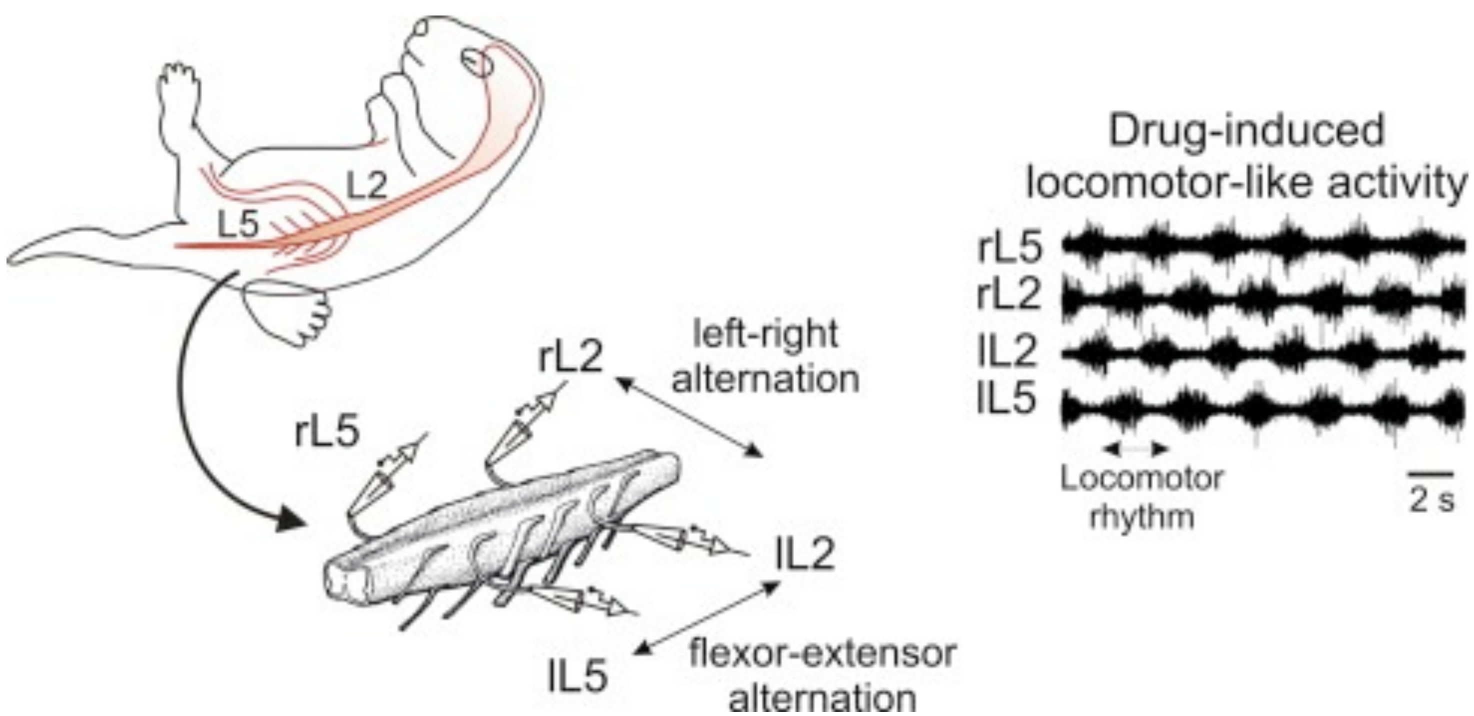
Circular statistics:

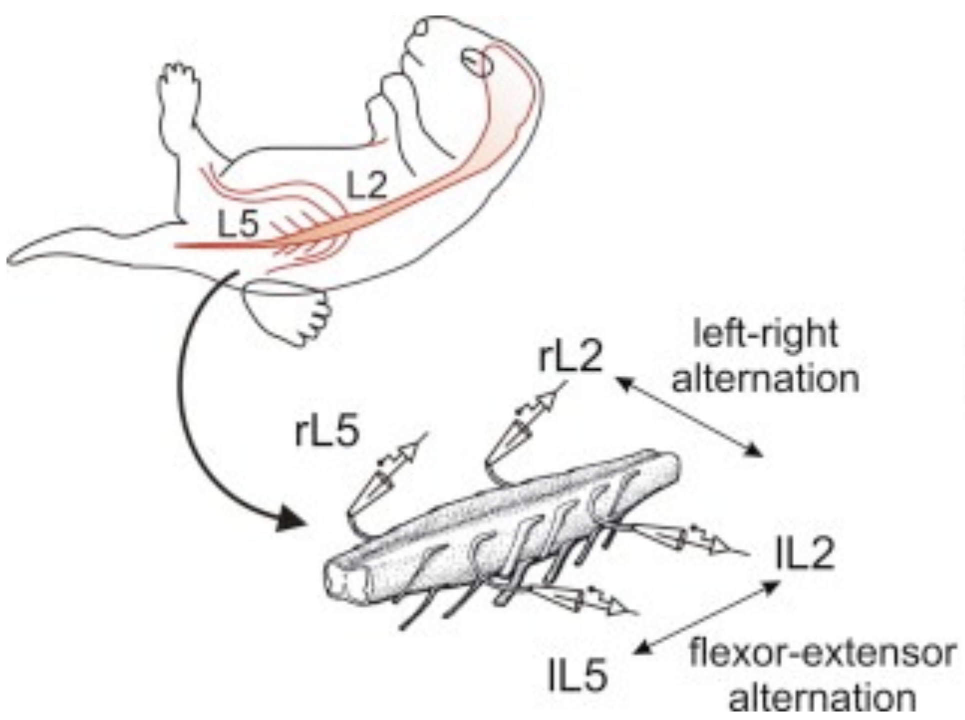
Quantification of locomotor coordination in electrophysiological recordings

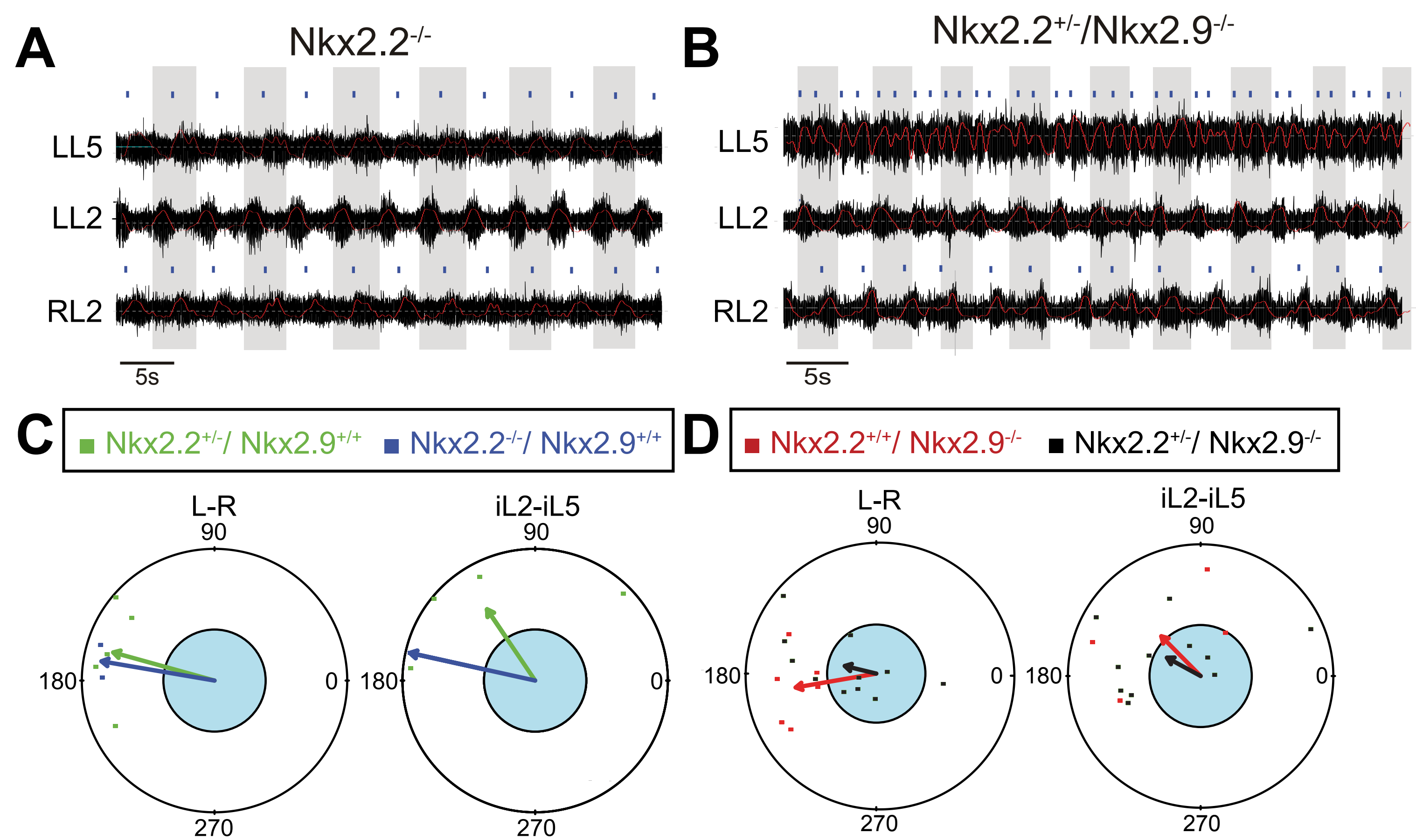
Jesper Ryge
Karolinska Institutet, Mammalian locomotor Lab
&
EPFL BMI LNMC

