

## USING THE ONCOTREE PACKAGE

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ABSTRACT. This paper shows a short example of building and exploring oncogenetic trees using the Oncotree package. A detailed description of the theory of oncogenetic trees can be found in

- Desper, R.; Jiang, F.; Kallioniemi, O.; Moch, H.; Papadimitriou, C. and Schäffer, A. A. “Inferring tree models for oncogenesis from comparative genome hybridization data.” *Journal of Computational Biology*, 1999, 6, 37-51.
- Szabo, A. and Boucher, K. “Estimating an oncogenetic tree when false negatives and positives are present.” *Mathematical Biosciences*, 2002, 176, 219-236.
- Szabo, A. and Boucher, K. “Oncogenetic trees” in *Handbook of cancer models with applications* Tan, Hanin (ed.) World Scientific, 2008.

A short introduction is given in doc/Oncotree.pdf.

We start by loading a dataset. The package contains an example dataset:

```
> library(Oncotree)
> data(ov.cgh)
> str(ov.cgh)
'data.frame':      87 obs. of  7 variables:
 $ 8q+: int  0 0 1 1 0 1 1 0 0 1 ...
 $ 3q+: int  0 0 1 0 0 1 1 1 1 0 ...
 $ 5q-: int  0 0 1 0 0 1 1 1 0 1 ...
 $ 4q-: int  0 1 1 0 0 1 1 0 0 1 ...
 $ 8p-: int  0 0 0 0 0 1 1 0 0 1 ...
 $ 1q+: int  1 1 0 0 0 0 0 0 0 1 ...
 $ Xp-: int  0 0 0 0 0 0 1 0 1 1 ...
```

Based on these data, we construct the oncogenetic tree using the default  $\ell_2$ -distance error function to estimate the false-positive and false-negative error rates.

```
> ov.tree <- oncotree.fit(ov.cgh)
```

The fitted tree can be examined several ways: printing it produces a quick summary, but the result of plotting is easier to interpret (the plots are shown in Figure 1).

```
> ov.tree
```

Oncogenetic tree from 7 events

Parent function:

```
8q+ <- Root
3q+ <- 8q+
5q- <- Root
4q- <- 5q-
8p- <- 5q-
1q+ <- Root
Xp- <- 8p-
```

Estimated error rates: epos= 0.2084556 , eneg= 0.0267696

```
> plot(ov.tree, edge.weights="est")
```

```
> pstree.oncotree(ov.tree, edge.weights="est", shape="oval")
```

We can compare the observed and fitted marginal occurrence frequencies of the mutations (the distance between these two was minimized for the error-rate estimation). The plot is shown in Figure 2.

```
> print(obs <- colMeans(ov.tree$data))
```

	Root	8q+	3q+	5q-	4q-	8p-	1q+	Xp-
1.0000000	0.7011494	0.5517241	0.5287356	0.5057471	0.4712644	0.4367816	0.4252874	

```
> print(est <- marginal.distr(ov.tree, with.errors=TRUE))
```

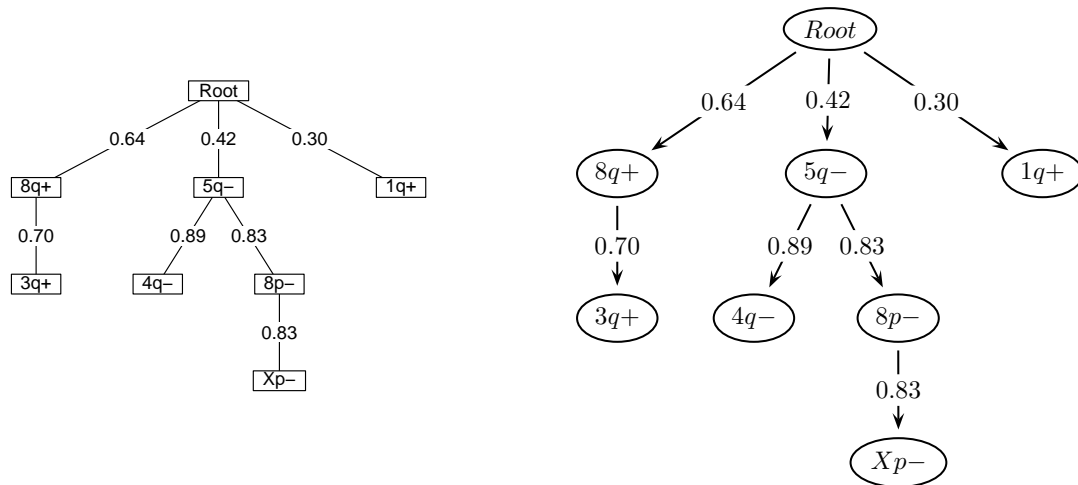
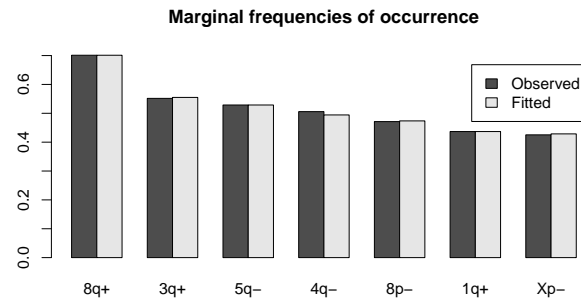
FIGURE 1. Fitted oncogenetic tree for the `ov.cgh` data set.

