

SEQUENCHER[®]

Tutorial for Windows and Macintosh

Translated Variance Table

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Translated Variance Table

The **Translated Variance Table** enables you to compare the differences between your exemplar (model or primary) sequence and other sequences you select. It is the ideal tool for scanning and evaluating your findings at the amino acid level. The **Translated Variance Table** directs your attention to the putative effects a base difference has on a codon and its associated amino acid. It saves you time and increases your accuracy in analyzing SNPs, mutations, heterozygotes or other significant coding changes.

You use the **Translated Variance Table** to compare differences among translations of sequences in the same contig. This type of **Variance Table** will be explored in the first part of the tutorial.

You use the **Translated Variance Table** to compare differences between the translations of consensus from multiple contigs that have all been assembled to a common Reference Sequence. This type of **Variance Table** will be explored in the second part of the tutorial.

In this tutorial, you will learn how to use the **Translated Variance Table** and how to interpret its results. We will describe how to perform these functions using **Sequencher's** menu commands and button bars. You may also invoke commands using your right mouse button. The sample data provided in the tutorial project has already been trimmed and Assembled by Name to a Reference Sequence using the command To Reference by Name.

CREATING A TRANSLATED VARIANCE TABLE FROM A SINGLE CONTIG

The first table you are going to generate will show you what happens when you compare sequences within the same contig to an exemplar. Once you start working with your data you will need to decide which sequence will act as the exemplar and which sample sequences you want to compare to it.

- Launch **Sequencher** to open an Untitled Project.
- Go to the **File** menu and select **Import > Sequencher Project...**
- Navigate to the **Sample Data** folder inside the **Sequencher** application folder.
- Select the **Translated Variance Table** project from the **Sample Data** folder and select **Open**.

The **Translated Variance Table** project in this example contains 13 contigs. One contig contains exon data from 10 individuals. The other contigs contain multiple exon data from 12 individuals and will be used in the second half of this tutorial.

Sequencher gives you three choices for the exemplar sequence. You can choose the Reference Sequence, the Consensus sequence, or the sequence at the top of your contig sequence list.

- Choose **Contig > Consensus Inclusively**.

- Select the contig called **Exon 11**.
- Go to the **Sequence** menu and select **Compare Translation To > Reference Sequence**.

The **Translated Variance Table** appears. Note that in some cases, if the names of the individual sequences are long, they appear truncated in the table view.

Description: 11 sequences compared to Reference Reference with 3 exons of contig Exon 11.

Comparison Range: Unfiltered

Base Positions: 14587..18495

Amino Acid Positions: 1..1303

Reference		Re...	94	138	152	93	116	130	150	128	156	131	Total
17,458 958	AGC S	AGC S	AGC S	AGC S	AGG R	AGC S	AGS ?	AGS ?	AGS ?	AGS ?	AGG R	AGG R	7
17,770 1,062	ACT T	ACT T	ACT T	AMT ?	ACT T	ACT T	ACT T	ACT T	AMT ?	AMT ?	ACT T	ACT T	3
17,785 1,067	TGG W	TGG W	TGA .	TGA .	TGA .	TGR ?	TGA .	TGA .	TGA .	TGA .	TGA .	TGA .	10
⬆	Total	0	1	2	2	1	2	2	3	3	2	2	20

In the table, the column order is the same as the original contig order of your sequences. To interpret the table, you need to remember that each row represents a base position and each column represents a sample. The first row contains the names of the sequences. The first column shows the position of each variant found as a base position and a codon position. The cell in the equivalent position in the last column of the table indicates the total number of variants for that position. The first row, position 17,458 and codon 958, has 7 variants out of 11 samples the second has 3 and the third has 10. The last row of the table shows the total number of variants for each sample.

The total number of variants in the entire table is given in the cell at the bottom right hand corner of the table. In this table, the total number of variants is 20.

You will notice pink headers in your **Translated Variance Table**. These indicate that the sequences in those columns do not span the entire length of the exemplar sequence. As you explore the tables you generate with your own data, you will notice that any time a sample sequence does not have a base for the equivalent position in the exemplar, the cell will have a pink cross in it.