Circular statistics for quantification of locomotor coordination

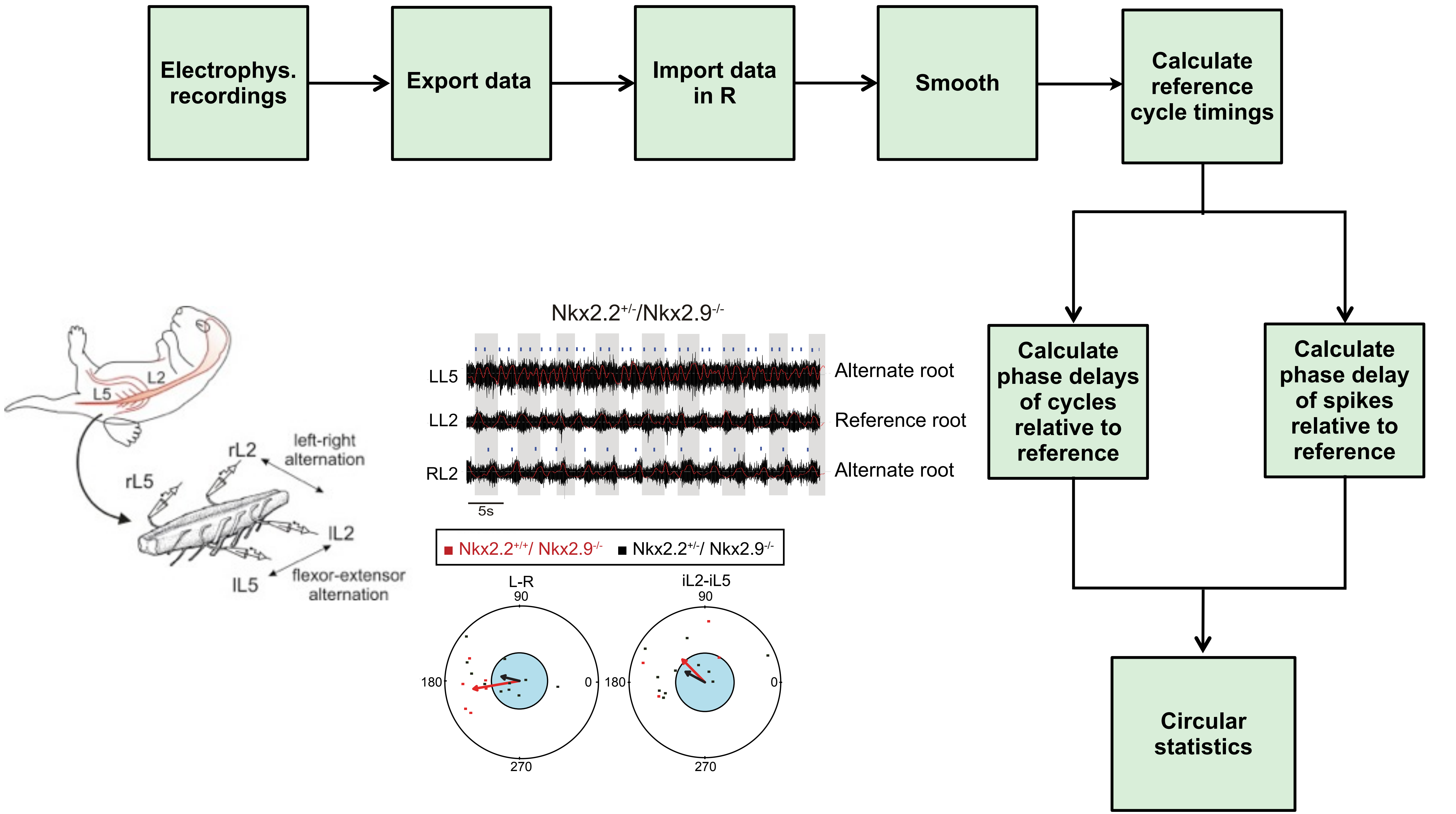


Circular statistics for quantification of locomotor coordination





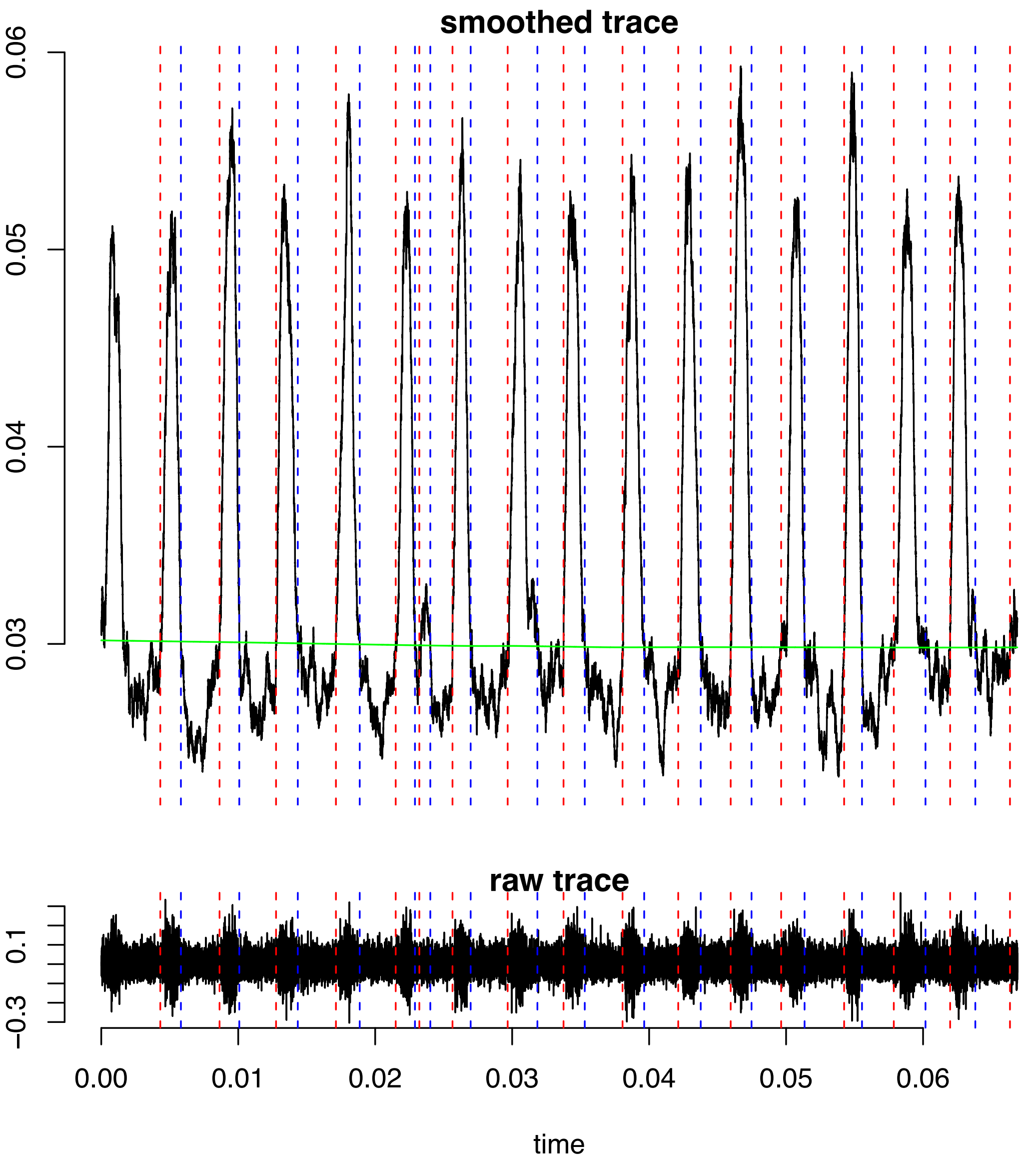
Workflow



Electrophys. recordings
Export data
Import data in R
Smooth
Calculate reference cycle timings
Calculate phase delays of cycles relative to reference
Calculate phase delay of spikes relative to reference
Circular statistics
L2
L5
rL2
rL5
lL2
lL5
left-right alternation
flexor-extensor alternation
Nkx2.2+/-/Nkx2.9-/-
LL5
LL2
RL2
Alternate root
Reference root
Alternate root
5s
Nkx2.2+/+/ Nkx2.9-/-
Nkx2.2+/-/ Nkx2.9-/-
L-R
iL2-iL5
90
180
0
270

Cycle extraction

```
> source(c("electrophys_stats.R","plot_period_stats.R"))
> phase.obj <- phase.cycle(data.file)
Read 5 items
Read 10 items
[1] "sample frequency of raw trace :  10.000"
[1] "resampling data to 1.000 Hz sampling rate, every 10 points used"
Read 3412355 items
[1] "sample frequency after re-sampling:  1.000 Hz"
[1] "length of recording 0.0668819 sec"
[1] "number of traces :  4 "
[1] "the traces are as follows: "
[1] "LL2 (mV)"   "LL5 (mV)"   "RL2 (mV)"   "R  L5 (mV)"
[1] "choose traces to rectify"
rectify LL2 (mV) (yes/no)? yes
rectify LL5 (mV) (yes/no)? no
rectify RL2 (mV) (yes/no)? no
rectify R  L5 (mV) (yes/no)? yes
[1] "choose traces to smooth"
smooth LL2 (mV) (yes/no)? yes
smooth LL5 (mV) (yes/no)? no
smooth RL2 (mV) (yes/no)? no
smooth R  L5 (mV) (yes/no)? yes
[1] "window size is 2e-04 secs with 200 points"
press enter for next plot :
re-plot traces with re-defined time axis, yes/no : no
smooth with different window size, yes/no? : no
[1] "define reference cycle trace : "
Extract cycle periodes from trace: LL2 (mV)
[1] "noise window is 3e-04 secs with 300 points"
```

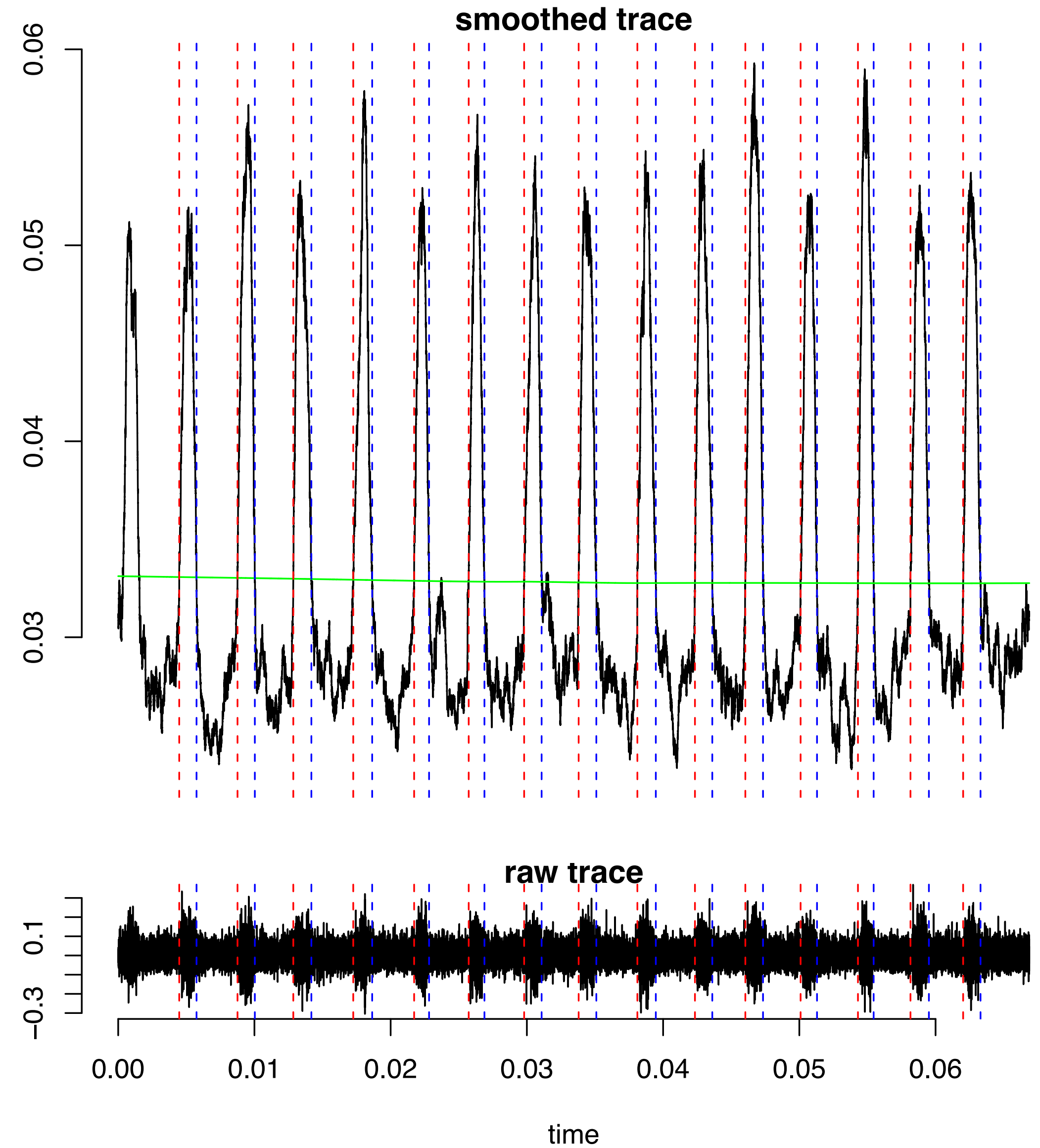


phase.cycle wrapper:

```
> data.ts.raw <- read.atf.file ()
> data.ts <- smooth(data.ts.raw)
> cycle.ref <- extract.period(data.ts, ref.root)
> cycle.alt <- extract.period(data.ts, alt.root)
> phase.delay <- extract.phase(cycle.alt, cycle.ref)
> return(list (all.data))
```

