

PESTICA2 introduction

- Physiologic EStimation by Temporal ICA
- Find slice-acquisition-sampled noise source in data using prior spatial information and ICA decomposition
 - PESTICA1 uses subject-dependent spatial prior, requires one separate scan with parallel physiologic monitoring; this will not be discussed in this tutorial, but is part of pestica...
- Requires: EPI timeseries and Anatomic data
- Software required: Bash, FSL, MATLAB

Steps overview

- 1) Prior spatial information -> setup script
- 2) Data -> ANALYZE format (origins=0,0,0)
- 3) MATLAB scripts, how to setup and run
- 4) QA of output, how to tell if estimators are accurate representation of noise sources

1) Setup Script

- Un-tar PESTICA distribution
 - *tar xvfz pestica.tar.gz*
 - *cd pestica*
- Edit *bash* script inside PESTICA distribution
 - Edit *setup_pestica.sh*, choose 1) data directory, 2) voxel volume and 3) orientation (contact me if you use *tcs*h or another shell)
- Run setup ONLY once from pestica directory
 - *./setup_pestica.sh*
 - Sets up links to the appropriate orientation geometry, check with viewer against your data, creates a file “.firsttime” so it doesn’t relink files during future runs of this setup script.
 - » If you later need to setup with different orientation, remove .firsttime and re-run again from inside pestica directory
- Put setup into your startup script
 - Edit .bashrc or similar startup script in your home dir, add:
 - *source /home/user/pestica_directory/setup_pestica.sh*
 - Or run *source* line manually every time you need PESTICA

```
fmri@pub2:/mnt/netScratch/fmri/pestica2mm - Shell - Konsole
Session Edit View Bookmarks Settings Help

[fmri@pub2 ~]$ cd /mnt/netScratch/fmri/pestica2mm
[fmri@pub2 pestica2mm]$ ls
apply_PESTICA1.m          prepare_ICA_decomp.m
apply_PESTICA2.m          prepare_PESTICA1.m
assemble_slices_to_timeseries.m  README
disassemble_timeseries_to_slices.m  register_ep2d_stddev_to_MNI.m
eeglab                    register_epi2epi_PESTICA1.m
example_runfile.m         setup_pestica.sh
get_correlation_threshold.m  tfilter_fft.m
optimize_pmu.m            view_and_correct_estim_for.m
pestica_volumes
[fmri@pub2 pestica2mm]$ emacs setup_pestica.sh

[1]+  Stopped                  emacs setup_pestica.sh
[fmri@pub2 pestica2mm]$ bg
[1]+  emacs setup_pestica.sh &
[fmri@pub2 pestica2mm]$ . /mnt/autofs/netHome/scratch/fmri/pestica2mm/setup_pestica.sh
[fmri@pub2 pestica2mm]$ env | grep PESTICA
PESTICA_ORIENT=RAI
PESTICA_DIR=/mnt/autofs/netHome/scratch/fmri/pestica2mm
PESTICA_VOL=16mm3
[fmri@pub2 pestica2mm]$ ls
apply_PESTICA1.m          MNI_T1.hdr          resp_mean_mni.hdr
apply_PESTICA2.m          MNI_T1.img          resp_mean_mni.img
assemble_slices_to_timeseries.m  optimize_pmu.m      setup_pestica.sh
card_mean_mni.hdr         pestica_volumes     stddev_avg_mni.hdr
card_mean_mni.img         prepare_ICA_decomp.m  stddev_avg_mni.img
disassemble_timeseries_to_slices.m  prepare_PESTICA1.m  tfilter_fft.m
eeglab                    README              view_and_correct_estim_for.m
example_runfile.m         register_ep2d_stddev_to_MNI.m
get_correlation_threshold.m  register_epi2epi_PESTICA1.m
[fmri@pub2 pestica2mm]$
```