As you browse through the table, you will notice that some cells have lettering in black while others are in gray. Cells with black lettering contain variants. Cells with gray lettering are identical to the Reference Sequence. The difference in letter color helps you quickly focus on the variants as you scan the table.

- Scroll to the top of the table.
- For Sample Column 152, locate the variant at base position 17,458, which is equivalent to residue 958, and select the cell.

The sample sequence contains the codon AGG, as opposed to the Reference Sequence, which has an AGC codon at this position. This change substitutes an Arginine for the original Serine. This difference could prove to be interesting since Serine is a smaller polar amino acid while Arginine is a larger strongly basic amino acid.

Reference		Re...	94	138	152
17,458 958	AGC S	AGC S	AGC S	AGC S	AGG R

You can sort the table to see which sequence has the most variants.

- At the bottom left-hand corner of the table is a **Total** button. Click on it once.

When you do this, the table will be rearranged. The sequences with the most variants will be on the left-hand side. Those with the least will be on the right-hand side.

Reference		128	150	131	156	130	116	152	Total
17,458 958	AGC S	AGS ?	AGS ?	AGG R	AGG R	AGS ?	AGS ?	AGG R	7
17,770 1,062	ACT T	AMT ?	AMT ?	ACT T	ACT T	ACT T	ACT T	ACT T	3
17,785 1,067	TGG W	TGA .	TGA .	TGA .	TGA .	TGA .	TGA .	TGA .	10
⬆	Total	3	3	2	2	2	2	2	20

Now suppose you wanted to see which sequence had the fewest variants.

- Click on the same **Total** button again.

The table will be rearranged again, only this time the sequences with the fewest variants will be on the left-hand side.

Reference		Ref...	94	93	138	152	116	130	Total
17,458 958	AGC S	AGC S	AGC S	AGC S	AGC S	AGG R	AGS ?	AGS ?	7
17,770 1,062	ACT T	ACT T	ACT T	ACT T	AMT ?	ACT T	ACT T	ACT T	3
17,785 1,067	TGG W	TGG W	TGA .	TGR ?	TGA .	TGA .	TGA .	TGA .	10
⬆	Total	0	1	1	2	2	2	2	20

You can sort the rows of the table the same way clicking on the **Total** button in the top right hand corner of the table.

It is often the case that you have very long sequence names. You can resize the column widths all at once or individually.

- At the bottom left-hand corner of the table is a resize button . Click on it once.

Reference		Reference with 3 exons	94	93	138	152	Total
17,458 958	AGC S	AGC S	AGC S	AGC S	AGC S	AGG R	7
17,770 1,062	ACT T	ACT T	ACT T	ACT T	AMT ?	ACT T	3
17,785 1,067	TGG W	TGG W	TGA .	TGR ?	TGA .	TGA .	10
↕	Total	0	1	1	2	2	20

- Click on the resize button again.

Reference		...	94	93	Total
17,458 958	AGC S	AGC S	AGC S	AGC S	AGC S	AGG R	AGS ?	AGS ?	AGG R	AGG R	AGS ?	AGS ?	7
17,770 1,062	ACT T	ACT T	ACT T	ACT T	AMT ?	ACT T	ACT T	ACT T	ACT T	ACT T	AMT ?	AMT ?	3
17,785 1,067	TGG W	TGG W	TGA .	TGR ?	TGA .	TGA .	TGA .	TGA .	TGA .	TGA .	TGA .	TGA .	10
↕	Total	0	1	1	2	2	2	2	2	2	3	3	20

Notice how all the columns were resized each time you clicked the button. Use this button any time you want to check on the sequence name in the column headers or to squeeze more columns into the current table view.

- Move your cursor over the line between two column headers until the cursor changes shape.
- Click and hold down the mouse button.
- Move the mouse left or right while holding down the mouse button.

Notice how the column on the left-hand side was resized as you moved the cursor.

