

La familia PriA

PriA es una familia de enzimas de Actinobacteria homóloga a la familia HisA en Enterobacteria. PriA es una familia promiscua según las definiciones de este trabajo ya que se han caracterizado experimentalmente varios de sus miembros con la capacidad de realizar las funciones tanto de HisA como de TrpF. Es decir PriA participa en las rutas de síntesis de Histidina y Triptofano, en varias Actinobacterias, como por ejemplo *Streptomyces coelicolor*. Dentro de la ruta de histidina HisA convierte ProFAR en PRFAR como parte de la ruta de la histidina. PriA además de la isomerización de PROFAR también convierte PRA en CdRP actuando análogamente a la función TrpF en la ruta del triptófano. La mayoría de las actinobacterias han perdido el gen trpF en la ruta del triptofano,

PriA ha mostrado un gradiente funcional en Actinobacteria que divide esta familia en varias subfamilias según su funcionalidad. Entre las subfamilias PriA están PriB en *Streptomyces* que muestra poca actividad trpF y un gen TrpF fuera de contexto. Otra subfamilia es subHisA en *Corynebacteria* y *Actinomycetes* que ha perdido actividad trpF, y finalmente hay subTrpF en *Actinomycetes* que ha perdido su actividad A.

Algunas de estas enzimas se seleccionaron en busca de nuevas funciones promiscuas, las reacciones catalizadas por el candidato se investigaron mediante la exploración de sustratos químicamente similares a los nativos.

EvoMining ha detectado un parólogo de PriA en Actinobacteria y una expansión conservada en un grupo de saxitoxinas en Cianobacteria [@ moustafa_origin_2009]. PriA

PriB

SubHisA

subTrpF

PriA como modelo de familia enzimática donde las expansiones no son condición necesaria para la promiscuidad.

PriA en EvoMining

Se investigaron las expansiones de la familia PriA en los linajes Actinobacteria, Cyanobacteria, Pseudomonas y Archaea. En Actinobacteria, donde se sabe que PriA es promiscua no se detectaron copias extra.

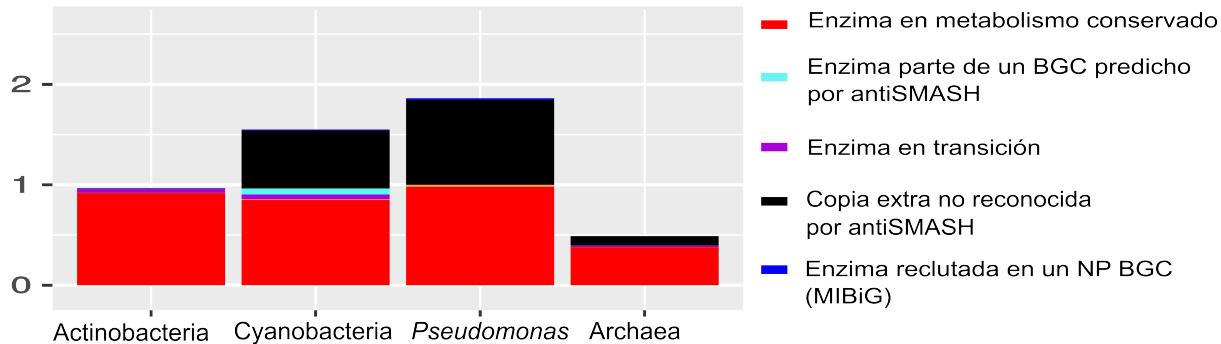


Figure 1: Expansiones de PriA en Actinobacteria, Cyanobacteria, Pseudomonas y Archaea

Los reclutamientos que tuvieron estas expansiones son los siguientes

Table 1: Reclutamientos de expansiones de PriA en MIBiG

Compuesto	Actinobacteria	Cyanobacteria	Pseudomonas	Archaea	BGC origen	Clase
saxitoxin	x	x	x	x	Cyanobacteria	Alkaloid
lipopoly saccharide	x	x	x	x	Proteobacteria	Saccharide
toxin	x	x	x	x	Cyanobacteria	Other T3
2'-chloro pentostatin	x	-	-x	-	Actinobacteria	Other
2'-amino-2'- deoxy adenosine						

Finalmente, los árboles que se produjeron por EvoMining son:

Table 2: Árboles EvoMining de PriA en MicroReact

Linaje	Link al árbol de EvoMining en Microreact
Actinobacteria	7g2IGflkv9
Cyanobacteria	qF6jWRMox
PriA_Pseudomonas	ydff6DWqs
Archaea	Ig-m9Cm6f

PriA CORASON

Todos los Streptomyces tienen el cluster de PriA parcialmente conservado respecto a *Streptomyces coelicolor*. Además *_Streptomyces_CT34* has a secondary copy whose best hit on NCBI is Lentzea's PriA with 50% identity 98% coverage

TrpF1 TrpF1 queries gave hits with TrpC enzyme present on every Streptomyces, additionally *S. rimosus*, *S. coelicolor*, *S. venezuelae* and *S. NRRL S-1813* had an extra copy. *S rimosus* TrpC vicinity has PKS and

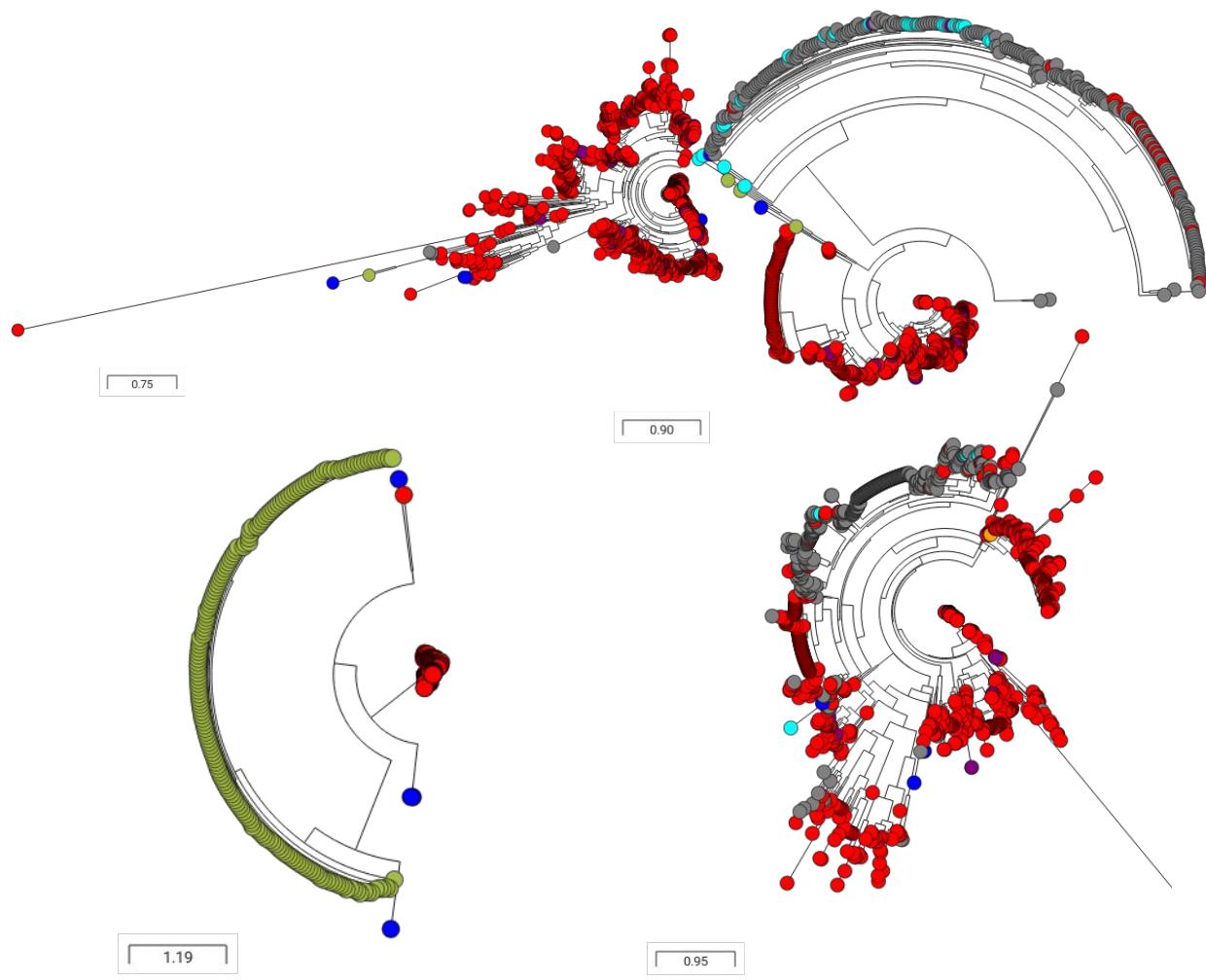


Figure 2: Árboles de destino metabólico de PriA en Actinobacteria, Cyanobacteria, Pseudomonas y Archaea según EvoMining

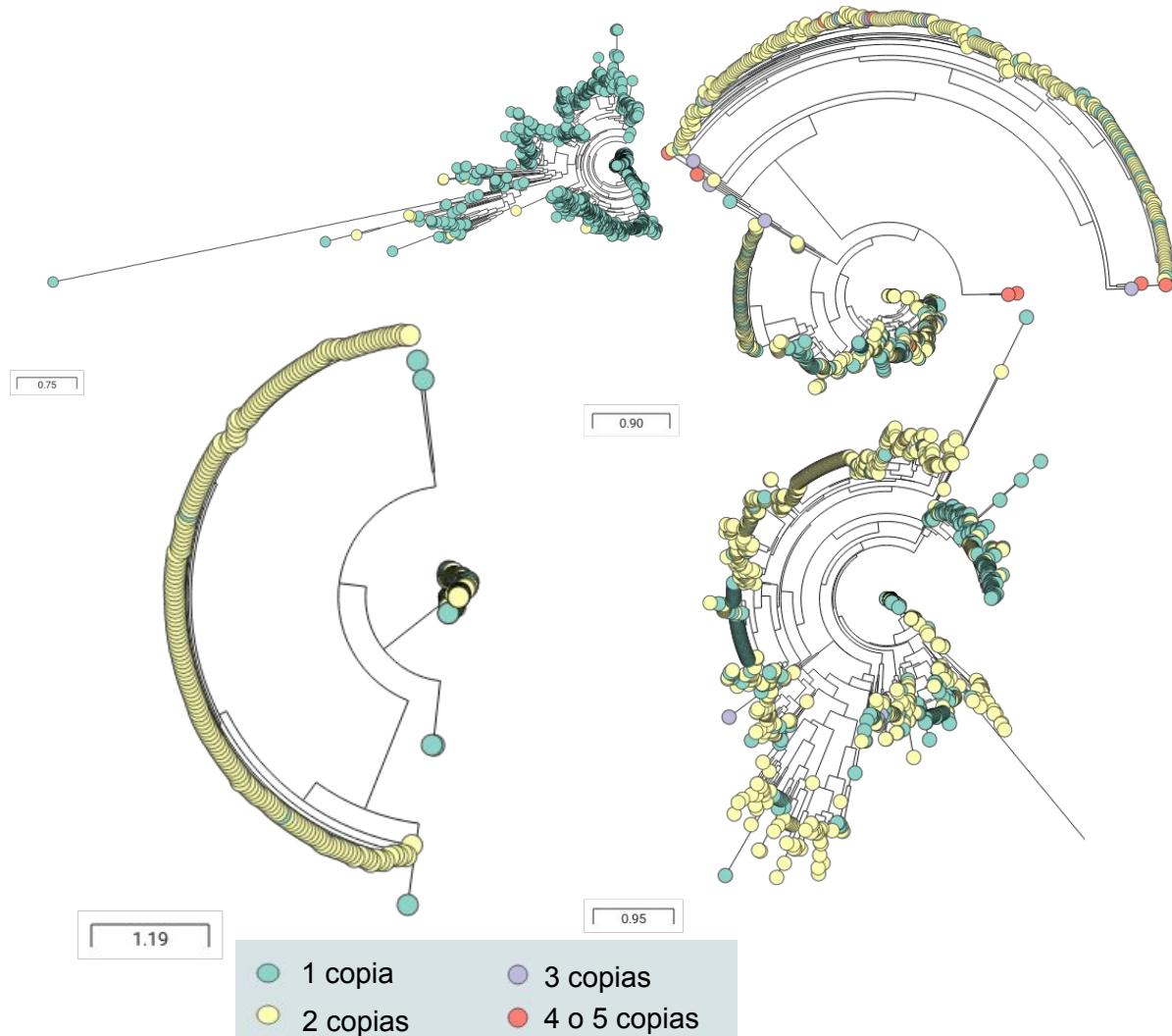


Figure 3: Copias extras de PriA en Actinobacteria, Cyanobacteria, Pseudomonas y Archaea

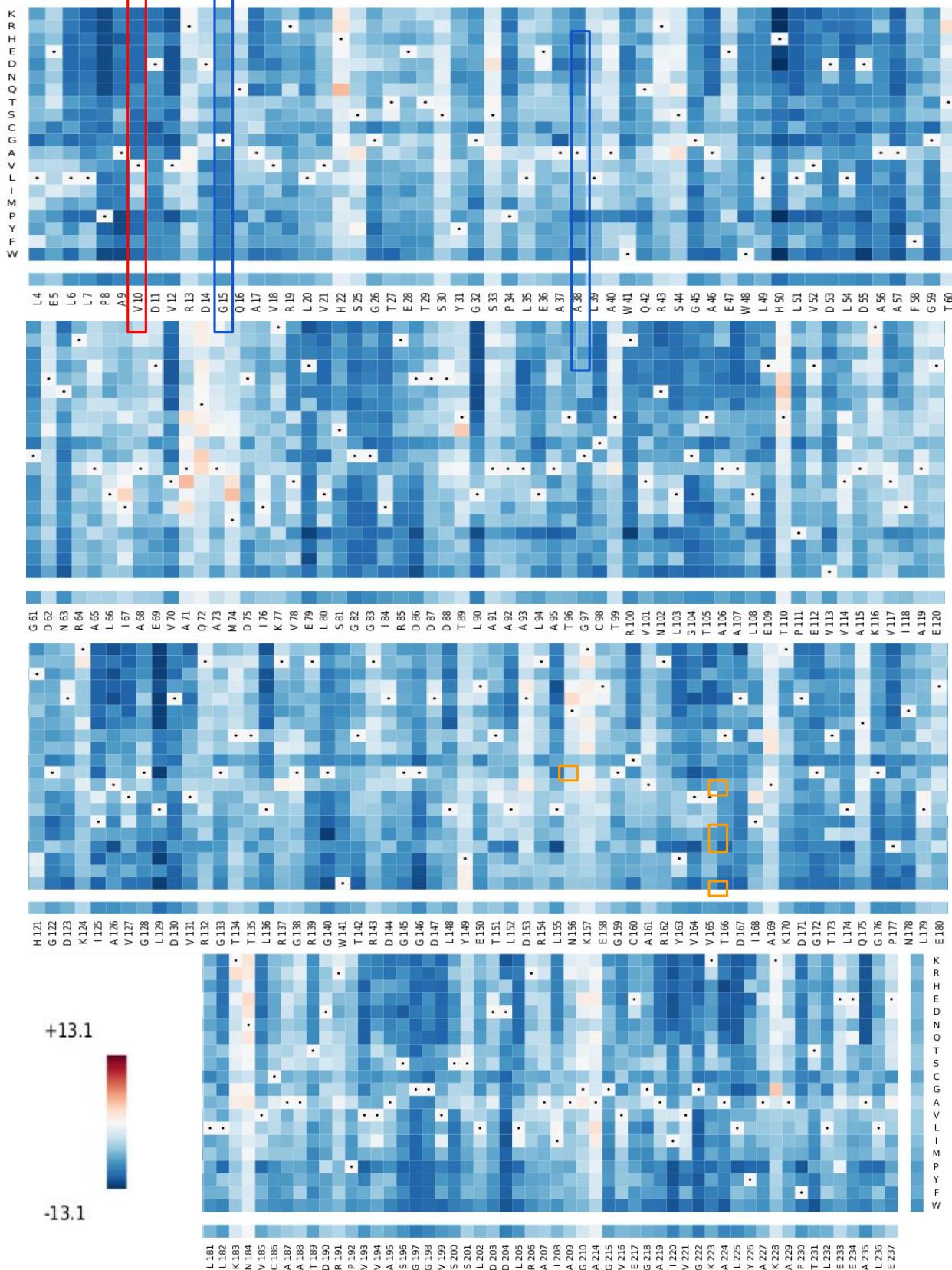


Figure 4: Tendencia de mutaciones observada en secuencias de PriA

siderophore genes.