1. Choose directory for pestica
2. Edit setup_pestica.sh
3. Source the setup file for first time (first time MUST be in directory)
4. Check environment and files (links are made only during the first time setup_pestica.sh is run, subsequent runs only setup environment)

```
emacs@localhost.localdomain
File Edit Options Buffers Tools Insert Help

# edit the hard-coded base directory containing the matlab code and the averaged vol
export PESTICA_DIR="/mnt/autofs/netHome/scratch/fmri/pestica2mm"

# edit your matrix size (approximate, decide which is closer to your voxel volume, 16mm^3 or 64mm^3)
# one set was created using 2x2x4mm voxels (16mm^3), the other with 4x4x4mm voxels (64mm^3)
export PESTICA_VOL="16mm3"
#export PESTICA_VOL="64mm3"

# edit the hard-coded orientation depending on your scanner's reconstruction orientation
export PESTICA_ORIENT="RAI"

-- setup_pestica.sh (Shell-script[bash])--L1--Top-----
Minibuffer window is not active
```

- 1) Edit directory, pick best
- 2) matrix size (16 or 64mm³),
- and 3) choose orientation

2) Data setup

- Data must be in ANALYZE format
 - `avwcreatehd <xd> <y> <z> <td> <dx> <dy> <dz> <dt> 0 0 0 4 epi`
 - Where the zeros refer to origin, leave these zeroed, not all apps recognize these fields. The last numeric input is datatype; 4=short (more common), 16=float.
- Mask out non-brain areas, eyes if possible by hand
 - `bet epi epi -n -m`
- If not already done, or not in startup files, source the setup script:
 - `source /home/user/pestica_directory/setup_pestica.sh`
- Check environment variables:
 - `env | grep PEST`
 - Output should show PESTICA_DIR, PESTICA_ORIENT, PESTICA_VOL setup correctly
- Check mask, data with viewer application
 - `fslview` or `afni`

```
fmri@pub2:/mnt/netScratch/fmri/test_pestica - Shell - Konsole
Session Edit View Bookmarks Settings Help

[fmri@pub2 fmri]$ mkdir test_pestica
[fmri@pub2 fmri]$ cd test_pestica/
[fmri@pub2 test_pestica]$ avwcreatehd 128 128 31 132 2 2 4 2800 0 0 0 4 epi
[fmri@pub2 test_pestica]$ cp /mnt/netRAID03/Trio/study893/S50sdt/S50vol.sdt epi.img
[fmri@pub2 test_pestica]$ cp /mnt/netRAID03/Trio/study893/S50sdt/resp_r1.dat .
[fmri@pub2 test_pestica]$ cp /mnt/netRAID03/Trio/study893/S50sdt/card_r1.dat .
[fmri@pub2 test_pestica]$ bet epi epi -n -m
[fmri@pub2 test_pestica]$ ls -l
total 132476
-rw-rw-r-- 1 fmri users 221713 May 1 13:33 card_r1.dat
-rw-rw-r-- 1 fmri users 348 May 1 13:32 epi.hdr
-rw-rw-r-- 1 fmri users 134086656 May 1 13:34 epi.img
-rw-rw-r-- 1 fmri users 348 May 1 13:33 epi_mask.hdr
-rw-rw-r-- 1 fmri users 1015808 May 1 13:33 epi_mask.img
-rw-rw-r-- 1 fmri users 145700 May 1 13:33 resp_r1.dat
[fmri@pub2 test_pestica]$ ./mnt/autofs/netHome/scratch/fmri/pestica2mm/setup_pestica.sh
[fmri@pub2 test_pestica]$ env | grep PEST
PESTICA_ORIENT=RAI
PESTICA_DIR=/mnt/autofs/netHome/scratch/fmri/pestica2mm
PESTICA_VOL=16mm3
[fmri@pub2 test_pestica]$ matlab -nojvm -nosplash

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>> addpath /mnt/autofs/netHome/scratch/fmri/pestica2mm/
>> prepare_ICA_decomp(15,'epi','epi_mask');
>>
>> !cp -R /mnt/netRAID03/Trio/study893/S50sdt/pest
pest_resamples.mat pestica/
>> !cp -R /mnt/netRAID03/Trio/study893/S50sdt/pestica/ .
>> ls pestica/
pestica_15comps_slice10.mat pestica_15comps_slice20.mat pestica_15comps_slice30.mat
pestica_15comps_slice11.mat pestica_15comps_slice21.mat pestica_15comps_slice31.mat
pestica_15comps_slice12.mat pestica_15comps_slice22.mat pestica_15comps_slice32.mat
pestica_15comps_slice13.mat pestica_15comps_slice23.mat pestica_15comps_slice33.mat
pestica_15comps_slice14.mat pestica_15comps_slice24.mat pestica_15comps_slice34.mat
pestica_15comps_slice15.mat pestica_15comps_slice25.mat pestica_15comps_slice35.mat
pestica_15comps_slice16.mat pestica_15comps_slice26.mat pestica_15comps_slice36.mat
pestica_15comps_slice17.mat pestica_15comps_slice27.mat pestica_15comps_slice37.mat
pestica_15comps_slice18.mat pestica_15comps_slice28.mat pestica_15comps_slice38.mat
pestica_15comps_slice19.mat pestica_15comps_slice29.mat pestica_15comps_slice39.mat
pestica_15comps_slice1.mat pestica_15comps_slice2.mat

>> !avwcreatehd 256 256 120 1 1 1 1.2 0 0 0 0 4 mprage
>> !cp /mnt/netRAID03/Trio/study893/S2sdt/S2vol.sdt mprage.img
>> register_ep2d_stddev_to_MNI('mprage','epi');
```