Reference		...	94	93	...	152	↕	Total
17,458 958	AGC S	AGC S	AGC S	AGC S	AGC S	152 589 BPs, AutoSeq Frag, ABI		AGG R	AGG R	AGG R	AGG R	AGS ?	AGS ?	7
17,770 1,062	ACT T	ACT T	ACT T	ACT T	AMT ?	ACT T		ACT T	ACT T	ACT T	ACT T	AMT ?	AMT ?	3
17,785 1,067	TGG W	TGG W	TGA .	TGR ?	TGA .	TGA .		TGA .	TGA .	TGA .	TGA .	TGA .	TGA .	10
↕	Total	0	1	1	2	2		2	2	2	2	3	3	20

- Click on the resize button  one more time to return the column widths to the original state.

CHANGING THE TRANSLATION RANGE

The great power of the **Translated Variance Table** lies in its ability to use sequence features to restrict the range of comparison between the exemplar and sample sequences. In the first section of this tutorial, the

comparisons were performed along the entire length of the Reference Sequence. In the next section of this tutorial, you will use a sequence feature to restrict the Comparison Range.

The Reference Sequence being used here, as the exemplar, is a stretch of genomic sequence from chromosome 22. It has a number of features, represented by different feature keys, marked on it. Each feature has several attributes, including a name, a location, and a feature key. You will use the CDS or coding sequence feature to restrict the Translation Range and direct your focus to variants that result in changes to the translation.

- Click on the **Translation Range** button in the **Translated Variance Table** button bar.
- Click on the **Filter Translation by** radio button.
- Choose **CDS** from the **Feature Key:** drop-down menu.
- Choose **HPS4a** from the window below the drop-down menu.
- Click **OK**.

You will see that a new **Translated Variance Table** has been generated. This table is very different from the first table. There are now 5 rows of differences rather than the original unfiltered 3. The differences reported in this new filtered table accurately reflect the joined reading frames of the three exon features in the reference.

Whenever you import a sequence from GenBank, it will include a feature table that **Sequencher** can read. You may have marked features on your exemplar yourself. In both cases, you can use the Translation Range function to restrict the comparison and focus on differences between your sequences in detail.

Notice that in some cells there is a question mark (?) instead of an amino acid residue. If you examine the codon in these cells you will see that one of the normal bases (A,G,C,T) has been replaced by an ambiguity code. For example, in several samples at base position 17, 586 there is an M in the first position of the codon. M is the IUPAC code for A or C. The equivalent codons would be ACG or CCG. Later in the tutorial you will learn how to review this data and distinguish between true SNPs and base calling errors.

- Close the **Translated Variance Table**.

CHANGING THE EXEMPLAR SEQUENCE

Sequencher gives you three choices for your exemplar: the Reference Sequence, the consensus or the top sequence in your list. You can use any sequence as your exemplar simply by simply dragging it to the top of your list of sequences in the Contig Editor and choosing the Compare Translation To > Top Sequence command.

- Double-click on contig **Exon 11** to open the **Contig Editor**.
- Click on the **Bases** button.
- Maximize the **Contig Editor**.
- Locate the sequence **131** in the list at the bottom left-hand side of the **Contig Editor**.
- Click on the name of this sequence. Note that the hand icon closes as you click.
- While holding the mouse button down, drag the sequence name to the top of the list, above the Reference.

- From the **Select** menu, choose the **Select All** menu item to select the names of all of the sequences.
- Go to the **Sequence** menu and select **Compare Translation To > Top Sequence**.

The **Translated Variance Table** opens. Notice that the Description at the top of the table tells you which sequences you used and which sequence was the exemplar in this case.

Description: 11 sequences compared to sequence 131 of contig Exon 11.

Comparison Range: Unfiltered

Base Positions: 17288..17847

Amino Acid Positions: 1..186

Top Sequence		131	Ref...	94	138	152	93	116	130	Total
17,459 58	GGT G	GGT G	GCT A	GCT A	GCT A	GGT G	GCT A	GST ?	GST ?	8
17,585 100	CAC H	CAC H	CAC H	CMC ?	CMC ?	CAC H	CAC H	CAC H	CAC H	4
17,648 121	TCT S	TCT S	TCT S	TCT S	TYT ?	TCT S	TCT S	TCT S	TCT S	3
17,714 142	CAG Q	CAG Q	CAG Q	CAG Q	CMG ?	CAG Q	CAG Q	CAG Q	CAG Q	1
⚡	Total	0	2	2	5	0	2	1	1	21

You are now able to explore putative differences from the perspective of your new exemplar. The ability to change to a different exemplar is particularly useful in any type of comparative sequence analysis.