FIGURE 2. Observed and fitted frequencies of occurrence of each event.

```

Root      8q+      3q+      5q-      4q-      8p-      1q+      Xp-
1.0000000 0.7011494 0.5550202 0.5287356 0.4943105 0.4736503 0.4367816 0.4286807
> #plot is in Figure 2
> barplot(rbind(obs[-1],est[-1]), beside=T, legend.text=c("Observed","Fitted"),
+         main="Marginal frequencies of occurrence")

```

In addition to the marginal frequencies, it is possible to estimate the entire joint distribution generated by the tree:

```

> dd <- distribution.oncotree(ov.tree, with.errors=TRUE)
> head(dd)

```

	Root	8q+	3q+	5q-	4q-	8p-	1q+	Xp-	Prob
1	1	0	0	0	0	0	0	0	0.029222901
2	1	1	0	0	0	0	0	0	0.027992097
3	1	0	1	0	0	0	0	0	0.009202964
4	1	1	1	0	0	0	0	0	0.062160896
5	1	0	0	1	0	0	0	0	0.008323722
6	1	1	0	1	0	0	0	0	0.007973145

Observed – Expected probabilities of co-occurrence of events

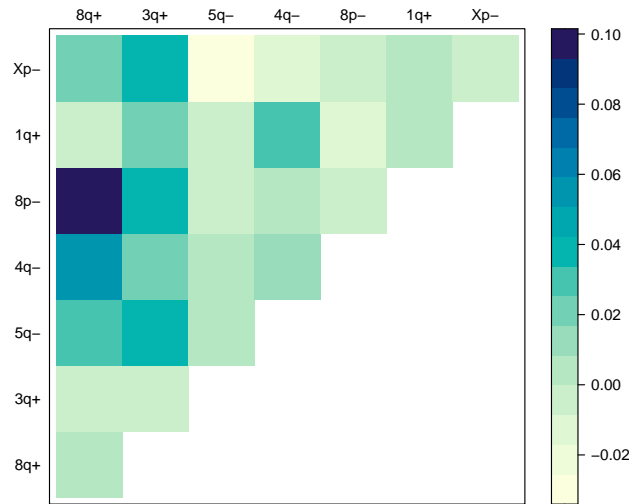


FIGURE 3. Goodness-of-fit plot: difference between observed and expected probabilities of two events being observed.

Using the overall joint distribution, it is straightforward to obtain marginal joint distributions (2- or higher way) if needed (the plot is shown in Figure 3).

```
> #estimated probabilities of 2 events
> print(est2way <- t(data.matrix(dd[2:8])) %*% diag(dd$Prob) %*% data.matrix(dd[2:8]))
      8q+      3q+      5q-      4q-      8p-      1q+      Xp-
8q+ 0.7011494 0.4834457 0.3707227 0.3465855 0.3320996 0.3062492 0.3005693
3q+ 0.4834457 0.5550202 0.2934589 0.2743523 0.2628855 0.2424226 0.2379265
5q- 0.3707227 0.2934589 0.5287356 0.3884206 0.3683135 0.2309420 0.3245477
4q- 0.3465855 0.2743523 0.3884206 0.4943105 0.3393380 0.2159057 0.2992688
8p- 0.3320996 0.2628855 0.3683135 0.3393380 0.4736503 0.2068817 0.3130649
1q+ 0.3062492 0.2424226 0.2309420 0.2159057 0.2068817 0.4367816 0.1872399
Xp- 0.3005693 0.2379265 0.3245477 0.2992688 0.3130649 0.1872399 0.4286807

> #observed probabilities of 2 events
> print(obs2way <- t(ov.tree$data[,-1]) %*% ov.tree$data[,-1]/nrow(ov.tree$data))
      8q+      3q+      5q-      4q-      8p-      1q+      Xp-
8q+ 0.7011494 0.4827586 0.4022989 0.4022989 0.4252874 0.2988506 0.3218391
3q+ 0.4827586 0.5517241 0.3333333 0.2988506 0.2988506 0.2643678 0.2758621
5q- 0.4022989 0.3333333 0.5287356 0.3908046 0.3678161 0.2298851 0.2988506
4q- 0.4022989 0.2988506 0.3908046 0.5057471 0.3448276 0.2413793 0.2873563
8p- 0.4252874 0.2988506 0.3678161 0.3448276 0.4712644 0.1954023 0.3103448
1q+ 0.2988506 0.2643678 0.2298851 0.2413793 0.1954023 0.4367816 0.1954023
Xp- 0.3218391 0.2758621 0.2988506 0.2873563 0.3103448 0.1954023 0.4252874

> oe.diff <- obs2way-est2way
> oe.diff[lower.tri(oe.diff)] <- NA #clear half of symmetric matrix for plotting
> require(lattice) #the plot is in Figure 3
> levelplot(oe.diff, xlab="", ylab="", scales=list(x=list(alternating=2), tck=0),
+           main="Observed - Expected probabilities of co-occurrence of events")
```