1. Put data in ANALYZE format, mask
2. Source the setup file prior to MATLAB (after first time, run in any directory)
3. Check environment variables
4. Start matlab, addpath



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3) PESTICA scripts

- Steps: ICA decomposition, registration to spatial priors, application of spatial priors.

- 1) Slicewise ICA decomposition

prepare_ICA_decomposition(components, epi, epi_mask)

- 2) Registration of anatomy, EPI to template

register_ep2d_stddev_to_MNI(anatomy,epi)

- 3) Apply spatial priors to ICA decomposition to retrieve noise sources and re-order in slice-acquisition order

apply_PESTICA2(components,epi,epi_mask,slice_timing,TR)

Very important: Before proceeding, look at co-registered priors in new subdir *./pestica/resp/card_PESTICA2* should look like Figure 11 in our manuscript when overlain on anatomic data.


```
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Session Edit View Bookmarks Settings Help

[fmri@pub2 fmri]$ mkdir test_pestica
[fmri@pub2 fmri]$ cd test_pestica/
[fmri@pub2 test_pestica]$ avwcreatehd 128 128 31 132 2 2 4 2800 0 0 0 4 epi
[fmri@pub2 test_pestica]$ cp /mnt/netRAID03/Trio/study893/S50sdt/S50vol.sdt epi.img
[fmri@pub2 test_pestica]$ cp /mnt/netRAID03/Trio/study893/S50sdt/resp_r1.dat .
[fmri@pub2 test_pestica]$ cp /mnt/netRAID03/Trio/study893/S50sdt/card_r1.dat .
[fmri@pub2 test_pestica]$ bet epi epi -n -m
[fmri@pub2 test_pestica]$ ls -l
total 132476
-rw-rw-r-- 1 fmri users 221713 May 1 13:33 card_r1.dat
-rw-rw-r-- 1 fmri users 348 May 1 13:32 epi.hdr
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-rw-rw-r-- 1 fmri users 1015808 May 1 13:33 epi_mask.img
-rw-rw-r-- 1 fmri users 145700 May 1 13:33 resp_r1.dat
[fmri@pub2 test_pestica]$ ./mnt/autofs/netHome/scratch/fmri/pestica2mm/setup_pestica.sh
[fmri@pub2 test_pestica]$ env | grep PEST
PESTICA_ORIENT=RAI
PESTICA_DIR=/mnt/autofs/netHome/scratch/fmri/pestica2mm
PESTICA_VOL=16mm3
[fmri@pub2 test_pestica]$ matlab -nojvm -nosplash

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>> addpath /mnt/autofs/netHome/scratch/fmri/pestica2mm/
>> prepare_ICA_decomp(15,'epi','epi_mask');
>>
>> !cp -R /mnt/netRAID03/Trio/study893/S50sdt/pest
pest_resamples.mat pestica/
>> !cp -R /mnt/netRAID03/Trio/study893/S50sdt/pestica/ .
>> ls pestica/
pestica_15comps_slice10.mat pestica_15comps_slice20.mat pestica_15comps_slice30.mat
pestica_15comps_slice11.mat pestica_15comps_slice21.mat pestica_15comps_slice31.mat
pestica_15comps_slice12.mat pestica_15comps_slice22.mat pestica_15comps_slice32.mat
pestica_15comps_slice13.mat pestica_15comps_slice23.mat pestica_15comps_slice33.mat
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pestica_15comps_slice16.mat pestica_15comps_slice26.mat pestica_15comps_slice36.mat
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>> register_ep2d_stddev_to_MNI('mprage','epi');
```