- Close the **Translated Variance Table**.
- Quit **Sequencher**.

CREATING A TRANSLATED VARIANCE TABLE FROM MULTIPLE CONTIGS

In many instances, you may want to work with data from different sample sources. Perhaps you are in a laboratory setting and are screening samples from different sources. Perhaps you are studying evolutionary changes among related species. You may be simply re-sequencing a cloned gene and want to make sure that no changes have occurred in a coding region. The commonality among these cases is that you have an established prototype sequence and are working with the consensus sequences from different contigs.

In the next section of the tutorial, you will generate a **Translated Variance Table** using the **Compare Translation to Reference** command from the **Contig** menu. You will be working with an established prototype sequence and are working with the consensus sequences from different contigs.

- Launch **Sequencher**.
- Go to the **File** menu and select **Import > Sequencher Project...**
- Navigate to the **Sample Data** folder, choose the **Translated Variance Table** project and select **Open**.

The project contains 12 contigs from separate individuals. The auto-sequencing data comes from different human samples and spans three exons of a gene in chromosome 22.

When you import a **Sequencher** project, all of the imported items are selected.

- Use **Apple+Click** on a Mac or **Ctrl+Click** on a PC to deselect the **Exon 1 1** contig.

Remember that the original sample sequences were assembled To Reference by Name so each contig already carries a copy of the Reference Sequence.

- Go to the **Contig** menu.
- Choose **Compare Translation to Reference**.

The **Translated Variance Table** appears in a new window. Each column represents a contig. The contig name is contained in the column header. To interpret the table, you need to remember that each row represents a consensus position but only displays this in black where there is a difference. The two left-most columns display the base position of the first base in the variant codon and the number of the variant amino acid. The right-most column displays the total number of variants for that position.

Notice the Description tells you how many consensus sequences are in the table and indicates the Reference Sequence to which they were compared. In most respects, the **Translated Variance Table** has the identical descriptions, sorting and navigation functions as the **Variance Table**.

Description: 12 consensus sequences compared to Reference Reference with 3 exons.
Comparison Range: Unfiltered
Base Positions: 14587..18495
Amino Acid Positions: 1..1303

The information included in the table header provides information about the contents of the **Translated Variance Table**. The Description gives the total number of samples, sequences or contigs, included in the comparison and indicates which sequence is the exemplar. In this example, the exemplar sequence is called "Reference with 3 exons". The Comparison Range provides the status of the comparison range filter. The default setting for all **Translated Variance Tables** is Unfiltered. The table header also displays both the base and amino acid positions included in the comparison.

REVIEWING THE DATA

Now look at your data in detail. You will notice that each of the header cells is shaded in pink. This indicates that the sequence does not cover the full comparison range. In addition, some of the cells contain a pink X. This indicates that there is no base coverage for the sequence at this position.

Description: 12 consensus sequences compared to Reference Reference with 3 exons. Comparison Range: Unfiltered Base Positions: 14587..18495 Amino Acid Positions: 1..1303											
Reference		116	128	130	131	138	150	152	156	35	Total
15,400	TGG		AGT	AGT	AGT	AGT	AGT	AGT	AGT	AGT	10
272	W		S	S	S	S	S	S	S	S	
15,403	CTA		GST	GCT	GCT	GCT	GCT	GCT	GCT	GST	10
273	L		?	A	A	A	A	A	A	?	
15,406	AGT		AAG	AAG	AAG	AAG	AAG	AAG	AAG	AAG	10
274	S		K	K	K	K	K	K	K	K	
15,412	ATG	ATG	TAT	TAT	TAT	TAT	TAT	TAT	TAT	TAT	10
276	M	M	Y	Y	Y	Y	Y	Y	Y	Y	
⚡	Total	97	177	170	172	176	175	177	174	170	1928

In the first few rows of the table, such as base position 15,400, each sample varies from the Reference. You can see this even without scrolling across the table because the gray Total column in the table has a ten (10) in each of the equivalent rows.