Cycle extraction

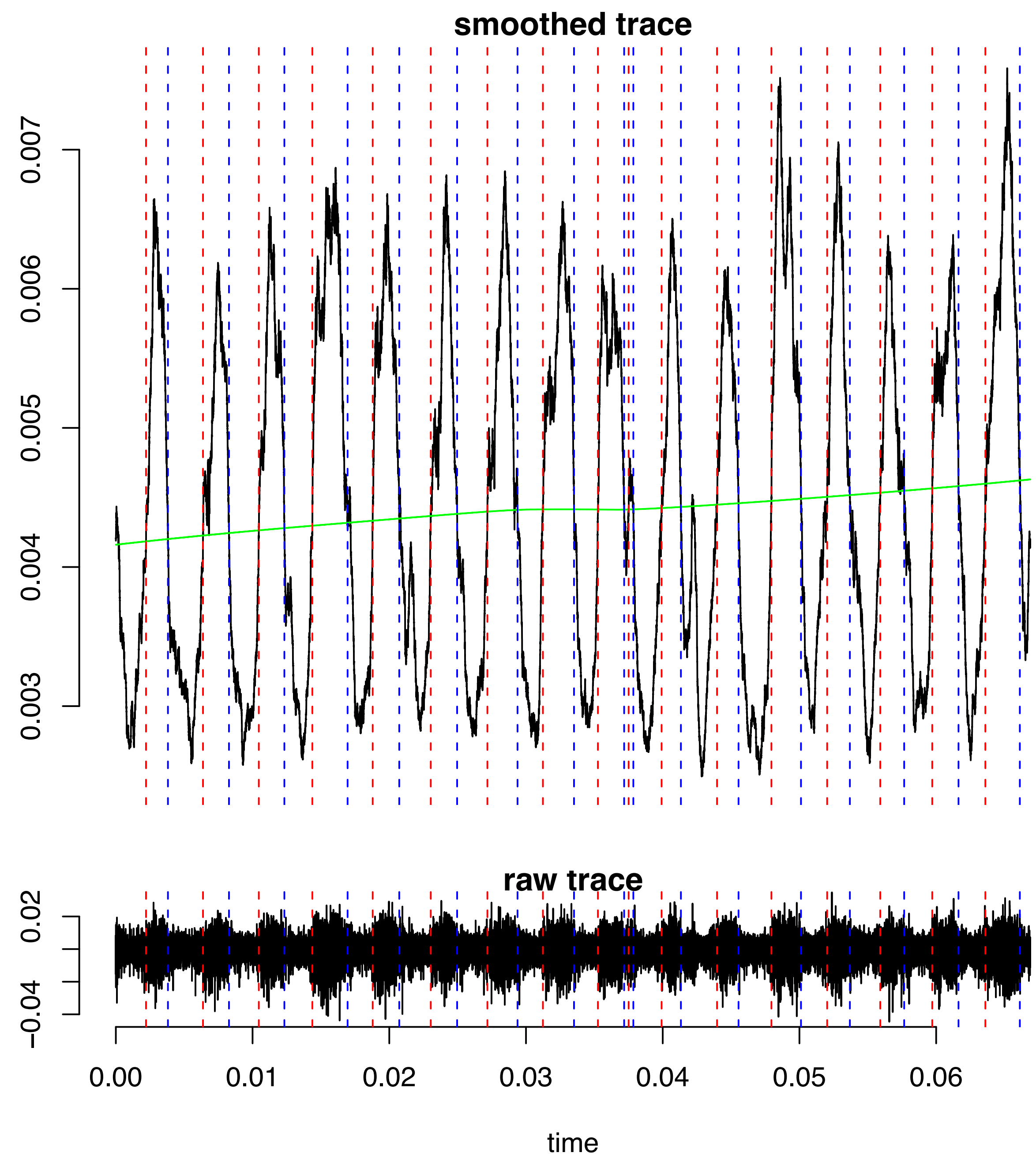
re-plot traces with re-defined time axis, yes/no : **no**
recalculate period times with new parameter values, yes/no? **yes**
choose threshold curvature {0;1} - default=0.9, press enter for no additional curvature :
choose offset [-1:1] for threshold - press enter for no offset : **0.1**
choose noise level - default = 300 ,press enter to use default value:**350**
[1] "noise window is 0.00035 secs with 350 points"
re-plot traces with re-defined time axis, yes/no : **no**
recalculate period times with new parameter values, yes/no? **no**
[1] "define test cycle trace"
Extract cycle periodes from trace: **R L5 (mV)**
[1] "noise window is 3e-04 secs with 300 points"



phase.cycle wrapper:
> data.ts.raw <- read.atf.file ()
> data.ts <- smooth(data.ts.raw)
> cycle.ref <- extract.period(data.ts, ref.root)
> cycle.alt <- extract.period(data.ts, alt.root)
> phase.delay <- extract.phase(cycle.alt, cycle.ref)
> return(list (all.data))

Cycle extraction

re-plot traces with re-defined time axis, yes/no : no
recalculate period times with new parameter values, yes/no? yes
choose threshold curvature {0;1} - default=0.9, press enter for no additional curvature :
choose offset [-1:1] for threshold - press enter for no offset :
choose noise level - default = 300 ,press enter to use default value: **500**
[1] "noise window is 5e-04 secs with 500 points"
re-plot traces with re-defined time axis, yes/no : no
recalculate period times with new parameter values, yes/no? no

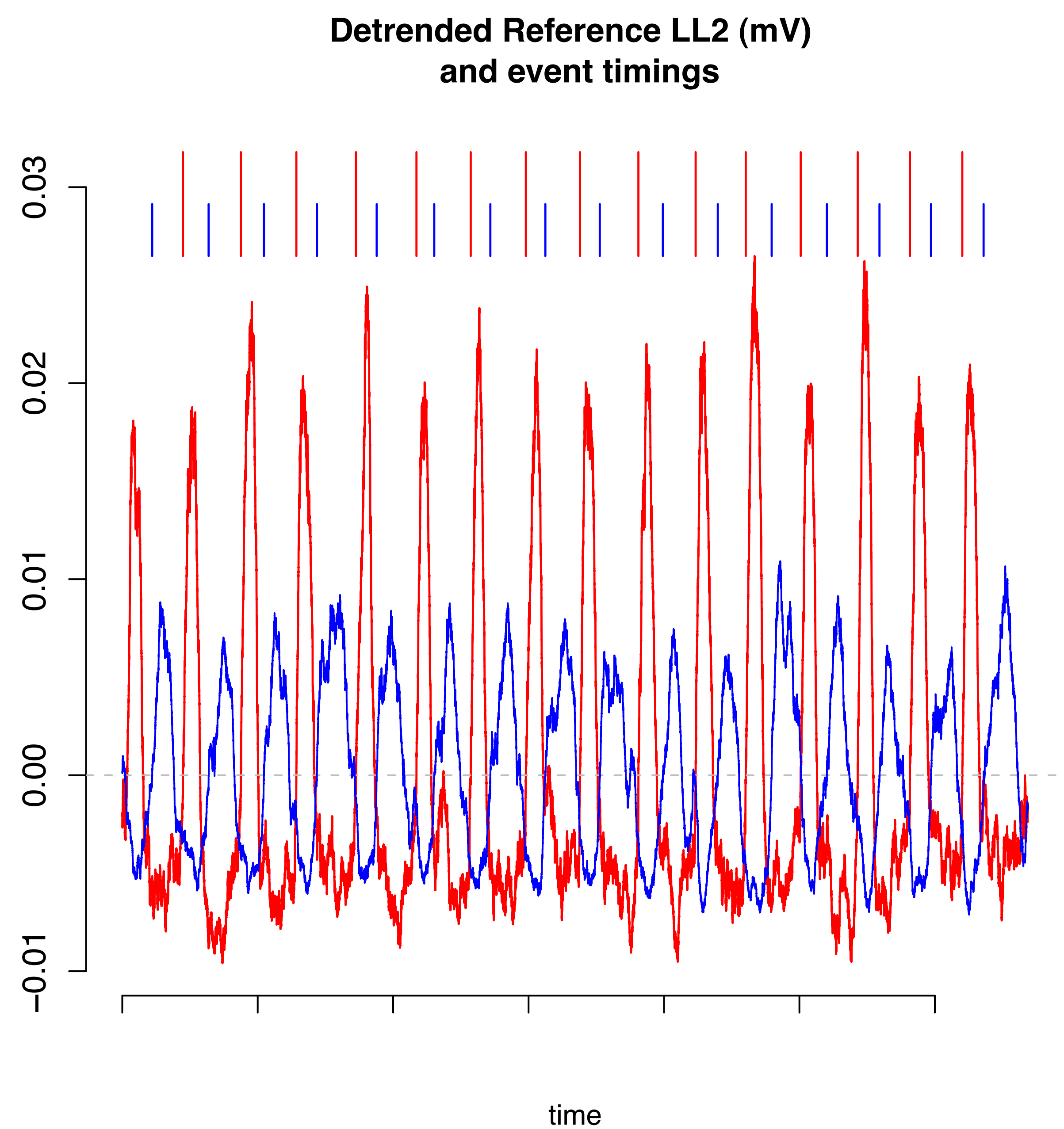


phase.cycle wrapper:

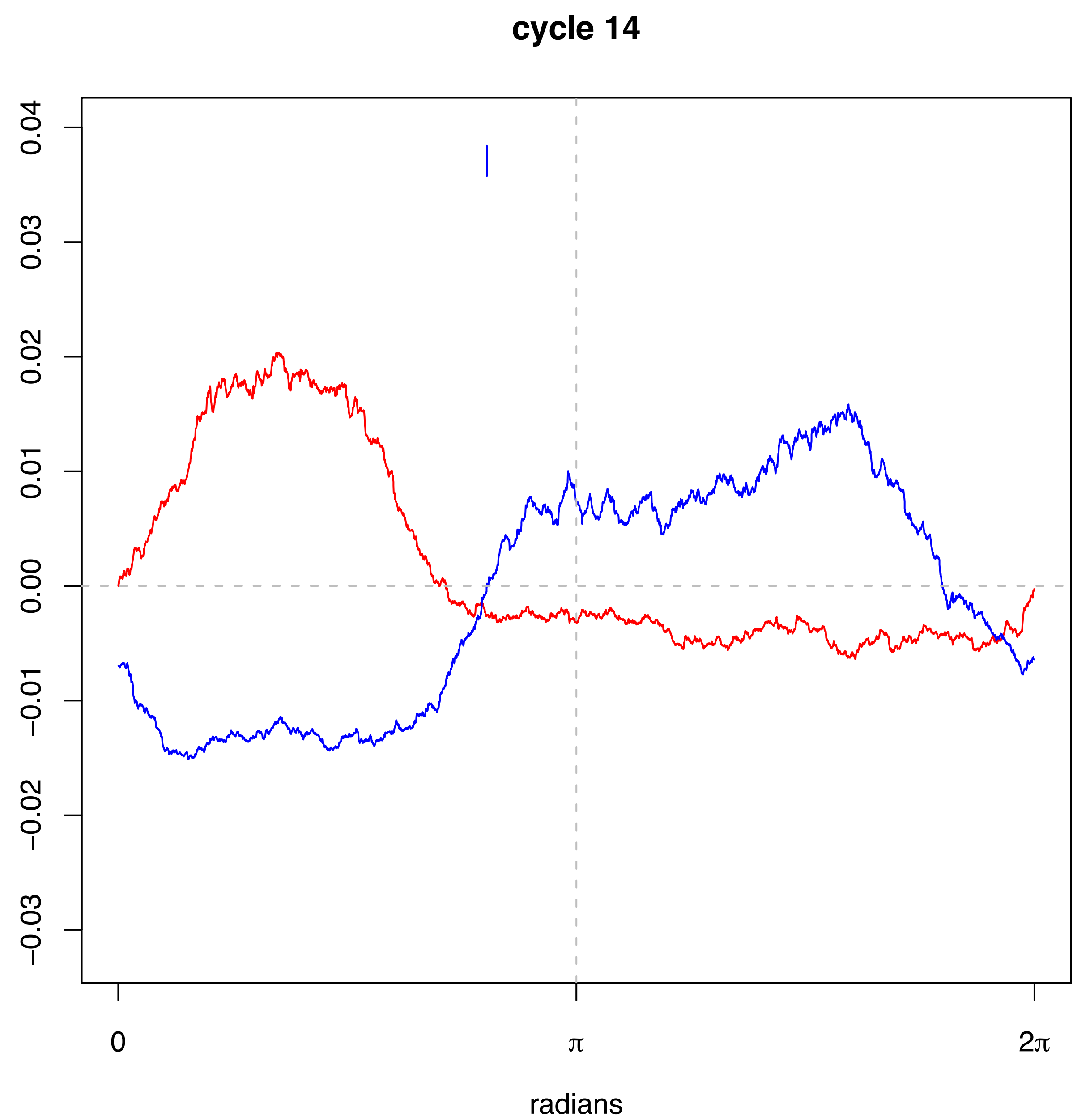
```
> data.ts.raw <- read.atf.file ()
> data.ts <- smooth(data.ts.raw)
> cycle.ref <- extract.period(data.ts, ref.root)
> cycle.alt <- extract.period(data.ts, alt.root)
> phase.delay <- extract.phase(cycle.alt, cycle.ref)
> return(list (all.data))
```

