
http://genome.ucsc.edu/cgi-bin/hgHubConnect



G-OnRamp

Galaxy workflow turning data like RNA-Seq into Assembly Hubs

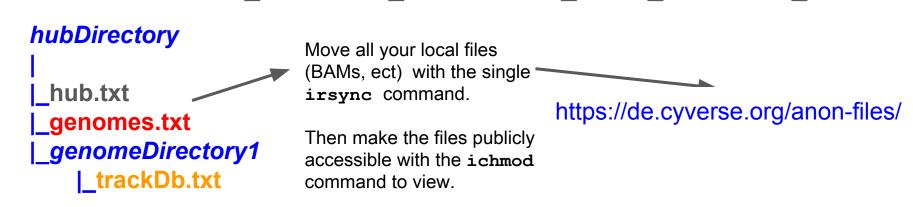
http://gonramp.wustl.edu/



CyVerse Discovery Environment Storage Solution

CyVerse iCommands (rysnc becomes irsync) allows transfer of 2-100GB files and recursive hub directory structures with commands like irsync:

```
$ irsync -r local_directory_name i:data_store_directory_name
```



\$ ichmod read anonymous data_store_directory_name



Thank You!