TrpF2 Conserved cluster with NRPS sequences flanking TrpF2

TrpF3 Non conserved cluster

TrpF4 *purpeofuscus* and *S. bikiniensis*

Selected enzymes to investigate affinity of substrates similar to PRA and PRO-FAR

Dado que en *Streptomyces* se han encontrado representantes de las familias PriA y PriB, se seleccionaron 39 secuencias homólogas para realizar el análisis del sustrato de la enzima de acoplamiento. Estas secuencias seleccionadas de PriA / PriB pertenecen a *Streptomyces* uniformemente distribuidas en un árbol de especies RpoB con diferentes condiciones sobre la presencia / ausencia de TrpF. En este estudio se incluyeron otros homólogos de PriA Actinobacterial caracterizados químicamente, y finalmente se agregaron HisA de *Escherichia coli*, *Arthrobacter Aurescens*, *Salmonella enterica* y *Acidimicrobium ferrooxidans* y Actinobacterial TrpF como controles.

Cuando existían estructuras de cristal, de lo contrario, se generaban estructuras homólogas utilizando como plantilla la enzima más cercana disponible con estructura de cristal.

Controls

HisA Enterobacteria enzymes from *Salmonella enterica* (PDB:5AHE), *Escherichia coli K12 Acidimicrobium ferrooxydans* (PDB:4WD0)

TrpF Actinobacteria *Jonesia denitrificans* and *Streptomyces* sp Mg1 sequences.

Chemically characterized Actinobacterial enzymes

PriA

Mycobacterium tuberculosis (Mtub PDB:2Y88,2Y89,2Y85,3ZS4) *Streptomyces coelicolor* (Scoe PDB:2VEP,2X30,1VZW), *Streptomyces globisporus*, *Actinomyces urogenitalis* 4X2R *Corynebacterium jeikeum*

subHisA

Corynebacterium diphtheriae *Actinomyces car* (PDB:4X2R)

subTrpF

Arthrobacter aurescens (PDB:4WD0)

PriB

Streptomyces ipomoeae, *Streptomyces sviecius* (PDB:4U28,4TX9)

TrpF controls *Jonesia denitrificans* (PDB:4WUI) *Chlamydia trachomatis*, *Streptomyces* sp. Mg1 TrpF and *Actinomyces odontolyticus* were included

PriA cristal structures on PDB

```
table <- read.csv("chapter4/EstructurasPDB", row.names = 1,sep="\t")
kable(table, caption = "Enzyme PDB \\label{tab:Enzyme PDB}",caption.short = "Enzyme PDB ")
```

Table 3: Enzyme PDB

	Organismo	Family	Observations	Resolution	Year
4WUI	Jonesia denitrificans	TrpF		1.09	2014
4X9S	Streptomyces sp. MG1	PriB		1.60	2014
5DN1	Streptomyces coelicolor	PriA		1.95	2015
1DL3	Thermotoga maritima	TrpF		2.70	1999
1LBM	Thermotoga maritima	TrpF	RCDRP	2.80	2002
1NSJ	Thermotoga maritima	TrpF		2.00	1996
1V5X	Thermus thermophilus	TrpF		2.00	2003
1VZW	Streptomyces coelicolor	PriA		1.80	2004
2VEP	Streptomyces coelicolor	PriA		1.80	2007
2X30	Streptomyces coelicolor	PriA	R139N	1.95	2010
2Y85	Mycobacterium tuberculosis	PriA	RCDRP	2.40	2011
2Y88	Mycobacterium tuberculosis	PriA	D11N PRFAR	1.33	2011
2Y89	Mycobacterium tuberculosis	PriA	D11N	2.50	2011
3ZS4	Mycobacterium tuberculosis	PriA	PRFAR	1.90	2012
4AAJ	Pyrococcus furiosus	TrpF		1.75	2012
4TX9	Streptomyces sviceus	PriB	ProFAR	1.60	2014
4U28	Streptomyces sviceus	PriB		1.33	2014
4W9T	Streptomyces sp. Mg1	PriB		1.57	2014
4WD0	Arthrobacter aurescens	PriB		1.50	2014
4X2R	Actinomyces urogenitalis			1.05	2014
4AXK	Corynebacterium efficiens	SubHisA		2.25	2013
5AHE	Salmonella enterica	HisA		1.70	2015
5AB3	Salmonella enterica	HisA	D7N, D10G, dup13-15, Q24L, G102A	1.80	2016
5ABT	Salmonella enterica	HisA	D7N, G102A, V106M, D176A	1.65	2016
5AC7	Salmonella enterica	HisA	D7N, D10G, dup13-15	1.90	2016
5AC8	Salmonella enterica	HisA	D10G, dup13-15, G102A	1.70	2016
5AC6	Salmonella enterica	HisA	D7N, D10G, dup13-15, Q24L, G102A	1.99	2016
5A5W	Salmonella enterica	HisA	HisA D7N D176A with ProFAR	NA	2015
5AHF	Salmonella enterica	HisA	HisA D7N with ProFAR	NA	NA
4GJ1	Campylobacter jejuni	HisA		2.15	2012
2W79	Thermotoga maritima	HisA		1.85	2008
1QO2	Thermotoga maritima	HisA		1.85	2000
5LHE	Thermococcus kodakaraensis	TrpF		1.85	2016
5LHF	Thermococcus kodakaraensis	TrpF		1.75	2016

Similar substrates to PRA and PROFAR suggested by Tanimoto distance or previously tested

S1,S2,...S20 substrates were collected from literature and chemoinformatics predictions. S3 PRA and S7 PROFAR are native substrates, S13-S16 are light activated substrates, S17 PRAP, S18 Compound V, were found on literature, S6 GMP, S11 GTP and other were suggested by chemoinformatics.

```
table <- read.csv("chapter4/Substrate.data", row.names = 1,sep="\t")
kable(table,  caption = "Substrates \label{tab:substrates}",caption.short = "Substrates ")
```

Table 4: Substrates

id	Number	Kind	Reference	Names
S13	dte6_open	LUZ		
S15	dte13_open	LUZ		
S14	dte6_closed	LUZ		
S16	dte13_closed	LUZ		
S10	C04376	James		5'-Phosphoribosyl-N-formylglycinamide
S12	C03838	James		5'-Phosphoribosylglycinamide
S9	C04640	James		2-(Formamido)-N1-(5'-phosphoribosyl)acetamidine
S18	CompoundV	Adams	CompundV	This compound is an intermediary between GTP and H2NMP
S5	C05923	James		2,5-Diaminopyrimidine nucleoside triphosphate
S4	C05922	James		Formamidopyrimidine nucleoside triphosphate
S8	C01268	James		5-Amino-6-(5'-phosphoribosylamino)uracil
S17	PraP	Verduzco	PraP	
S7	C04302	James		PRA
S6	C00144	James		GMP
S11	C00044	James		GTP
S1	C01253	James		ADP-D-ribosyl-[dinitrogen reductase]
S2	C01201	James		N(omega)-(ADP-D-ribosyl)-L-arginine
S3	C04896			ProFAR
S19	S_17146	Due et al	17146	2,5-dimethyl-N-(4-oxocyclohexa-2,5-dienylidene)benzenesulfonam
S20	S_16827	Due et al	16827	(E)-N-(3-chloro-5-methyl-4-oxocyclohexa-2,5-dienylidene) benzen

On next figure we can see their chemical structures.

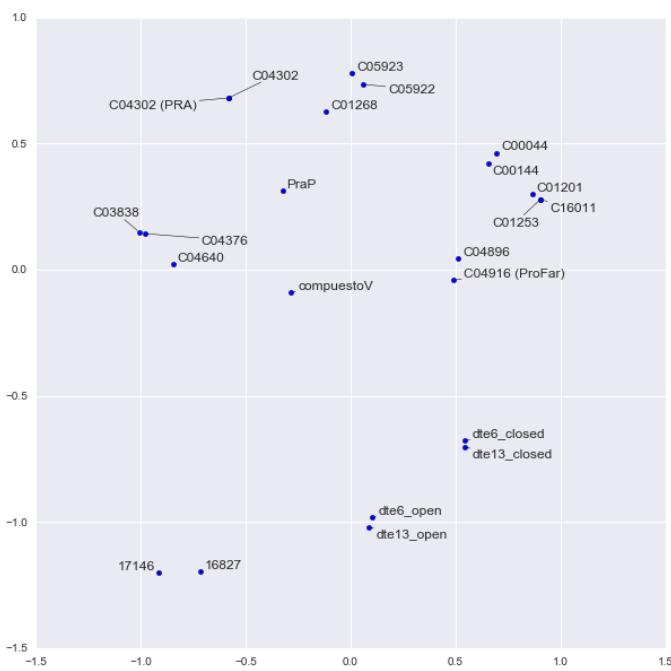
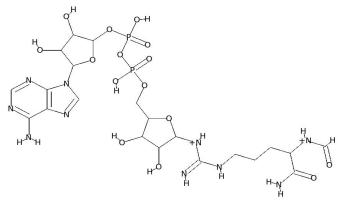
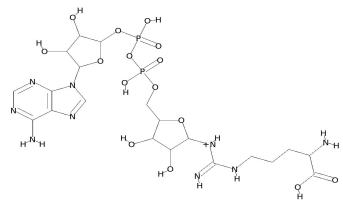


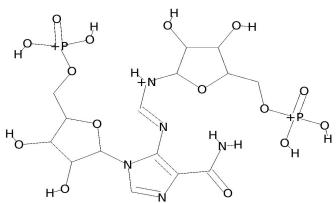
Figure 5: Substrates clustering according to Tanimoto distance



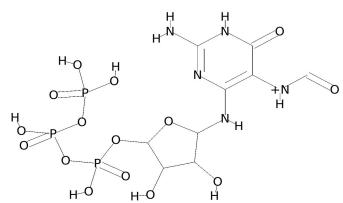
1
ADP-D-ribosyl-[dinitrogen reductase]



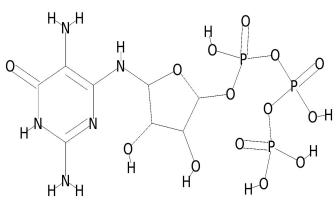
2
N(omega)-(ADP-D-ribosyl)-L-arginine



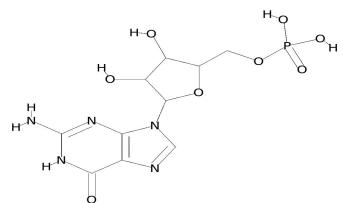
3
5-(5-Phospho-D-ribosylaminoformimino)-1-(5-phosphoribosyl)-imidazole-4-carboxamide



4
Formamidopyrimidine nucleoside triphosphate

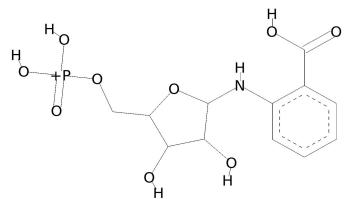


5
2,5-diaminopyrimidine nucleoside triphosphate

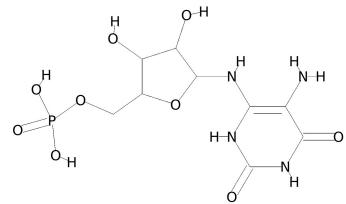


6
Guanosine monophosphate

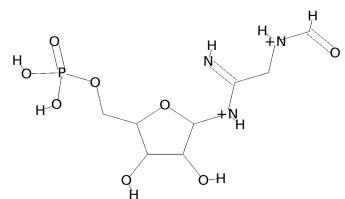
Figure 6: Substrates 1



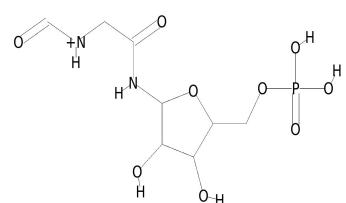
7
N-(5-phospho-D-ribosyl) anthranilate



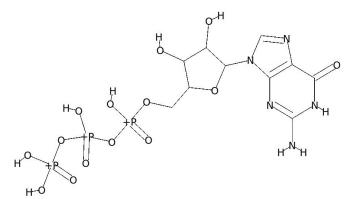
8
5-Amino-6-(5'-phosphoribosylamino)uracil



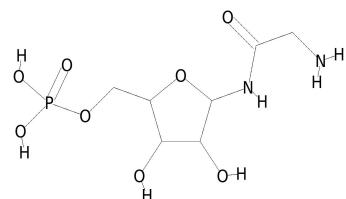
9
2-(Formido)-N1-(5'-phosphoribosyl)acetamidine



10
5'-Phosphoribosyl-N-formylglycinamide

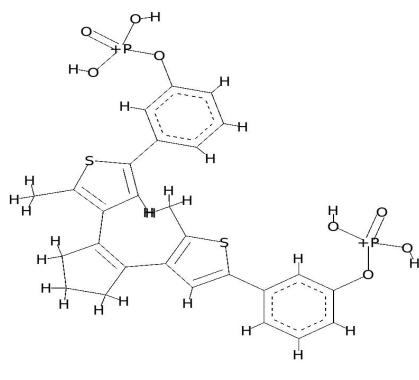


11
Guanosine 5'-triphosphate



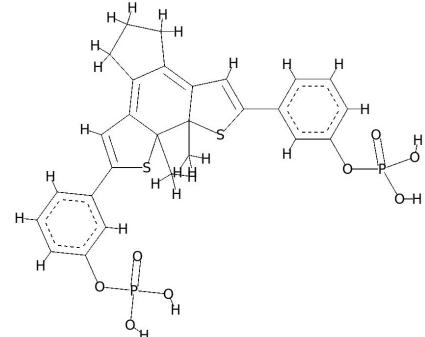
12
5'-Phosphoribosylglycinamide

Figure 7: Substrates 2



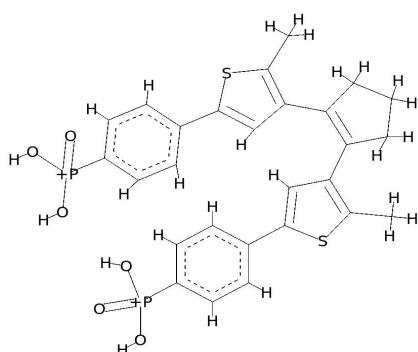
13

DTE-meta-phosphate(dte6_Open form)



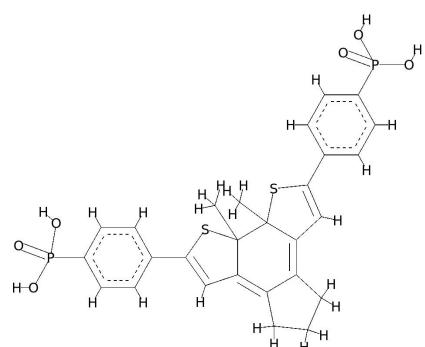
14

DTE-meta-phosphate(dte6_Closed form)



