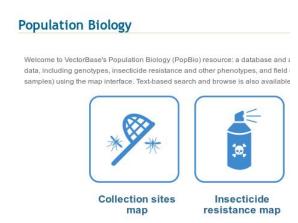
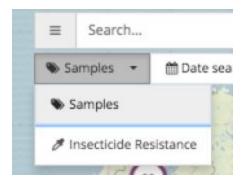
Population Biology (PopBio)

Map-based search and analysis of insecticide resistance (IR) data

In the "tools" menu click on Population Biology, then click on 'Collection sites map' -->



Notice this link takes you to the 'Samples views'



Then, if you have a small screen, it's a really good idea to click on the "full screen" icon, top right.



Search

Only the samples that match the current search will be shown on the map.

Start typing "Anopheles darlingi" in the search box. Stop when you see "Anopheles darlingi in Taxonomy" in the autocomplete suggestions below where you are typing. Choose this option and see how the display changes. The total number of displayed items (samples) is shown at the bottom of the map. It should say "5134 visible samples summarized by Species".

5134 visible samples summarized by Species ©2017 MA

Clear the search by clicking the small "x" next to "*Anopheles darlingi*" or the larger "X" (which becomes red if you hover over it) next to the search box.

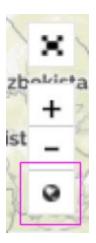


Try the same thing again, but this time just start typing "darlingi" and you should get the same autocomplete suggestion.

Cancel/clear the search again and reset the zoom level (globe icon below the "+" and "-" icons, top-right).

Now we will learn what the Global/Local suggestions switch below the search box does.

Type "darl" into the search box again and you will see the "Anopheles darlingi" suggestion. Now zoom the map in to show only India or Africa. Click the globe icon (so that it goes blue).



Try typing "darl" again - there are no search term suggestions any more. If you start typing the name of a species endemic to India or Africa you will see suggestions.

Reset the search box and zoom into data points in China until no other countries are visible.

2. Type 'sin' in the search box. What is the first autocomplete suggestion in the two modes?

Global suggestions	
Local suggestions	

Reset the search and zoom level so that you can view the whole world, and make sure the globe icon is grey (Global suggestions).

- 3. Now we're going to search for two different species at the same time. Clear the search again and reset the zoom level. Start typing and selecting the "in Taxonomy" suggestions for both of these species names (do one and then the other):
 - Anopheles culicifacies
 - Anopheles fluviatilis

Note that when searching for two different things within the same category (in this case "in Taxonomy") then the search terms are combined with a logical "or". Later we will search for terms in different categories - which are combined with "and".

4. Assuming that all the data shown on the map was obtained in a comprehensive and unbiased way, where do the two species occur?

	Answer
Eastern India, North Pakistan and Southern Iran	
South East Asia and India	
Sri Lanka only	

Digging deeper

5. Keep the two-species search from the previous section. Zoom into the southern tip of India until you see several markers with two "samples", one from each species. What is going on here? Why is there exactly one of each species at these collection sites? See the image to the right.

We can find out more about what data is shown on the map by clicking (once) on a marker. By default, a species pie chart is



shown on the left hand side. Click the table icon (second from the top on the left hand side) so it says "Details for selected samples" and shows information panels about each sample.



An important piece of information is shown in parentheses after the sample stable ID. Most of the samples in this part of India/Sri Lanka are either "population" or "pool" samples. Click around on various markers until you see both of these sample types.

You will notice that the "population" samples come from project VBP0000029. You can click on the link-out icon and it will open up the project page in another browser tab. There you can see that this is the Asia data from the <u>Malaria Atlas Project</u>. Click through to the publication page ("Journal" link is the quickest) and you could read there that each data point represents the *presence* of a local population of a particular species on a particular date (or date range) using one or more specific collection protocols.

Publications

A global map of dominant malaria vectors.

