# **MycoCosm:** Comparative analysis of Gene Families

# **Objective:** Compare genomes of wood decay fungi to identify gene families which can be used to distinguish white rot and brown rot fungi

Many fungi of the phylum Basidiomycota are capable of degrading wood, including the recalcitrant polymer lignin, which gives wood its structural strength and resistance to microbial attack (Floudas et al. 2012; Riley et al. 2014). These wood decaying fungi are often classified as either white rot, in which lignin is completely degraded and cellulose is left somewhat intact; or brown rot, in which cellulose is degraded and lignin left somewhat intact. While the precise enzymatic mechanisms vary from one fungus to another, in general the white rot fungi's genomes encode class II peroxidase enzymes (CAZy: AA2) to break down lignin; carbohydrate-binding motifs (CAZy: CBM1) to bind cellulose; and glycoside hydrolases of families 6 and 7 (CAZY: GH6 and GH7) to break down cellulose. The genome of a brown-rot fungus tend to lack genes encoding these enzymes, or have them in reduced numbers compared to white rot fungi.

Suppose we are comparing the genomes of four wood decaying fungi: *Auricularia subglabra*, *Calocera cornea*, *Gloeophyllum trabeum*, *Phanerochaete chrysosporium* RP-78. Suppose, also, that we don't know which of them are white-rot or brown-rot fungi. How can we use MycoCosm to make predictions about their mode of decay?

Start by going to the genome group page created for this example (in real life we would use a similar genome group page, but with a larger, ecologically- or phylogenetically-relevant selection of organisms):

SEAR	СН	BLAST	ANNOTATIONS -	MCL CLUSTERS	DOWN	LOAD	NFO HELP!	
Grou	ıp Nan	ne:	White rot/br	rown rot examp	le 2017			
Rele	ase Da	ate:	2017-04-03	3				
##								
##	Name	)		Assembly	y Length	# Genes	Published	
			g <u>labra v2.0</u>		y Length 5,853,599		Published	II. <u>, 2012</u>
1	Auricu		<u> </u>	76		25,459		
1	Auricu Caloco	ilaria sub era corne	<u> </u>	76	6,853,599	25,459 13,177	Floudas D et a	<u>, 2016</u>

https://genome.jgi.doe.gov/WR\_BR\_example\_2017/

## CAZy browser

Click on the CAZYMES item under ANNOTATIONS in the Main menu.

				CAZymes • White rot/brown rot example 2017
SEARCH BLAST	ANNOTATIONS -	MCL CLUSTERS	DOWNLOAD INFO H	ELP!
To Default Searc		BOLISM CLUSTERS		Show only filtered results Show only filtered totals
	PEPTIDASES			
	TRANSPORTERS			
Annotations/Genor	TRANSCRIPTION FA	ACTORS	Annotation Description	
	Aurc Calc Glot	Phot	_	
⊿ CAZy	<u>802 350 362</u>	450 <u>1,964</u> CA2	ý	
▶ <u>AA</u>	<u>118 27 41</u>	89 <u>275</u> Aux	liary Activities family	
▶ <u>CBM</u>	<u>123 18 19</u>	71 231 Car	oohydrate-Binding Module fami	ly
▶ <u>CE</u>	<u>65 15 17</u>	20 <u>117</u> Car	oohydrate Esterase family	

Here you will see a table representation of the predicted CAZymes (Levasseur et al. 2013). The organisms are labeled along the top. The CAZymes are organized by family and labeled along the sides. The numbers in the table tell you how many proteins from each organism's gene catalog were annotated with a given CAZyme. There is also a totals column. Notice that the CAZymes are hierarchically organized: you can see the total number of genes assigned to the general enzyme category (e.g. 'AA'). To expand top level assignment, click on the small arrow left of the catergory, or use the "Expand All" button at the top. Family designations ('AA1', 'AA2', etc.), and to subfamilies ('AA1\_1', 'AA1\_2', etc.) will then show up. In the image below, arrows have been added in grey for white rot fungi and brown for brown rot fungi.