Another way to evaluate goodness-of-fit is through bootstrap resampling of the data. Two approaches are implemented: a parametric bootstrap that assumes that the model is correct and a non-parametric bootstrap. The plot is shown in Figure 4.

```
> set.seed(43636)
> ov.boot <- bootstrap.oncotree(ov.tree, type="nonparam", R=1000)
> ov.boot
```

Out of the 1000 replicates there are 279 unique trees with frequencies from 98 down to 1  
The bootstrap process found the original tree 98 times

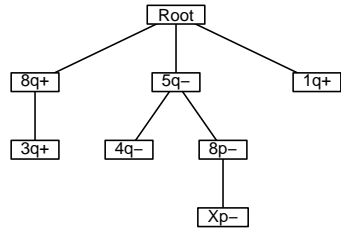
```
> opar <- par(mfrow=c(3,2)) #the plot is in Figure 4
> plot(ov.boot, minfreq=45)
> par(opar)
```

The non-parametric bootstrap gives an estimate of the reconstruction confidence: the original tree was obtained 83 times out of 1000 resamples, so the estimated confidence is 8.3%.

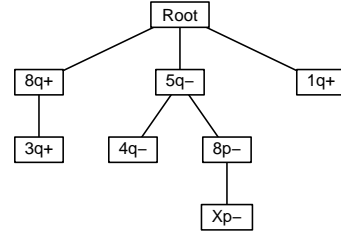
We can look at the frequency of edge occurrences in the bootstrapped trees:

```
> ov.boot$parent.freq
      Child
Parent Root 8q+ 3q+ 5q- 4q- 8p- 1q+ Xp-
      1000  0  0  0  0  0  0  0  0
Root   0 998  53 524 232  3 808  66
8q+   0  0 943  96 131 386  21  11
3q+   0  2  0  35  5  0 100  44
5q-   0  0  4  0 540 419  9 175
4q-   0  0  0 261  0 139  51 119
8p-   0  0  0  80  85  0  1 584
1q+   0  0  0  0  1  0  0  1
Xp-   0  0  0  4  6  53 10  0
```

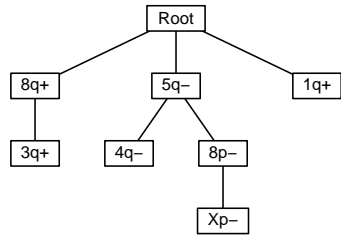
It is clear that some edges are really stable: Root  $\rightarrow$  8q+, 8q+  $\rightarrow$  3q+, root  $\rightarrow$  1q+, all with confidence  $> 80\%$ , while other edges are less stable (for example, 8p- is the child of 8q+ about as often as of 5q-).



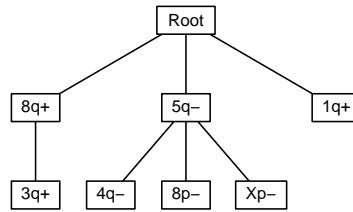
Original Tree



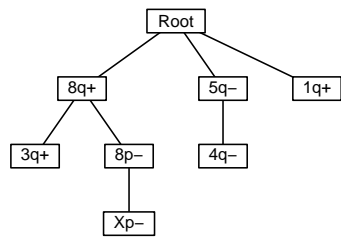
Tree based on most frequent parent



Observed Frequency = 98



Observed Frequency = 52



Observed Frequency = 51

FIGURE 4. The most frequently occurring bootstrap trees.