Reimplementation of Lonnberg et al GPFates (from their example)

In [112]:
```
import pandas as pd
import numpy as np
from GPfates import GPfates
import matplotlib.pyplot as plt
from scipy.linalg import svd
import GPy
```

Load in the expression data, filter and log normalize

In [35]:
```
etpm = pd.read_table('tapio_tcell_tpm.txt', index_col=0)
etpm = etpm[(etpm > 2).sum(1) > 2]
logexp = np.log10(etpm + 1)
```

Load metadata

In [36]:
```
tcells = pd.read_csv('tcells_rebuttal.csv', index_col=0)
```

Create a GPFates object

In [45]:
```
mydata = GPfates.GPfates(sample_info=tcells, expression_matrix=logexp)
```

Use a GPLVM to infer a low-dimensional representation of the data

In [46]:
```
mydata.dimensionality_reduction()
```

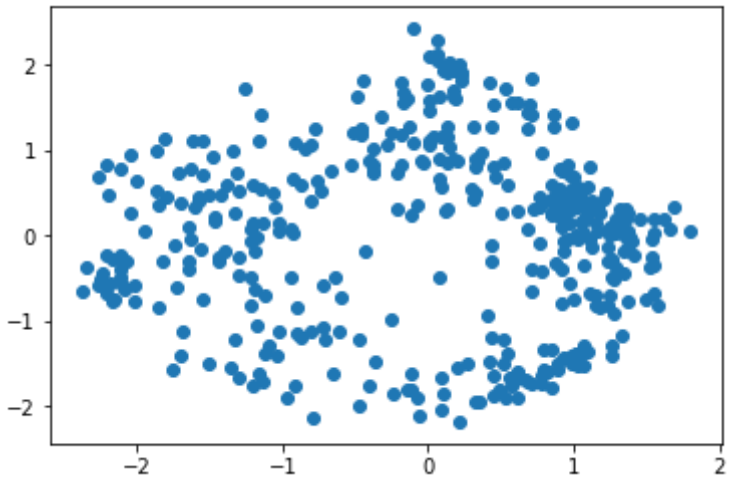
```
 /scratch/groups/abattle4/josh/gp_fates/GPfates/GPfates.py:62: FutureWa
rning:Method .as_matrix will be removed in a future version. Use .value
s instead.
```

This is very cool. Their dimensionality reduction calls GPy.models.BayesianGPLVM - note to self, check that out. I am now going to store these results, overwriting the previous data they previously had stored in their metadata

In [47]:
```
mydata.store_dr()
```

What does this dimensionality reduction look like?

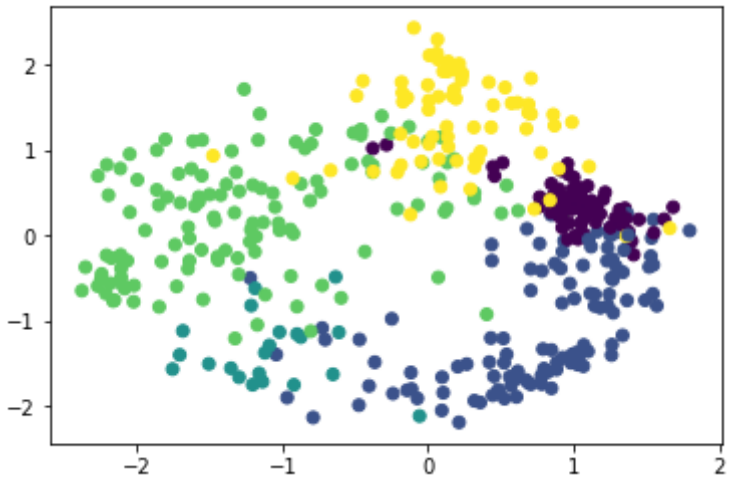
In [52]:
```
dim1 = mydata.s['bgplvm_0']
dim2 = mydata.s['bgplvm_1']
plt.scatter(dim1, dim2)
plt.show()
```



... neat? Try coloring by day

In [56]:
```
fig, ax = plt.subplots()
days = np.unique(mydata.s['day'])
colors = np.linspace(0, 1, len(days))
colordict = dict(zip(days, colors))

day2col = mydata.s['day'].apply(lambda x: colordict[x])
ax.scatter(dim1, dim2, c=day2col)
fig.show()
```

```
 /software/apps/anaconda/5.2/python/3.6/lib/python3.6/site-packages/mat
plotlib/figure.py:459: UserWarning:matplotlib is currently using a non-
GUI backend, so cannot show the figure
```



Not going to worry about a legend here, but the color assignments are:
day 0: purple
day 2: dark blue
day 3: dark teal
day 4: green
day 7: yellow
So this is showing clear separation by day. How does this dimensionality reduction compare to what PCA would give us?