						CAZymes • Wł
SEARCH BLAST	NNOT	ATION	IS <del>-</del>	M	CL CLUST	ERS DOWNLOAD INFO HELP!
To Default Search fo	or:				Any <b>T</b> Exact	Keywords       Filter         ✓       Filter         ✓       Clear         ✓       Show only filtered to
Annotations/Genomes	Aurde3_1	Calco1	Glotr1_1	Phchr2	Total	Annotation Description
CAZy	802	<u>350</u>	<u>362</u>	<u>450</u>	<u>1,964</u>	CAZy
AA AA	<u>118</u>	<u>27</u>	<u>41</u>	<u>89</u>	275	Auxiliary Activities family
<u>AA1</u>	<u>10</u>	<u>5</u>	<u>5</u>	<u>5</u>	<u>25</u>	Auxiliary Activity Family 1
<u>AA1_1</u>			<u>4</u>		<u>4</u>	Auxiliary Activity Family 1 / Subf 1
<u>AA1_2</u>		2	1	1	<u>4</u>	Auxiliary Activity Family 1 / Subf 2
<u>AA1_3</u>	7				<u>Z</u>	Auxiliary Activity Family 1 / Subf 3
<u>AA2</u>	<u>19</u>			<u>15</u>	<u>34</u>	Auxiliary Activity Family 2
<u>AA3</u>	<u>50</u>	<u>15</u>	<u>24</u>	<u>39</u>	<u>128</u>	Auxiliary Activity Family 3
<u>AA3_1</u>	1		1	1	<u>3</u>	Auxiliary Activity Family 3 / Subf 1
<u>AA3_2</u>	38	<u>13</u>	<u>20</u>	<u>34</u>	<u>105</u>	Auxiliary Activity Family 3 / Subf 2

If we read Levasseur et al. 2013 we know that the AA2 family consists of class II peroxidases that may degrade lignin. Browsing the table, we see that for AA2, we see that *P*. *chrysosporium* and *A. subglabra* possess 15 and 19 copies of AA2, whereas *G. trabeum* and

*C. cornea* possess no AA2s. This might suggest that the former two are white rot fungi and the latter two brown rot fungi!

What about the carbohydrate binding motifs, CBM1? Let's say we don't want to scroll through the entire list of CAZymes. Type 'CBM1' into the 'CAZY terms' search box. This will limit the view to only those CAZymes that have a CBM1. Why do so many CAZymes besides CBM1 show up? Because CBM1 co-occurs on the same protein chain with many other CAZymes of diverse function. The numbers in the table will now show, for each CAZyme's row, the number of proteins that also have a CBM1.

SEARCH BLAST ANNOTATIONS - MCL CLUSTERS DOWNLOAD INFO HELP!											
To Default Sea	o Default Search for: Any V Keywords V Filter Show only filtered results										
	BM1	t T Clear									
Annotations/Gen	omes	Aurde3_1	Calco1	Glotr1_1	Phchr2	Total	Annotation Description				
CAZy	8	<u>33</u>	2	2	<u>68</u>	<u>155</u>	CAZy				
AA 🔊	1	<u>8</u>			<u>7</u>	<u>15</u>	Auxiliary Activities family				
<u>AA3</u>		2				2	Auxiliary Activity Family 3				
<u>AA3_2</u>		2				2	Auxiliary Activity Family 3 / Subf 2				
<u>AA8</u>					1	1	Auxiliary Activity Family 8				
<u>AA9</u>	1	<u>5</u>			<u>6</u>	<u>11</u>	Auxiliary Activity Family 9				
<u>AA12</u>		1				1	Auxiliary Activity Family 12				
⊿ <u>CBM</u>	4	<u>48</u>	1	1	<u>36</u>	<u>86</u>	Carbohydrate-Binding Module family				
CBM1	4	<u> 18</u>	1	<u>1</u>	<u>36</u>	<u>86</u>	Carbohydrate-Binding Module Family				
<u>CE</u>		7			<u>4</u>	<u>11</u>	Carbohydrate Esterase family				
<u>CE1</u>		1			2	<u>3</u>	Carbohydrate Esterase Family 1				
CE5	1	2				2	Carbohydrate Esterase Family 5				
<u>CE15</u>		<u>3</u>			1	4	Carbohydrate Esterase Family 15				
<u>CE16</u>		1			1	2	Carbohydrate Esterase Family 16				
⊿ <u>GH</u>	2	<u>20</u>	1	1	<u>21</u>	<u>43</u>	Glycoside Hydrolase family				
GH3					1	1	Glycoside Hydrolase Family 3				
⊿ <u>GH5</u>		4	1		<u>4</u>	<u>9</u>	Glycoside Hydrolase Family 5				
<u>GH5_5</u>		<u>3</u>	1		2	<u>6</u>	Glycoside Hydrolase Family 5 / Subf				

Notice the abundance of CBM1-encoding genes in *P. chrysosporium* and *A. subglabra*, while *G. trabeum* and *C. cornea* have only a single CBM1-encoding gene each (co-occuring with GH5\_5 and GH10 proteins). All of this indicates that we might be looking at two white-rot and two brown-rot fungi.

