

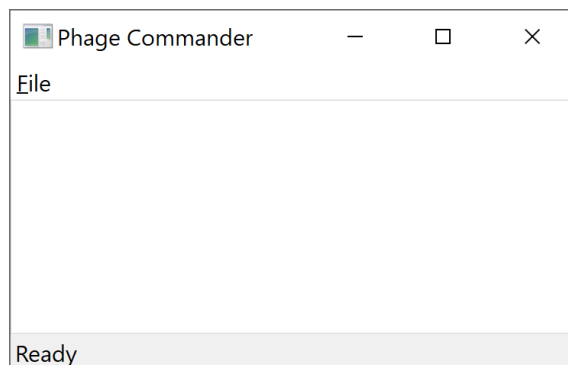
Introduction to Phage Commander

- **Phage Commander** is a software tool for sequencing phage genomes.
- Phage Commander runs a phage's DNA sequence through gene sequencing software tools and outputs a spreadsheet of potential genes. These tools include:
 - Glimmer, Genemark, Genemark HMM, Genemark S, Genemark S2, Genemark Heuristic, Prodigal, RAST, and Metagene
- Phage Commander's output can be exported as a spreadsheet.
- These slides describe how to use Phage Commander
- A draft paper describing Phage Commander in detail is here:
<https://www.biorxiv.org/content/10.1101/2020.11.11.378802v1>

Opening Phage Commander

How to open Phage Commander:

- Download Phage Commander from GitHub repository:
<https://github.com/sarah-harris/PhageCommander>
- Run Phage Commander:
 - **Windows:** Navigate to phagecommander/bin. Click on the executable: phagecom-windows.exe
 - **Linux or Mac:** Open a shell. Navigate to phagecommander. Type `py phagecom.py`. If you need to install any of the supporting packages, first type: `'pip install .'` (in the PhageCommander-master directory – the directory that contains setup.py)
- A small Phage Commander window will appear:



Using Phage Commander

How to use Phage Commander:

- In the Phage Commander Window, click on the **File** menu, and select **New**.
- A Select Query Tools window will open (see next slide).
 - If using **GeneMark.hmm** you will need to select your phage's bacterial host from the drop down menu under **Species**
 - To use **RAST**, you will need to create a login and password at <https://rast.nmpdr.org>
- Press **Open** and select your phage's fasta file – you may use the example fasta file included in the GitHub repository: `Patience.fasta`
- Press **Query** to run Phage Commander. Phage Commander will now run the sequence through the selected sequencing tools – this may take several minutes.
- When Phage Commander completes, press **OK**

Select Query Tools Window

The screenshot shows a window titled "Select Query Tools" with a close button (X) in the top right corner. The window contains several sections of tools, each with a checkbox and a label. The "Genemark" section has three checkboxes: "Genemark" (checked), "HMM" (checked), and "Heuristic" (checked). The "GMS" section has two checkboxes: "GMS" (checked) and "GMS2" (unchecked). The "Glimmer" section has three checkboxes: "Glimmer" (checked), "Prodigal" (checked), and "RAST" (checked). The "Metagene" section has two checkboxes: "Metagene" (checked) and "Aragorn" (checked). Below these sections is a "Species:" label followed by a dropdown menu showing "Mycobacterium_smegr". Below that is a "Fasta File:" label followed by a text input field and an "Open..." button. At the bottom are two buttons: "Query" and "Cancel". Red arrows point from text instructions to specific elements: one from "Leave GMS2 unchecked, all others checked" to the "GMS2" checkbox; one from "Select host from drop down menu if 'HMM' box is checked (GeneMark.hmm)" to the "Species:" dropdown; one from "Press open select phage fasta file" to the "Open..." button; and one from "Press query" to the "Query" button.

Section	Tool	Checked
Genemark	Genemark	✓
	HMM	✓
	Heuristic	✓
GMS	GMS	✓
	GMS2	✗
Glimmer	Glimmer	✓
	Prodigal	✓
	RAST	✓
Metagene	Metagene	✓
	Aragorn	✓

Species: Mycobacterium_smegr

Fasta File: Open...