In [65]:
```
mymat = mydata._gene_filter(None).as_matrix().T
u, s, vh = svd(mymat)
loadings = np.matmul(mymat, vh.T)
```

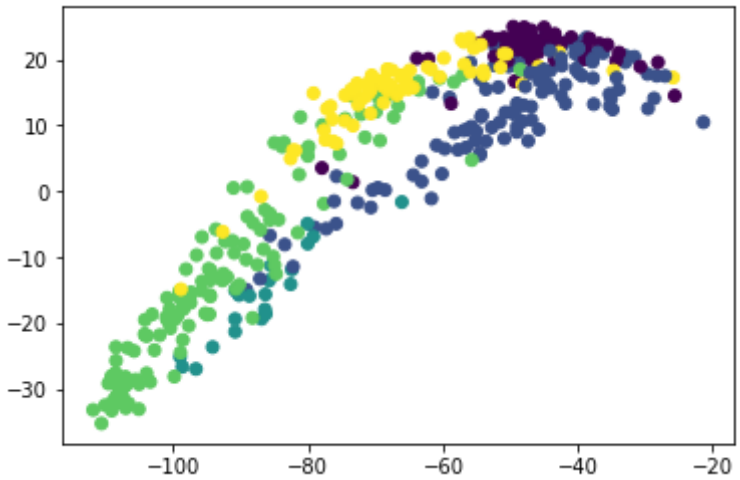
```
 /software/apps/anaconda/5.2/python/3.6/lib/python3.6/site-packages/ipy
kernel/__main__.py:1: FutureWarning:Method .as_matrix will be removed i
n a future version. Use .values instead.
```

In [66]:
```
loadings.shape
```

Out[66]:
```
(408, 13931)
```

In [68]:
```
fig, ax = plt.subplots()
days = np.unique(mydata.s['day'])
colors = np.linspace(0, 1, len(days))
colordict = dict(zip(days, colors))

day2col = mydata.s['day'].apply(lambda x: colordict[x])
ax.scatter(loadings[:,0], loadings[:,1], c=day2col)
fig.show()
```

```
 /software/apps/anaconda/5.2/python/3.6/lib/python3.6/site-packages/mat
plotlib/figure.py:459: UserWarning:matplotlib is currently using a non-
GUI backend, so cannot show the figure
```



Interestingly enough it looks like PCA may capture the exact same trend as the GPLVM did, though maybe with slightly worse resolution. I don't have a great understanding of how the GPLVM works, but this is surprising to me. I'll read up about Gaussian processes and GPLVMs and think about whether this should be a surprising result, or if this is what we should expect. I'm going to store these results.

In [70]:
```
mydata.s['pca_0'] = loadings[:,0]
mydata.s['pca_1'] = loadings[:,1]
```

Infer pseudotime

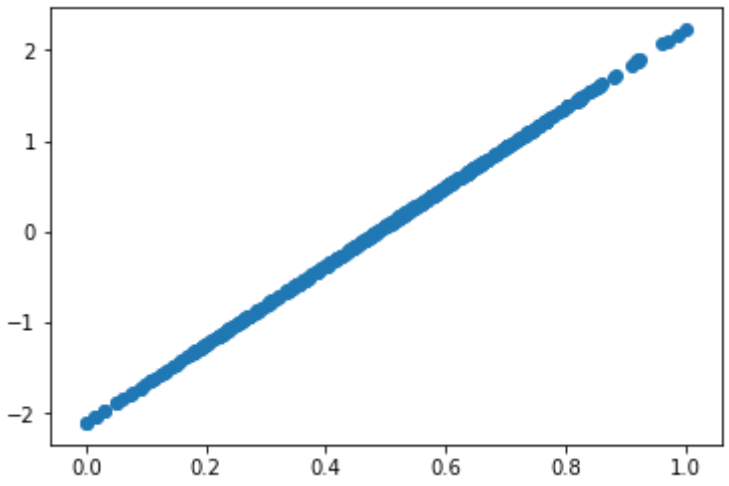
In [107]:
```
mydata.infer_pseudotime(priors=mydata.s.day_int, s_columns=['bgplvm_0',
'bgplvm_1'])
```

```
 /scratch/groups/abattle4/josh/gp_fates/GPfates/GPfates.py:36: FutureWa
rning:Method .as_matrix will be removed in a future version. Use .value
s instead.
```

This converged extremely quickly. At this point, they mention scaled_pseudotime, which I'm going to assume is just this inferred pseudotime scaled to be from zero to 1