Circular stats and plots

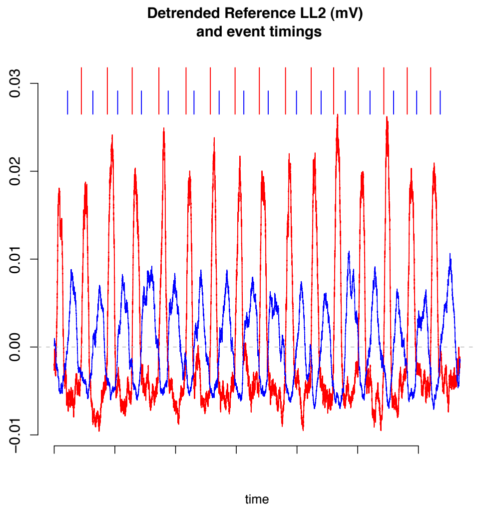
```
> circular.stats <- plot.period.stats(phase.obj)
> plot.period.stats(test, plot.periods=T)
pres to plot figure  2
pres to plot figure  3
...
pres to plot figure  15
[1] "data.ts"      "event.timing" "ref.period"   "event.period" "thres"
[6] "thresE"       "ref.trace"    "event.trace"  "is.root"
```



Circular stats and plots

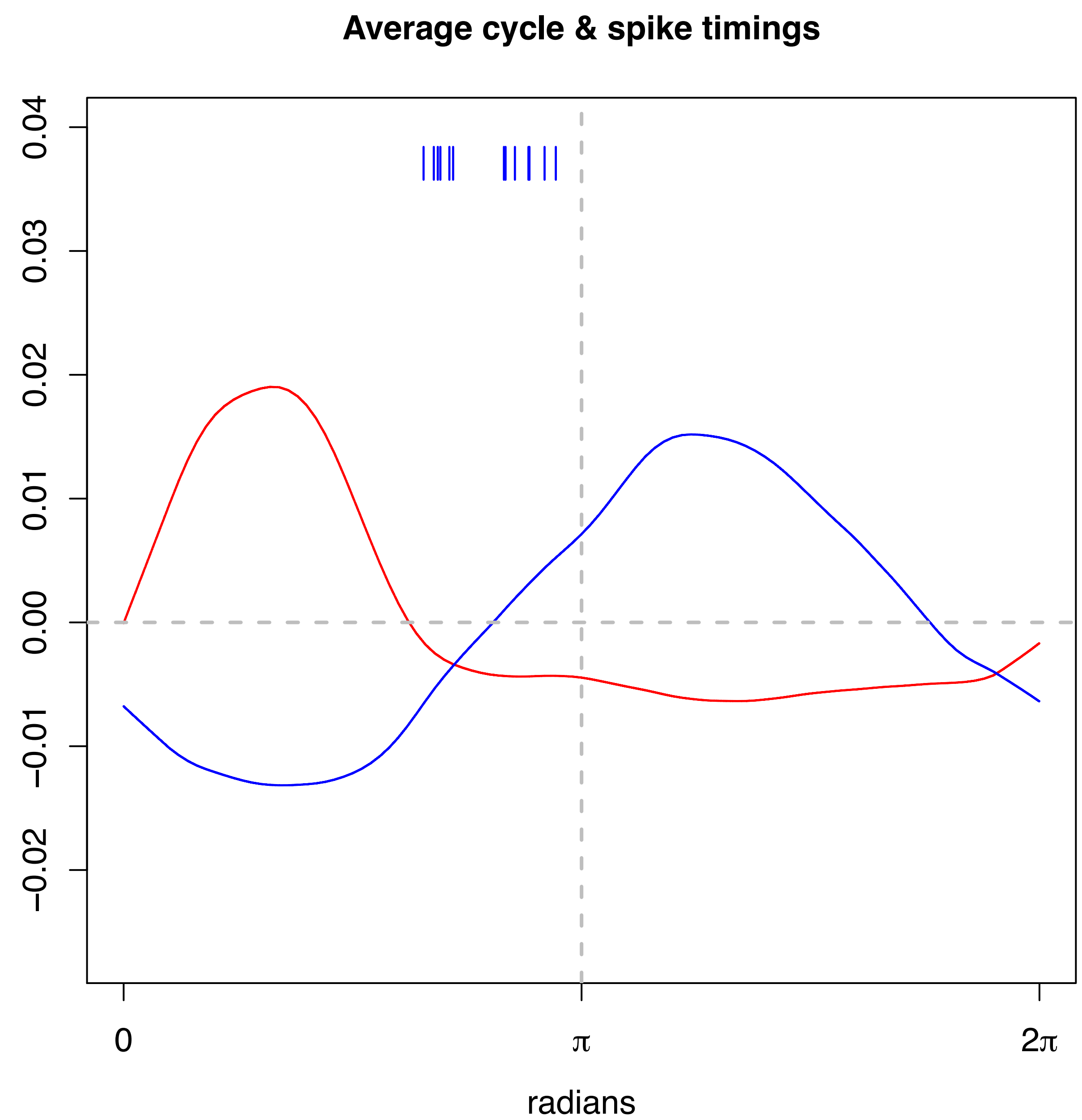


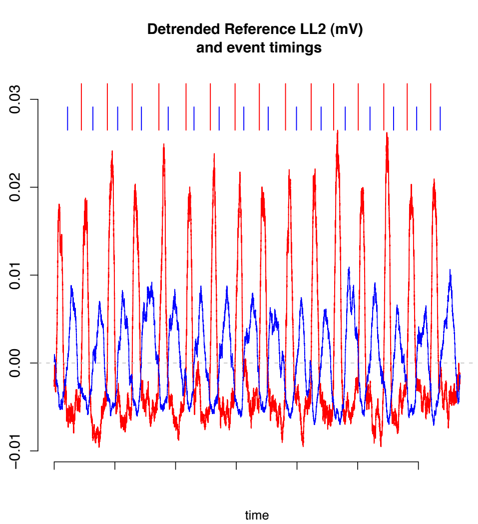

```
> circular.stats <- plot.period.stats(phase.obj)
> plot.period.stats(test, plot.periods=T)
pres to plot figure  2
pres to plot figure  3
...
pres to plot figure  15
[1] "data.ts"      "event.timing" "ref.period"   "event.period" "thres"
[6] "thresE"       "ref.trace"    "event.trace"  "is.root"
```

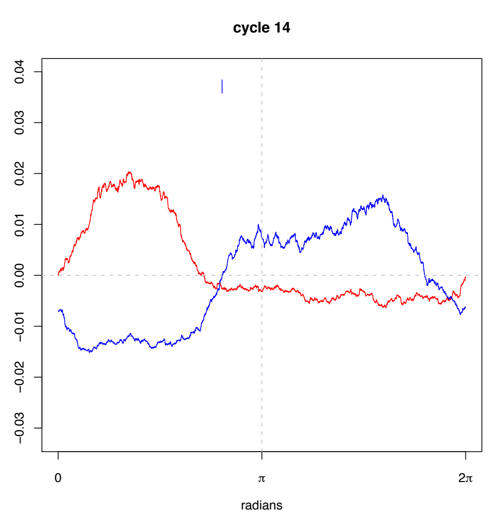


Circular stats and plots

```
> circular.stats <- plot.period.stats(phase.obj)
> plot.period.stats(test, plot.periods=T)
pres to plot figure  2
pres to plot figure  3
...
pres to plot figure  15
[1] "data.ts"      "event.timing" "ref.period"   "event.period" "thres"
[6] "thresE"       "ref.trace"    "event.trace"  "is.root"
```