Sinka ME, Bangs MJ, Manguin S, Rubio-Palis Y, Ch Hay SI.

published PubMed Journal

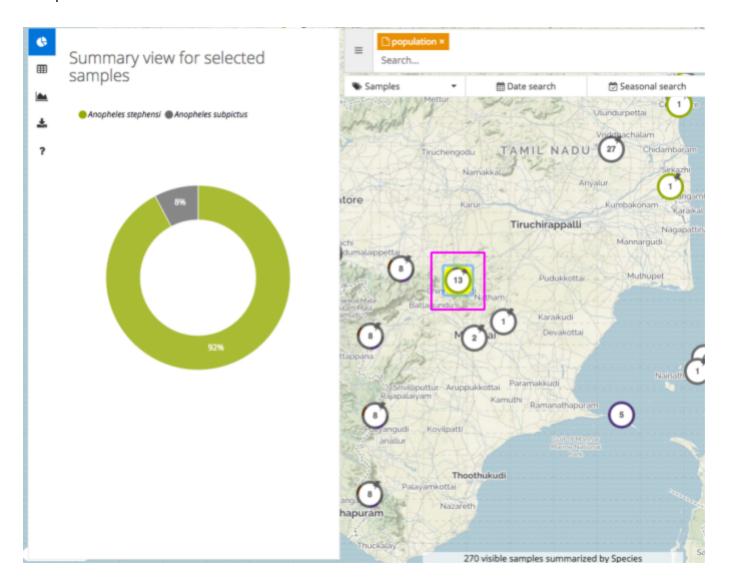
Let's go back to the main map:

If you wanted to show only the "population" samples in the PopBio map display, you can refine your search by typing "population" into the search box and selecting "population in Sample type" from the autocomplete suggestions. Stay zoomed in to southern India and cancel the two species search terms (click the small 'x' next to them). When no species are searched for, *all* species are shown.

6. Zoom in and out of Southern India and Sri Lanka, clicking on the markers to show the pie charts. What is the *largest number of different species* you can see in *one pie chart* for Southern India or Sri Lanka?

	Answer
4	
8	
9	

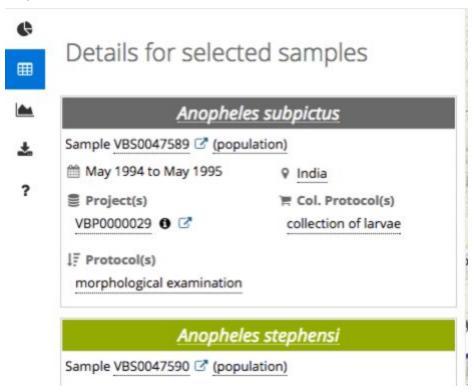
7. As shown below, the marker near the town of Tiruchirappalli contains 13 population samples, but only two species.



Which is the correct explanation of the existence of 12 An. stephensi samples?

	Answer
The habitat is perfect for Anopheles stephensi.	
If you zoom in far enough you can see that there are 12 different collection sites.	
The 12 samples were collected on different dates.	

Hint:



Insecticide resistance data

Clear the search, reset the zoom and switch to **Insecticide resistance phenotypes view**. Or you can go back to the PopBio home page and click on the insecticide spray can icon.



Zoom in to Central and South America and make sure you can see a blue marker with 76 samples over Puerto Rico, and a red marker with 6 samples in North East Brazil.

Click the "i" icon in the bottom right corner of the map. This shows the species color key (also available in Sample view mode) and a resistance color scale from blue (susceptible) to red (resistant). More info on the color scale in the panel below:

RESISTANCE COLOR SCALE

VectorBase's PopBio resource contains insecticide resistance data from different assay protocols and reported in a variety of measures and units, such as percent mortality, lethal concentration (e.g. LC50) and lethal time (e.g. LT95). Not all of these protocols and measures have WHO-recognized thresholds for categorizing into "susceptible" or "resistant" classes.

To aid the user in discovering geographical regions of resistance we have rescaled all comparable data (see below) between 0 (susceptible) and 1 (resistant) after discarding data outside the 2nd and 98th percentiles, and inverting value ranges where appropriate. These rescaled values are used to color the map markers (from blue to red).

COMPARABLE DATA

By comparable data we mean all phenotypes recorded with the same measurement type and units. For example, LC50 measured in parts per million or LT95 measured in minutes. Note that comparable data currently includes assays performed using different concentrations of insecticides. Thus the data is not strictly comparable. However the insecticide concentrations are displayed in info panel popups for plots showing individual data points. The search facility to limit displayed data to particular insecticide concentrations can be accessed with the three line icon on the left hand side of the search box.

Mouse-over the Puerto Rican '76' marker. The grey box that appears shows the GPS coordinates of the region in which the 76 samples are found. Double click on the marker to zoom in to show the 76 markers... You may want to hide the color legend panel if it is hiding some markers.

8. How many different species from Puerto Rico have IR phenotypes in VectorBase at the moment?

9. Briefly switch to "Samples view". This shows you where all PopBio samples have been collected, not just those assayed for insecticide resistance phenotypes.