1. Run slicewise ICA decomposition
2. Get anatomic data in ANALYZE
3. Registration to MNI + variance map of EPI registration to template



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3) Temporally filter estimators

- *view_and_correct_estimators(input,fs)*
- This script plots the frequency spectrum of the estimator, it is up to operator to pick a frequency range to accept.
- There is substantial heart and breathing rate variability, nevertheless it should be possible to identify major peaks.
- Avoid sidebands if clearly separated from main peak

```
fmri@pub2:/mnt/netScratch/fmri/test_pestica - Shell - Konsole
Session Edit View Bookmarks Settings Help

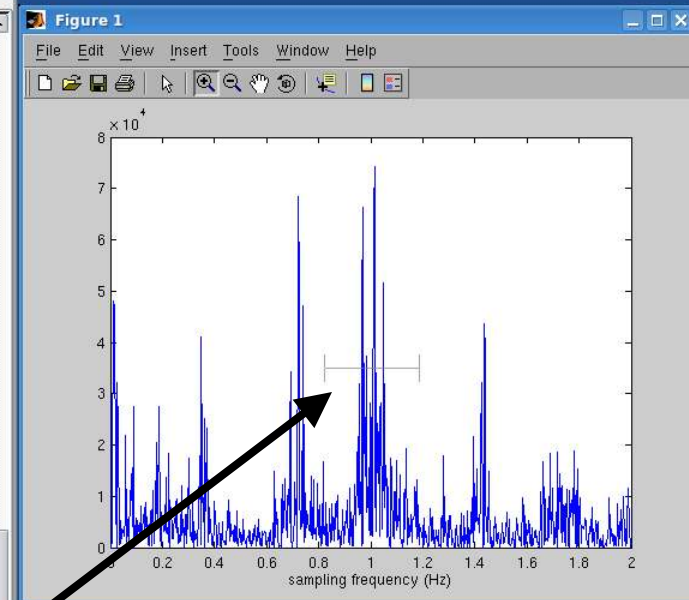
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pestica_15comps_slice12.mat pestica_15comps_slice22.mat pestica_15comps_slice32.mat
pestica_15comps_slice13.mat pestica_15comps_slice23.mat pestica_15comps_slice33.mat
pestica_15comps_slice14.mat pestica_15comps_slice24.mat pestica_15comps_slice34.mat
pestica_15comps_slice15.mat pestica_15comps_slice25.mat pestica_15comps_slice35.mat
pestica_15comps_slice16.mat pestica_15comps_slice26.mat pestica_15comps_slice36.mat
pestica_15comps_slice17.mat pestica_15comps_slice27.mat pestica_15comps_slice37.mat
pestica_15comps_slice18.mat pestica_15comps_slice28.mat pestica_15comps_slice38.mat
pestica_15comps_slice19.mat pestica_15comps_slice29.mat pestica_15comps_slice39.mat
pestica_15comps_slice1.mat pestica_15comps_slice2.mat

>> !avwcreatehd 256 256 120 1 1 1 1.2 0 0 0 0 4 mprage
>> !cp /mnt/netRAID03/Trio/study893/S2sdt/S2vol.sdt mprage.img
>> register_ep2d_stdev to MNIT('mprage', 'epi');
>> [card_est2,resp_est2]=apply_PESTICA2(15,'epi','epi_mask','alt-asc');
>> slice_sample_rate 31/(2800/1000);
>> fcard2=view_and_correct_estimator(card_est2,slice_sample_rate);
hit enter when you have zoomed on a reasonable pmu peak (cardiac typically near 1Hz, resp near 0.1Hz)
[]
```