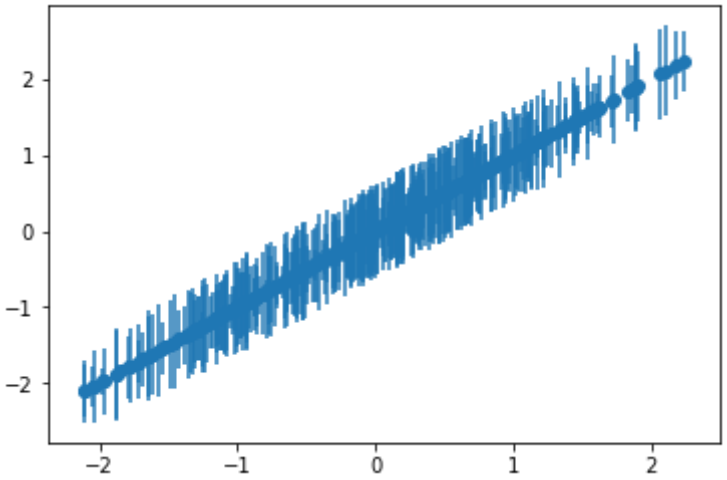
In [81]:
```
mydata.s.scaled_pseudotime = mydata.s.pseudotime+abs(min(mydata.s.pseudo
time))
mydata.s.scaled_pseudotime = mydata.s.scaled_pseudotime/max(mydata.s.sca
led_pseudotime)
```

In [83]:
```
plt.scatter(mydata.s.scaled_pseudotime, mydata.s.pseudotime)
plt.show()
```



I guess this is just meant to show what the scaling did? There's also a method to show uncertainty (psuedotime is their typo not mine)

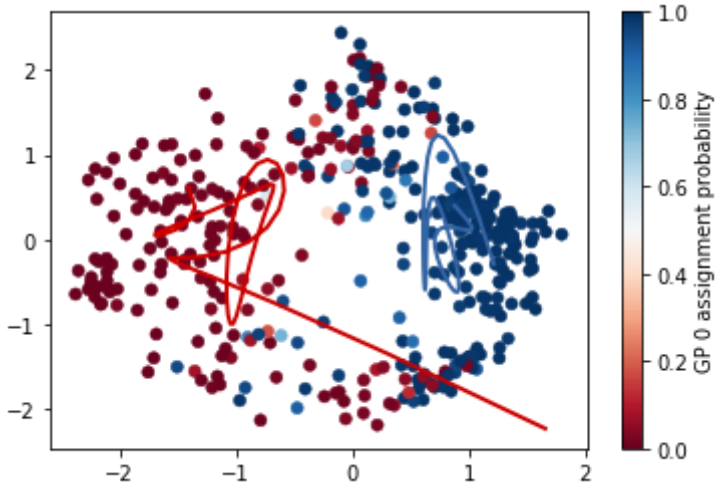
In [91]:
```
mydata.plot_psuedotime_uncertainty()
```



Now that we've inferred pseudotime, it's time to use the Overlapping Mixture of Gaussian Processes model to take a look at bifurcation - we assume that before bifurcation, the two processes are the same, and afterwards they are not

In [94]:
```
mydata.make_fates_viz(['bgplvm_0', 'bgplvm_1'])
mydata.fates_viz.plot()
plt.show()
```

```
 /scratch/groups/abattle4/josh/gp_fates/GPfates/GPfates.py:91: FutureWa
rning:Method .as_matrix will be removed in a future version. Use .value
s instead.
```

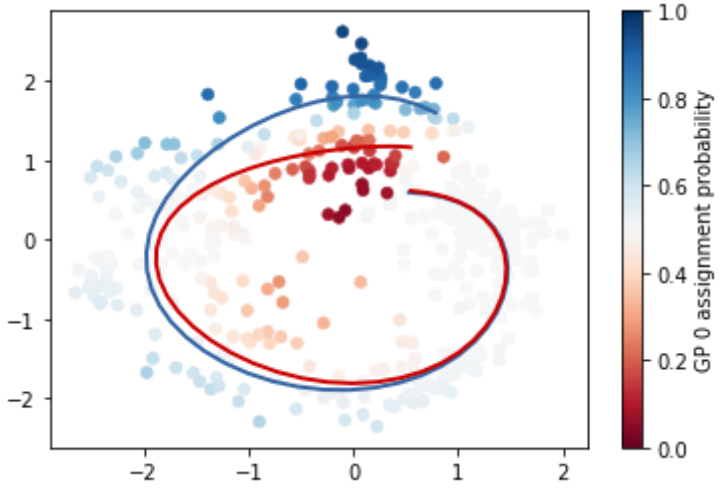


This is not looking great. Running blindly through their example worked fine, so I'm going to do that to see where the discrepancy is.