Click on the number (e.g. 48 for Aurde3\_1) to see the the CBM1-containing proteins of A. suglabra in more detail. Notice a variety of CAZymes co-occur with CBM1, including GH5 (various subfamilies), GH6, and many others.

	ia subglabra v2.0 » CBM1 Carl at 25 • rows per page	bohydrate	e-Binding	Module Family 1		
Protein Id	Location	Gene Length	Protein Length	CAZy Annotations	Domains	Models Domains
Aurde3_1 1329318	scaffold_34:305.088-301.173	3,916	1,144	Carbohydrate-Binding Module Family 1      Carbohydrate-Binding Module Family 1	• <u>Eungal cellulose</u> <u>binding domain</u> • <u>WSC</u> <u>domain</u>	
Aurde3_1 1201389	scaffold_2:1.437.648-1.439.395	1,748	402	Carbohydrate-Binding Module Family 1      Carbohydrate-Binding Module     Family 1	Eungal cellulose     binding domain     WSC     domain	
Aurde3_1 1352721	scaffold_27:183.041-181.410	1,632	320	Auxiliary Activity Family 9 Carbohydrate-Binding Module Family 1	• Eungal cellulose binding.domain • Glycosyl hydrolase family. 61	<b>132 68 38 78 74 29 72 67 6 94 48 184 52 1</b> <b>50 47 54 50 53 54 52 61 53 51 44 52 51</b>
Aurde3_1 162872	scaffold_29:136.999-137,991	993	280	<u>Carbohydrate-Binding Module Family 1</u> <u>Glycoside Hydrolase Family 11</u>	Glycosyl hydrolases family 11      Fungal cellulose binding domain	248 184 297 111 47 53 53
Aurde3_1 1175661	scaffold_43:408.689-407.097	1,593	442	Carbohydrate-Binding Module Family 1      Glycoside Hydrolase Family 6	Eungal cellulose     binding_domain     Glycosyl hydrolases     family_6	- 85 15 188 96 261 681 53 52 59 50 53
Aurde3_1 124125	scaffold_32:254.008-255.583	1,576	396	Carbohydrate-Binding Module Family 1      Glycoside Hydrolase Family 5 / Subf 5	Cellulase (glycosyl hydrolase family 5)     Fungal cellulose binding domain	- 91 - 111 - 161 - 78 - 265 - 188 - 221 - 73 54 55 52 64 - 53 57 53
Aurde3_1 140513	scaffold_37:362.657-364.080	1,424	314	Auxiliary Activity Family 9      Carbohydrate-Binding Module Family 1	Eungal cellulose     binding_domain     Glycosyl hydrolase family     61	- <mark>72 63 184 74 69 39 66 118 197 66</mark> 52 58 55 54 52 55 55 55 48
Aurde3_1 1335548	scaffold_7:812,688-814,432	1,745	447	Carbohydrate-Binding Module Family 1      Glycoside Hydrolase Family 6	Eungal cellulose binding domain • Glycosyl hydrolases family 6	- 246 206 96 1,011 67 62 57
Aurde3_1 1228030	scaffold_1:2.029.582-2.027.770	1,813	520	Carbohydrate-Binding Module Family 1      Glycoside Hydrolase Family 7	• Eungal cellulose binding domain • Glycosyl hydrolase family. Z	- <u>136 69 401 114 767 75</u>
Aurde3_1 138514	scaffold_66:143.034-141.170	1,865	411	Carbohydrate-Binding Module Family 1      Glycoside Hydrolase Family 5 /      Subf 5	Cellulase (glycosyl hydrolase family 5)     Fungal cellulose binding domain	<b>79 80 166 187 117 37 75 65 36 59 253</b> 77 <b>48 119 54 50 53 61 46 52 44 56 49</b>