15

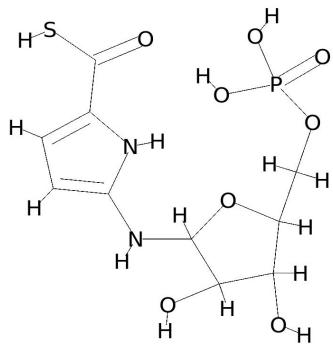
DTE-Para-Phosphonate(dte13_closed form)



16

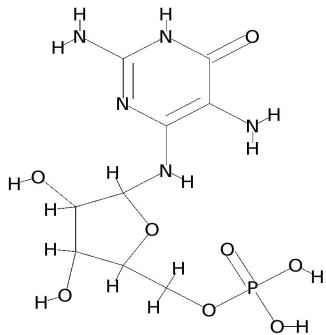
DTE-para-phosphonate(dte13_closed form)

Figure 8: Substrates 3



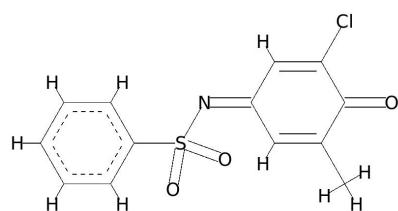
17

4N'-(5'-phosphoribosyl) 4-aminopyrrole-2-carboxilate



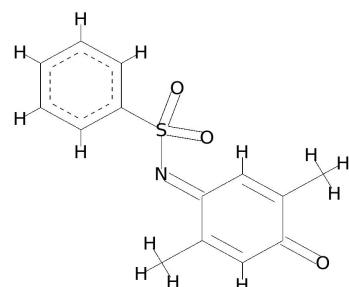
18

2,5-di-amino-6-ribosylamino-4(3H)-pyrimidinone 5'-phosphate



19

(E)-N-(3-chloro-5-methyl-4-oxocyclohexa-2,5-dienylidene)benzenesulfonamide



20

2,5 dimethyl-N-(4-oxocyclohexa-2,5-dienylidene)benzenesulfonamide

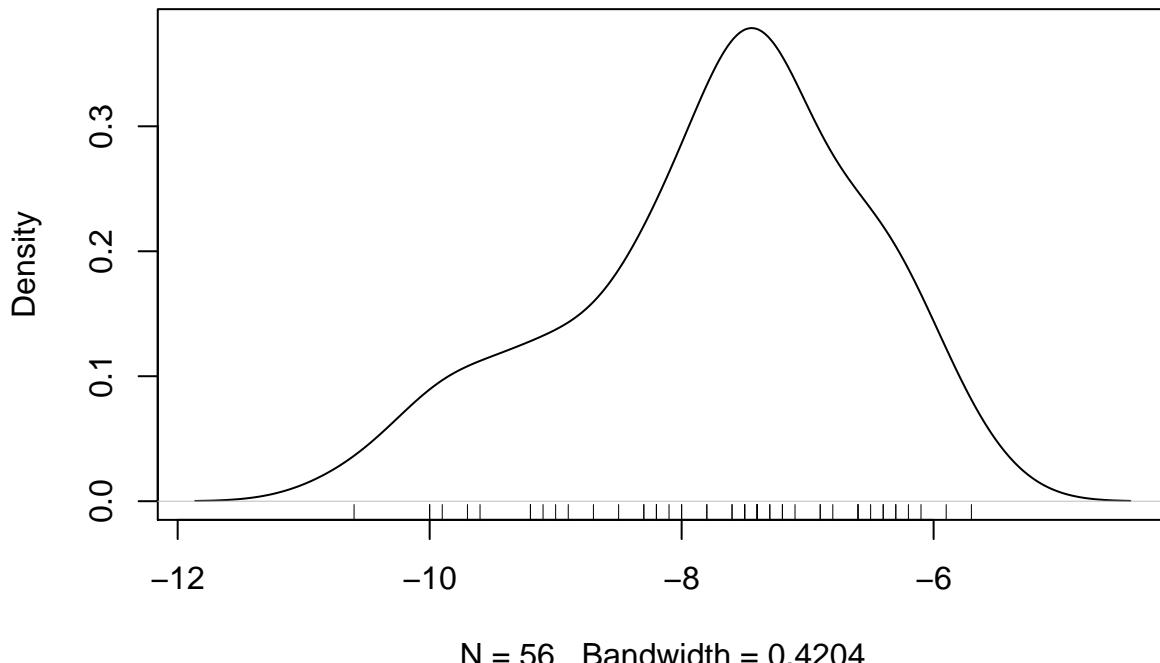
Figure 9: Substrates 4

Docking between PriA enzymes and selected substrates

Docking simulation were calculated for PriA *Streptomyces* enzymes. TrpF enzymes from *Streptomyces Mg1*, *Jonesia denitrificans*, were added as controls

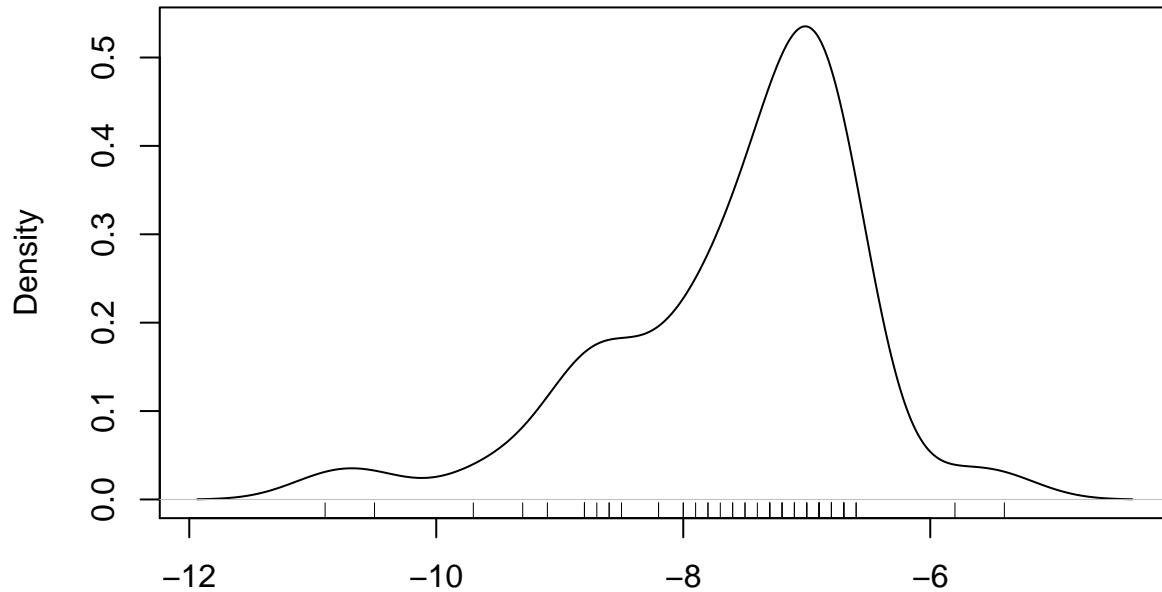
Procedures can be found at Docking Protocols

```
docking <- read.csv("chapter4/SmallHeat.data", header=TRUE, sep="\t")  
  
for (i in 2:21)  
{plot(density(docking[,i],na.rm=T)); rug(docking[,i]); browser()}  
  
density.default(x = docking[, i], na.rm = T)
```



```
## Called from: eval(expr, envir, enclos)
## debug en <text>#4: plot(density(docking[, i], na.rm = T))
```

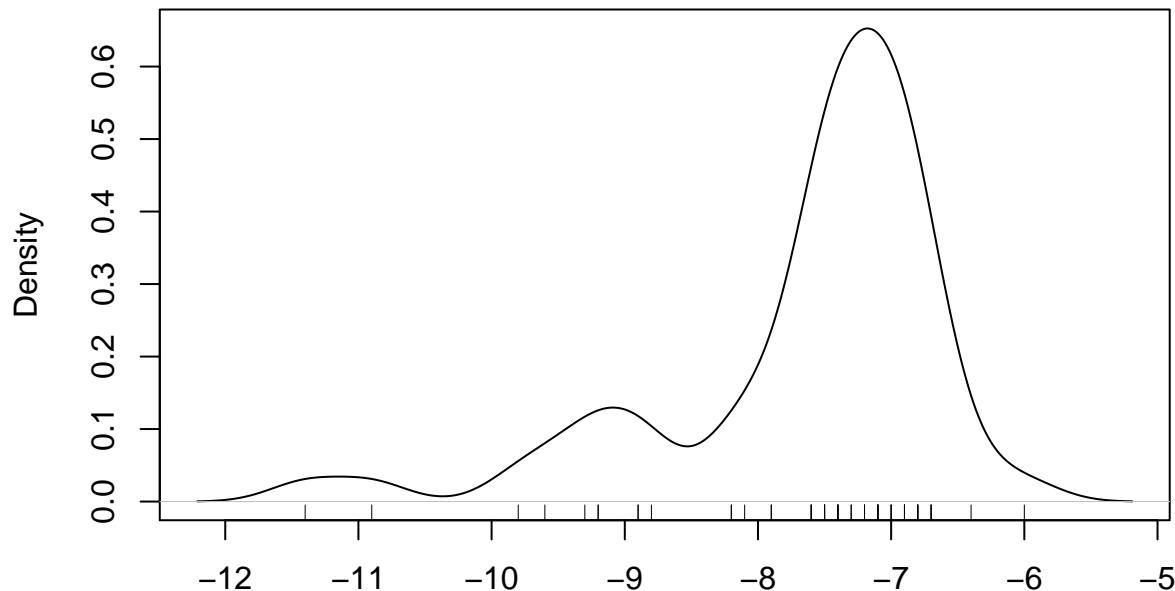
```
density.default(x = docking[, i], na.rm = T)
```



N = 56 Bandwidth = 0.3453

```
## debug en <text>#4: rug(docking[, i])
## debug en <text>#4: browser()
## debug en <text>#4: plot(density(docking[, i], na.rm = T))
```

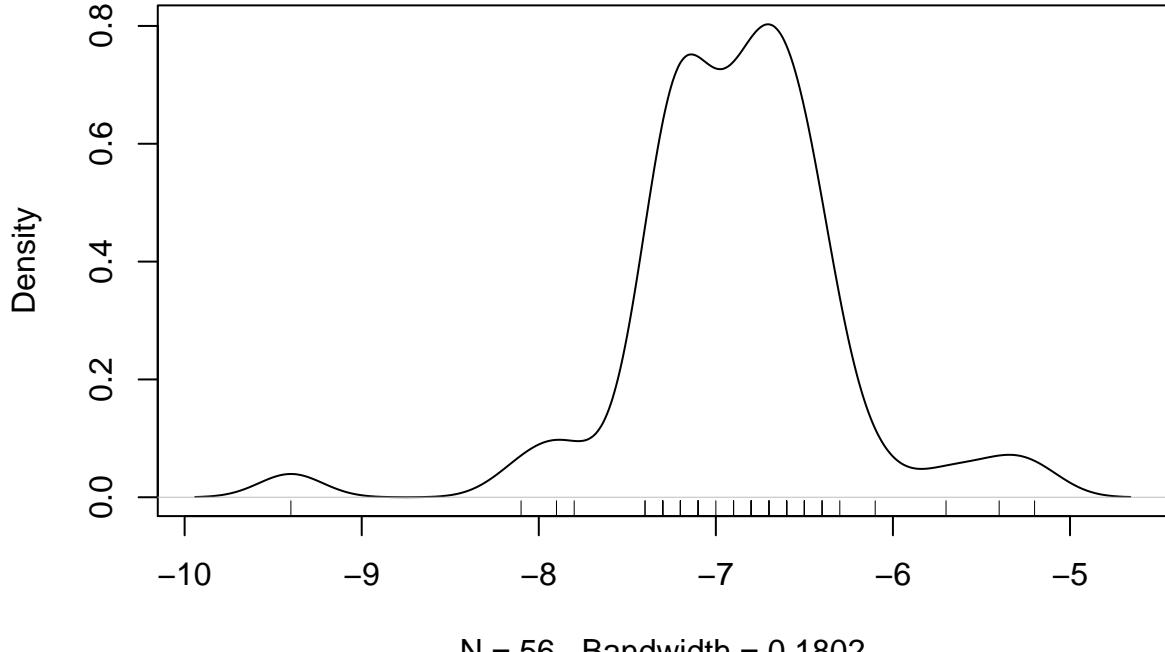
```
density.default(x = docking[, i], na.rm = T)
```



N = 56 Bandwidth = 0.2702

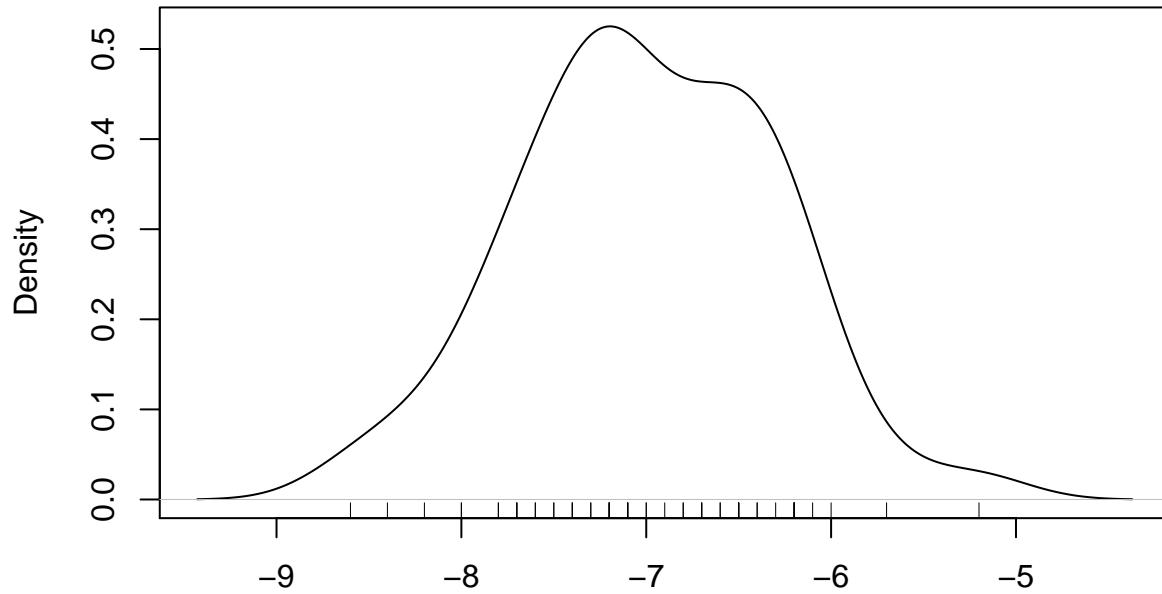
```
## debug en <text>#4: rug(docking[, i])
```

```
## debug en <text>#4: browser()
## debug en <text>#4: plot(density(docking[, i], na.rm = T))
density.default(x = docking[, i], na.rm = T)
```



```
## debug en <text>#4: rug(docking[, i])
## debug en <text>#4: browser()
## debug en <text>#4: plot(density(docking[, i], na.rm = T))
```