In [102]:
```
etpm = pd.read_table('tapio_tcell_tpm.txt', index_col=0)
etpm = etpm[(etpm > 2).sum(1) > 2]
logexp = np.log10(etpm + 1)
tcells = pd.read_csv('tcells_rebuttal.csv', index_col=0)
m = GPfates.GPfates(tcells, logexp)
m.model_fates(X=['bgplvm_2d_1'])
m.make_fates_viz(['bgplvm_2d_0', 'bgplvm_2d_1'])
m.fates_viz.plot()
GPfates.plt.show()
```

```
iteration 271 bound=801.1580740422485 grad=2.8115992671508806e-05, beta
=0.9078922558507376
iteration 272 bound=801.1580794566942 grad=2.5147293109987035e-05, beta
=0.9061646825934879
iteration 273 bound=801.158084289666 grad=2.2499198192975316e-05, beta=
0.9041012425965143
iteration 274 bound=801.1580885953997 grad=2.0149393787036934e-05, beta
=0.9021298339542735
iteration 275 bound=801.1580924280307 grad=1.8072686419343334e-05, beta
=0.9009053438419835
iteration 276 bound=801.1580958418717 grad=1.6238922819426454e-05, beta
=0.9010144028569784
iteration 277 bound=801.1580988896069 grad=1.4613809505173855e-05, beta
=0.9026267651784781
iteration 278 bound=801.1581016192335 grad=1.3162402236517862e-05, beta
=0.9052861009701758
iteration 279 bound=801.1581040712516 grad=1.1853638566479064e-05, beta
=0.9080165915195133
iteration 280 bound=801.1581062774967 grad=1.066393741160049e-05, beta=
0.9097449026264491
iteration 281 bound=801.1581082620602 grad=9.578501447782435e-06, beta=
0.9098021498826571
iteration 282 bound=801.1581100437412 grad=8.59000100348354e-06, beta=
0.9082136676077254
iteration 283 bound=801.1581116389499 grad=7.695345803843171e-06, beta=
0.9056527804306899
iteration 284 bound=801.1581130639222 grad=6.891962779561014e-06, beta=
0.9031268614144754
iteration 285 bound=801.1581143356128 grad=6.17510582057254e-06, beta=
0.9015391552598945
iteration 286 bound=801.1581154712719 grad=5.537108642321339e-06, beta=
0.9012800357240888
iteration 287 bound=801.15811648721 grad=4.968468768864458e-06, beta=0.
9020184107364831
iteration 288 bound=801.1581173975476 grad=4.459830590338401e-06, beta=
0.9028361841065322
vb converged (ftol)
```

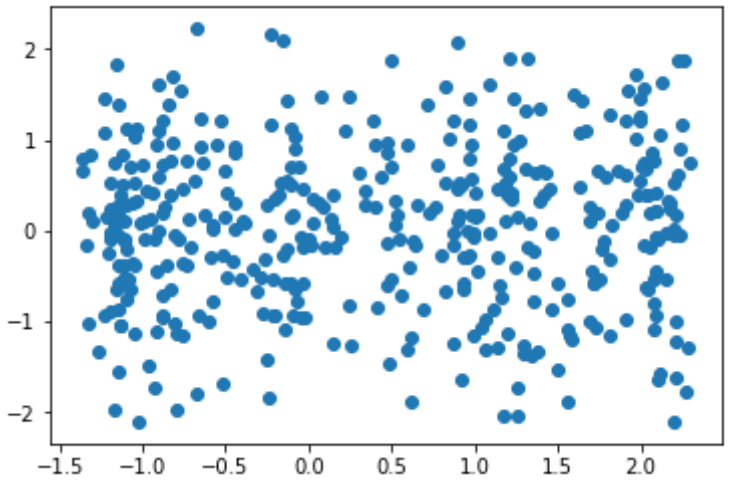
```
 /scratch/groups/abattle4/josh/gp_fates/GPfates/GPfates.py:91: FutureWa
rning:Method .as_matrix will be removed in a future version. Use .value
s instead.
```



In [104]:
```
assert((m.s.day==mydata.s.day).all())
```

In [106]:
```
plt.scatter(m.s.pseudotime, mydata.s.pseudotime)
```

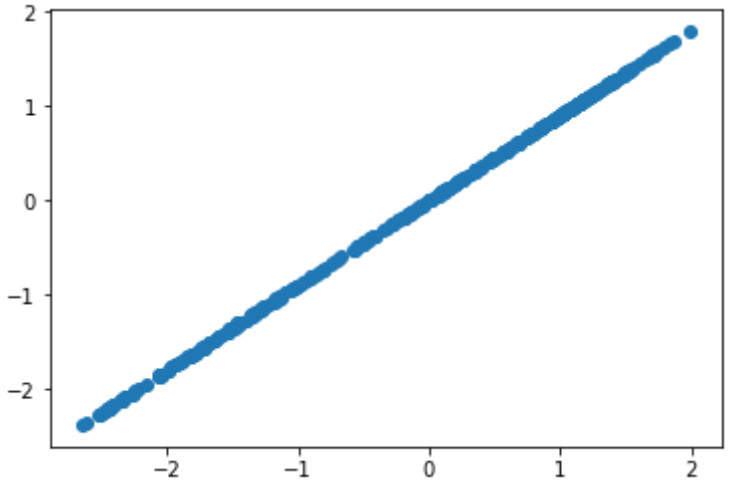
Out[106]: <matplotlib.collections.PathCollection at 0x2ae899bc9e80>



Well, that'll do it.