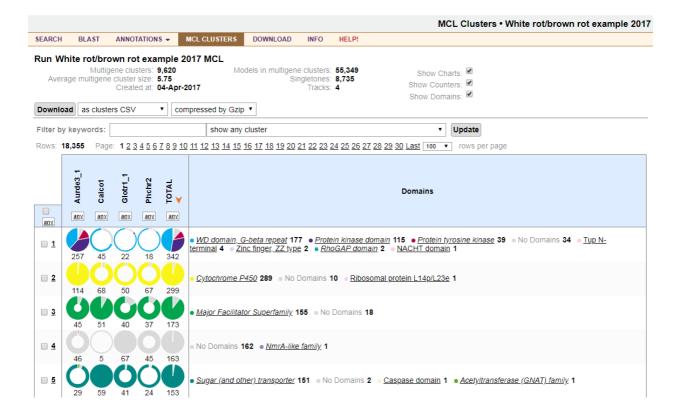
White rot/brown rot example 2017

As an exercise, repeat the same search with GH6, GH7, and also the AA9 family of lytic polysaccharide monoxygenases, which may oxidatively act on lignin (Levasseur et al. 2013). Do the presence/absence patterns of these genes indicate the same conclusions about these fungi's mode of decay as we found with AA2 and CBM1? Is it a strict dichotomy, or are there some grey areas in the distribution of these genes?

(Answer: *P. chrysosporium* and *A. subglabra* induce white rot wood decay; *G. trabeum* and *C. cornea* brown rot. Notice that brown rot *G. trabeum* has a few AA9 genes, however, indicating that these genes may play a role in brown rot, not just white rot, where AA9s are expanded.)

### **Cluster page**

Now that we have an idea which fungus uses which decay mode, let's ask the reverse question: what are the genes present in one lifestyle, and absent in the other? To do this, click the 'MCL CLUSTERS' item of the Main menu. Here you will see the results of protein sequence clustering by the MCL algorithm (Enright et al. 2002). You can think of clusters as protein families. As with the CAZy browser, the columns indicate organisms. The rows indicate a protein cluster, one cluster per row, with the number of proteins each organism contributes to a cluster. See the HELP Menu for a full explanation of the cluster page.



Notice that under each organism label is a button 'any' that can be used to filter clusters by the number of proteins that organism contributes to a cluster, and thus limit which clusters are shown. As an experiment, set the white rot fungi (Aurde3\_1 and Phchr2) to "1+" and the brown rot fungi (Calco1 and Glotr1\_1) to "=0". Doing this returns only those clusters which are present in Aurde3\_1/Phchr2 and absent in Calco1/Glotr1\_1.



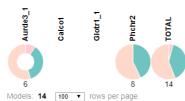
Some 150 clusters fit these criteria. These clusters might include genes important to the white rot decay mode, because they are present in white rot fungi and absent in brown rot fungi. But some of these clusters might have no functional connection to wood decay mode - they are present/absent from the respective kinds of wood decay fungi merely by chance.

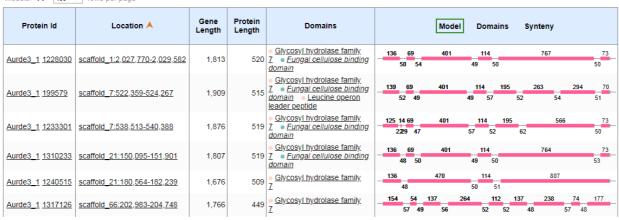
These clusters nevertheless represent candidates for further analysis of possible connections to decay mode.

How does one begin interpreting the results? To help with this, each cluster row shows the Pfam domains (http://pfam.xfam.org) that are found in that cluster. Notice that the third row has a "Peroxidase" (PF00141) domain. Notice that the numbers are very close to what we found for the AA2 class II peroxidases in the CAZy browser. It turns out that PF00141 is a superfamily that includes the AA2 enzymes, but it is important to note that not all members of PF00141 can degrade lignin - some have other functions.