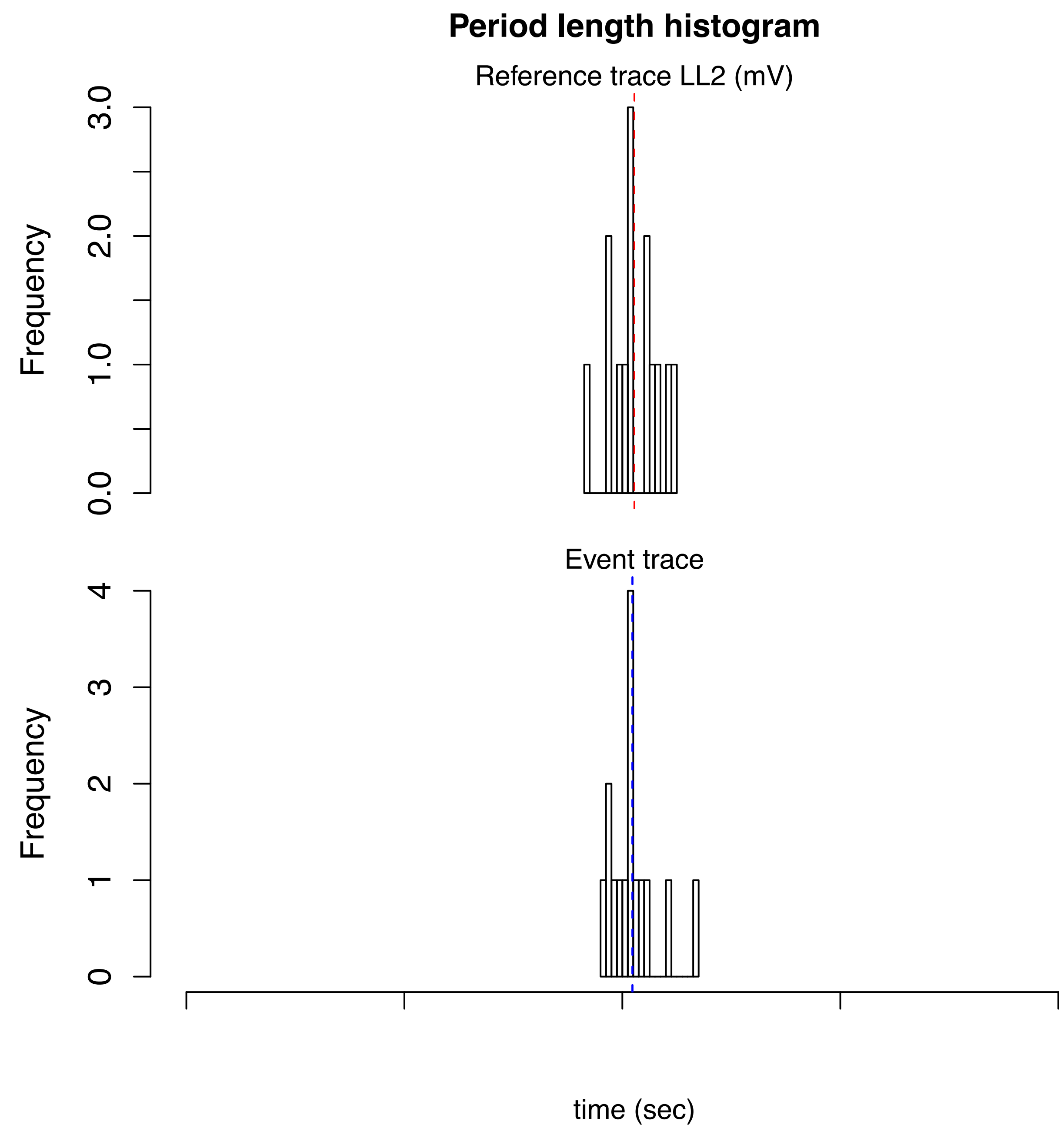


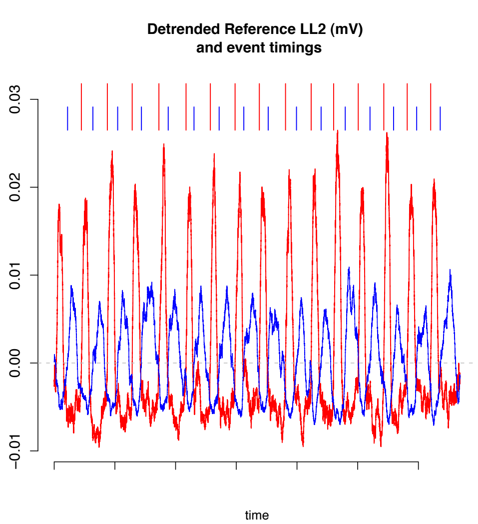


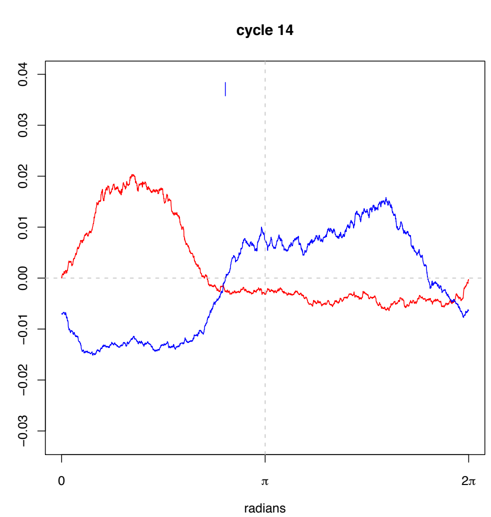


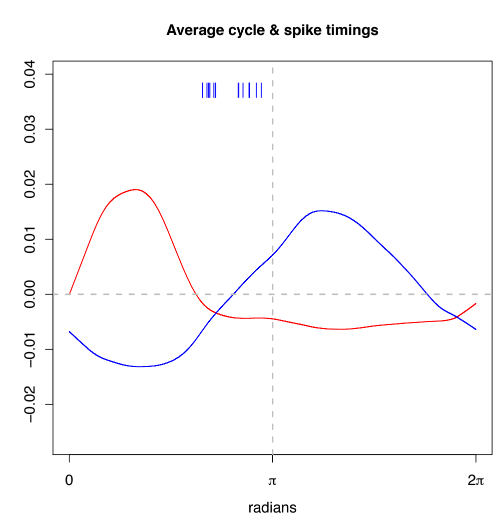
Circular stats and plots

```
> circular.stats <- plot.period.stats(phase.obj)
> plot.period.stats(test, plot.periods=T)
pres to plot figure  2
pres to plot figure  3
...
pres to plot figure  15
[1] "data.ts"      "event.timing" "ref.period"   "event.period" "thres"
[6] "thresE"       "ref.trace"    "event.trace"  "is.root"
```

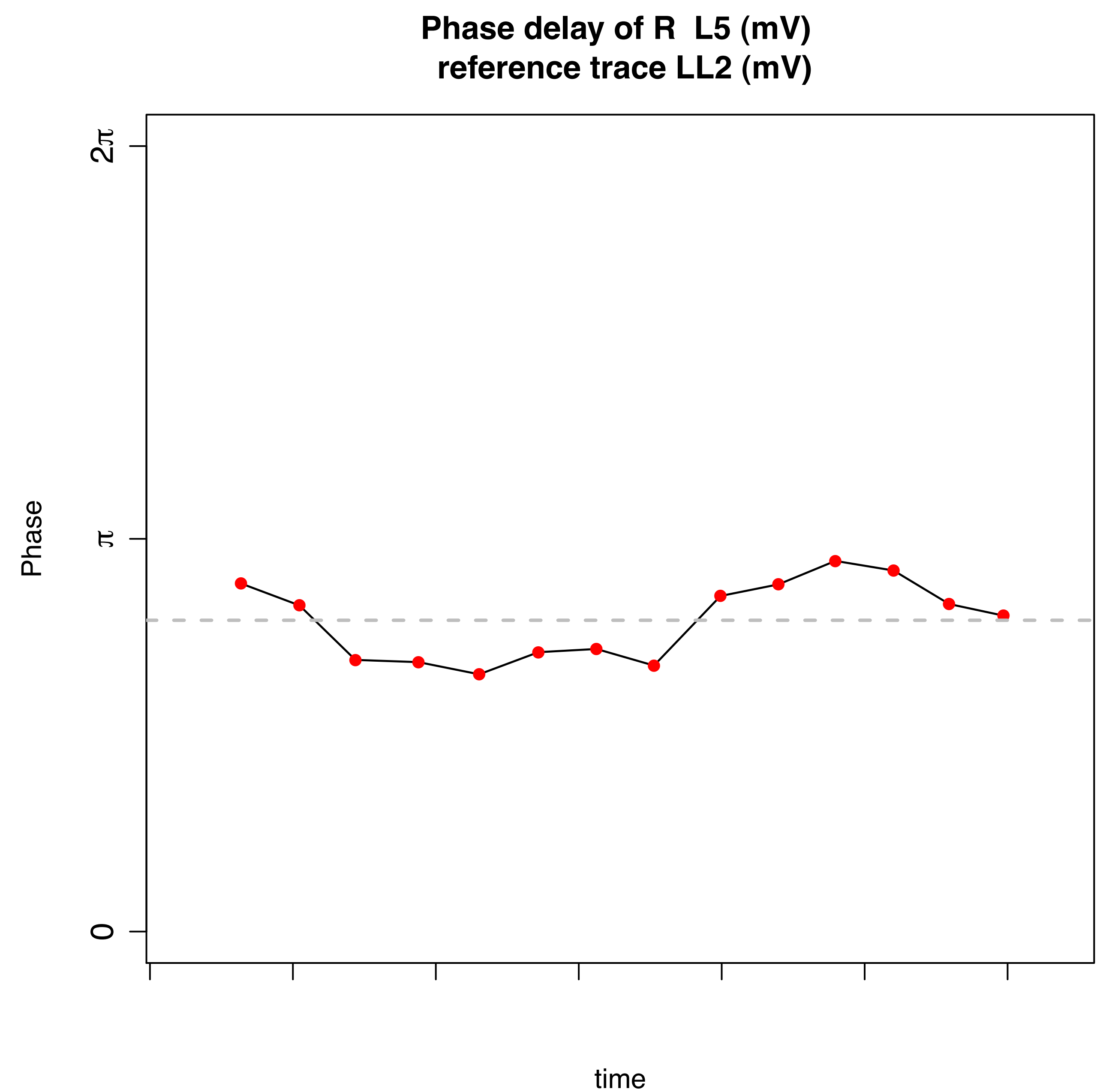




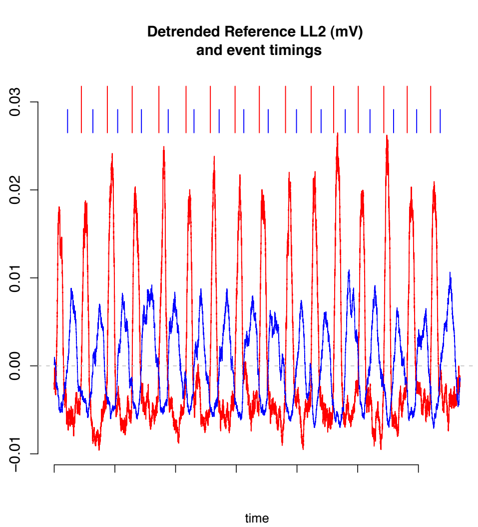


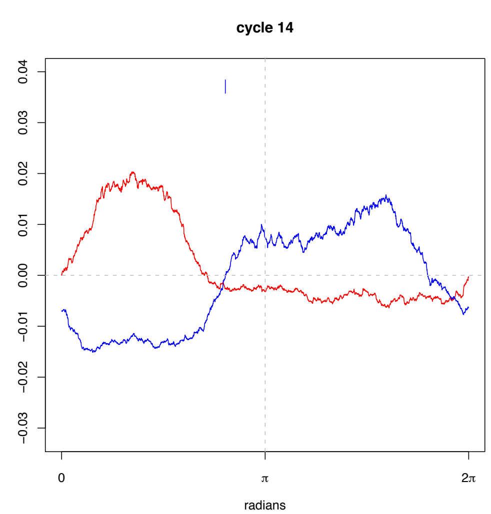


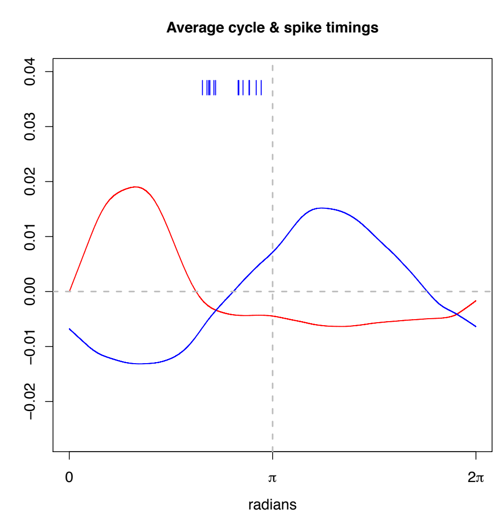
Circular stats and plots

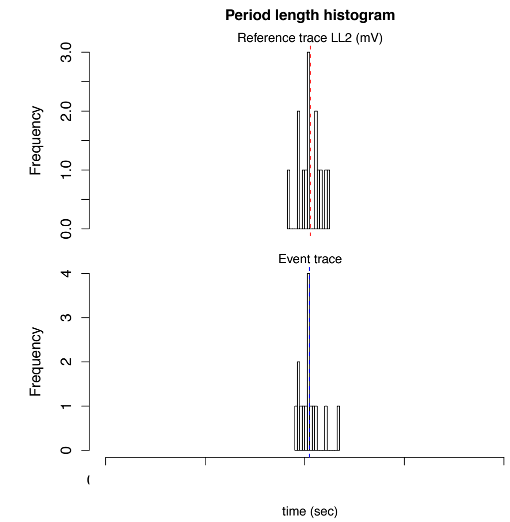



```
> circular.stats <- plot.period.stats(phase.obj)
> plot.period.stats(test, plot.periods=T)
pres to plot figure  2
pres to plot figure  3
...
pres to plot figure  15
[1] "data.ts"      "event.timing" "ref.period"    "event.period" "thres"
[6] "thresE"        "ref.trace"      "event.trace"   "is.root"
```









Circular stats and plots

[1] "Stack points in circular plots with 150 bins"

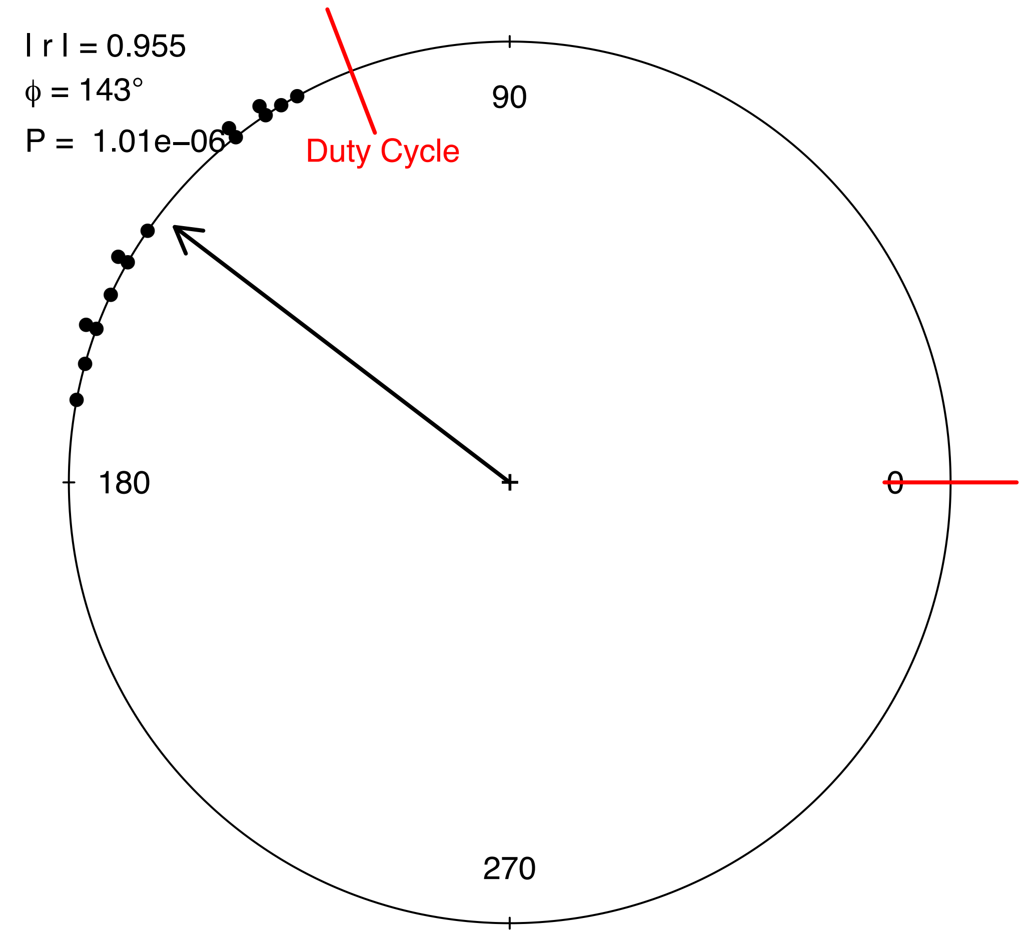
Rayleigh Test of Uniformity
General Unimodal Alternative