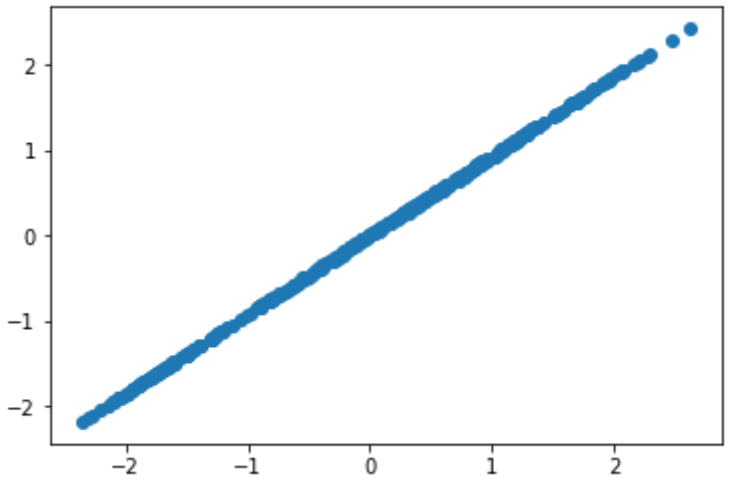
In [108]:
```
plt.scatter(m.s.bgplvm_2d_0, mydata.s.bgplvm_0)
```

Out[108]: <matplotlib.collections.PathCollection at 0x2ae899c2ba58>



In [109]:
```
plt.scatter(m.s.bgplvm_2d_1, mydata.s.bgplvm_1)
```

Out[109]:
```
<matplotlib.collections.PathCollection at 0x2ae899c69cf8>
```



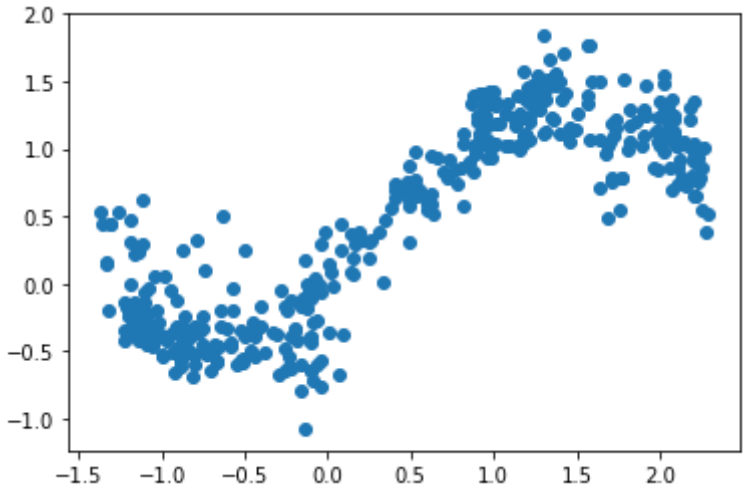
Considering how quickly pseudotime inference was converging, I think the error must be somewhere in pseudotime inference, because the dimensionality reduction being used is the same

In [119]:
```
Y = mydata.s[['bgplvm_0', 'bgplvm_1']].as_matrix()

mydata.time_model = GPy.models.BayesianGPLVM(Y, 1, init='random')

mydata.time_model.rbf.lengthscale.constrain_fixed(2., warning=False)
mydata.time_model.rbf.variance.constrain_fixed(200., warning=False)

# priors = mydata.s.day_int+np.ones(len(mydata.s.day_int),dtype=int)
priors = mydata.s.day_int
if priors is not None:
    for i, p in enumerate(priors):
        prior = GPy.priors.Gaussian(p, 2.)
        mydata.time_model.X.mean[i, [0]].set_prior(prior, warning=False)

mydata.time_model.optimize(max_iters=2000, messages=True)
new_pseudotime = mydata.time_model.X.mean[:, [0]]
```

```
 /software/apps/anaconda/5.2/python/3.6/lib/python3.6/site-packages/ipy
kernel/__main__.py:1: FutureWarning:Method .as_matrix will be removed i
n a future version. Use .values instead.
```