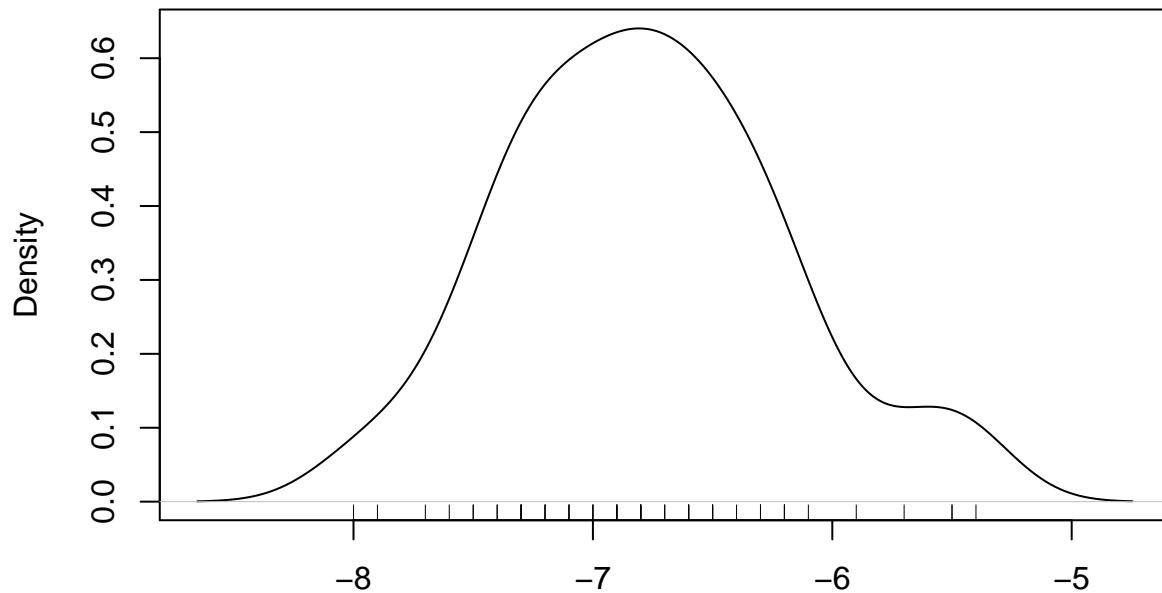
```
density.default(x = docking[, i], na.rm = T)
```



N = 56 Bandwidth = 0.2764

```
## debug en <text>#4: rug(docking[, i])
## debug en <text>#4: browser()
## debug en <text>#4: plot(density(docking[, i], na.rm = T))
```

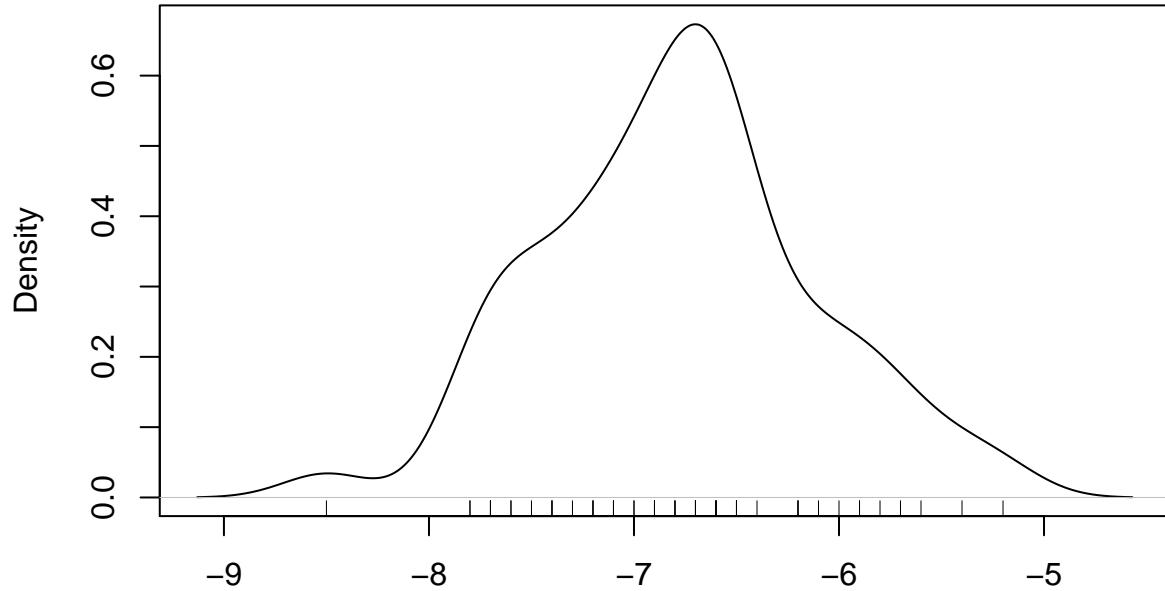
```
density.default(x = docking[, i], na.rm = T)
```



N = 56 Bandwidth = 0.2177

```
## debug en <text>#4: rug(docking[, i])
```

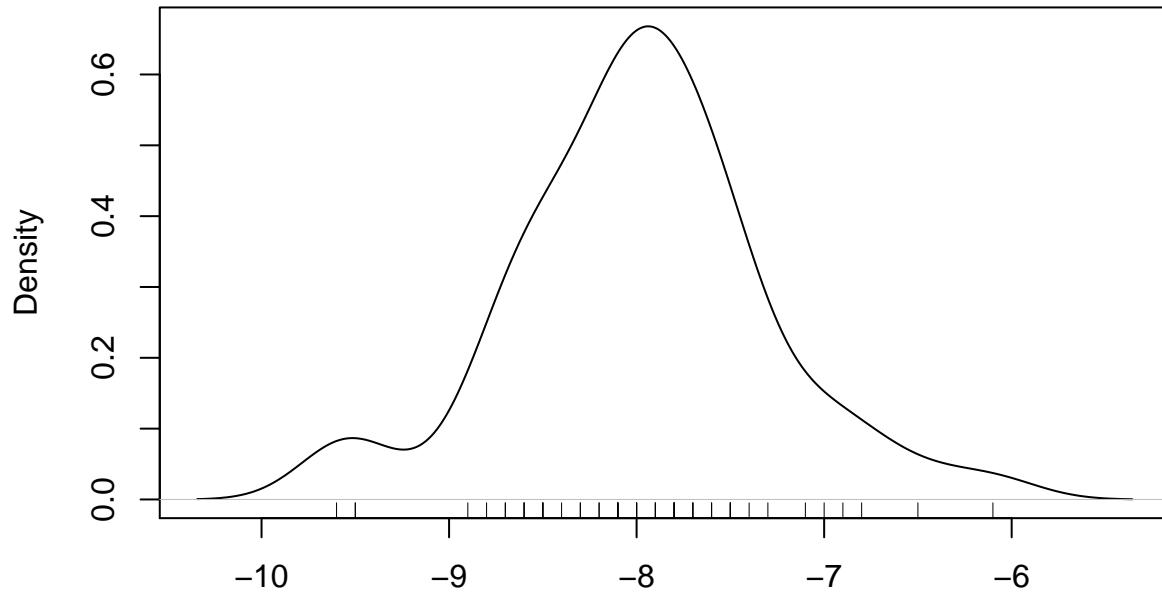
```
## debug en <text>#4: browser()
## debug en <text>#4: plot(density(docking[, i], na.rm = T))
density.default(x = docking[, i], na.rm = T)
```



N = 56 Bandwidth = 0.2102

```
## debug en <text>#4: rug(docking[, i])
## debug en <text>#4: browser()
## debug en <text>#4: plot(density(docking[, i], na.rm = T))
```

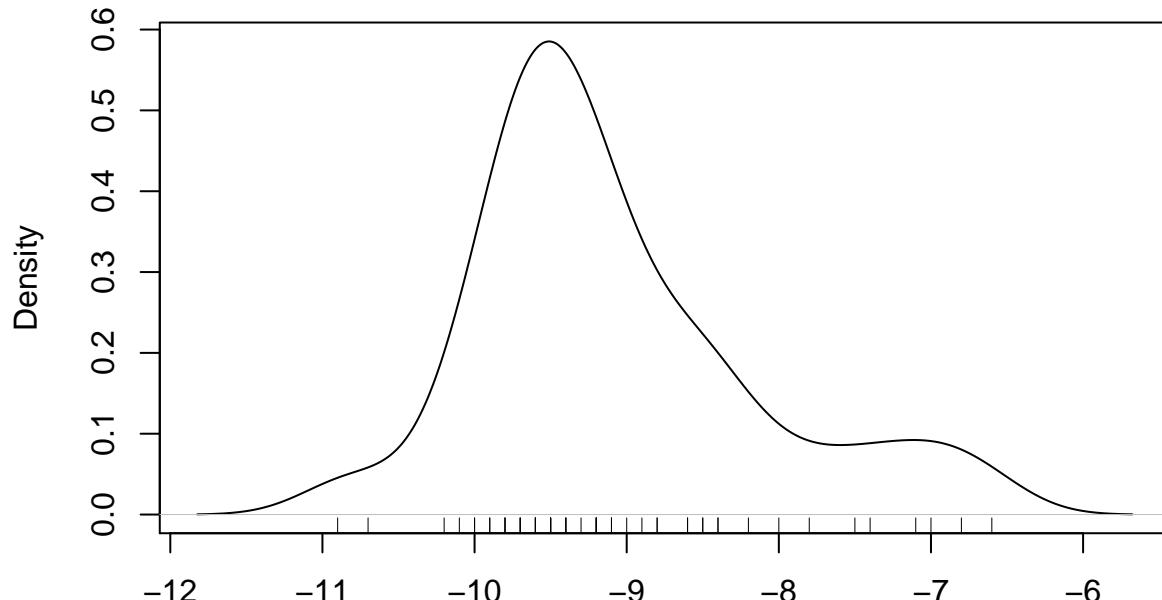
```
density.default(x = docking[, i], na.rm = T)
```



N = 56 Bandwidth = 0.2477

```
## debug en <text>#4: rug(docking[, i])
## debug en <text>#4: browser()
## debug en <text>#4: plot(density(docking[, i], na.rm = T))
```

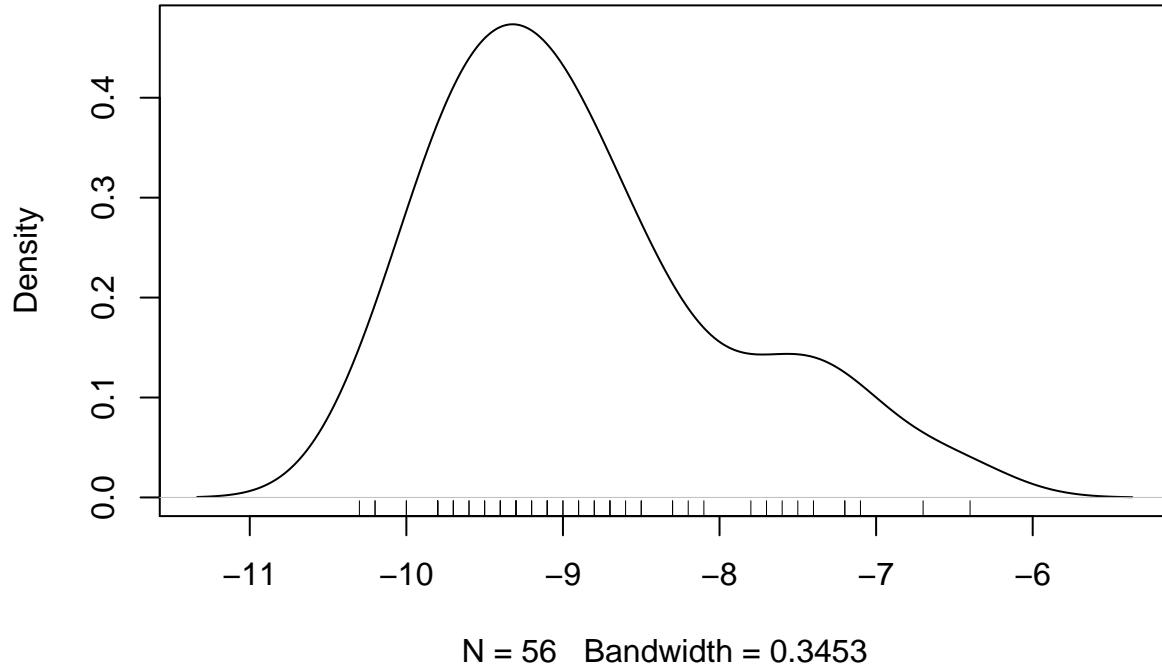
```
density.default(x = docking[, i], na.rm = T)
```



N = 56 Bandwidth = 0.3078

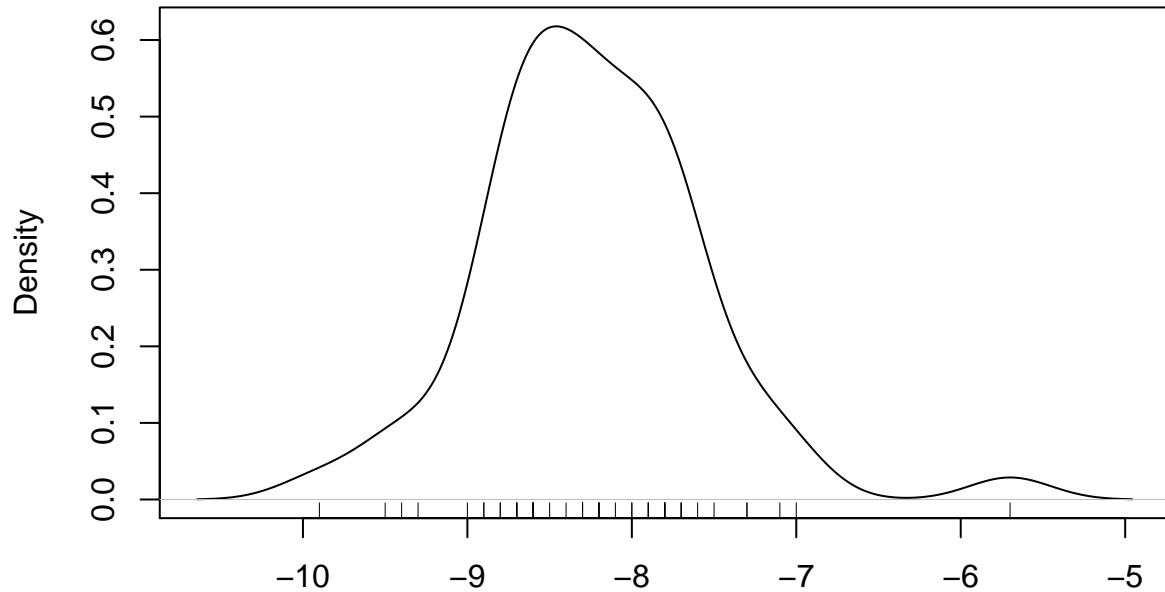
```
## debug en <text>#4: rug(docking[, i])
```

```
## debug en <text>#4: browser()
## debug en <text>#4: plot(density(docking[, i], na.rm = T))
density.default(x = docking[, i], na.rm = T)
```



