Annotation

- Associating a Spreadsheet with an Annotation File
- **Building an Annotation File**

Associating a Spreadsheet with an Annotation File

For Partek Genomics Suite to recognize an annotation spreadsheet, it must meet several requirements. First, there must be a column header row in the annotation file. Second, there must be a column in the annotation file that matches the identifiers in your data spreadsheet. Third, any text field above the column header row must start with #. Fourth, the text fields must be tab or comma delimited.

We will illustrate associating a spreadsheet with an annotation file using an imported .txt data file from an Illumina HumanHT-12 v4.0 Gene Expression BeadChip array and the HumanHT-12 v4.0 Whole-Genome Manifest File (TXT Format) from Illumina.

Open the annotation file with a text editor such as Notepad++/WordPad/TextEdit (Microsoft Excel is not recommended to edit text files, for instance when used default settings, it converts gene names to dates and floating-point numbers)

Microsoft Excel is not recommended for viewing text files because on default settings it converts some gene names to dates and others to floating-point numbers

- · Verify that a column in the annotation file matches the identifier in your data spreadsheet, e.g probe ID, the identifier must be unique to each row
- Remove the text before the first column header (Figure 1) or add # to each text box
- ٠ Save the annotation file as a .txt file



this annotation file in Partek Genomics Suite, we delete any rows prior to the column headers row.

Right-click the spreadsheet you want to annotate in the spreadsheet tree panel, select Properties from the pop-up menu (Figure 2) or select Prop erties from the File menu on the main toolbar

Partek Genomics Suite - 1 (GSM1823575)	8H_1.txt)		-		×
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			Workflows Choose		~
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1 (GSM1823575 8H 1.txt)	Current Selec	tion ILMN_17	762337		^
Revert to	Last Saved State	2.			
Save As		L 1762337 60			
Clone		2055271 78	84		-
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	5. ILMIN	1806310 7 4	4.4		-
	6. ILMN	1779670 6 9	9.8		-
	7. ILMN	1653355 7 4	4.6		-
	8. ILMN	1717783 51	1.6		
	9. ILMN	1705025 62	2		-
	10. ILMN	N_1814316 70	0.8		
	11. ILMN	N_2359168 6 4	4		-
	12. ILMN	N_1731507 48	8.1		
	13. ILMN	N_1787689 69	9.2		
	14. ILMN	N_3241953 88	8		
	15. ILMN	1745607 55	5.1		
	16. ILMN	N_2136495 5 4	4.6		
	17. ILMN	V_1668111 56	6.7		
	18. ILMN	1_2295559 68	8.8		
	19. ILMN	N_1735045 12	20.7		_
	20. ILMN	N_1680754 89	9.4		
	21. ILMN	N_2375184 5 9	9		_
	✓ 22. ILMN	1659452 91	1.2		
<	> Rows: 47322	Columns: 2	<	>	×

Figure 11. Changing the spreadsheet properties

Depending on how you imported the data, you may see a *Configure Spreadsheet* dialog (Figure 3). Select the most appropriate option for your data; here we have chosen **Genomic microarray**.

🤣 Configure Spreadsheet 1 🛛 🗙
Choose the category which best describes the data in this spreadsheet.
Genomic microarray List of genomic region (eg: .bed file) Result of copy number segmentation List of samples in bam files High throughput screening Other
Close

Figure 12. The Configure Spreadsheet dialog may appear depending on how you imported your data

The Configure Genomic Properties dialog will now open.

• Select the appropriate option for Choose the type of genomic data; here we have chosen Gene Expression (Figure 4).

Ø Configure Genomic Properties of 1		×
Choose the type of genomic data		
Gene Expression		
Location of genomic features in spreadsheet		
Gene symbol instead of Marker ID		
C Feature in column label		
Feature in column 1. ID_REF ✓		
Choose chips/references and annotation files		
Chip/Reference Annotation file		
×	Browse	Download
		Add
Annotation column with gene symbols or microRNA names Set Column:		
Species and Genome Build		
Other ~		~
Advanced	Cancel	ОК

Figure 13. Selecting the type of genomic data

• Select the appropriate options for Location of genomic features in spreadsheet

Selecting *Gene Symbol instead of Marker ID* allows biological interpretation tasks like GO Enrichment or Pathway Enrichment to be performed without an annotation file because the gene symbol can be used to look up the gene set or pathway database.

Location of genomic features in spreadsheet allows you to specify whether genomic features (e.g. genes, miRNAs, probes, SNPs, CpGs etc) are represented by columns or rows. For *Feature in column label*, each feature is on a column, each row is a sample. For *Feature in column*, each feature is on a row and the feature ID for each feature is located in the column chosen with the drop-down menu.

Choose chips/reference and annotation files allows you to specify an annotation file to associate with the spreadsheet.