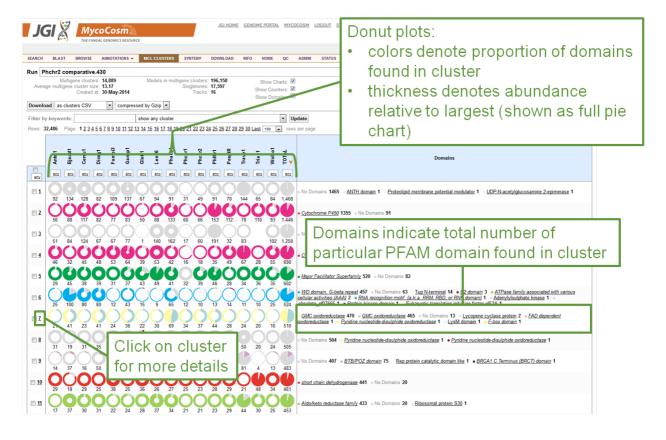
Scroll through the rest of the 150 clusters and you will see domains such as Glycosyl hydrolase family 7 and Fungal cellulose binding domain in cluster 507, which roughly overlap with the CAZy GH7 and CBM1 families. Click the '507' to explore that cluster in more detail. On the cluster detail page, a table is presented with one protein per row. Click the 'Domains' view on the rightmost column to see the domain structure of each protein. Notice that all of the proteins have the GH7 domain, and that most, but not all, have a single CBM1 motif at the C-terminus.

Run White rot/brown rot example 2017 MCL » Cluster 507





Let's look at what other proteins have the CBM1 carbohydrate-binding motifs in them. Returning to the cluster run page (click the back button, or click the CLUSTERS Menu). Enter the phrase "fungal cellulose binding domain" (be sure to include the quotes) into the "filter by keywords" field. This returns some 26 clusters, all of which have the Pfam domain CBM\_1 (PF00734). We see that CBM1 motifs occur in a wide array of domain combinations: often with GMC oxidoreductases, AA9 lytic polysaccharide monoxygenases (formerly GH61), and many hydrolytic enzymes such as GH5, GH6, and GH7. Notice that while these proteins typically are found in expanded copy number in the white rot fungi (Aurde3\_1 and Phchr2) they are sometimes found, albeit in lower copy number, in the brown rot fungi (Calco1 and Glotr1\_1). As additional exercises you can (a) search for gene families absent in both white rot fungi; (b) find gene families absent in white rot but present in both brown rot fungi and look at functional domains associated with these families; (c) check if any of these domains are present only in brown rot fungi by resetting filters back to 'any' and searching for names of these domains.



A summary of tools available in MCL clustering are shown below.

Clicking in Cluster number provides additional tools as shown below.