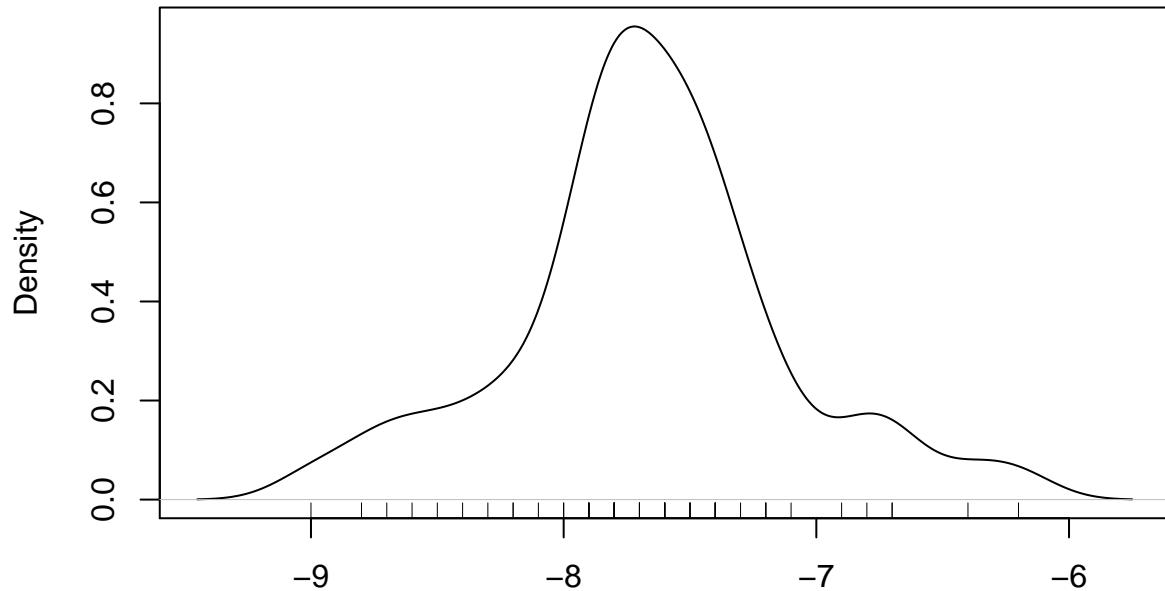
```
## debug en <text>#4: rug(docking[, i])
## debug en <text>#4: browser()
## debug en <text>#4: plot(density(docking[, i], na.rm = T))
```

```
density.default(x = docking[, i], na.rm = T)
```



N = 56 Bandwidth = 0.2477

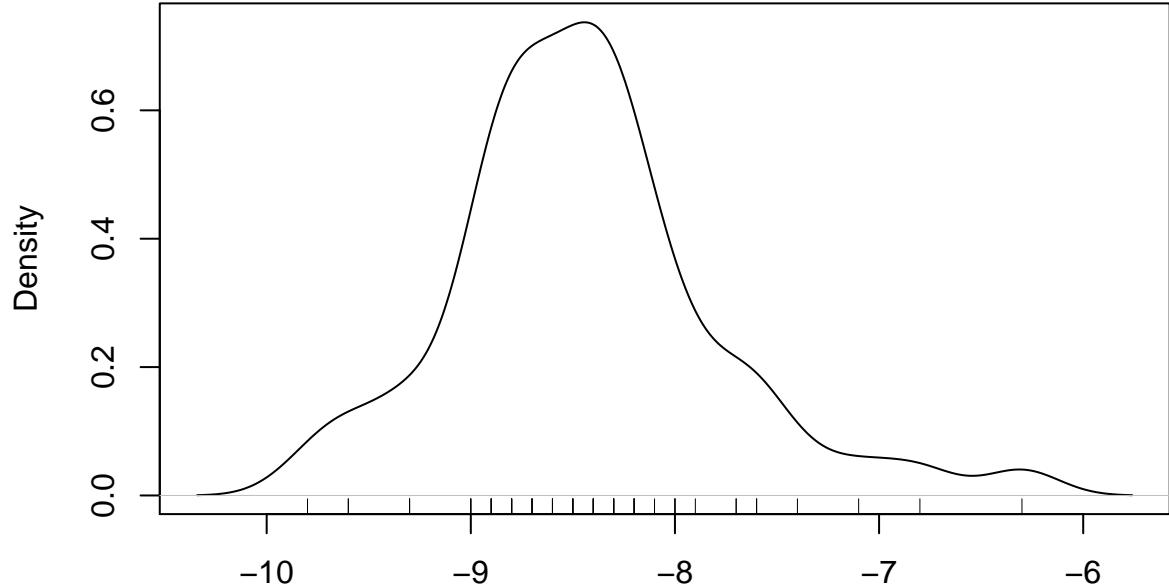
```
## debug en <text>#4: rug(docking[, i])
## debug en <text>#4: browser()
## debug en <text>#4: plot(density(docking[, i], na.rm = T))
density.default(x = docking[, i], na.rm = T)
```



N = 56 Bandwidth = 0.1501

```
## debug en <text>#4: rug(docking[, i])
```

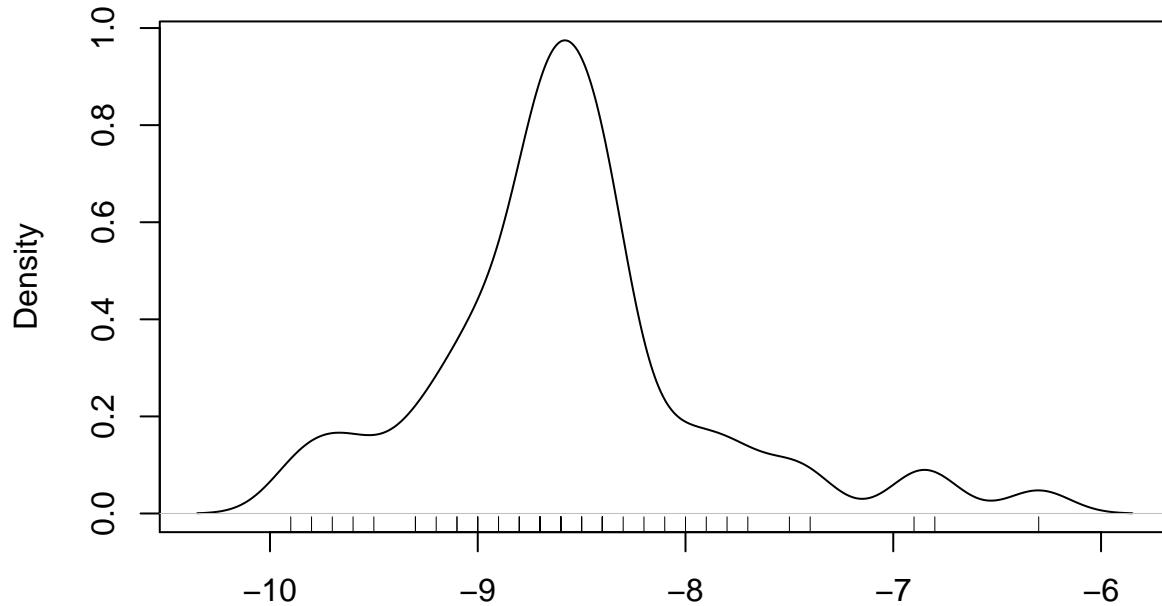
```
## debug en <text>#4: browser()
## debug en <text>#4: plot(density(docking[, i], na.rm = T))
density.default(x = docking[, i], na.rm = T)
```



N = 56 Bandwidth = 0.1802

```
## debug en <text>#4: rug(docking[, i])
## debug en <text>#4: browser()
## debug en <text>#4: plot(density(docking[, i], na.rm = T))
```

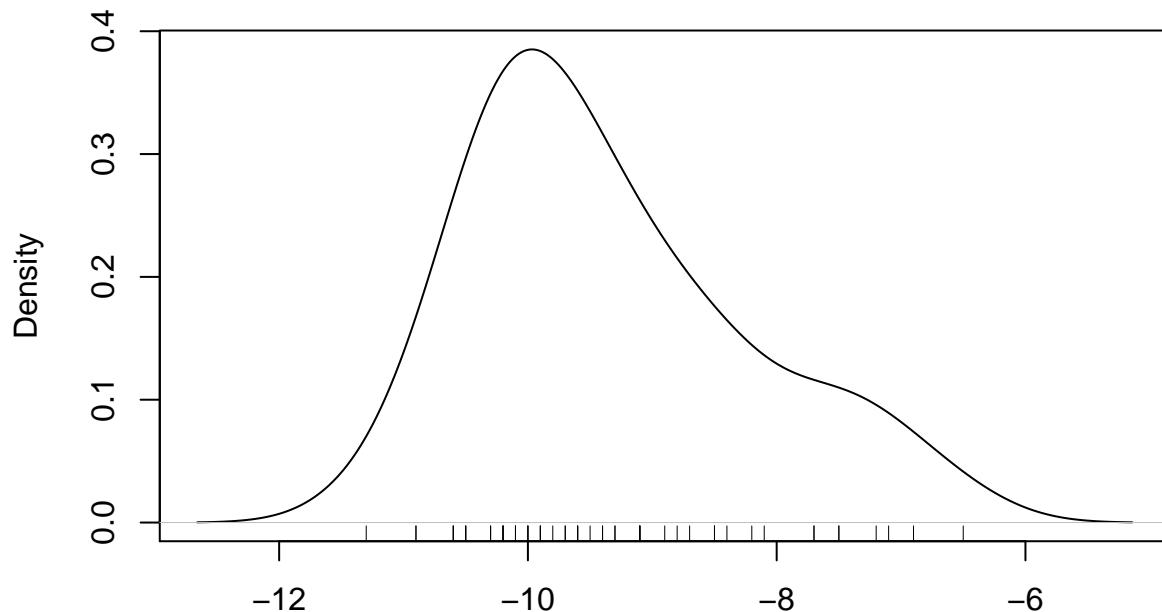
```
density.default(x = docking[, i], na.rm = T)
```



$N = 56$ Bandwidth = 0.1501

```
## debug en <text>#4: rug(docking[, i])
## debug en <text>#4: browser()
## debug en <text>#4: plot(density(docking[, i], na.rm = T))
```

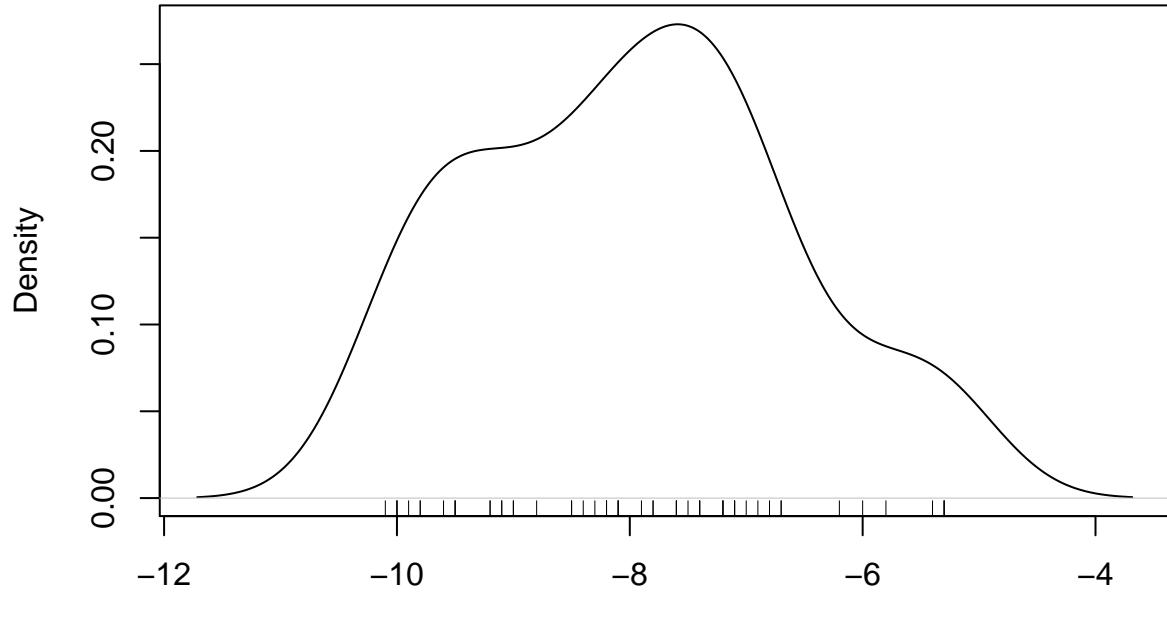
```
density.default(x = docking[, i], na.rm = T)
```



$N = 56$ Bandwidth = 0.453

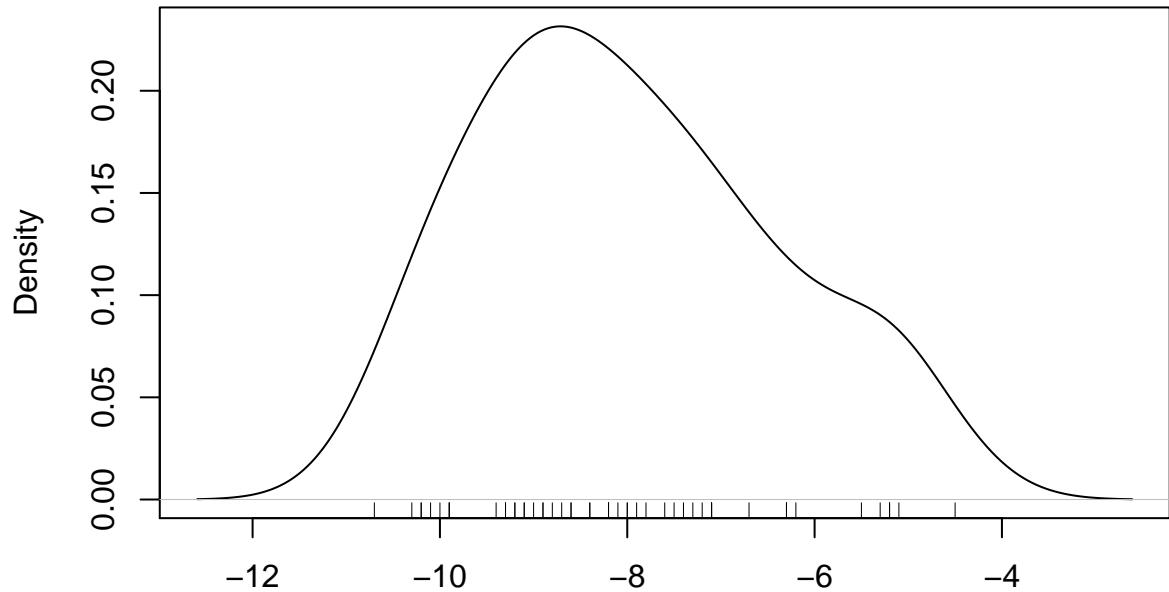
```
## debug en <text>#4: rug(docking[, i])
```

```
## debug en <text>#4: browser()
## debug en <text>#4: plot(density(docking[, i], na.rm = T))
density.default(x = docking[, i], na.rm = T)
```