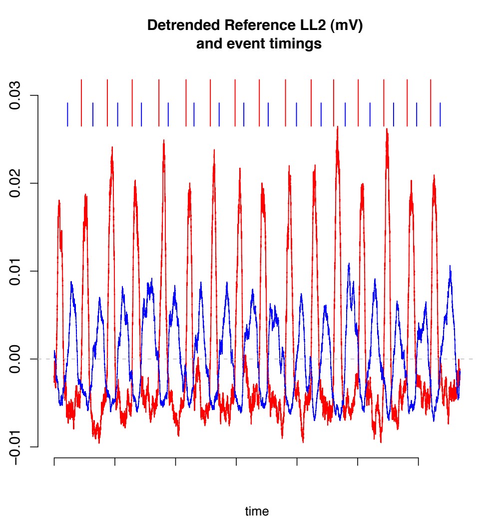
Test Statistic: 0.9551
P-value: 0

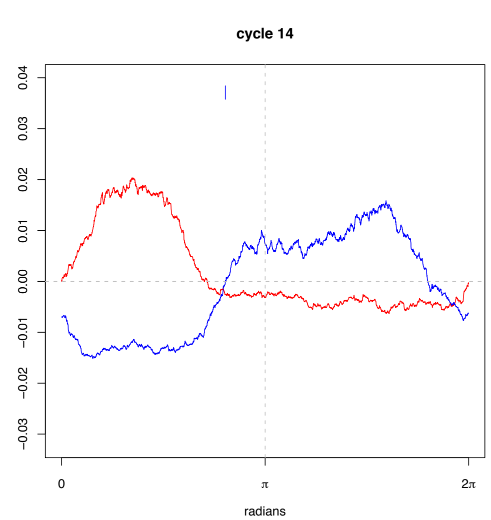
[1] "average firing phase in degrees : 143 "
[1] "active phase duty cycle of reference trace, from degrees 0: 111.08"
$r.length
[1] 0.955

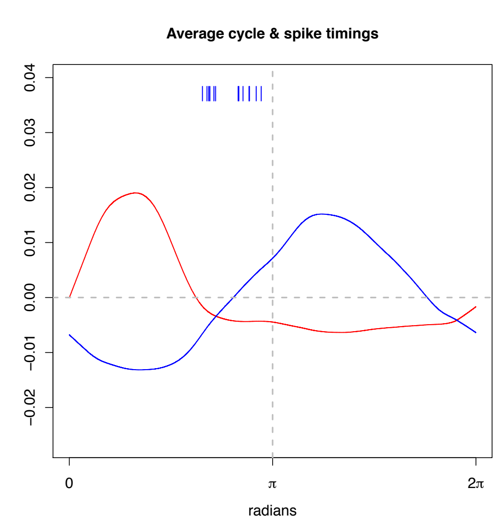
$r.angle
Circular Data:
Type = angles
Units = radians
Template = none
Modulo = asis
Zero = 0
Rotation = counter
[1] 2.490117

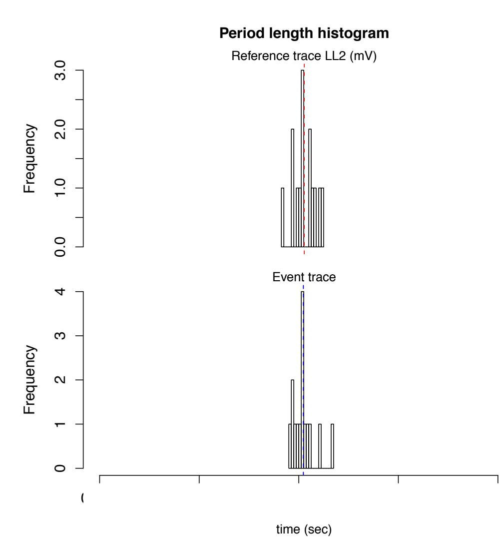
$p.value
[1] 1.013014e-06

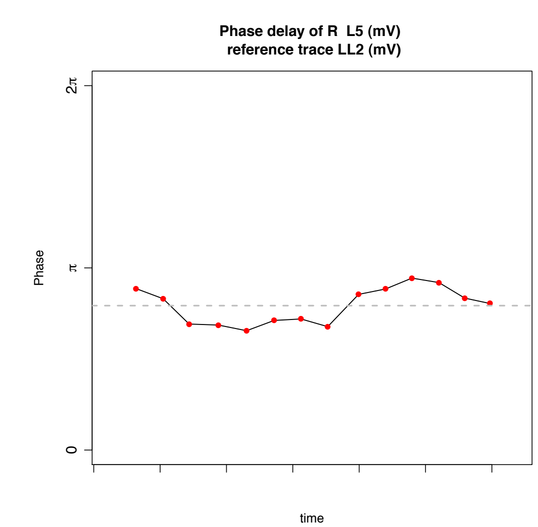












Circular stats and plots

[1] "Stack points in circular plots with 150 bins"

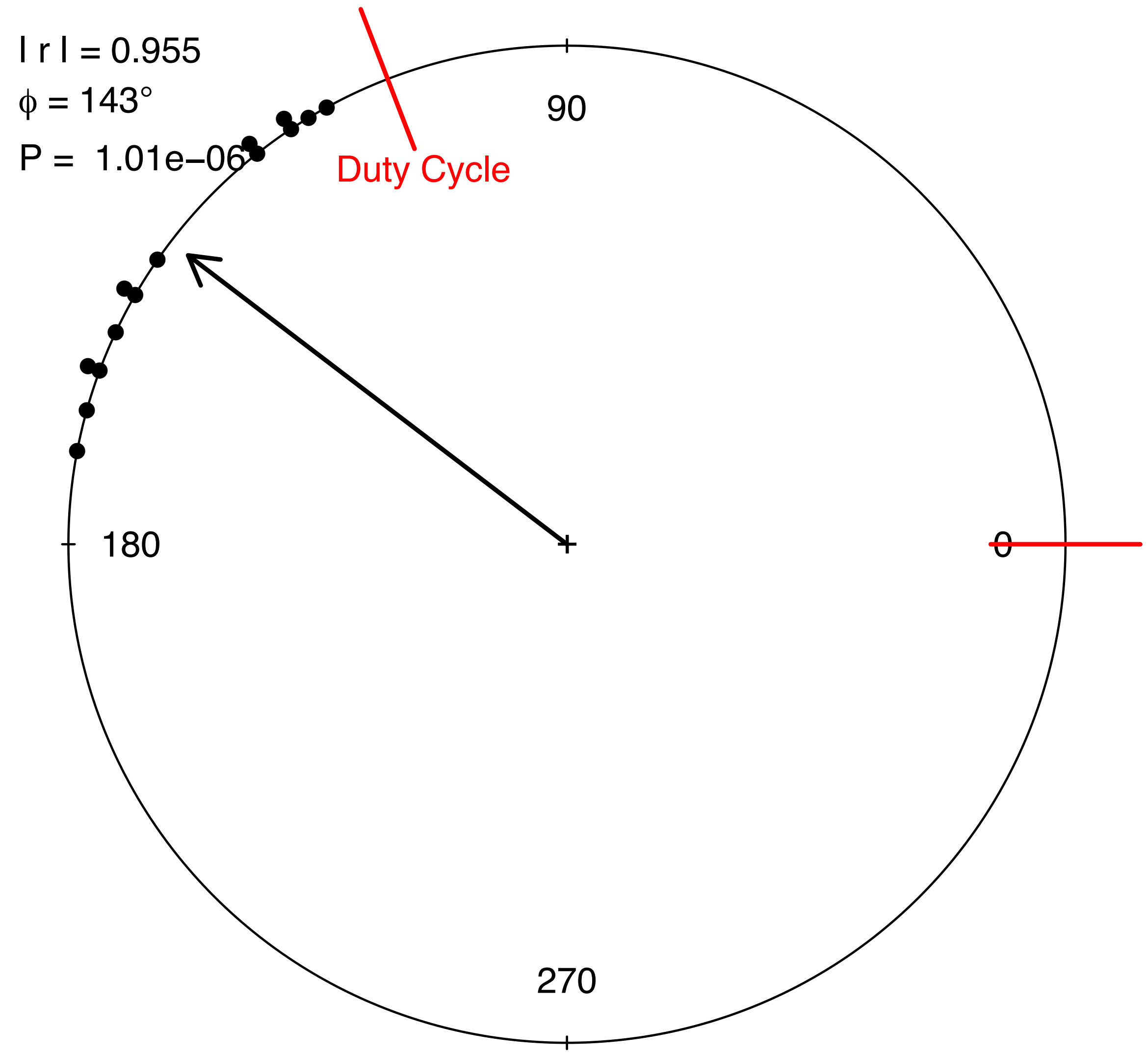
Rayleigh Test of Uniformity
General Unimodal Alternative

Test Statistic: 0.9551
P-value: 0

[1] "average firing phase in degrees : 143 "
[1] "active phase duty cycle of reference trace, from degrees 0: 111.08"
$r.length
[1] 0.955