Switch back to IR phenotypes view. Don't be alarmed that the species color scheme changes. It is supposed to do this.

Now click on the "10" marker on Puerto Rico's northern coast and **select the graph/plot icon on the left hand panel**. You will see IR phenotype data for the clicked "selection" marker plotted against "background" data. The background data can be defined in three different ways but we will come back to that. Leave it on the default "phenotypes matching search".



4

The marker contained 10 phenotypes but only 5 are plotted - LC50 milligram per liter. The second drop-down menu takes you to another plot for LC90 milligram per liter. You can see that in total there are plots for 10 phenotypes.

10. Mouse-over the points on the right hand plot for LC50. There are five different insecticides used in the assays. Fill in the missing insecticide.

malathion	fenitrothion	chlorpyrifos	temephos	
		-		

11. Now you are going to restrict all displayed data to only those assays using malathion as the insecticide.

Which of the following methods achieve this?

	Works	Does not work
Shout "malathion" at the computer.		
Type "malath" in the query box and use autocomplete.		
Find the details of a malathion assay in "Details for selected samples" and click on the word "malathion".		
Click on a malathion data point in the plot and then click on the word "malathion" in the popup data window.		

Now the map shows the relative picture of malathion resistance in the area.

You can also search for insecticides using higher order chemical classes such as "organophosphate" and "pyrethroid". Try that out before going back to a simple search for "malathion".

View the plots (LC50 and LC90) for the two phenotypes with the highest resistance (the red marker). You can now see that the background plots only show malathion data. There is less data than before, so now the plots can show individual data points.

12. At least for LC50, you can see that this assay suggests relatively high resistance. Only two other samples in the database have higher LC50 values. Where and when are those samples from? Fill in the missing table entries:

Place	Date
	1989

Trinidad and Tobago

Note that clicking on the data points makes the popup info panels sticky.

Note that the LC50 background data points include assays performed on other species.

13. Restrict the search to "Aedes aegypti in Taxonomy"

Now you can see that there are relatively few (n=) background data points with the same combination of measurement type (LC50, milligrams per liter), species and insecticide.

Click on the 3 marker at the eastern end of the island. Now you can see data for LC50, LC90 and LC95, all reported in parts per million. Again this assay suggests relatively high levels of resistance and this is reflected by the lighter blue coloring of the marker.

At this point you may be wondering if it is meaningful to compare Central American assay results with those from Australia. Luckily, you can restrict the background data to those shown in the current map view (and therefore also matching the current search) by selecting "phenotypes visible on map" in the upper menu of the plot panel. As expected, this also reduces the number of data points plotted quite drastically. A third background data option "all phenotypes" ignores both the search query and the current view and shows all "compatible data".

Now take a moment to reflect on how the IR plots can change when you move around the map and change your search terms.

14. Clear the search and zoom out to show Brazil again. Zoom into the area around the red 6 until the orange 7 below it splits up into smaller markers. Which insecticide is showing high levels of resistance? **<u>Hint</u>**: mouse-over the points in the plot to see this or consult the marker table display

Note: we have noticed a technical problem with the display of insecticide concentrations (they are missing in some cases, please let us know if this is your case).

Restrict the search to this insecticide and also *Aedes aegypti* and zoom out to see the global picture.

Reset the search and add *Aedes albopictus* as a search term. You can also search/filter on the type of insecticide resistance assay performed. Enter "dose" and select the general term "dose response test *in Protocols*". This will now show only lethal concentration-type (e.g. LC50) results.

As you can see, after a few keystrokes and mouse clicks, you can get a picture of insecticide resistance for the specific species and/or insecticides of interest. Current data includes the <u>President's Malaria Initiative (PMI)</u> and other sources, future plans include adding more *Anopheles* and *Aedes* data. These data, the date-based search and filtering, makes it a very simple and powerful tool.

Other map features you can try

1. You can change the map background style (see the layers icon). In the future we will add layers for vegetation type, climate and epidemiology.



- 2. Zoom to a rectangle with shift-click-drag
- 3. Search on collection protocols (try typing "catch" or "human")
- 4. Text-based search on geographical regions e.g. "Europe" (but note that some data points may be missed due to issues with data annotation quality)
- 5. Color coded pie charts based on other annotations, not just species.



6. Integration with VectorBase site search

If you need help with any question and its answer contact us at info@vectorbase.org. Because VectorBase data, tools and resources are updated every two months (6 release cycles per year), answers to these exercises will change too.