In [120]:
```
plt.scatter(m.s.pseudotime, mydata.s.pseudotime)
```

Out[120]: <matplotlib.collections.PathCollection at 0x2ae899cbe2e8>

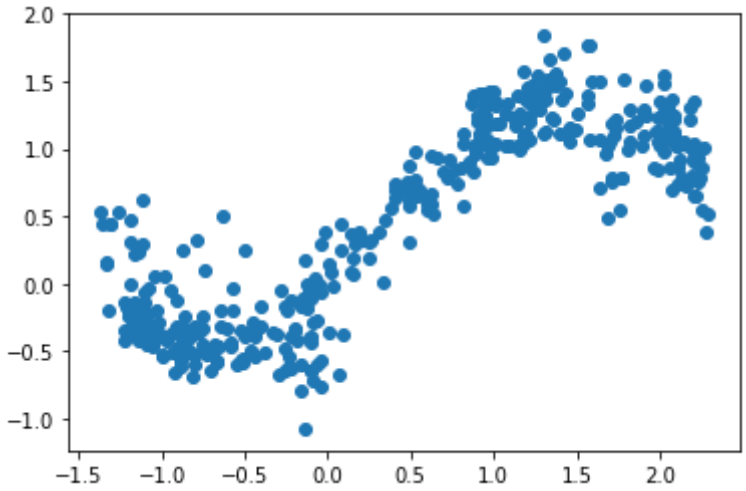


In [121]:
```
Y = mydata.s[['bgplvm_0', 'bgplvm_1']].as_matrix()

mydata.time_model = GPy.models.BayesianGPLVM(Y, 1, init='random')

mydata.time_model.rbf.lengthscale.constrain_fixed(2., warning=False)
mydata.time_model.rbf.variance.constrain_fixed(200., warning=False)

# priors = mydata.s.day_int+np.ones(len(mydata.s.day_int),dtype=int)
priors = mydata.s.day_int
if priors is not None:
    for i, p in enumerate(priors):
        prior = GPy.priors.Gaussian(p, 2.)
        mydata.time_model.X.mean[i, [0]].set_prior(prior, warning=False)

mydata.time_model.optimize(max_iters=2000, messages=True)
new_pseudotime2 = mydata.time_model.X.mean[:, [0]]
```

```
 /software/apps/anaconda/5.2/python/3.6/lib/python3.6/site-packages/ipy
kernel/__main__.py:1: FutureWarning:Method .as_matrix will be removed i
n a future version. Use .values instead.
```

In [122]:
```
plt.scatter(m.s.pseudotime, mydata.s.pseudotime)
```

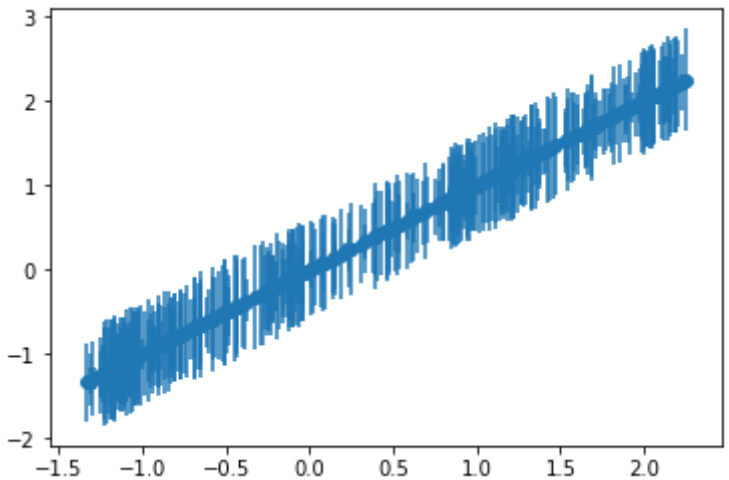
Out[122]:
```
<matplotlib.collections.PathCollection at 0x2ae899dbfb38>
```



Now there's at least a relationship between the two pseudotime metrics, although I don't immediately know why the relationship would be sinusoidal. This seems like it'd have something to do with the fact that the pattern is circular. Traversing a unit circle, as x moves from 1 to 1 clockwise (like our data does with respect to latent component 1), y follows a sinusoidal pattern very much like this one. In their example where they don't actually do any learning, just using saved metadata, that metadata is labeled 'bgplvm_2d_0/1' rather than 'bgplvm_0/1' - I think there's some way to incorporate the two-dimensional nature of the data that I am not accounting for, but that'll have to wait until after my 4 hours. Either way, this should be better than the total randomness observed earlier. Does this improve results?