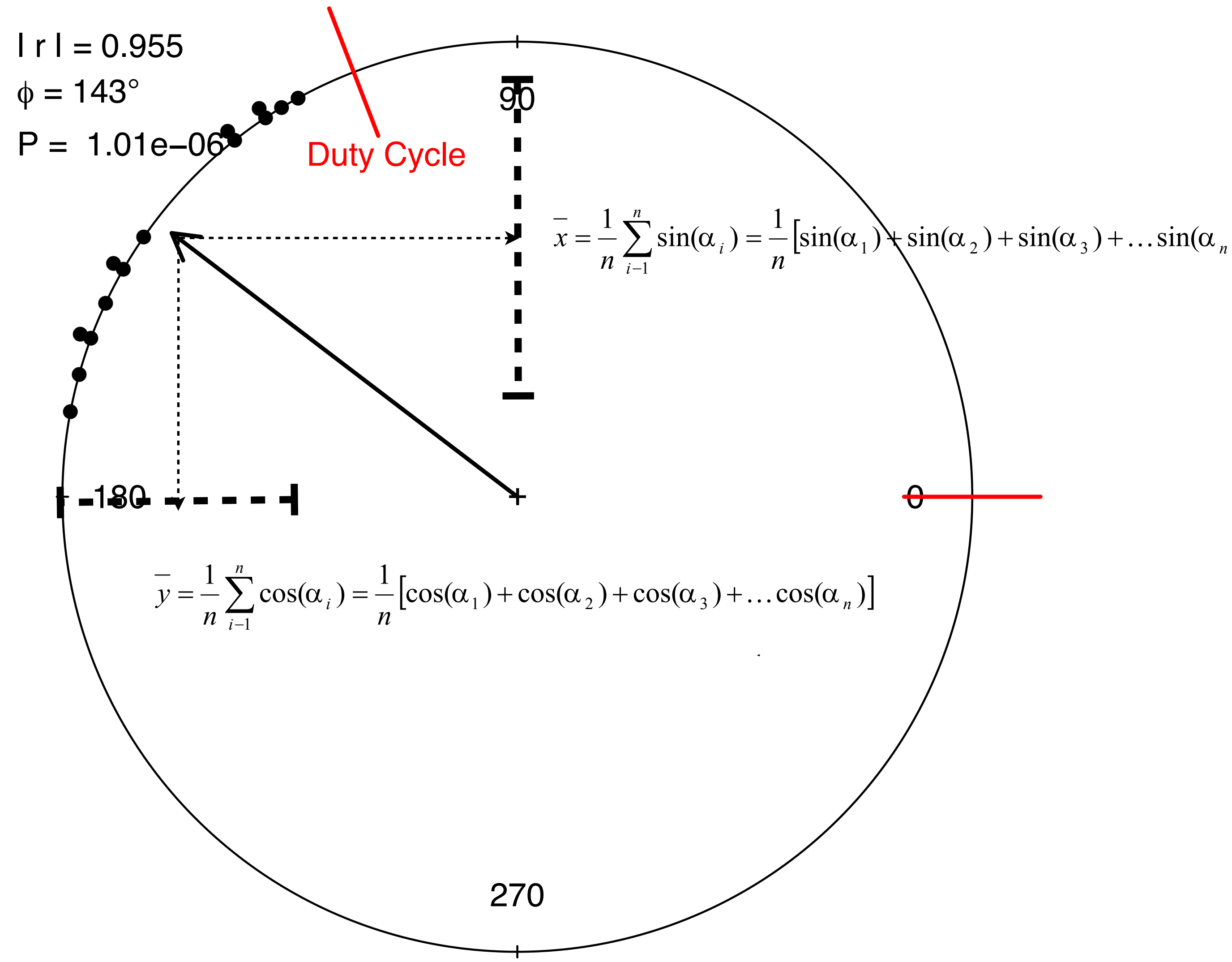
$r.angle
Circular Data:
Type = angles
Units = radians
Template = none
Modulo = asis
Zero = 0
Rotation = counter
[1] 2.490117

$p.value
[1] 1.013014e-06



Circular stats and plots

```
[1] "Stack points in circular plots with 150 bins"

       Rayleigh Test of Uniformity
       General Unimodal Alternative

Test Statistic:  0.9551
P-value:  0

[1] "average firing phase in degrees :  143 "
[1] "active phase duty cycle of reference trace, from degrees 0:  111.08"
$r.length
[1] 0.955

$r.angle
Circular Data:
Type = angles
Units = radians
Template = none
Modulo = asis
Zero = 0
Rotation = counter
[1] 2.490117

$p.value
[1] 1.013014e-06
```



where n is the number of observations and α_i is the i^{th} azimuth or observation. The length or magnitude of the mean vector is :

$$r = \sqrt{\bar{x}^2 + \bar{y}^2}$$

To obtain the angle of the mean direction solve the following equations.

$$\sin(\Theta) = \frac{\bar{x}}{r} \rightarrow \Theta = \sin^{-1}\left(\frac{\bar{x}}{r}\right) \quad \text{AND} \quad \cos(\Theta) = \frac{\bar{y}}{r} \rightarrow \Theta = \cos^{-1}\left(\frac{\bar{y}}{r}\right)$$

Circular stats and plots

[1] "Stack points in circular plots with 150 bins"

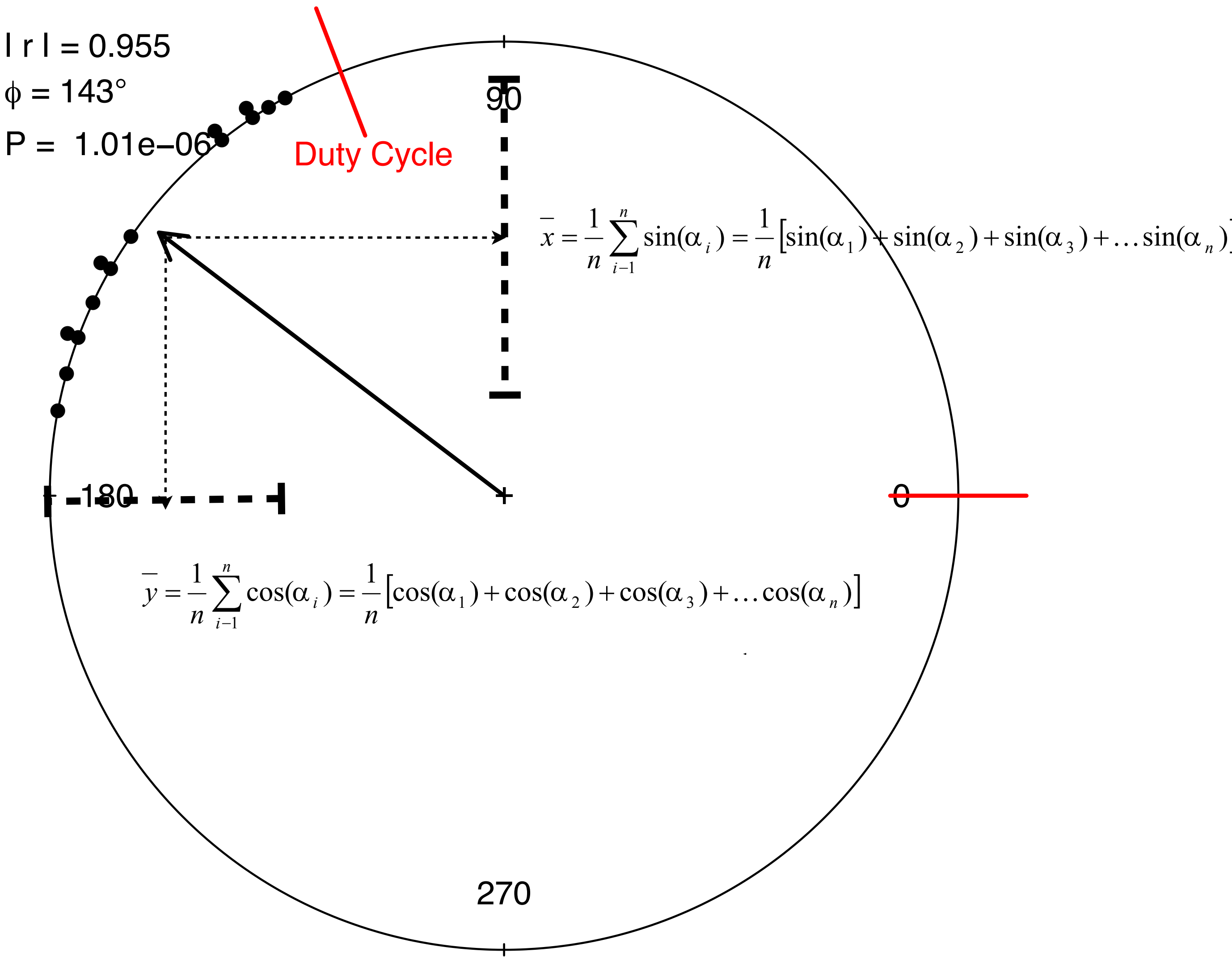
Rayleigh Test of Uniformity
General Unimodal Alternative

Test Statistic: 0.9551
P-value: 0

[1] "average firing phase in degrees : 143 "
[1] "active phase duty cycle of reference trace, from degrees 0: 111.08"
$r.length
[1] 0.955

$r.angle
Circular Data:
Type = angles
Units = radians
Template = none
Modulo = asis
Zero = 0
Rotation = counter
[1] 2.490117

$p.value
[1] 1.013014e-06



RAYLEIGH TEST

The Rayleigh test is a statistical procedure for determining whether a circular distribution is random or non-random. That is, are the azimuths of a distribution clumped in a particular direction? Calculate a critical value (the test statistic), Z, for the Rayleigh test using the following formula:

$$Z = nr^2$$

where Z is the critical value, n is the number of observations or azimuths, and r is the magnitude of the mean vector (determined as above). The **null** (H_o) and **alternative** (H_a) hypotheses for the test are:

H_o = the bearings are randomly distributed.

H_a = the bearings are distributed nonrandomly.

Use Table B.34 (Zar 1999) to determine whether to **accept or reject** the null hypothesis.

where n is the number of observations and α_i is the ith azimuth or observation. The length or magnitude of the mean vector is :

$$r = \sqrt{\bar{x}^2 + \bar{y}^2}$$

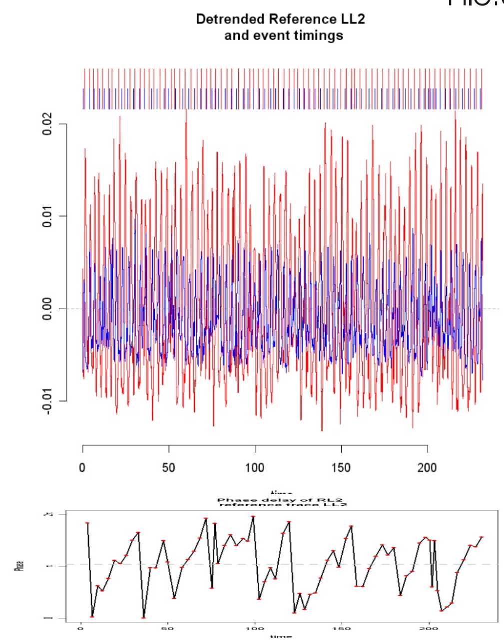
To obtain the angle of the mean direction solve the following equations.

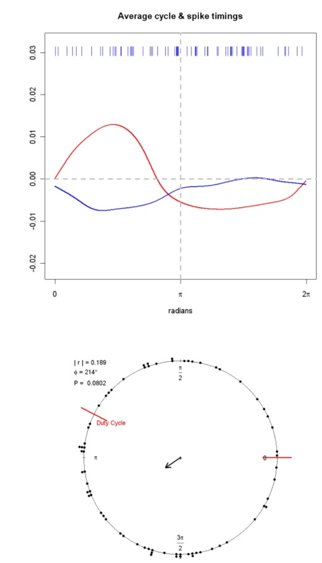
$$\sin(\Theta) = \frac{\bar{x}}{r} \rightarrow \Theta = \sin^{-1}\left(\frac{\bar{x}}{r}\right) \quad \text{AND} \quad \cos(\Theta) = \frac{\bar{y}}{r} \rightarrow \Theta = \cos^{-1}\left(\frac{\bar{y}}{r}\right)$$