1. Run PESTICA2 corr procedure
2. view_and_correct_estimator
3. Pick a range of frequencies with the mouse (my heart rate is 1Hz, it can be difficult sometimes due to sidebands as seen above; regardless, pick the largest freq near where you expect the physio cycle to be)

```
fmri@pub2:/mnt/netScratch/fmri/test_pestica - Shell - Konsole
Session Edit View Bookmarks Settings Help

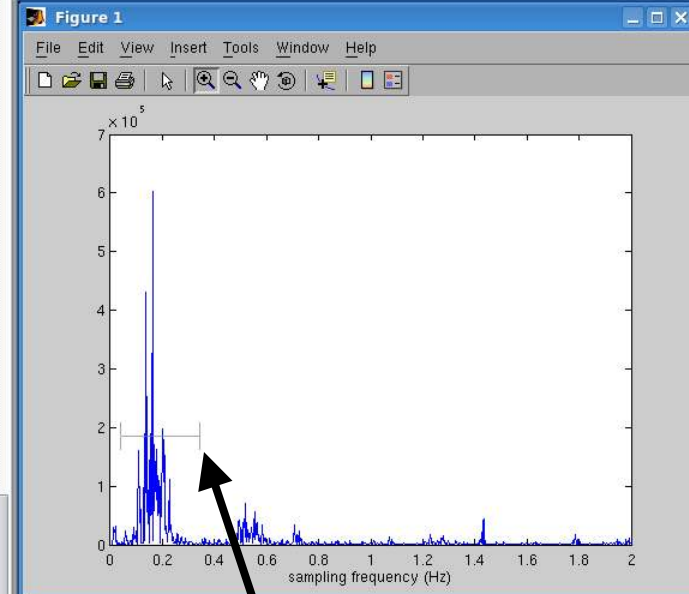
[fmri@pub2 test_pestica]$ avwcreatehd 128 128 31 132 2 2 4 2800 0 0 0 4 epi
[fmri@pub2 test_pestica]$ cp /mnt/netRAID03/Trio/study893/S50sdt/S50vol.sdt epi.img
[fmri@pub2 test_pestica]$ cp /mnt/netRAID03/Trio/study893/S50sdt/resp_r1.dat .
[fmri@pub2 test_pestica]$ cp /mnt/netRAID03/Trio/study893/S50sdt/card_r1.dat .
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[fmri@pub2 test_pestica]$ ls -l
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[fmri@pub2 test_pestica]$ env | grep PEST
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PESTICA_DIR=/mnt/autofs/netHome/scratch/fmri/pestica2mm
PESTICA_VOL=16mm3
[fmri@pub2 test_pestica]$ matlab -nojvm -nosplash

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>>
>> !cp -R /mnt/netRAID03/Trio/study893/S50sdt/pest
pest_resamples.mat pestica/
>> !cp -R /mnt/netRAID03/Trio/study893/S50sdt/pestica/ .
>> ls pestica/
pestica_15comps_slice10.mat pestica_15comps_slice20.mat pestica_15comps_slice30.mat
pestica_15comps_slice11.mat pestica_15comps_slice21.mat pestica_15comps_slice31.mat
pestica_15comps_slice12.mat pestica_15comps_slice22.mat pestica_15comps_slice32.mat
pestica_15comps_slice13.mat pestica_15comps_slice23.mat pestica_15comps_slice33.mat
pestica_15comps_slice14.mat pestica_15comps_slice24.mat pestica_15comps_slice34.mat
pestica_15comps_slice15.mat pestica_15comps_slice25.mat pestica_15comps_slice35.mat
pestica_15comps_slice16.mat pestica_15comps_slice26.mat pestica_15comps_slice36.mat
pestica_15comps_slice17.mat pestica_15comps_slice27.mat pestica_15comps_slice37.mat
pestica_15comps_slice18.mat pestica_15comps_slice28.mat pestica_15comps_slice38.mat
pestica_15comps_slice19.mat pestica_15comps_slice29.mat pestica_15comps_slice39.mat
pestica_15comps_slice1.mat pestica_15comps_slice2.mat

>> !avwcreatehd 256 256 120 1 1 1 1.2 0 0 0 0 4 mprage
>> !cp /mnt/netRAID03/Trio/study893/S2sdt/S2vol.sdt mprage.img
>> register_ep2d_stddev_to_MNI('mprage','epi');
>> [card_est2,resp_est2]=apply_PESTICA2(15,'epi','epi_mask','alt-asc');
>> slice_sample_rate=31/(2800/1000);
>> fcard2=view_and_correct_estimator(card_est2,slice_sample_rate);
hit enter when you have zoomed on a reasonable pmu peak (cardiac typically near 1Hz, resp near 0.25Hz)
>> fresp2=view_and_correct_estimator(resp_est2,slice_sample_rate);
hit enter when you have zoomed on a reasonable pmu peak (cardiac typically near 1Hz, resp near 0.25Hz)
[]
```