diffe usef	detail pag rent tabs ful inform	s pro		JOLHOME GENOME PORTAL	do si	omains: Co omain diagr gnificance ( cid lengths o	ams v top) a	vith e-va Ind amin	lue o	Synteny:
Line data	THE FUNGAL GENOMICS RESOL	MCLCLU Todau es 35 22	diagr and i	、	n exo		MCL Clus	ters - Phanerochaete chrys	sporium RP-78 v2.2	Scaffold context for genes, color coded by function
Protein Id	Location A	Gene Length	Protein Length	Domains		Model Domains Sy	nteny	Model Doma	ains Synteny	Model Domains Synteny
Antsi1 727117	scaffold_41:84.682-87.896	3,215	607	GMC oxidoreductase - GMC	coxidoreductase	55 8 100 624474 70 74 59391289 179 143 79 238 6850 59 5956 55 58 69 565454 58 64 51 58		4.8E-53 198 34 211526 24 26 2013423 60 4	3.7E-39 9 <sup>'</sup> 27 <sup>'</sup> 80 <sup>'</sup> 25 23 <sup>'</sup> 26181222 28	
Antsi1 788439	scaffold_60:90.716-91.775	1,060	177	GMC oxidoreductase		<u>     35     230     90     49     52     56     55 </u>	438	12 78 2.56-22	31 17 43	- + +-
Antsi1 727990	scaffold_71:66.928-69.471	2,544	694	GMC oxidoreductase • GMC	; oxidoreductase		80 237 53 225 59 55 58 55	1.0E-55	2.3E-32	
Antsi1 709516	scaffold_85:23.345-26.934	3,590	606	GMC oxidoreductase GMC		00109 221 141 79 130 184 111 143 131 91 251 2			26 105 28 80 18 7	
Antsi1 710585	scaffold_115:9.777-12.373				; oxidoreductase	52 47 59 58 58 77 74 65 67 53 75 68		7.3E-81 21 47 27 44 62 37 48 4	1.2E-36	
		2,597	582	GMC oxidoreductase GMC		52 47 59 58 58 77 74 60 67 53 75 60 286 91 289 52 357 79 264 58 53 53 59 46 58		21' 47' 27' 44 ' 62 ' 37' 48 ' 4 3.8E-68 '27' 31' 101 ' 18' 120	1 2E-38 5 31 85 91923 43 33 3 2.5E-33 27 88 175	
Antsi1 710971	scaffold_127:3.598-6.241	2,597	582 s			52.47         59.58         57.7         74.65         67.53         75.         66           266         91         269         62         367         79.         264           56         53         55.56         46.56         56.57         79.         264	83588664 131 58 53 783 50 792 47	21' 47' 27' 44' 62' 37' 48' 4 3.8E-68 27' 31' 101' 18' 120 1.0E-52 27' 31' 101' 18' 120	1 2E-36 6 '31' 85 91 823 '43 '33 ' 2.5E-33 27' 88 '175 27' 88 '175 27' 88 '176	
Antsi1 710971 Antsi1 722239	scaffold_127:3.598-6.241 scaffold_140.56.265-58.911			GMC oxidoreductase • GMC	C oxidoreductase	52         47         59         58         57         74         65         67         53         75         64           269         91         269         62         357         79         264           58         53         53         59         46         54         53         59         46         54           354         91         269         52         357         79         264           364         91         269         52         357         79         264           350         91         269         52         357         79         264           350         91         269         52         357         79         264           36         91         269         52         357         79         264           30         93         93         94         95         33         33         34         35           312         79         40         158         106         67         923         173           47         56         53         54         97         51         47         66         53	ECODO 64 131 58 53 783 50 792 47 154 86 163 60 189 51 56 55 63	21' 47' 27' 44' 62' 37' 48' 4 3.85-68 27' 31' 101' 16' 120 105-62 27' 31' 101' 16' 120 4.35-75 '44' 27' 29' 231520' 30' 1925' 108	128-36 6 31 86 91923 43 33 266-33 27 88 176 128-35 27 88 176 2.68-38 68 52 28 65 20 6	
		2,644	583	<u>GMC oxidoreductase</u> = <u>GMC</u> <u>GMC oxidoreductase</u> = <u>GMC</u>	Coxidoreductase Coxidoreductase	224         99         326         97         74         60         67         53           235         91         226         92         357         79         244           344         91         226         92         357         79         244           344         91         226         92         357         79         244           345         91         226         92         357         79         244           345         91         226         93         377         70         245           345         95         53.46         49         53.57         60         53.57         60         53.57         60         53.57         60         53.57         60         53.57         60         53.57         60         53.57         50         50         53.57         50         50         53.57         50         50         53.57         50         50         53.57         50         50         50         50         50         50         50         50         50         50         50         50         50         50         50         50         50         50         50	80600 64 131 58 53 783 50 792 47 154 85 163 60 189 51 60 55 63 209 69 331 92 123 6 55 61 54 61	21         47         27         44         62         37         48         4           27         31         101         15         120         106         52         106         52         106         52         106         52         52         52         53         106         52 <t< td=""><td>12E-36         12E-36           31         85         91822         43         33           27         88         175         12E-36           27         88         175         2.6E-33           27         88         176         2.6E-36           27         83         12E-36         2.6E-36           68         52         29         56         20           7.7E-46         9         87         70         24</td><td></td></t<>	12E-36         12E-36           31         85         91822         43         33           27         88         175         12E-36           27         88         175         2.6E-33           27         88         176         2.6E-36           27         83         12E-36         2.6E-36           68         52         29         56         20           7.7E-46         9         87         70         24	