In [123]:
```
mydata.s['pseudotime'] = new_pseudotime
mydata.s.scaled_pseudotime = mydata.s.pseudotime+abs(min(mydata.s.pseudo
time))
mydata.s.scaled_pseudotime = mydata.s.scaled_pseudotime/max(mydata.s.sca
led_pseudotime)
```

In [124]:
```
mydata.plot_psuedotime_uncertainty()
```

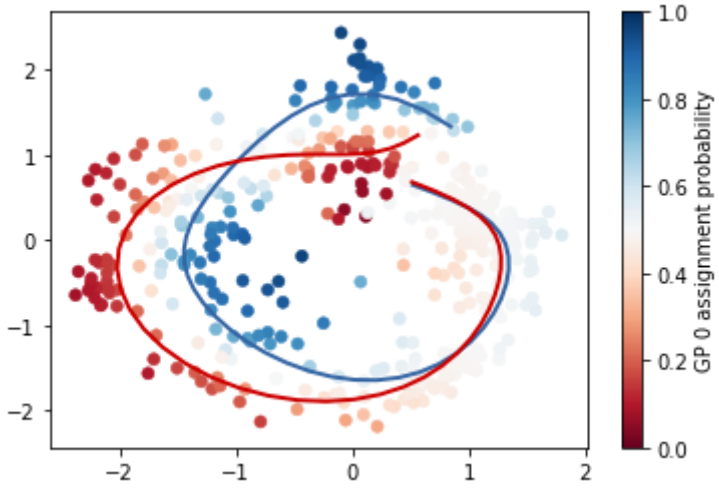


In [125]:
```
mydata.model_fates(t='pseudotime', X=['bgplvm_0', 'bgplvm_1'], C=2, step
_length=0.01)
```

```
iteration 380 bound=811.799680525923 grad=2.3669643316142378e-05, beta=
0.839629293695068
iteration 381 bound=811.7996825995381 grad=1.9983629385461894e-05, beta
=0.8436080786429118
iteration 382 bound=811.799684329364 grad=1.687889816244994e-05, beta=
0.8477773579394488
iteration 383 bound=811.7996857766884 grad=1.4231861371508297e-05, beta
=0.8501248430130088
iteration 384 bound=811.7996869864276 grad=1.1967007147124746e-05, beta
=0.849390849231977
iteration 385 bound=811.7996879938785 grad=1.0041823582431597e-05, beta
=0.8457835377864468
iteration 386 bound=811.7996888298226 grad=8.427347070984388e-06, beta=
0.8409028514562867
vb converged (ftol)
```

In [126]:
```
mydata.make_fates_viz(['bgplvm_0', 'bgplvm_1'])
mydata.fates_viz.plot()
plt.show()
```

```
 /scratch/groups/abattle4/josh/gp_fates/GPfates/GPfates.py:91: FutureWa
rning:Method .as_matrix will be removed in a future version. Use .value
s instead.
```



WOOHOO!! We have trajectories. They seem to bifurcate somewhat prematurely, but this looks pretty good overall. Many questions to dig into, but I believe my big problems came from the infer_pseudotime method being far from robust. Running it several times, I sometimes ran into errors, sometimes saw it converge immediately, etc. I think that is the reason the first results were essentially garbage. Why my inferred pseudotime is a sinusoidal transformation of theirs, and where they got their bgplvm_2d latent variables, I will need to think about to figure out. One last question - does PCA work for trajectory inference?