All of the samples have a Serine (S) residue at position 15,400 instead of a Tryptophan (W) residue. If this alteration were in a coding region, it would indicate a significant change in the protein sequence since the base difference is affecting the aromatic Tryptophan residue.

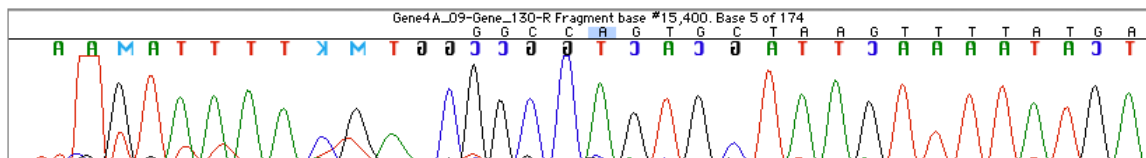
- Double-click in the cell containing a Serine at base position 15,400 in sample 130.

Two more windows appear. They are the **Contig Editor** and the **Contig Chromatogram**. Arrange the windows to suit your viewing preference. If you would like to preserve the Review function with the windows in the same relative positions, do the following.

- Go to the **Window** menu.
- Choose **Remember Window Layout > Single Variance Table Review**.

Now that the three windows are arranged on your screen, explore the data. In the **Translated Variance Table**, you have **AGT** representing the codon for Serine. Looking at the **Contig Editor** you can see your Reference Sequence at the top of the editor window. The codon **TGG** is highlighted. Notice that a gap has been introduced to maintain the maximum number of matching bases.

For this contig, there is only one sequence covering this position and therefore only one trace at this position. This base falls at the 5' end of the sequence close to where the data has been trimmed.



In addition to the trace data, the blue highlighting of the bases also contributes to the analysis. The darker the shade of blue, the lower the reliability of the base call. In summary, there is a degree of uncertainty for this position

because there is no trace in the opposite direction to confirm the finding, and because there is a higher concentration of bases with low confidence. Note also that the codon for the consensus sequence, **AGT**, and for the Reference, **TGG** are highlighted in yellow. In this codon, there are two base differences that result in a single amino acid difference.

TGTGGTTGGGACAAGCTTTCTGGGCC TG :GCTAAGTTTTATGAACAGATGGCAC
GGCCAGTGCTAAGTTTTATGAACAGATGGCAC
70 15380 15390 15401.1 15410 15420
.....GGCC AGT GCTAAGTTTTATGAACAGATGGCAC

- Use the left arrow key to move through the table.

As you do this, you will see the information in the **Contig Editor** and the **Contig Chromatograms** change. Remember each column represents the consensus for a single contig. As you move from cell to cell, you are seeing the underlying data for each contig at that base position. Cells that contain a pink X will have no chromatogram data.

- Note the similarity in the data in each of the samples with coverage at base position 15,400.
- Close the **Contig Editor** and **Contig Chromatogram** windows.

CHANGING THE REGION OF INTEREST

When a GenBank sequence is imported into **Sequencher** for use as a Reference Sequence, its Feature Table is read and any joined features such as CDSs or Exons are noted. When you first construct a **Translated Variance Table**, the default comparison range is the full length of the exemplar sequence. In some instances, this will be exactly what you want. However, the sequences used in this tutorial example are derived from genomic DNA and the Reference Sequence is marked up with the relevant coding regions. It would be more scientifically relevant to restrict the table to these regions.

- Click on the **Translation Range** button in the **Translated Variance Table** button bar.
- Click on the **Filter Translation by** radio button.
- Choose **CDS** from the **Feature Key**: drop-down menu.

Feature Key:

HPS4alight ear protein isoform a CDS, 3 features

HPS4alight ear protein isoform a CDS:
15501..15537 16212..16308 16937..17846

The only CDS feature is selected in the Feature list.

- Hover your mouse over feature **HPS4alight ear protein isoform a CDS, 3 features** to trigger a tooltip that describes the feature and the location of the three coding regions.
- Click **OK**.