Antsi1 722239	scaffold_140.56.265-58.911 scaffold_141:37.281-40.616	2,644 2,647	583 613 681	<u>GMC oxidoreductase</u> = <u>GMC</u> <u>GMC oxidoreductase</u> = <u>GMC</u>	Coxidoreductase Coxidoreductase Coxidoreductase Coxidoreductase	202         40         428         77         416         62         63         79         24           25         41         25         52         52         57         7         24           26         41         25         52         57         7         24           26         11         20         52         32         7         7         24           26         11         20         53         34         40         33         16         45         35         16         45	KK00064         131         58         53           783         783         792         792           47         56         60         163         60         163           59         56         66         60         331         52         123           50         56         65         64         61         269         69         269         60         702           60         60         60         60         702         60         60         702         65         61         60         702         60         60         702         60         60         702         60         60         702         703<	21'         47'         22'         44'         62'         37'         48'         48'           27'         31'         10         18'         120'         106:62           27'         31'         101'         18'         120'         106:62           4         27'         29'         21'         10'         14'         120'           64'         27'         29'         21'         20'         100'         100'         100'           90'         27'         44'         100'         20'         100'         100'         100'         100'           90'         27'         44'         100'         20'         100' </td <td>6         31         85         M 223         43         33           2</td> <td></td>	6         31         85         M 223         43         33           2	
Antsi1 722239 Antsi1 82024 Antsi1 729557 Antsi1 722519	acaffold_140.56.265.58.911 acaffold_141.37.281.40.616 acaffold_141.41.160.43.966 acaffold_164.21.899.24.981	2,644 2,647 3,336 2,807 3,083	583 613 681 713 648	GMC oxidoreductase = GMK GMC oxidoreductase = GMK GMC oxidoreductase = GMK GMC oxidoreductase = GMK GMC oxidoreductase = GMK	2 oxidoreductase 2 oxidoreductase 2 oxidoreductase 2 oxidoreductase 2 oxidoreductase 2 oxidoreductase	22         40         50         56         77         74         60         67         57         60           28         10         200         20         50         50         50         66         50           30         10         200         50         50         50         66         50           30         10         200         50         50         66         50           30         10         200         50         50         66         50           30         10         200         50         50         66         50           30         50         54         66         51         70         220         100         30         70         230         100         70         240         70         240         70         240         70         240         70         240         70         240         70         240         70         240         70         240         70         240         70         240         70         240         70         240         70         240         70         240         70         240         70         240         70<	KK00064         131         58         53           783         783         792         792           47         56         60         163         60         163           59         56         66         60         331         52         123           50         56         65         64         61         269         69         269         60         702           60         60         60         60         702         60         60         702         65         61         60         702         60         60         702         60         60         702         60         60         702         703<	21         47         22'         44         62         27         48         63           27         31         10         14         120         106/62           27         31         10         14         120/62         106/62           27         31         10         14         120/62         106/62         106/62         106/62         106/62         106/64         100/62         100         106/64         100/62         100         106/64         100/62         100         106/64         100/62         100         100/62         100         100/62         100         100/62         100         100/62         100         100/64 </td <td>128-30         128-30           5         31         85         91823         43         33           266-33         266-33         126-36         126-36           27         88         176         126-36           27         88         176         126-36           27         88         176         266-23           68         52         26         52         36           9         87         70         24         111           1486-31         1486-36         11         1486-31</td> <td></td>	128-30         128-30           5         31         85         91823         43         33           266-33         266-33         126-36         126-36           27         88         176         126-36           27         88         176         126-36           27         88         176         266-23           68         52         26         52         36           9         87         70         24         111           1486-31         1486-36         11         1486-31	