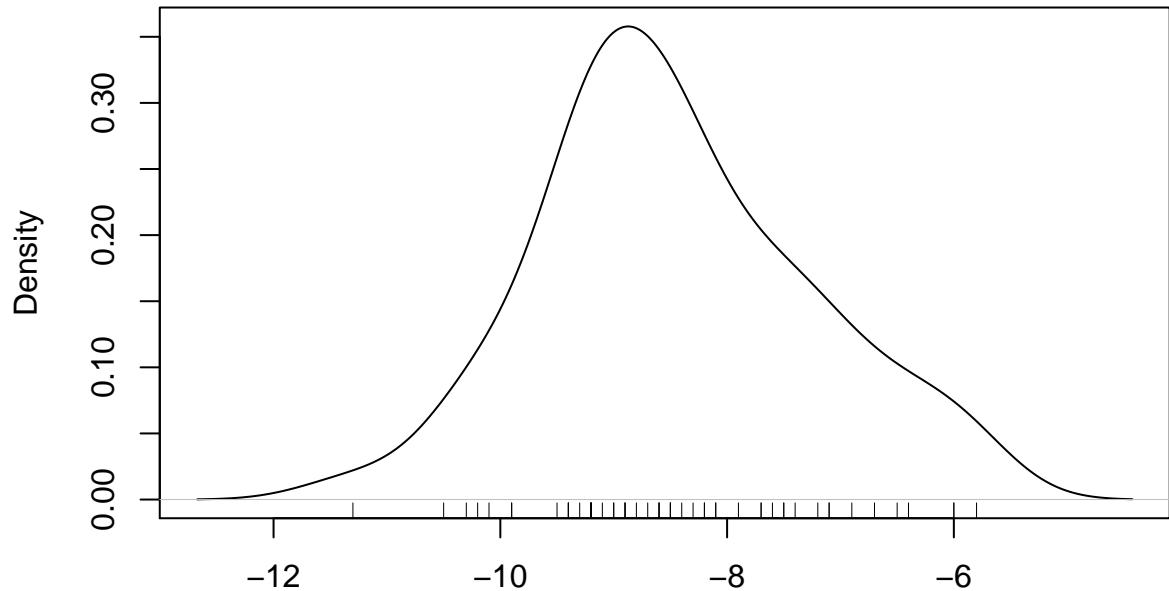
```
## debug en <text>#4: rug(docking[, i])
## debug en <text>#4: browser()
## debug en <text>#4: plot(density(docking[, i], na.rm = T))
```

```
density.default(x = docking[, i], na.rm = T)
```



N = 56 Bandwidth = 0.6305

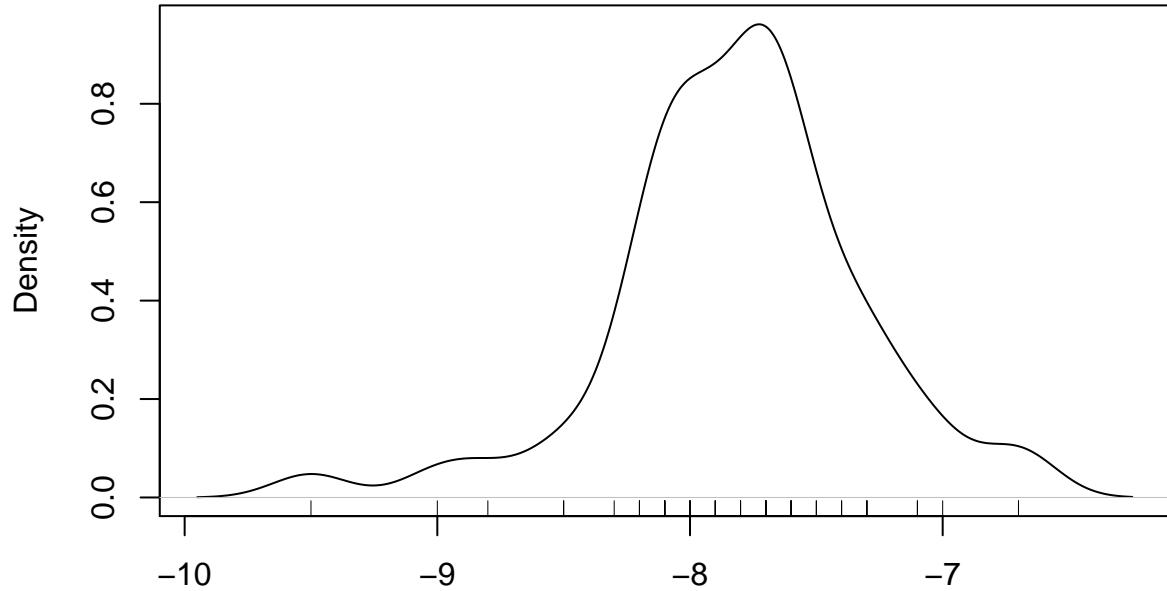
```
## debug en <text>#4: rug(docking[, i])
## debug en <text>#4: browser()
## debug en <text>#4: plot(density(docking[, i], na.rm = T))
density.default(x = docking[, i], na.rm = T)
```



N = 56 Bandwidth = 0.4579

```
## debug en <text>#4: rug(docking[, i])
```

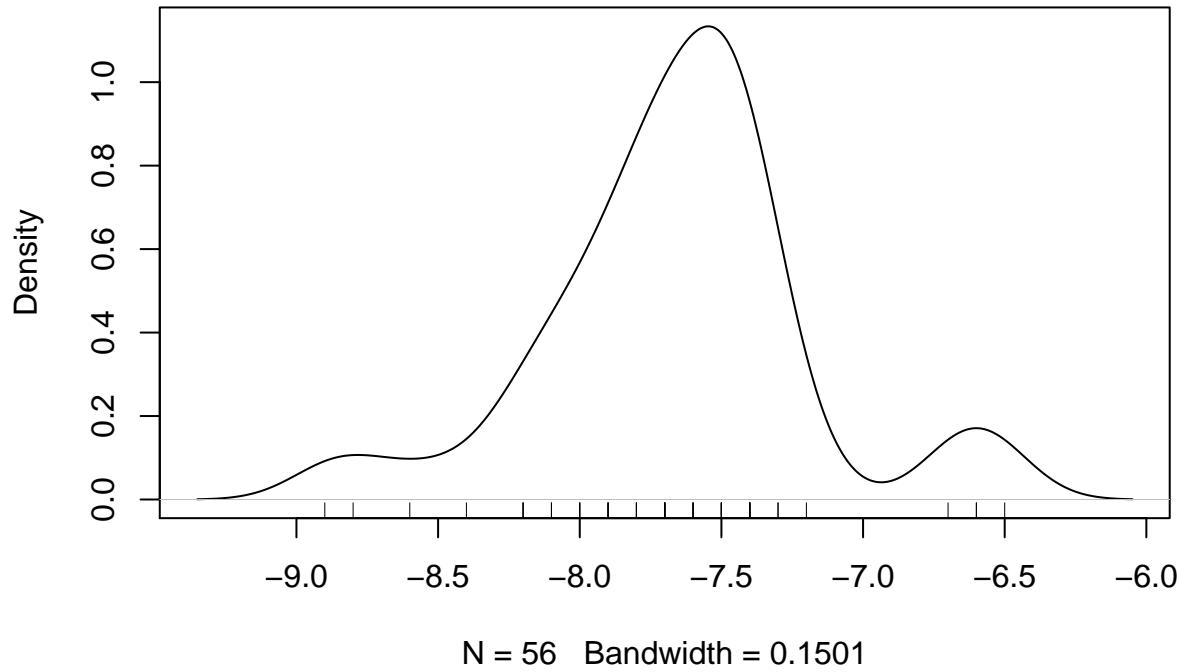
```
## debug en <text>#4: browser()
## debug en <text>#4: plot(density(docking[, i], na.rm = T))
density.default(x = docking[, i], na.rm = T)
```



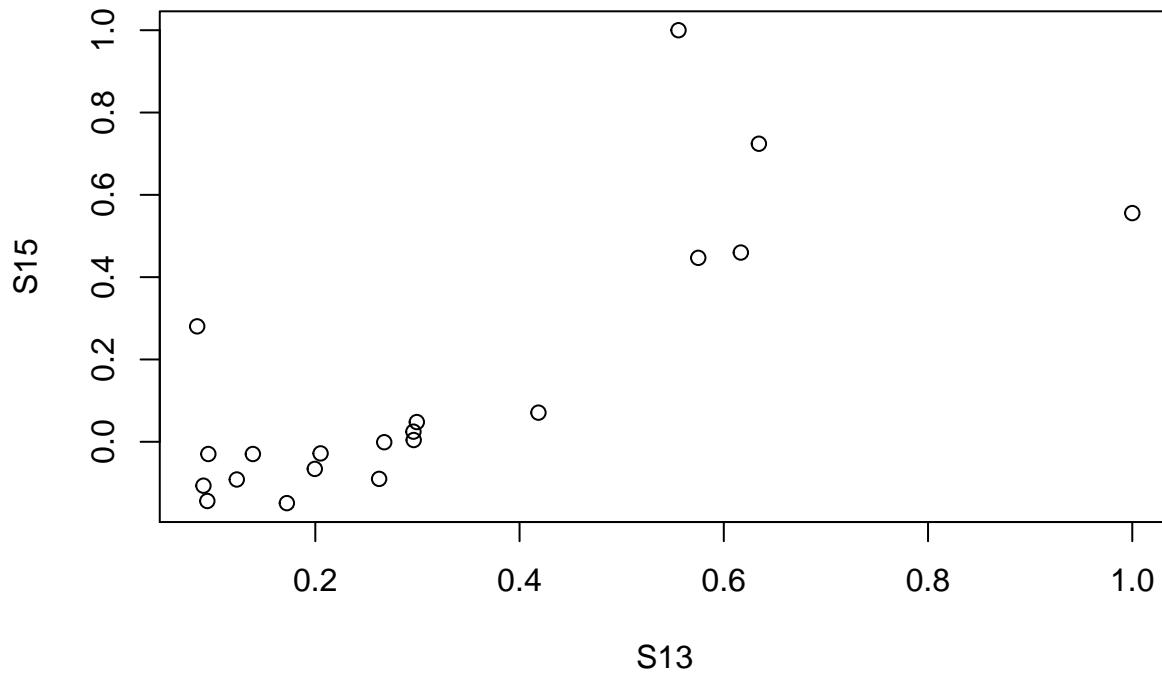
N = 56 Bandwidth = 0.1501

```
## debug en <text>#4: rug(docking[, i])
## debug en <text>#4: browser()
## debug en <text>#4: plot(density(docking[, i], na.rm = T))
```

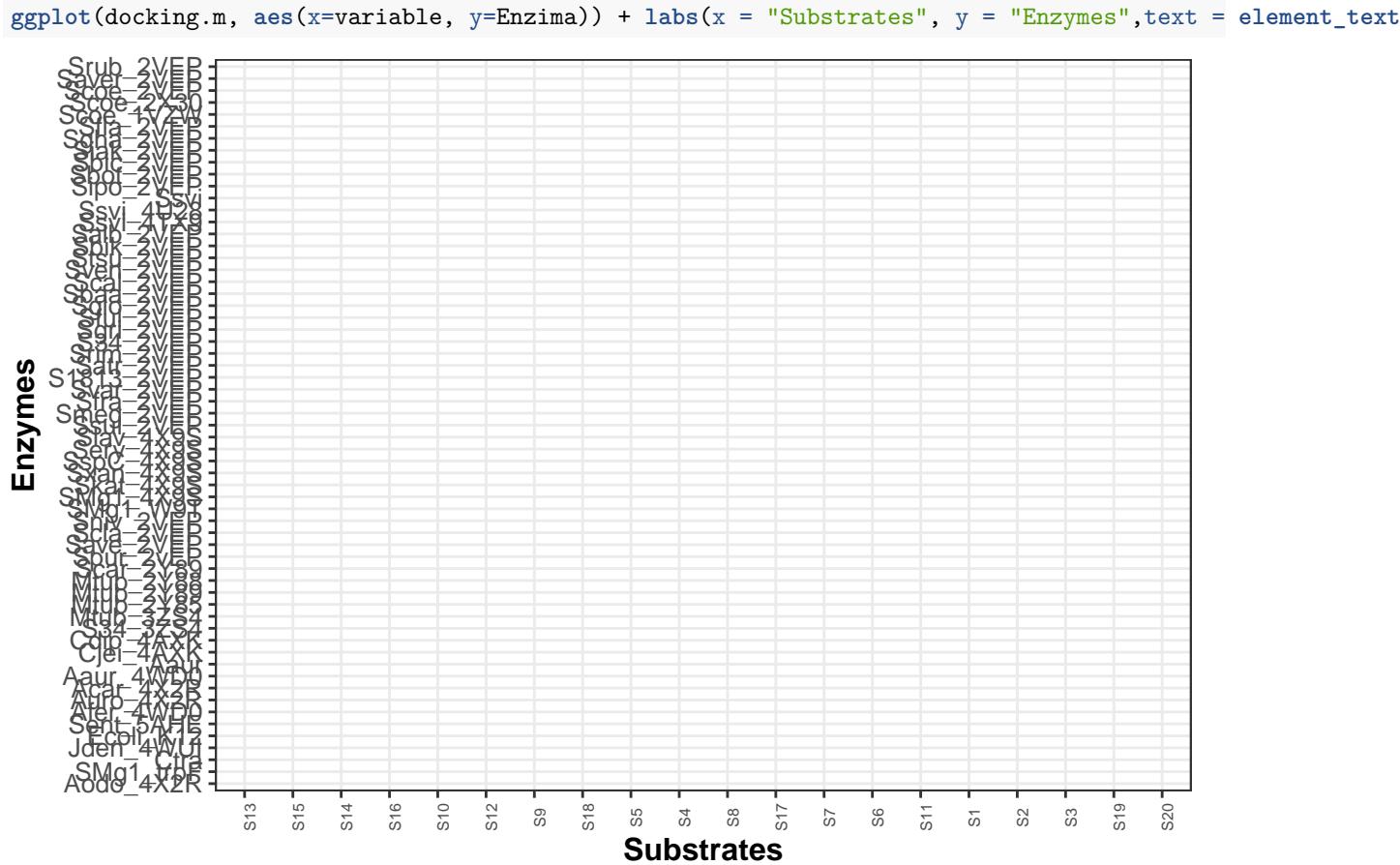
```
density.default(x = docking[, i], na.rm = T)
```



```
## debug en <text>#4: rug(docking[, i])
## debug en <text>#4: browser()
plot(cor(docking[-c(5,12,13,38,51), -1]))
```



```
## Leer sobre la incertidumbre del 2 y explicarla
## Y leer el paper de Julian y el de mauricio sobre reportes de docking
```



GTP is the substrate with best PriA affinity

```
## boxplot de los sustratos
ggplot(docking.m, aes(x=variable, y=value)) + labs(x = "Substrates", y = "Affinity",text = element_text
## Warning: Removed 100 rows containing non-finite values (stat_boxplot).
```