- · Select Browse... from Choose chips/references and annotation files
- Select your annotation spreadsheet file using the file selection interface

If the genomic position information from the annotation file cannot be automatically parsed, the *Configure Annotation* dialog will launch. This dialog allows you to choose which columns in the annotation file give the identity and genomic location of the features in your data spreadsheet. There are four options depending on if and how chromosome coordinates are described in the annotation file.

 Select the appropriate option for your annotation file; we have selected Chromosome is in one column and the physical position is in another column (eg: chr1, 100 or chr1, 100-200)

The Choose the columns section displays the annotation file spreadsheet with options to choose which columns are the Marker ID, Chromosome, and Physi cal Position (Figure 4).

- · Select the column that matches the feature IDs in your data spreadsheet for Marker ID; we have chosen Probe_Id for Marker ID.
- Select the column(s) that matches the chromosome location data; we have chosen Chromosome for Chromosome and Probe_Coordinates for P hysical Position.
- Select Close to return to the Configure Genomic Properties

An index file for the genomic location data of the annotation file is generated in the same folder as the annotation file; it has the same file name as the annotation file, but the file extension .idx. If you need to re-configure the genomic location field in the annotation file, first manually delete the .idx file and re-do the above steps to generate a new index file for the annotation file.

🧐 Configure A	nnotation					2	×
Partek was unable to locate genomic positions within the annotation file. Specify the columns that contain the genomic locations of markers.							
	inni configurati						
O The chromos	ome and coordi	nates are in one c	olumn (eg:	chr1:100-20	0)		
Chromosome	e is in one colum	nn and the physic	al position is	s in another	column (eg: chr1,100 or chr	1,100-20	00)
O Chromosome	e, start, and stop	are in separate co	olumns (eg:	chr1,100,2	00)		
 The annotation 	on file does not (contain genomic	coordinates				
Choose the colu	Imns	-					
Marker ID	۲	0	0	0	0	0	^
Chromosome	0	0	0	0	0	۲	
Physical Position	õ	õ	õ	õ	õ	Õ	
	t Probe Id	Arrav Address Id	Probe Type	Probe Start	Probe Sequence	Chromo	
	ILMN 3166687	0005270161	S	12			
	ILMN_3165566	0004260594	S	224	GGATTAACTGCTGTGGTGTGT		
	ILMN_3164811	0007610424	S	868	GACCACGCCTTGTAATCGTAT		
	ILMN_3165363	0005260356	S	873	CTGCAATGCCATTAACAACCT		
	ILMN_3166511	0002030196	S	130	CGTGCAGACAGGGATCGTAAG		
	ILMN_3164754	0003170364	S	14	GGCAGAGGCGTTTGTATCTGC		
	ILMN_3166432	0006550376	S	161	AGTTGCTGGAGTAGAGCTTGG		
	ILMN_3165742	0001400451	S	642	CGGTATCTACCAGCAAAACAT		
	ILMN_3164913	0002320341	S	808	GTCAATACGATAATGCGCCCA		
	ILMN_3165408	0001820014	S	732	ACGCGAGTTAGACCGTATCGT		
	ILMN_3165203	0002190670	S	631	CTTCCTCGCGCATGGCTTGAA		
	ILMN_3166630	0002750053	S	193	CCCGAGGTAGACTATCTTCCA		¥
	<					>	
						Close	

Figure 14. Specifying the columns that contain the genomic locations of markers in the annotation file

The Chip/Reference text field will be populated with the annotation file name. You can edit this text field this if you wish.

For the Annotation column with gene symbols or miRNA names section, if Gene symbol instead of Marker ID is selected, this field is used automatically populated with the gene symbol column; however, if it is not selected, you will need to manually specify the column in the annotation file that corresponds with gene symbols or miRNA names.

- Select Set Column:
- Select the appropriate column from the dialog; here we have selected ILMN_gene (Figure 5)
- Select OK

🧐 Choose column conta	aining gene symbol/micr	×
O Species	ILMN Controls	^
O Source	ILMN_Controls	
O Search_Key	ERCC-00162	
 Transcript 	ILMN_333737	
ILMN_Gene	ERCC-00162	
O Source_Reference_ID	ERCC-00162	
RefSeq_ID		Υ.
<	>	
	OK Cance	9

Figure 15. Choosing the annotation column with gene symbols

Species and gene symbol information is required for biological interpretation analysis.

- Select the correct species and genome build from the drop-down menus; we have chosen Homo sapiens and hg19 (Figure 6)
- Select **OK** apply the annotation file to your data spreadsheet

Configure Genomic Properties of 1		×
Choose the type of genomic data		
Gene Expression V		
Location of genomic features in spreadsheet		
Gene symbol instead of Marker ID		
C Feature in column label		
Feature in column 1. ID_REF ✓		
Choose chips/references and annotation files Chip/Reference Annotation file		
X HumanHT-12_V4_0_R2_1500 V HumanHT-12_V4_0_R2_1500	Browse	Download
		Add
Annotation column with gene symbols or microRNA names		
Set Column: ?		
Species and Genome Build		
Homo sapiens V	hg 19	~
Advanced	Cancel	ОК

Figure 16. Choosing annotation file using the Configure Genomic Properties dialog

To verify that the annotation has been added, we can try to add annotation information to the spreadsheet when the feature are on rows in the spreadsheet.