Antsi1 722239 Antsi1 82024 Antsi1 722557 Antsi1 722519 Antsi1 791044	scaffold_140.56.265.58.911 scaffold_141.37.281.40.616 scaffold_141.41.160.43.966 scaffold_164.21.899.24.991 scaffold_152.9.515.12.300	2,644 2,647 3,336 2,807 3,083 2,796	583 613 681 713 648 597	GMC axidoxeductase = GMK GMC axidoxeductase = GMK GMC axidoxeductase = GMK GMC axidoxeductase = GMK GMC axidoxeductase = GMK	2 oxidoreductase 2 oxidoreductase 2 oxidoreductase 2 oxidoreductase 2 oxidoreductase 2 oxidoreductase 2 oxidoreductase 2 oxidoreductase	1000         100 <td>KK00064         131         58         53           783         783         792         792           47         56         60         163         60         163           59         56         66         60         331         52         123           50         56         65         64         61         269         69         269         60         702           60         60         60         60         702         60         60         702         65         61         60         702         60         60         702         60         60         702         60         60         702         703&lt;</td> <td>21' 47' 22' 44         62         23' 45'         62           27' 31' 10' 15'         106' 42'         106' 42'           27' 31' 10' 15'         120'         436' 65'           42' 22' 22' 65' 05' 152'         15' 65' 65'           64' 22' 22' 22' 65' 15' 16' 25'         5' 65' 65'           62' 22' 24' 7833' 31' 45' 26' 65' 76'         69' 55' 75'           12' 73.4078112' 65' 76' 79' 15' 16' 25'         5' 65' 76'</td> <td>126-00 6 31 65 9123 43 33 5 27 88 175 27 88 172 88 122-39 28 2 176 88 2 29 85 20 9 87 70 24 124 28274 9 87 70 24 124 28274 48 57 247 83 58 29</td> <td></td>	KK00064         131         58         53           783         783         792         792           47         56         60         163         60         163           59         56         66         60         331         52         123           50         56         65         64         61         269         69         269         60         702           60         60         60         60         702         60         60         702         65         61         60         702         60         60         702         60         60         702         60         60         702         703<	21' 47' 22' 44         62         23' 45'         62           27' 31' 10' 15'         106' 42'         106' 42'           27' 31' 10' 15'         120'         436' 65'           42' 22' 22' 65' 05' 152'         15' 65' 65'           64' 22' 22' 22' 65' 15' 16' 25'         5' 65' 65'           62' 22' 24' 7833' 31' 45' 26' 65' 76'         69' 55' 75'           12' 73.4078112' 65' 76' 79' 15' 16' 25'         5' 65' 76'	126-00 6 31 65 9123 43 33 5 27 88 175 27 88 172 88 122-39 28 2 176 88 2 29 85 20 9 87 70 24 124 28274 9 87 70 24 124 28274 48 57 247 83 58 29	
Antsi1 722239 Antsi1 82024 Antsi1 729557 Antsi1 722519 Antsi1 791044 Antsi1 713654	scaffold_140_56_265_58_911 scaffold_141_37_281_40_515 scaffold_141_41_50_43_966 scaffold_141_41_150_43_966 scaffold_164_21_899_24_901 scaffold_192_9_515_12_300 scaffold_229_13_914_17_039	2,644 2,647 3,336 2,807 3,083 2,786 3,126	583 613 681 713 648 597 635	GMC oxidereductase = GMK GMC oxidereductase = GMK	2 oxidoreductase 2 oxidoreductase 2 oxidoreductase 2 oxidoreductase 2 oxidoreductase 2 oxidoreductase 2 oxidoreductase 2 oxidoreductase 2 oxidoreductase	1000         100 <td>2000         64         131         56         53           783           782           782           783           783           66         66           66         66           66         67           66         66           120         20           120         20           66         66           66         66           66         66           120         20           131         62           120         21           120         121           120         123           7         76           73         69           7         65</td> <td>21         42         25         37         44         28         37         31&lt;</td> <td>1356 %         1266 %         2463 %         237 %         246 %         246 %         246 %         246 %         176 %</td> <td></td>	2000         64         131         56         53           783           782           782           783           783           66         66           66         66           66         67           66         66           120         20           120         20           66         66           66         66           66         66           120         20           131         62           120         21           120         121           120         123           7         76           73         69           7         65	21         42         25         37         44         28         37         31<	1356 %         1266 %         2463 %         237 %         246 %         246 %         246 %         246 %         176 %	
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Antsi1 722239 Antsi1 82024 Antsi1 729557 Antsi1 722519 Antsi1 791044 Antsi1 713654	scathol, 140,55,265-58,911 scathol, 141,37,201-40,616 scathol, 141,41,169-43,566 scathol, 161,41,999-24,181 scathol, 152,9,515-12,300 scathol, 152,9,515-12,300 scathol, 2021,3,914-17,039 scathol, 312,0,209-22,872 scathol, 345,11,120-13,781	2,644 2,647 3,336 2,807 3,083 2,786 3,126	583 613 681 713 648 597 635 627 633	GMC oxidereductase = GMK GMC oxidereductase = GMK	2 oxidoreductase 2 oxidoreductase	$\begin{array}{c c c c c c c c c c c c c c c c c c c $	Construction         13         10 <th10< th="">         10         10</th10<>	51         -27         44         62         37         34           77         31         34         364         364         37         34         364           77         31         34         364	1256 %           1         16         162 3 43 33           27         8         176           27         8         176           27         8         120 3           28         120 3         126           48         220 3         50 45 26           9         9         70 40 50         226 30           9         9         9         70 24         100           1000         200 50         100 50         100 50           3107 747         50 46         50 24         120 40           2107 749         50 43 48         324 48         324 48           2107 74         70 50 75 24         100 40         120 40	

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