Query Cancel

Leave GMS2
unchecked, all
others checked

Select host from
drop down menu if
"HMM" box is
checked
(GeneMark.hmm)

Press open select
phage fasta file

Press query

In the Event of an Error

- If Phage Commander runs successfully, you will see a window saying “**Done! Query Successful**”
- Press **OK** and you will see the Phage Commander output in the form of a spreadsheet (expand the window)
- If phage commander encounters an error, the error message will display which program is causing the error (e.g. Glimmer, Aragorn, GMS2, etc.)
- To work around this, re-run Phage Commander without including the program causing the error

Total Calls column
Is the number of
programs
identifying a gene

**“GM” is the genes
identified by
GeneMark**

**“HMM” is the
genes identified by
GeneMark.hmm**

**“Heuristic” is the
genes identified by
GeneMark Heuristic**

Phage Commander - untitled*

File

Genes

	TOTAL CALLS	ALL	ONE	GM	GM	GM	GM	HMM	HMM	HMM	HMM	HEURISTIC	HEURISTIC	HEURISTIC	HEURISTIC	GMS
1	2							+	2	94	93					
2	8	X		+	138	431	294	+	138	431	294	+	138	431	294	+
3	8	X		+	616	2076	1461	+	616	2076	1461	+	616	2076	1461	+
4	8	X		+	2135	3547	1413	+	2093	3547	1455	+	2135	3547	1413	+
5	8	X		+	3538	4875	1338	+	3538	4875	1338	+	3538	4875	1338	+
6	8	X		+	4879	5244	366	+	4879	5244	366	+	4879	5244	366	+
7	8	X		+	5281	6225	945	+	5281	6225	945	+	5281	6225	945	+
8	8	X		+	6225	6575	351	+	6225	6575	351	+	6225	6575	351	+
9	8	X		+	6579	6941	363	+	6579	6941	363	+	6579	6941	363	+
10	8	X		+	6941	7291	351	+	6941	7291	351	+	6941	7291	351	+
11	8	X		+	7307	7654	348	+	7307	7654	348	+	7307	7654	348	+
12	8	X		+	7654	7923	270	+	7654	7923	270	+	7654	7923	270	+
13	8	X		+	7923	8336	414	+	7923	8336	414	+	7923	8336	414	+
14	8	X		+	8333	8770	438	+	8333	8770	438	+	8333	8770	438	+
15	8	X		+	8862	9860	999	+	8862	9860	999	+	8862	9860	999	+
16	8	X		+	9987	10586	600	+	9987	10586	600	+	9987	10586	600	+
17	8	X		+	10687	10905	219	+	10687	10905	219	+	10687	10905	219	+
18	8	X		+	10919	14569	3651	+	10919	14569	3651	+	10919	14569	3651	+
19	8	X		+	14566	16311	1746	+	14566	16311	1746	+	14566	16311	1746	+
20	8	X		+	16311	18047	1737	+	16311	18047	1737	+	16311	18047	1737	+
21	8	X		+	18070	18963	894	+	18070	18963	894	+	18070	18963	894	+
22	8	X		+	18964	21225	2262	+	18964	21225	2262	+	18964	21225	2262	+
23	8	X		+	21225	22292	1068	+	21225	22292	1068	+	21225	22292	1068	+
24	8	X		+	22289	22465	177	+	22289	22465	177	+	22289	22465	177	+
25	1		X													
26	7			+	22477	22839	363	+	22477	22839	363	+	22477	22839	363	+
27	1		X													
28	8	X		+	22836	24017	1182	+	22839	24017	1179	+	22836	24017	1182	+
29	8	X		+	24014	25180	1167	+	24014	25180	1167	+	24014	25180	1167	+
30	8	X		+	25202	25450	249	+	25202	25450	249	+	25202	25450	249	+
31	8	X		+	25447	25806	360	+	25447	25806	360	+	25447	25806	360	+

Each row is a gene

Exporting Phage Commander Results

- To export as excel spreadsheet, select:
 - **File** → **Export** → **Excel**
- To export as GenBank (.gb) format file, select:
 - **File** → **Export** → **GenBank**
 - Make sure number in “Calls” box is maximum (should usually be 8)
 - Make sure “Less than or equal to” circle is filled
 - Press “Save as” and enter desired filename
 - Press “Export”

Exporting Phage Commander results in .gb format

Make sure this number is the maximum

Select "Less than or equal to"

Press "Save as" and enter desired filename

Press "Export"

51		+	6225	6575	351
53		+	6579	6941	363
51		+	6941	7291	351
48		+	7307	7654	348
70				7923	270
14				8336	414
88				8770	438
99				9860	999
00				10586	600
19				10905	219
51				14569	3651
46		+	14566	16311	1746
37		+	16311	18047	1737
94		+	18070	18963	894
62		+	18964	21225	2262
68		+	21225	22292	1068

Export to Genbank

Select genes with:

8 Calls

☒ Less than or equal to ☐ ALL

☐ Greater than ☐ ONE

☐ Exactly

Save as...

Export Cancel