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- Right-click on a column in the annotated data file spreadsheet
- Select Insert Annotation from the pop-up menu (Figure 5)

Partek Genomics Suite - 1 (GSM1823575_8H_1.txt)	-	- 0	×
File Edit Transform View Stat Filter Tools Window Custom Help	Workflows Choose		~
Analysis X	Choose in		
🖿 📁 🔜 😹 🖬 🚈 🛞 🌉 🔍 💙 🔞			
1 (GSM1823575_8H_1.txt) Current Selection ILMN_1762337			^
1. 2. ID. REF 81.1			
1. ILMN_176233 Copy			
2. ILMN_205527 Paste			
3. ILMN_173600 Plot			
4. ILMM_238322 Sort Ascending			4
 LDM_iotoSi sortuescenarg mailtane FillColumn 			-
Z. ILMI 16535 Splt Column			-
8. LLMN_12/27/3 Channe area			
9. ILMN_170502 Files to date			
10. ILMN_181431 Fite Exclude			
11. ILMN_235916 Insert			
12. ILMN_173150 Insert Annotation			
13. LLVM_179768 Insert Average			
14. ILMN 224199 Dette			
16. 11M2 27306 PT Columns			-
Create List from Column Labels			
18. LMN_22955 Create List			
19. ILMN_17350- Properties			
20. ILMN_1680754 89.4			
21. ILMM_2375184 59			_
✓ 22. [ILNN_1659452 91.2			4
Kows: 4/322 Columns: 2		2	~

Figure 17. Adding an annotation column to data spreadsheet

The Column Configuration section of the Add Rows/Columns to Spreadsheet dialog should contain all the feature annotations from the annotation file spreadsheet (Figure 6). Here we selected **ILMN_Gene**, which will add gene name information as a column next to 1. ID_REF.

Add Rows/Columns to Spreadsheet						
Add Rows Add Columns Add	Add Average					
Add to the Right v of Column 2.8H_1 v						
Maximum String Length	80 🜩					
Add as string O Add as call	ategorical					
Add selected to defaults			Edit Defaults			
Column Configuration						
Accession	Array_Address_Id	Chromosome	^			
Cytoband	Definition	Entrez_Gene_ID				
GI	ILMN_Gene	Obsolete_Probe_Id				
Ontology_Component	Ontology_Function	Ontology_Process				
Probe_Chr_Orientation	Probe_Coordinates	Probe_Id				
Probe_Sequence	Probe_Start	Probe_Type				
Protein_Product	RefSeq_ID	Search_Key				
			×			
		Set All Clear All	Set Defaults			
		ОК	Cancel			
0						

Figure 18. Annotations from the annotation spreadsheet file should appear as options in the Column Configuration section of the Add Rows/Columns to Spreadsheet

Building an Annotation File

Annotation files for most commercial arrays are available from the chip manufacturer. If you have a custom chip or want to use a customized annotation file, you can create an annotation file that will allow you to add annotations to your features (e.g. probe IDs) when the features are represented by rows on the spreadsheet. Your annotation file must meet the following criteria:

- The annotation file must have a column header with a label for each column
- A column in the annotation file must correspond to the feature ID column of your data spreadsheet
- Any comments before the header must start with # or the header will not be recognized
- The fields of the annotation file must be tab or comma delimited

To invoke a genome view of your data, your annotation file must also have one or more columns that contain the genomic location in a format that Partek Genomics Suite can recognize. The annotation file must also contain a column that has the chromosome and base pair location (start and stop or physical position). Cytoband and/or strand can also be included.

The table below provides possible column labels, a description of the format for that field, and an example.

Column label	Description of format	Example
chromosome	a chromosome label	3
start	an integer, the start position (in base pairs) of the feature	69871322
stop	an integer, the stop position (in base pairs) of the feature	70100176
genomic_coordinates	chromosome:start-stop	3:69871322-70100176
strand	+ for top, - for bottom	+
physical position	an integer, the position (in base pairs) of the feature	70100176

Here are a few examples of the first two rows of annotation files:

Using Agilent format

ProbeID	GeneName	GenomicCoordinates	Cytoband
A_44_P1025812	TC521361	chr12:2546883-2546824	rn 12p12

Using Affymetrix SNPs format

Probe Set ID	Chromosome	Physical Position	Strand	Cytoband
SNP_A-1512540	9	22205296	-	p21.3

• Using Affymetrix exons format

probeset_id	seqname	strand	start	stop
2315588	chr1	+	1155398	1155624

Additional Assistance

If you need additional assistance, please visit our support page to submit a help ticket or find phone numbers for regional support.