1. Do same for respiration, 0.2Hz
2. Store output in files for use later



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4) Estimator validation

- Two ways to validate: 1) correlate with parallel measured PMU (but we typically don't have this) or 2) look at the spatial distribution of coupling.
- Use script to perform RETROICOR algorithm on epi data using estimators, output is a spatial coupling map
matlab_retroicor(epi,card_est,resp_est,slice_timing,...)
- Examine variance-normalized coupling histogram: should show substantial deviation from 4th order chi-square distribution due to real coupling.


```

fmri@pub2:/mnt/netScratch/fmri/test_pestica - Shell - Konsole
Session Edit View Bookmarks Settings Help

>> load card_icaest2.dat
>> load resp_icaest2.dat
>> load card_r1.dat
>> load resp_r1.dat
>> card_pmu=assemble_slices_to_timeseries(disassemble_timeseries_to_slices(card_r1,31,132),31,132);
using interleaved ascending 'alt-asc' for the slice timing
using interleaved ascending 'alt-asc' for the slice timing
>> resp_pmu=assemble_slices_to_timeseries(disassemble_timeseries_to_slices(resp_r1,31,132),31,132);
using interleaved ascending 'alt-asc' for the slice timing
using interleaved ascending 'alt-asc' for the slice timing
>> card_pmu=(card_pmu-mean(card_pmu))/std(card_pmu);
>> resp_pmu=(resp_pmu-mean(resp_pmu))/std(resp_pmu);
>> whos

Name              Size              Bytes  Class      Attributes

card_icaest2       4092x1              32736  double
card_pmu           1x4092              32736  double
card_r1            18480x1             147840  double
resp_icaest2       4092x1              32736  double
resp_pmu           1x4092              32736  double
resp_r1            18480x1             147840  double

>> corr(card_icaest2,card_pmu')

ans =

    0.5327

>> corr(resp_icaest2,resp_pmu')

ans =

    0.6110

>> % run these estimators through retroicor and examine the coupling maps
>> addpath /mnt/netScratch/fmri/test_pestica/matlab_retroicor
>> tic
>> [retima,im_card,im_resp,cardph,respch]=matlab_retroicor('epi',card_icaest2,resp_icaest2,2,'alt-asc','epi_mask'); toc
Elapsed time is 50.875871 seconds.
>> whos

Name              Size              Bytes  Class      Attributes

ans               1x1                32736  double
card_icaest2       4092x1              32736  double
card_pmu           1x4092              32736  double
card_r1            18480x1             147840  double
cardph             4092x1              32736  double
im_card            128x128x31          5000000  double
im_resp            128x128x31          5000000  double
resp_icaest2       4092x1              32736  double
resp_pmu           1x4092              32736  double
resp_r1            18480x1             147840  double
respch             4092x1              32736  double
retima             4-D                 5000000  double

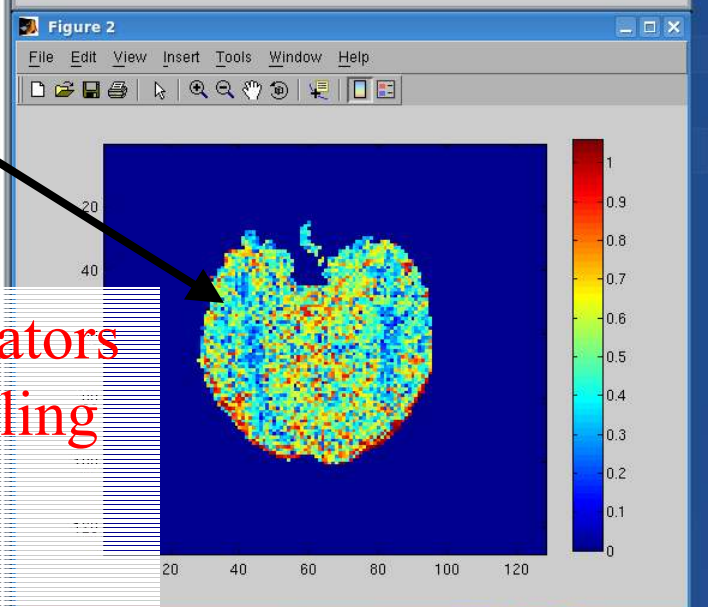
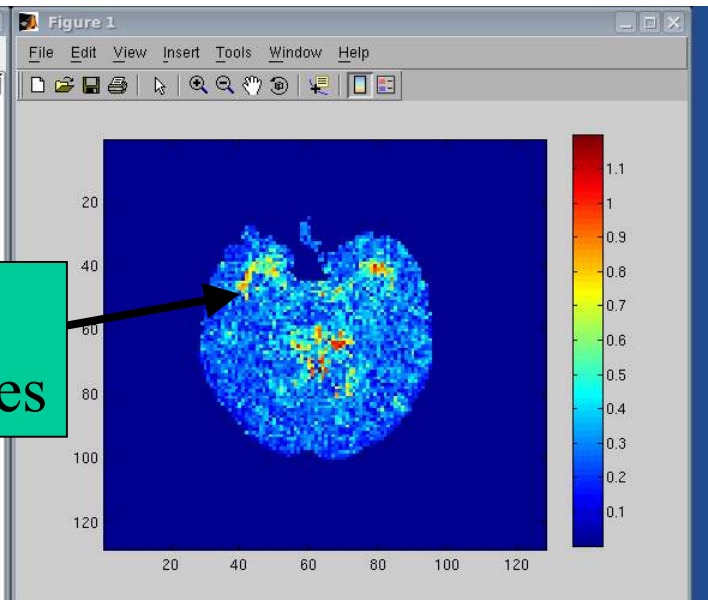
>> imagesc(im_card(:,:,12))
>> figure; imagesc(im_resp(:,:,12))
>>