In [129]:
```
Y = mydata.s[['pca_0', 'pca_1']].as_matrix()

mydata.time_model = GPy.models.BayesianGPLVM(Y, 1, init='random')

mydata.time_model.rbf.lengthscale.constrain_fixed(2., warning=False)
mydata.time_model.rbf.variance.constrain_fixed(200., warning=False)

# priors = mydata.s.day_int+np.ones(len(mydata.s.day_int),dtype=int)
priors = mydata.s.day_int
if priors is not None:
    for i, p in enumerate(priors):
        prior = GPy.priors.Gaussian(p, 2.)
        mydata.time_model.X.mean[i, [0]].set_prior(prior, warning=False)

mydata.time_model.optimize(max_iters=2000, messages=True)
mydata.s['pca_pseudotime'] = mydata.time_model.X.mean[:, [0]]
mydata.s['scaled_pca_pseudotime'] = mydata.s.pca_pseudotime+abs(min(myda
ta.s.pca_pseudotime))
mydata.s.scaled_pca_pseudotime = mydata.s.scaled_pca_pseudotime/max(myda
ta.s.scaled_pca_pseudotime)
mydata.model_fates(t='pca_pseudotime', X=['pca_0', 'pca_1'], C=2, step_l
ength=0.01)
mydata.make_fates_viz(['pca_0', 'pca_1'])
mydata.fates_viz.plot()
plt.show()
```

```
 /software/apps/anaconda/5.2/python/3.6/lib/python3.6/site-packages/ipy
kernel/__main__.py:1: FutureWarning:Method .as_matrix will be removed i
n a future version. Use .values instead.

 /scratch/groups/abattle4/josh/gp_fates/GPfates/GPfates.py:79: FutureWa
rning:Method .as_matrix will be removed in a future version. Use .value
s instead.
```

```
iteration 1 bound=-259948.12693189978 grad=429543.8626277946, beta=0
iteration 2 bound=-134714.57391608864 grad=25651835.295126826, beta=11.
050191627254565
iteration 3 bound=-122974.948897559 grad=4192037.882654677, beta=0.0
iteration 4 bound=-119262.41276722432 grad=23706740.787014797, beta=0.0
iteration 6 bound=-122962.76769021216 grad=10297425.926616227, beta=0.0
iteration 7 bound=-121402.90400990288 grad=33993761.67698123, beta=0.0
iteration 8 bound=-109888.17665712361 grad=15287752.625584178, beta=0.0
iteration 10 bound=-110547.3222258641 grad=2305852.6839771215, beta=0.0
iteration 11 bound=-105923.2618571575 grad=8366787.760247212, beta=0.0
iteration 13 bound=-106834.04289939435 grad=298936.2862746454, beta=0.0
iteration 14 bound=-106062.64856857264 grad=3002094.8544157743, beta=0.
0
iteration 15 bound=-105564.40637594758 grad=2766977.293421834, beta=0.0
iteration 17 bound=-114989.81539179625 grad=919925.1129157592, beta=0.0
iteration 18 bound=-109795.50139730792 grad=19318731.759122655, beta=0.
0
iteration 19 bound=-105449.22889530327 grad=20547918.39813802, beta=0.0
iteration 21 bound=-106775.17984018063 grad=371053.9690084466, beta=0.0
iteration 22 bound=-105492.61027018909 grad=7493941.792698596, beta=0.0
iteration 24 bound=-118213.92305148288 grad=1620844.28691742, beta=0.0
iteration 25 bound=-113385.01972628577 grad=12923362.162064698, beta=0.
0
iteration 27 bound=-114309.5594028435 grad=6019831.957124851, beta=0.0
iteration 28 bound=-107547.15100669512 grad=23473599.34464584, beta=0.0
iteration 30 bound=-109570.58736118762 grad=10150757.055844173, beta=0.
0
iteration 31 bound=-105516.75989370463 grad=17127373.34012263, beta=0.0
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iteration 787 bound=-109886.96323555149 grad=13241378.308990184, beta=
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iteration 789 bound=-105177.5375011299 grad=8588580.630748732, beta=0.0
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iteration 799 bound=-109450.18044735231 grad=1339580.4012452557, beta=
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iteration 811 bound=-106462.44139759992 grad=14225085.792202896, beta=
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iteration 814 bound=-105406.0016478035 grad=820554.4406527588, beta=0.0
iteration 816 bound=-109513.01635083131 grad=1282636.3296963046, beta=
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iteration 835 bound=-105675.67462250064 grad=12287422.381561816, beta=
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iteration 837 bound=-111546.25134757897 grad=963221.3699796535, beta=0.
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iteration 867 bound=-111360.79850418825 grad=9179041.657945575, beta=0.
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iteration 881 bound=-111730.03620729232 grad=679225.3338791541, beta=0.
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iteration 884 bound=-105332.05841944311 grad=26799026.060098737, beta=
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0.0
iteration 958 bound=-109446.86862038847 grad=13575222.616481224, beta=
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0.0
maxiter exceeded
```
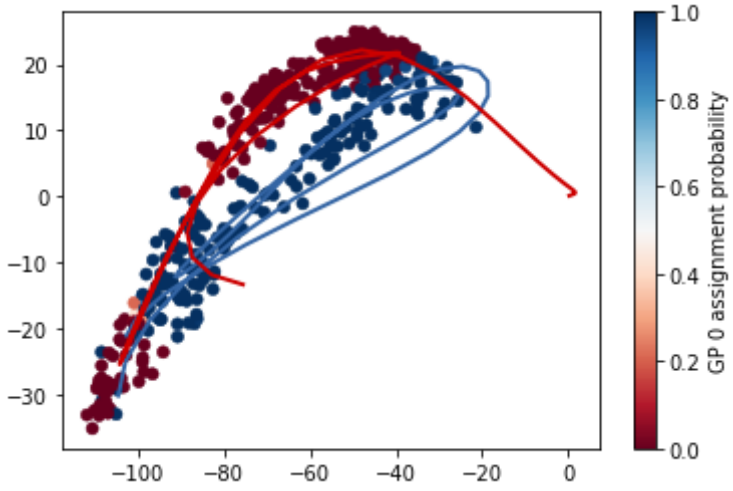

```
 /scratch/groups/abattle4/josh/gp_fates/GPfates/GPfates.py:91: FutureWa
rning:Method .as_matrix will be removed in a future version. Use .value
s instead.
```



Looks like PCA doesn't cut it here. However, since they do pseudotime inference first and bifurcation analysis later, in theory a different pseudotime metric would work okay