You will see that the **Translated Variance Table** has been refreshed. Note the changes in the header of the table. The Comparison Range has changed from Unfiltered to the name of the feature. Notice that Base Positions: now lists three discontinuous ranges, equivalent to the three CDS features in the Reference Sequence.

Description: 12 consensus sequences compared to Reference Reference with 3 exons.

Comparison Range: Filtered by HPS4alight ear protein isoform a CDS

Base Positions: 15501..15537 16212..16308 16937..17846

Amino Acid Positions: 1..348

The filtered **Translated Variance Table** reduces the original number of differences from 1928 to a total of 46 variants. Look at your data in detail. Find the base position with the most variants by looking at the right-most column.

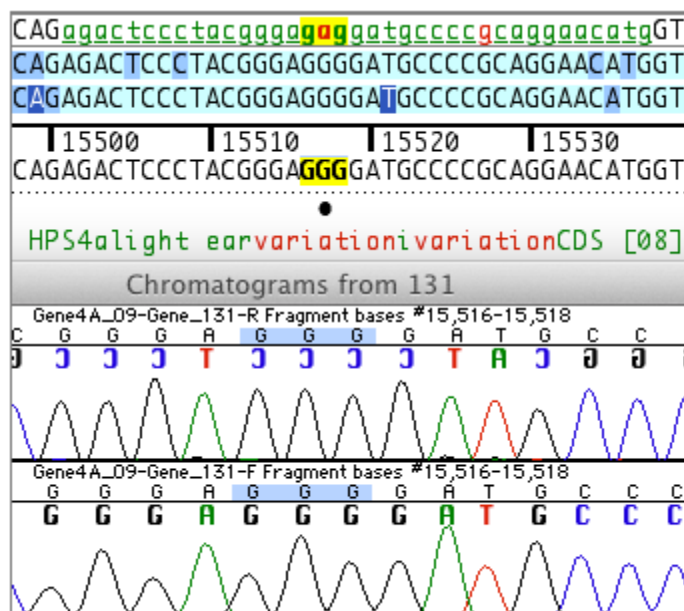
- Click on the **Total** button in the top right-hand corner.

The table is now sorted so that the position with the most variants is in the top row. The total number of variants for this position (base position 15,516) is 12. In the majority of cases, the predicted change is from Glutamic Acid to Glycine. Remember that, in this table, you are looking at consensus sequences.

- Select the top cell in sample 131 and double-click on it.

In the **Translated Variance Table**, you have **GGG** representing the codon for Glycine. This has been substituted for the original Glutamic acid residue in the Reference Sequence. A smaller, neutral, non-polar residue has replaced the acidic, polar residue.

Looking at the **Contig Editor**, you can see the Reference Sequence at the top of the editor window. The codon **GAG** (for Glutamic Acid) is highlighted in yellow, but the sample consensus sequence has **GGG** highlighted in yellow. Your selection extends from the first base of this codon down through the sequences providing coverage in the contig and through to the consensus sequence. You can see that the first "G" consensus call is supported by both sequences at this position in the contig and the Reference. In the second base of the codon, the Reference has an "A", but the sample sequence has a "G". The background for both supporting bases is pale blue, indicating that these are high quality base calls.



The traces in the **Contig Chromatograms** window provide further support. You have two overlapping sequences. One is in the forward orientation and one is in the reverse orientation. Notice that the original base calls in the top chromatogram, displayed in color below the current base calls, are in mirror image. This is a visual cue indicating that the sequence is in the reverse orientation.

There are three clear peaks for the “GGG” codon at this position for both sequences. You have confirmed the variant.

In the Reference Sequence, the variant base is in red, distinguishing this position as a previously characterized variant. More details on the variant are available from the **Sequence > Feature Listing** or from the **Sequence > Edit Features...** commands.

- In the **Contig Editor**, click on the name of the Reference Sequence.
- Choose **Sequence > Feature Listing**.
- Scroll to the **Variation** feature at position **15517**.

```
Name: variation
Key: variation
From: 15517 To: 15517 Strand: Complement Color: Red Style: Underline
/gene="HPS4"
/replace="c"
/replace="t" /db_xref="dbSNP:713998"
```

This Feature Listing indicates that you have found a new “G” variant. The previous variants at this position included only “C” and “T” substitutions.