```

Cardiac:
MCA and arteries

Respiration:
Edges and CSF

1. Compare PESTICA estimators with raw data (same sampling rate, use correlation)
2. OR, use matlab_retroicor routine and look at coupling maps



CentOS 5

Coupling maps

- Use RETROICOR to determine coupling to estimators and to random sinusoids in order to validate estimators. Output maps are variance normalized, so cardiac should show up in arteries...

- Random sinusoids for noise coupling:

```
[retimas,im_cards,im_resps]=matlab_retroicor('epi',sin(sort(unifrnd(0,2*pi,4092,1)*360)),  
sin(sort(unifrnd(0,2*pi,4092,1)*360/5)))
```

- Histogram and display coupling maps:

```
hist(im_card(find(im_card~=0)),0:0.01:2)←
```

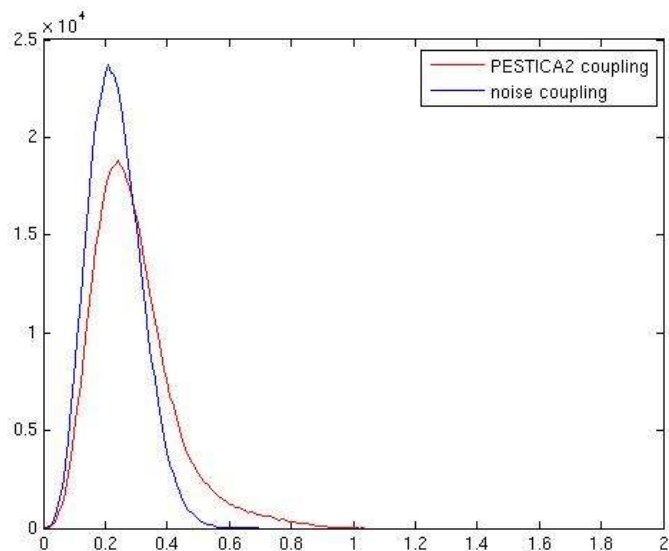
```
hist(im_cards(find(im_cards~=0)),0:0.01:2)←
```

```
imagesc(im_card(:, :, 12))←
```

```
imagesc(im_cards(:, :, 12))←
```