Ex. Nkx 2.2 KO 2.9 het

20070212 P1
Ll2 vs RL2

Lococmotion: 5-HT + NMDA
File:004

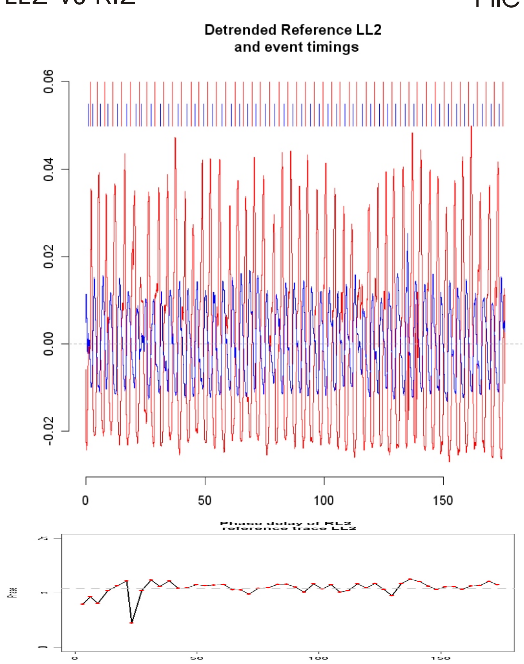


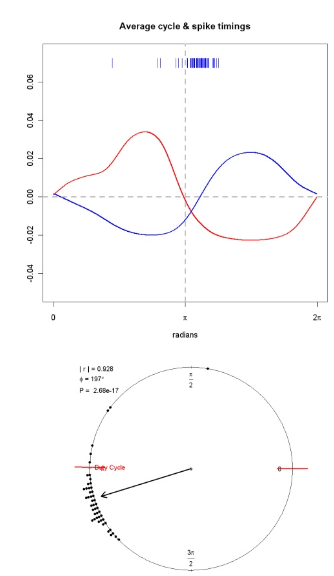


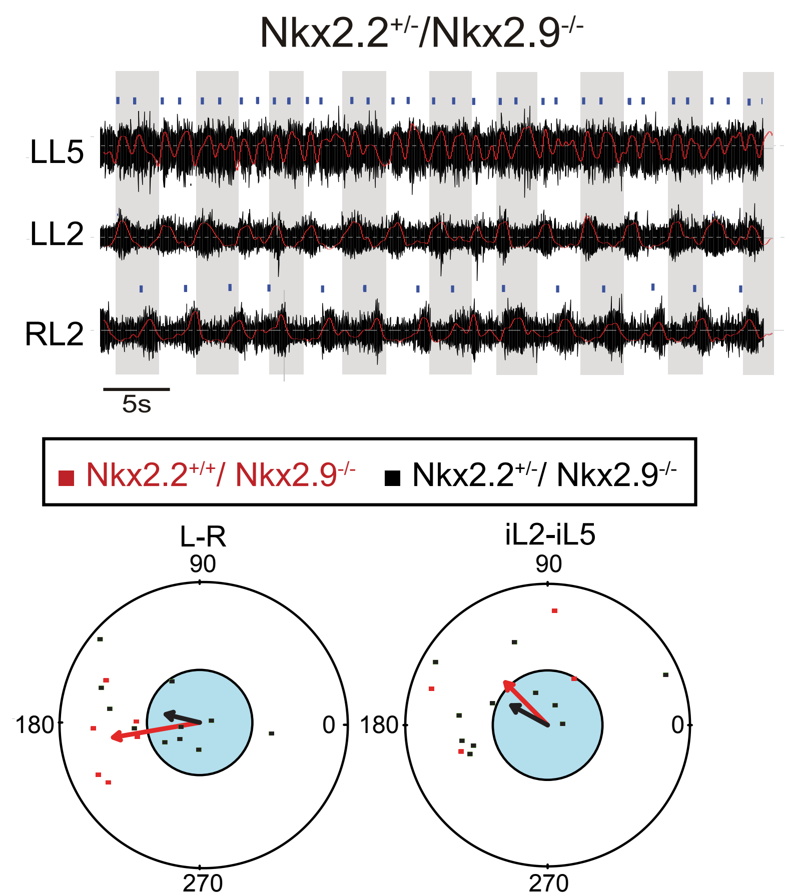
20070207 P1
LL2 vs Rl2

Lococmotion: 5-HT + NMDA
File:002

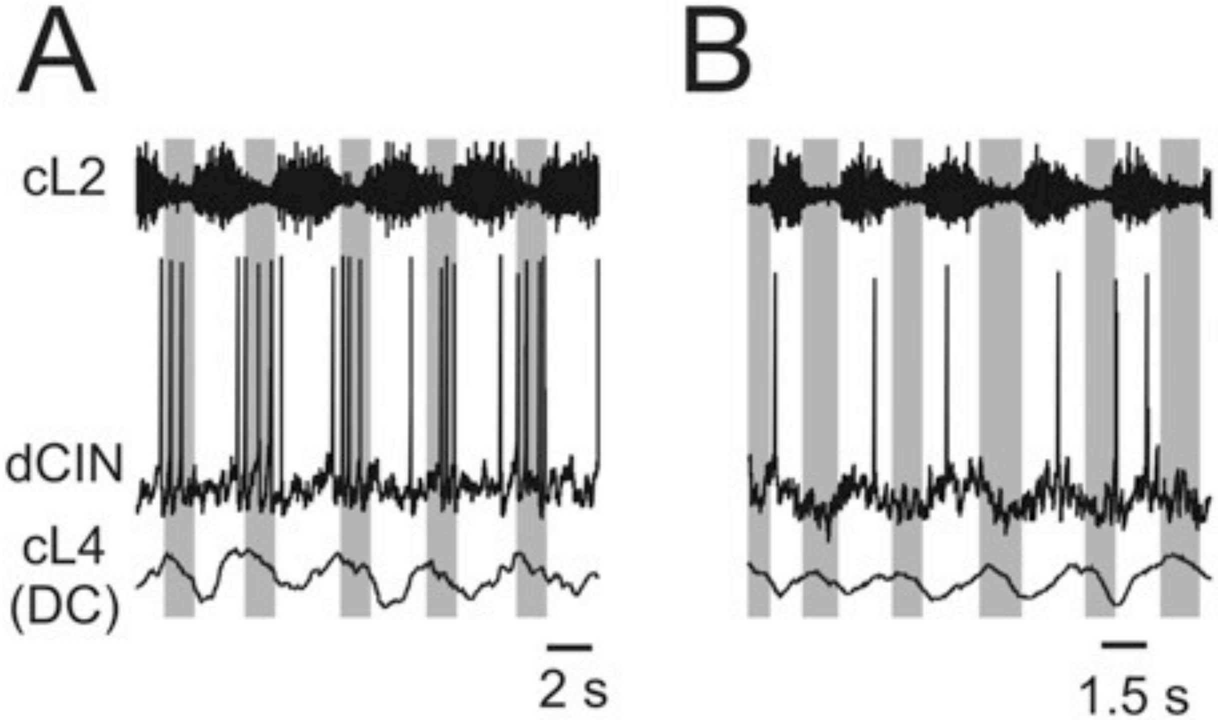
Nkx 2.2+/+ Nkx 2.9 +/-

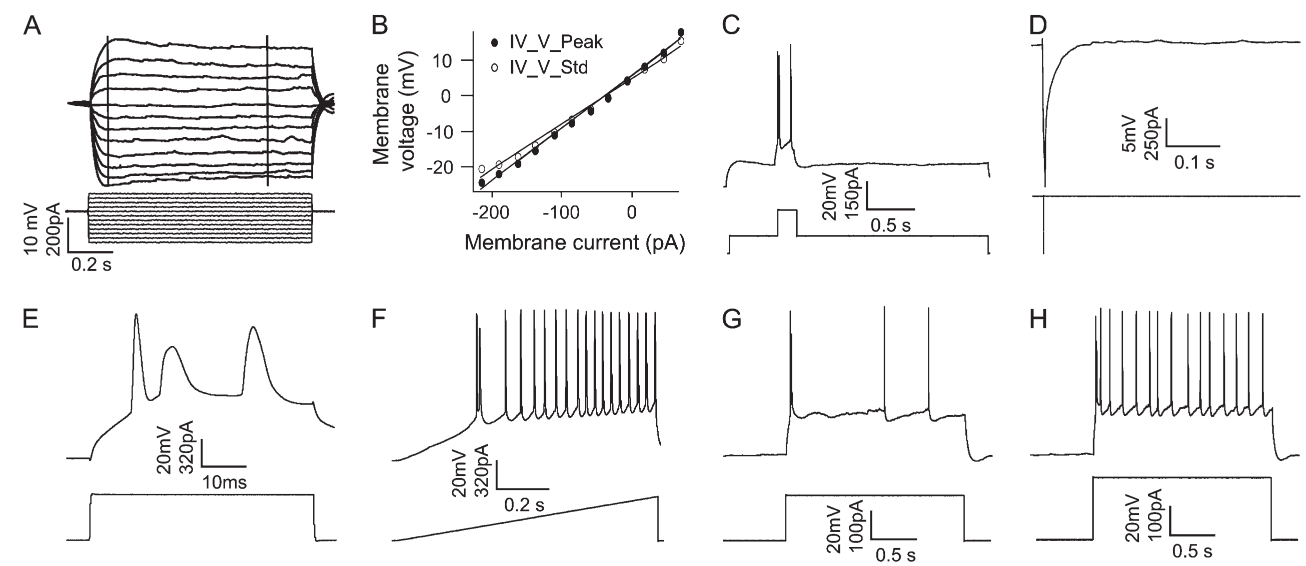


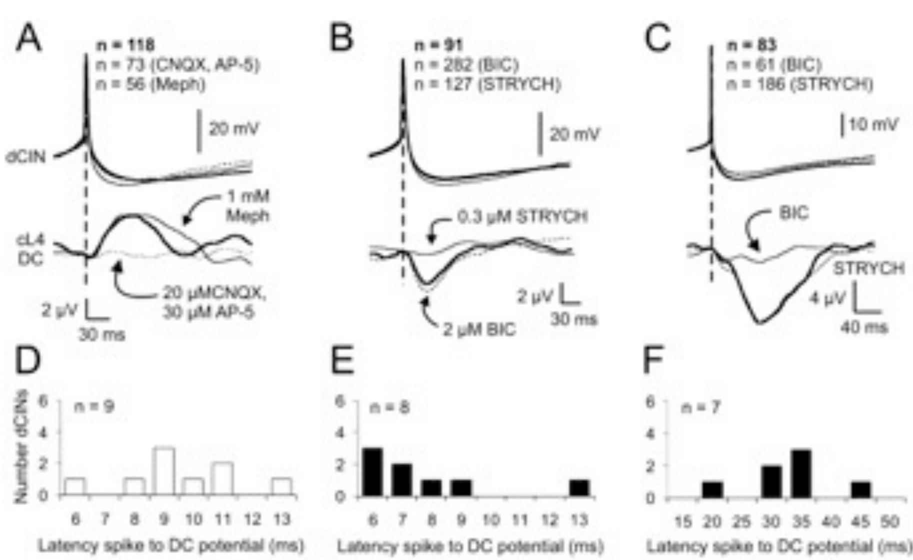




Additional features







	CCP ($n = 22$)	TTC ($n = 35$)
Access (MΩ)	23.7 ± 5.4	21.9 ± 4.9
First AP in train		
AP amplitude (mV)	59.6 ± 5.5	63.0 ± 5.3*
AP duration (ms)	4.29 ± 0.64	3.09 ± 0.52***
AP duration at half amplitude (ms)	1.93 ± 0.31	1.35 ± 0.24***
AP rise time (ms)	1.05 ± 0.16	0.80 ± 0.15***
AP fall time (ms)	3.24 ± 0.55	2.29 ± 0.41***
Fast AHP (mV)	−2.2 ± 4.1	3.7 ± 3.5***
Second AP in train		
AP amplitude (mV)	45 ± 11	63 ± 6***
AP duration (ms)	7.2 ± 1.2	6.0 ± 0.7***
AP duration at half amplitude (ms)	2.87 ± 0.88	2.06 ± 0.35***
AP rise time (ms)	1.99 ± 0.39	1.25 ± 0.26***
AP fall time (ms)	5.2 ± 1.1	4.7 ± 0.7
IV analysis		
Time to hyperpol peak (s)	0.194 ± 0.033	0.417 ± 0.430**
Input resistance for peak (MΩ)	228 ± 92	64 ± 19***
Input resistance for steady state (MΩ)	183 ± 67	51 ± 15***
Maximum sag (mV)	6.6 ± 3.9	5.8 ± 3.4
Decay time constant for Δ pulse (ms)	20.0 ± 5.8	17.5 ± 4.7
AP threshold (mV)	−32.3 ± 5.6	−39.4 ± 2.6***
Slow AHP (mV)	7.1 ± 3.2	8.6 ± 2.9
Step current		
Average delay to first spike (s)	0.031 ± 0.013	0.026 ± 0.012
Current-discharge slope (Hz nA^{-1})	27.6 ± 28.5	26.2 ± 5.7
Initial burst interval (s)	0.052 ± 0.047	0.081 ± 0.049*

Note: Mean ± SD, $*P < 0.05$, $**P < 0.01$, $***P < 0.001$, P value is from Student *t*-test; AHP = after hyperpolarization.

Summary

Present state

- Data formats and import
  - Read electrophys. txt file
- Data pre-processing
  - smooth
  - extract cycles/events
- Circular statistics and plots

Ongoing work and Future directions

- Make into package
- Optimize data import
- Implement interactive graphs
- Expand functions
  - Spike triggered average
  - Episodic stimulus protocol