- Close the **Feature Listing** window.

SISTER TABLES

Throughout this tutorial, you have used the **Contig Editor** and the **Contig Chromatograms** command to support the findings you have made using the **Translated Variance Table**. You can simultaneously view the sister **Variance Table** alongside the **Translated Variance Table**, and you can generate Sister Tables with or without the editors available in Review mode. Sister Tables share the same comparison range.

- Go to the **Translated Variance Table** button bar.
- Click on the **Bases** button.

Arrange the windows to suit your viewing preference. If you would like the windows to be arranged this way every time you use the Sister Tables in Review mode, follow these steps.

- Go to the **Window** menu.
- Choose **Remember Window Layout > Double Variance Table Review**.

Now you can see four windows on your screen, the **Variance Table**, the **Translated Variance Table**, the **Contig Editor**, and the **Contig Chromatogram**. There are several advantages to this layout. The **Variance Table** displays not only the individual bases but also shows any feature information. This is displayed as a color and text style. You control default styles through the Feature, Motif User Preferences. You can also apply individual styles.

- Go to the **Bases Variance Table**. Its window is titled **Compare Consensus to Reference**.
- Select the cell at base position 17,158 in sample 116.

This action opens the **Contig Editor** and the **Contig Chromatogram** for this sample and this position. The font of the base is lower case, underlined and green, indicating that the base is part of an exon, but the lack of red color indicates that this is the position of a new variant. With your selection still in the **Variance Table**, column 116, row 17,158:

- Choose **Sequence > Mark Selection As Feature**.
- Check the **Feature Key: variation**.
- Change the **Feature Name:** to **variation new**.
- Change the **Feature Color:** to **cyan**.
- Click **OK**.

The style is immediately applied to the selected base and to the underlying sequence data in contig 116. You can see the name of the feature appear below the consensus line in the **Contig Editor**.

gcccgcaggactgcaaacctctgccagggg									
GCCCCGAGGACTGCAAACTCTGCCAGGGG									
40				17150			17160		171
GCCCCGAGGACTGCAAACTCTGCCAGGGG									
.....variation new.....									

- Go to the **Translated Variance Table**. Its window is entitled **Compare Translation to Reference**.
- Locate the column header called 116. Click on the column header.
- Go to the **Edit** menu. Choose **Label > Label 2**.

This will change the text color of the header in both **Variance Tables**, giving you a visual cue about the status of the sample. You can set up names for the labels in the Label & Name User Preferences.

Reference		116	Total	Reference		116	Total
15,517	A	g	12	15,516	GAG	GGG	12
16,277	C	y	5	6	E	G	
17,061	G		1	16,277	CTC	YTC	5
17,158	A	r	1	35	L	?	
17,229	G		1	17,061	GAT	GAT	1
17,353	G	k	1	87	D	D	
17,419	C		1	17,157	CAC	CRC	1
17,460	C	s	7	119	H	?	
17,586	A		4	17,229	GAA	GAA	1
17,649	C		3	143	E	E	
17,715	A		1	17,352	AGC	AKC	1
17,771	C		3	184	S	?	
17,787	G	a	10	17,418	TCT	TCT	
⚡	Total	6	50	⚡	Total	6	46

- Close the project from the **File** menu by choosing the **Close Project** command.

CONCLUSION

The **Variance Table** gives you a summary analysis of your data that focuses on differences. Not every base difference will cause a change in the amino acid. The **Translated Variance Table** lets you see the impact of a base change immediately and check the validity of any codon change you observe in the underlying sequence data. Working with the Translation Range allows you to focus on a feature of scientific interest such as a coding sequence (CDS), even when it is interrupted by regions of non-coding sequence.

The **Translated Variance Table** saves you time and allows you to double-check your work as you go along, making your efforts to track variants more efficient and relevant to your project.

You can find out more about the **Variance Table** and the **Translated Variance Table** in the manual and other tutorials in this series.