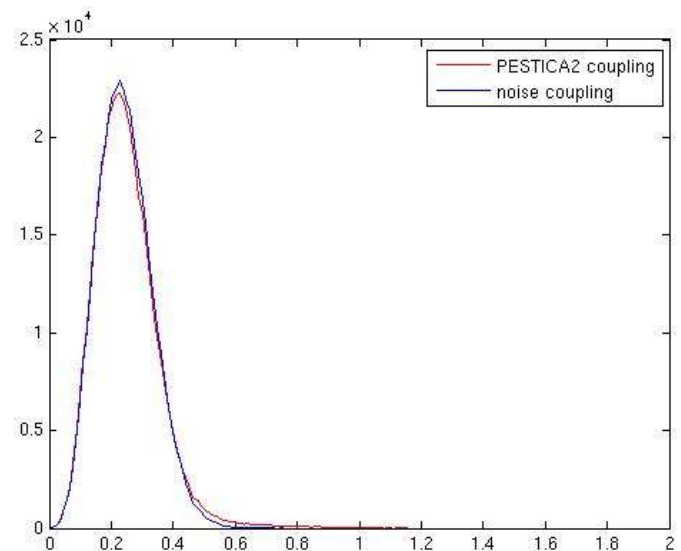
PESTICA coupling

Noise coupling



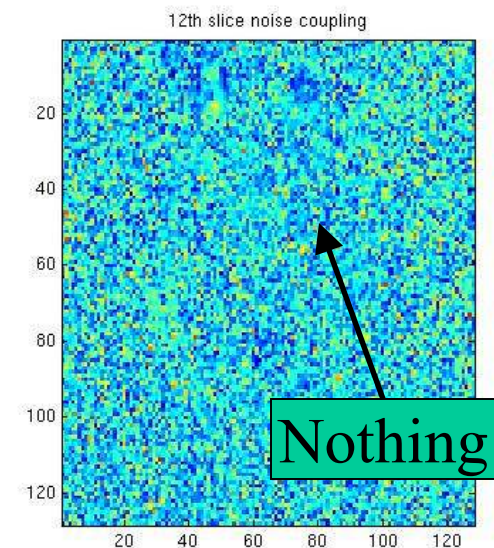
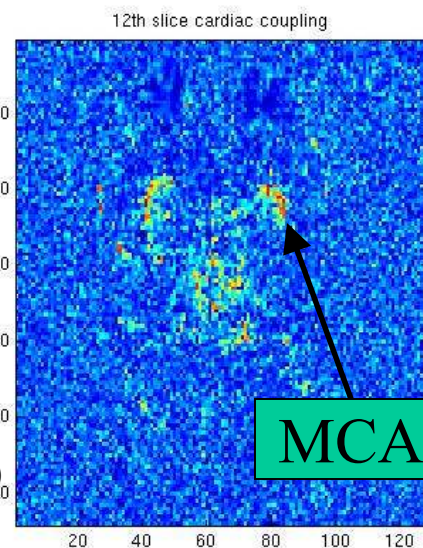
Respiratory coupling is
clearly greater than noise

hist(im_resp(find(im_resp~=0)),0:0.01:2)



But cardiac is tougher to
see from histogram...

Solution: look at the coupling
Map for cardiac validation:
should look like the PESTICA
cardiac prior map, when
compared with noise (cardiac is same
as that two pages earlier, but with no mask here)



Final Remark

- PESTICA2 works most of time at our site to do away with need for monitoring, but it may still be better to obtain parallel physiologic monitoring for one EPI per subject, for both PESTICA1 (subject-specific spatial priors, which may be more accurate) and for validation purposes.
- For advice and help: ebeall@gmail.com