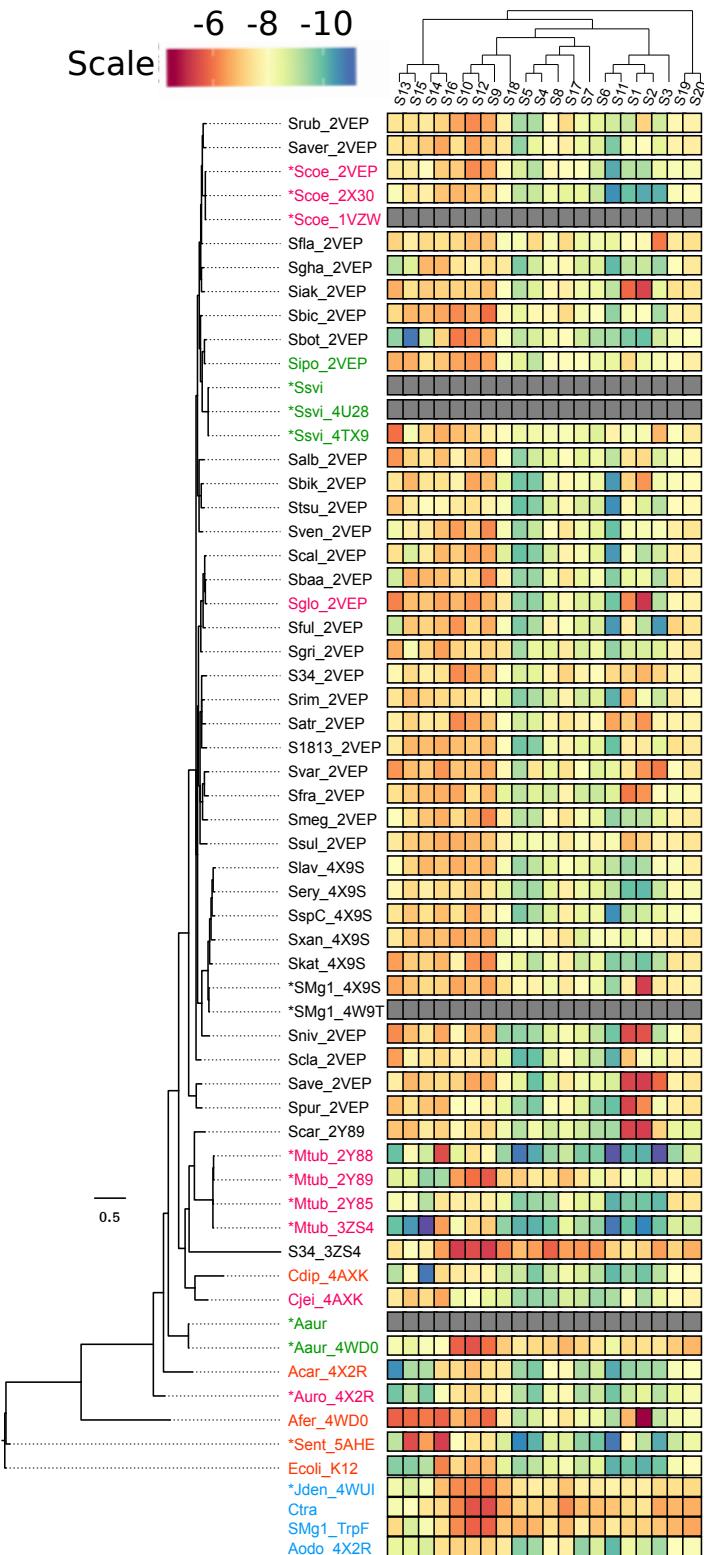


Figure 10: Heatplot docking PriA enzyme Family vs substrates
30

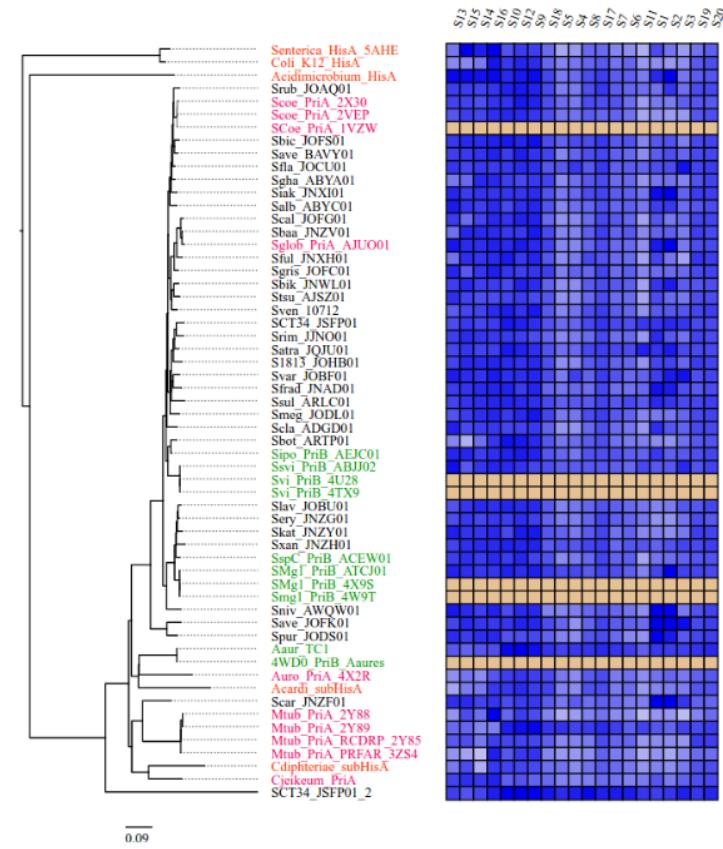
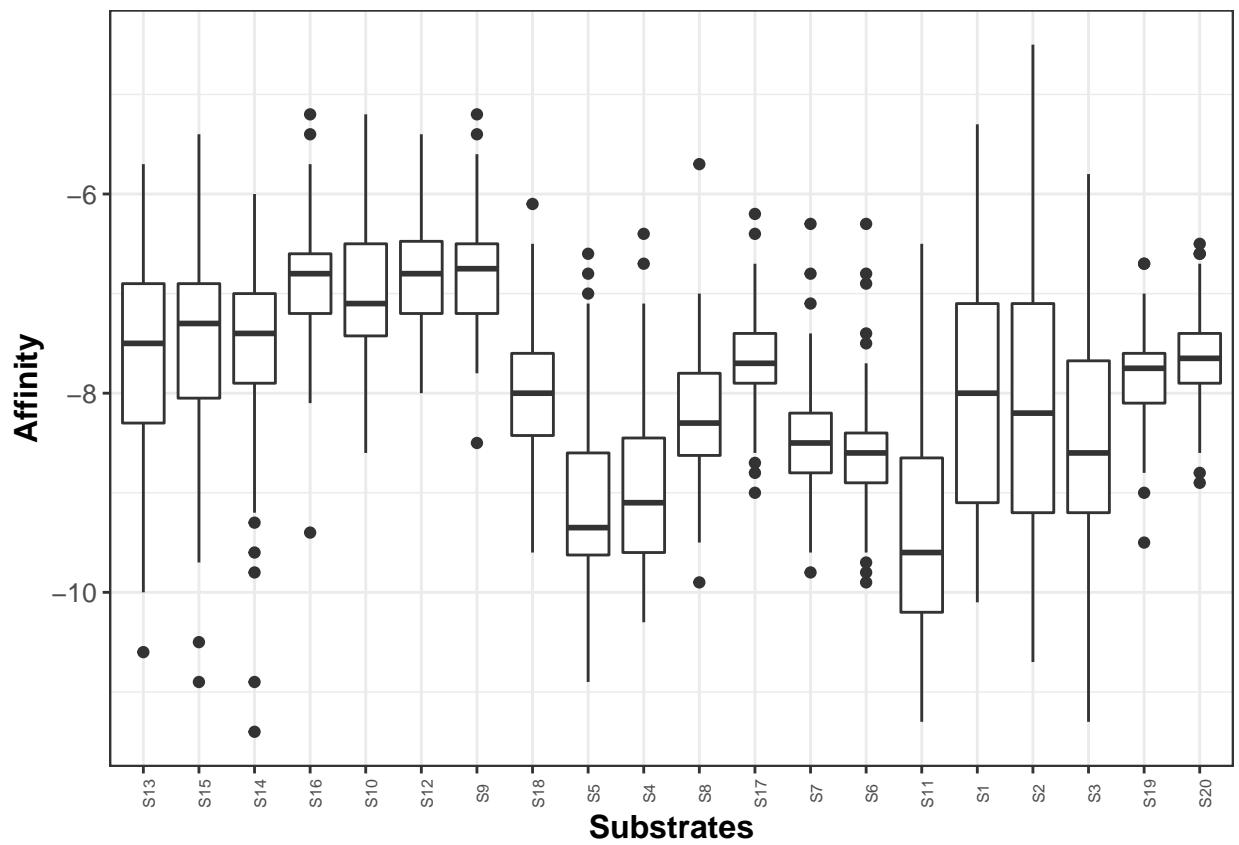


Figure 11: Heat Plot PriA Streptomyces vs other substrates



PriA Activity on GTP

Activity was measured fluorometrically in 96-well plates (Nuc 96-Well Optical Botto Plates) in a TECAN infinite M1000 plate reader (excitation at 286 nm and emission at 386 nm)

Preliminary activity assays were performed on an active PriA from *Streptomyces coelicolor* and an inactive mutant D11A.

Enzymes were cloned on coli V68 strains, overexpression were induced and protein were purified.

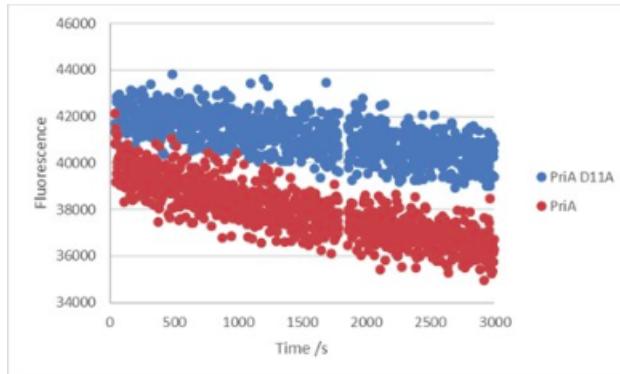


Figure 12: Scoe and non functional Scoe PriA acting on dGTP

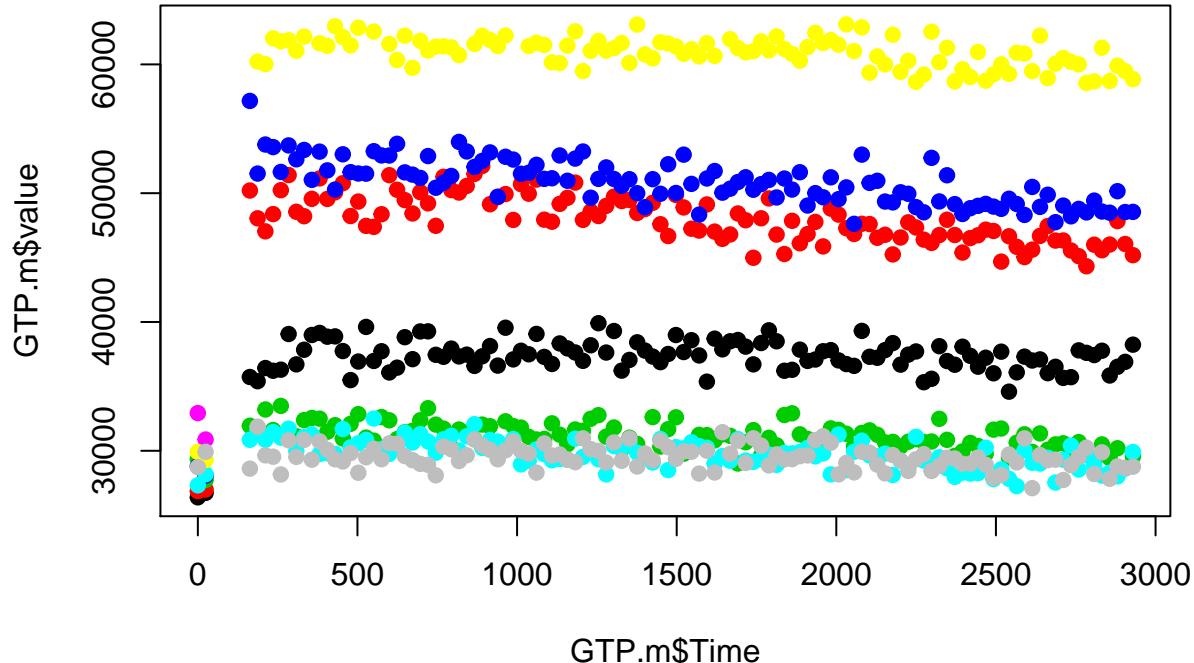
```

GTP <- read.table("chapter4/GTP.activity", header=TRUE, sep="\t")
dGTP <- read.table("chapter4/dGTP.activity", header=TRUE, sep="\t")
GTP.m <- melt(GTP,id="Time")

plot(GTP.m$Time, GTP.m$value, col=GTP.m$variable, pch=19)

## Warning in xy.coords(x, y, xlabel, ylabel, log): NAs introducidos por
## coerción

```



```

plot.new()
legend("center", legend = unique(GTP.m$variable), col = unique(GTP.m$variable), pch=19)

```

- AMg1
- Ssvi
- Jden
- Tcur
- Sros
- Smeg
- SspC
- Buffer

```

ggplot(GTP.m, aes(x=Time, y=value,color=variable)) + geom_point() + facet_wrap(~ variable) + geom_smooth()

## `geom_smooth()` using method = 'loess' and formula 'y ~ x'
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : span too small. fewer data values than degrees of freedom.
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : at 227.79
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =

```

```

## parametric, : radius 64.12
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : all data on boundary of neighborhood. make span bigger
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : pseudoinverse used at 227.79
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : neighborhood radius 8.0075
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : reciprocal condition number 1
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : at 1845.3
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : radius 64.12
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : all data on boundary of neighborhood. make span bigger
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : There are other near singularities as well. 64.12
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : zero-width neighborhood. make span bigger
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : zero-width neighborhood. make span bigger
## Warning: Computation failed in `stat_smooth()`:
## NA/NaN/Inf en llamada a una función externa (arg 5)
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : span too small. fewer data values than degrees of freedom.
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : at 474.62
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : radius 15.07
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : all data on boundary of neighborhood. make span bigger
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : pseudoinverse used at 474.62
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : neighborhood radius 3.882
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : reciprocal condition number 1
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : at 1258.8
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : radius 15.07
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : all data on boundary of neighborhood. make span bigger

```

```

## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : There are other near singularities as well. 15.07
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : zero-width neighborhood. make span bigger
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : zero-width neighborhood. make span bigger
## Warning: Computation failed in `stat_smooth()`:
## NA/NaN/Inf en llamada a una función externa (arg 5)
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : span too small. fewer data values than degrees of freedom.
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : at 859.79
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : radius 47.817
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : all data on boundary of neighborhood. make span bigger
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : pseudoinverse used at 859.79
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : neighborhood radius 6.915
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : reciprocal condition number 1
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : at 2256.6
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : radius 47.817
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : all data on boundary of neighborhood. make span bigger
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : There are other near singularities as well. 47.817
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : zero-width neighborhood. make span bigger
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : zero-width neighborhood. make span bigger
## Warning: Computation failed in `stat_smooth()`:
## NA/NaN/Inf en llamada a una función externa (arg 5)
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : span too small. fewer data values than degrees of freedom.
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : at 1084.3
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : radius 0.7208

```

```

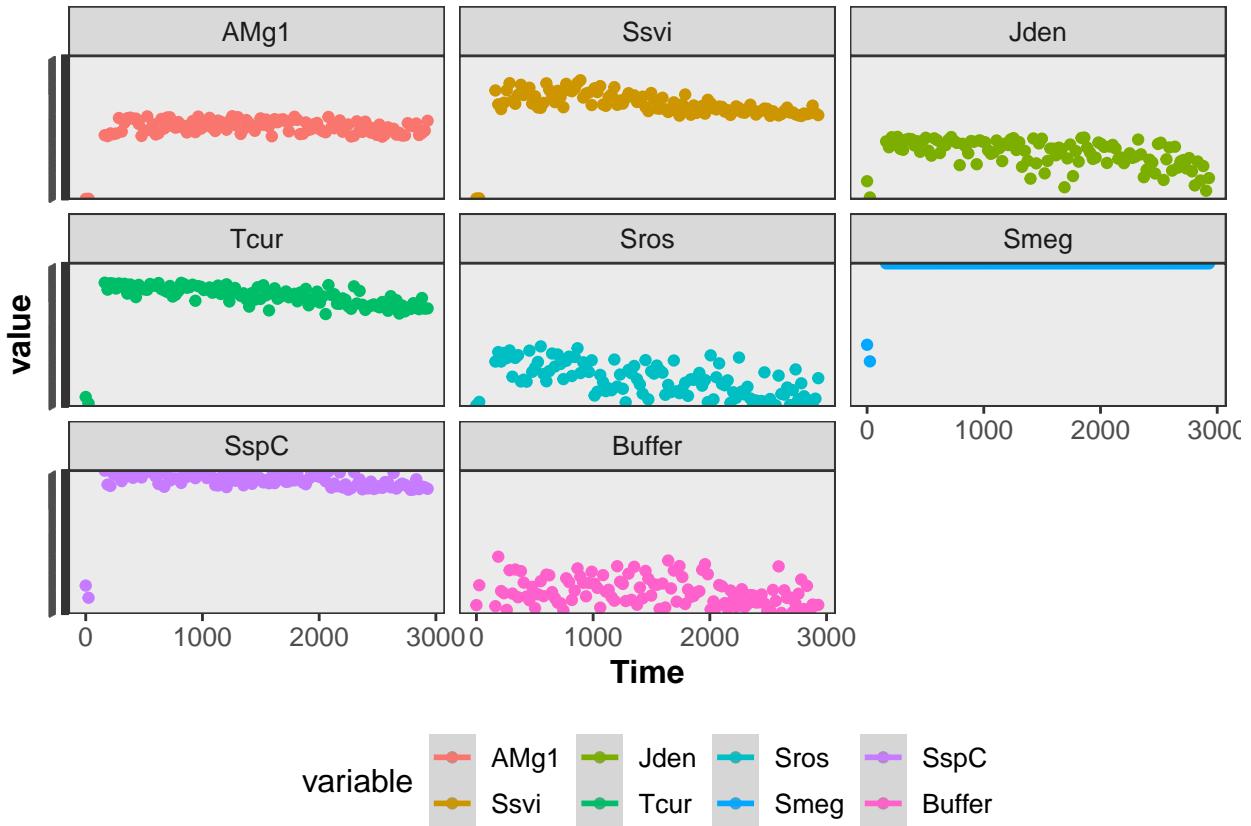
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : all data on boundary of neighborhood. make span bigger
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : pseudoinverse used at 1084.3
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : neighborhood radius 0.849
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : reciprocal condition number 1
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : at 1255.7
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : radius 0.7208
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : all data on boundary of neighborhood. make span bigger
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : There are other near singularities as well. 0.7208
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : zero-width neighborhood. make span bigger
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : zero-width neighborhood. make span bigger
## Warning: Computation failed in `stat_smooth()`:
## NA/NaN/Inf en llamada a una función externa (arg 5)
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : span too small. fewer data values than degrees of freedom.
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : at 1470.5
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : radius 7.7869
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : all data on boundary of neighborhood. make span bigger
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : pseudoinverse used at 1470.5
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : neighborhood radius 2.7905
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : reciprocal condition number 1
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : at 2034.2
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : radius 7.7869
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : all data on boundary of neighborhood. make span bigger

```

```

## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : There are other near singularities as well. 7.7869
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : zero-width neighborhood. make span bigger
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : zero-width neighborhood. make span bigger
## Warning: Computation failed in `stat_smooth()`:
## NA/NaN/Inf en llamada a una función externa (arg 5)
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : span too small. fewer data values than degrees of freedom.
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : at 2612.2
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : radius 2.1199
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : all data on boundary of neighborhood. make span bigger
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : pseudoinverse used at 2612.2
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : neighborhood radius 1.456
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : reciprocal condition number 1
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : at 2906.4
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : radius 2.1199
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : all data on boundary of neighborhood. make span bigger
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : There are other near singularities as well. 2.1199
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : zero-width neighborhood. make span bigger
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : zero-width neighborhood. make span bigger
## Warning: Computation failed in `stat_smooth()`:
## NA/NaN/Inf en llamada a una función externa (arg 5)

```



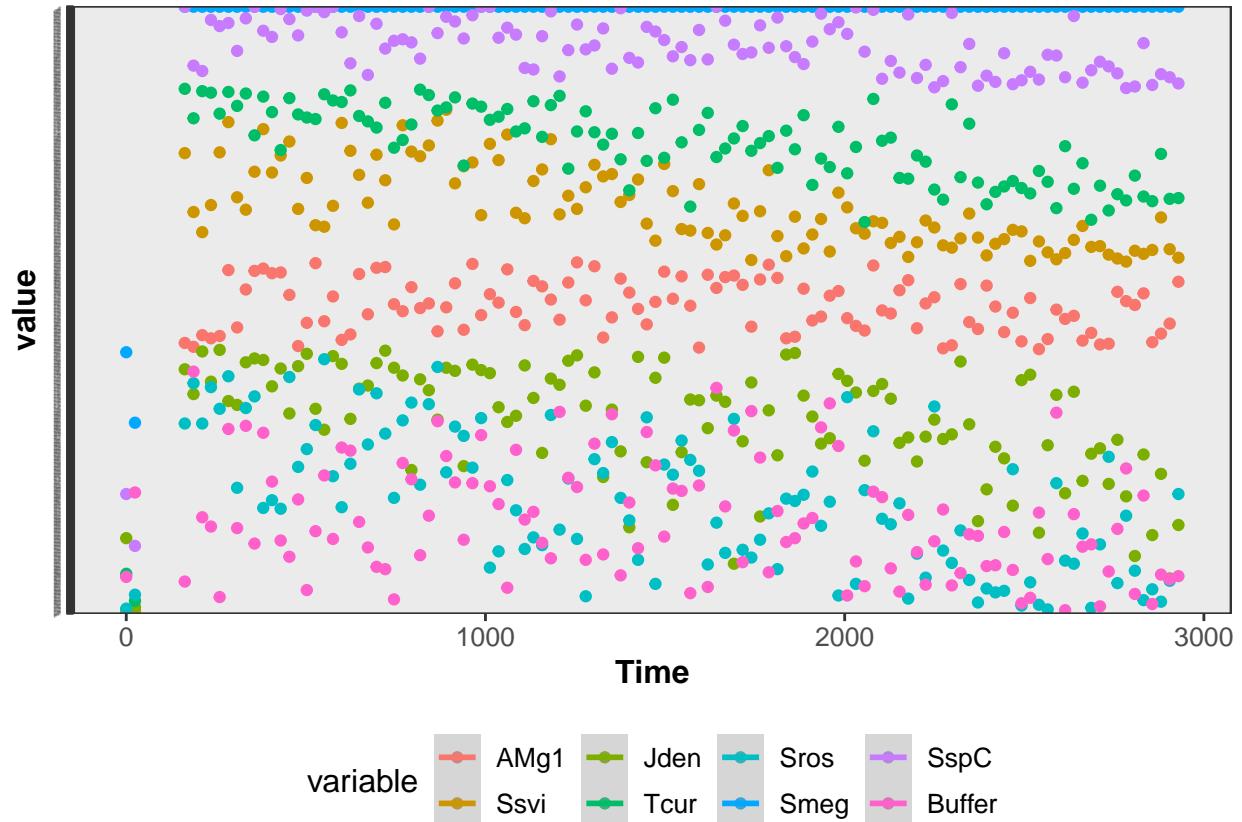
```
ggplot(GTP.m, aes(x=Time, y=value,color=variable)) + geom_point() + geom_smooth() + theme_bw() + theme(plot.title = element_text(size = 16), plot.subtitle = element_text(size = 14), axis.title.x = element_text(size = 14), axis.title.y = element_text(size = 14), axis.text.x = element_text(size = 12), axis.text.y = element_text(size = 12), legend.title = element_text(size = 14), legend.text = element_text(size = 12))

## `geom_smooth()` using method = 'loess' and formula 'y ~ x'
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : span too small. fewer data values than degrees of freedom.
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : at 1470.5
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : radius 7.7869
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : all data on boundary of neighborhood. make span bigger
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : pseudoinverse used at 1470.5
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : neighborhood radius 2.7905
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : reciprocal condition number 1
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : at 2034.2
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : radius 7.7869
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : all data on boundary of neighborhood. make span bigger
```

```

## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : There are other near singularities as well. 7.7869
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : zero-width neighborhood. make span bigger
## Warning in simpleLoess(y, x, w, span, degree = degree, parametric =
## parametric, : zero-width neighborhood. make span bigger
## Warning: Computation failed in `stat_smooth()`:
## NA/NaN/Inf en llamada a una función externa (arg 5)

```



the two enzymes with activity

Thermomonospora curvata thermophilic Actinobacteria from Thermomonosporaceae genus, it can be found in compost and participate in the active degradation of cellulose [@chertkov_complete_2011].

Jonesia trpF shows no activity *Jonesia denitrificans* is classified as a pathogenic organism for animals, reported genome was originally isolated from cooked ox blood [@pukall_complete_2009].

```

docking <- read.csv("chapter4/SmallHeat.data", header=TRUE, sep="\t")
kable(docking, caption = "Enzymes docking \\label{tab:docking}", caption.short = "Enzymes docking ")

```

Table 5: Enzymes docking

Enzima	S13	S15	S14	S16	S10	S12	S9	S18	S5	S4	S8	S17	S7	S6	S11	S1
Srub_2VEP	-7.4	-7.3	-7.5	-7.1	-6.5	-6.2	-6.5	-7.7	-9.4	-9.3	-7.9	-7.2	-8.3	-8.6	-8.9	-9.0
Saver_2VEP	-7.4	-7.2	-7.0	-6.5	-7.3	-6.4	-7.0	-7.5	-9.6	-8.5	-7.9	-7.6	-8.4	-8.7	-9.8	-8.3
Scoe_2VEP	-7.5	-7.5	-7.9	-7.0	-7.0	-6.2	-6.5	-7.8	-8.8	-9.2	-7.8	-7.9	-8.0	-8.9	-10.3	-9.2
Scoe_2X30	-8.1	-7.4	-7.6	-6.9	-6.7	-6.8	-7.1	-7.9	-9.1	-9.0	-8.3	-8.6	-8.5	-9.0	-10.6	-10.0

Enzima	S13	S15	S14	S16	S10	S12	S9	S18	S5	S4	S8	S17	S7	S6	S11	S1
Scoe_1VZW	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
Sfla_2VEP	-7.1	-7.6	-7.2	-7.3	-7.2	-6.8	-6.9	-8.1	-8.2	-7.2	-8.2	-7.2	-8.4	-8.3	-8.5	-7.9
Sgha_2VEP	-9.2	-8.7	-6.7	-6.7	-7.4	-7.7	-7.2	-7.5	-9.8	-8.9	-8.2	-7.8	-8.8	-8.7	-10.1	-9.1
Siak_2VEP	-6.6	-7.3	-7.0	-7.1	-7.1	-7.1	-6.8	-8.0	-9.2	-8.7	-7.8	-7.6	-8.3	-8.4	-9.1	-5.8
Sbic_2VEP	-7.2	-6.7	-6.8	-6.5	-6.2	-6.6	-5.9	-7.8	-8.5	-7.8	-7.8	-7.2	-8.2	-8.0	-9.6	-8.2
Sbot_2VEP	-9.6	-10.9	-8.9	-7.1	-6.0	-6.2	-6.7	-8.1	-9.1	-8.8	-8.3	-7.9	-8.9	-9.3	-9.4	-9.8
Sipo_2VEP	-6.6	-6.6	-7.3	-6.9	-6.5	-6.3	-6.5	-8.1	-8.6	-9.1	-8.0	-7.9	-8.0	-8.4	-8.5	-7.2
Ssvi	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
Ssvi_4U28	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
Ssvi_4TX9	-5.9	-8.2	-7.1	-6.6	-6.8	-7.0	-7.6	-7.9	-8.4	-8.3	-8.2	-8.1	-8.4	-8.7	-7.7	-8.2
Salb_2VEP	-6.3	-7.1	-7.4	-6.8	-7.7	-6.9	-6.6	-8.0	-9.6	-8.9	-8.6	-7.9	-8.6	-8.4	-9.1	-7.4
Sbik_2VEP	-7.4	-6.7	-7.4	-7.3	-7.7	-6.5	-6.8	-8.5	-9.9	-9.8	-8.3	-7.8	-8.2	-8.4	-10.5	-7.1
Stsu_2VEP	-6.9	-7.5	-7.9	-7.8	-7.5	-7.3	-7.7	-8.1	-10.0	-9.7	-8.7	-7.8	-8.8	-8.8	-10.6	-7.8
Sven_2VEP	-8.3	-7.6	-7.5	-6.8	-6.4	-7.0	-6.2	-8.2	-9.6	-9.0	-7.9	-7.4	-8.3	-8.6	-10.0	-8.2
Scal_2VEP	-7.3	-8.8	-7.5	-6.7	-7.0	-6.5	-6.5	-8.7	-10.0	-9.7	-8.8	-7.7	-8.2	-8.6	-10.5	-8.1
Sbaa_2VEP	-8.9	-6.6	-6.7	-6.8	-7.2	-7.2	-6.2	-7.7	-9.6	-9.5	-8.4	-7.4	-8.5	-8.2	-9.7	-8.5
Sglo_2VEP	-6.1	-6.8	-6.9	-6.5	-6.7	-6.3	-6.7	-7.5	-9.6	-9.6	-8.6	-7.8	-8.7	-8.7	-9.9	-6.2
Sful_2VEP	-9.0	-6.9	-7.1	-6.8	-6.3	-7.2	-6.6	-8.1	-9.8	-9.3	-7.7	-8.0	-8.7	-8.8	-10.5	-7.6
Sgri_2VEP	-6.6	-8.2	-7.1	-6.4	-7.1	-7.4	-7.1	-7.4	-9.4	-8.5	-8.6	-7.5	-8.8	-8.4	-9.6	-8.8
S34_2VEP	-7.8	-7.2	-7.6	-7.3	-6.2	-6.5	-6.9	-8.0	-8.8	-8.6	-7.7	-7.1	-7.7	-7.9	-7.2	-7.1
Srim_2VEP	-7.4	-6.8	-7.1	-7.2	-7.2	-7.4	-7.8	-8.6	-9.7	-9.3	-8.8	-7.7	-8.9	-8.7	-10.2	-6.8
Satr_2VEP	-7.6	-7.1	-7.5	-7.4	-6.2	-6.5	-6.9	-8.0	-8.9	-8.7	-7.7	-7.4	-7.7	-7.8	-6.5	-7.0
S1813_2VEP	-7.5	-6.7	-6.8	-6.6	-6.8	-7.0	-6.7	-7.9	-9.9	-9.7	-8.3	-7.7	-8.5	-8.6	-10.0	-7.6
Svar_2VEP	-6.3	-6.8	-7.0	-6.3	-6.9	-6.9	-6.6	-7.8	-9.3	-7.4	-8.5	-7.3	-8.0	-8.7	-8.5	-7.4
Sfra_2VEP	-7.3	-7.0	-6.8	-6.6	-6.5	-7.3	-6.6	-8.6	-9.1	-8.7	-8.9	-7.7	-8.9	-9.0	-8.7	-6.0
Smeg_2VEP	-8.0	-7.3	-6.9	-6.6	-7.2	-6.7	-6.1	-7.6	-9.2	-9.1	-7.8	-7.4	-8.2	-8.5	-9.6	-9.2
Ssul_2VEP	-7.5	-7.0	-6.9	-6.6	-6.6	-6.7	-6.9	-7.7	-8.4	-8.2	-8.2	-7.5	-8.1	-7.7	-8.2	-6.7
Slav_4X9S	-8.0	-7.1	-6.7	-6.9	-6.6	-6.8	-6.7	-8.2	-9.5	-9.1	-8.6	-8.0	-8.4	-8.7	-9.1	-9.5
Sery_4X9S	-7.8	-7.2	-7.6	-7.2	-7.4	-6.9	-7.4	-8.5	-9.4	-9.4	-8.6	-7.6	-8.4	-8.6	-9.1	-10.0
SspC_4X9S	-7.4	-6.9	-7.3	-6.8	-7.3	-6.6	-7.5	-8.0	-9.8	-9.3	-8.7	-7.6	-8.5	-8.4	-10.6	-9.1
Sxan_4X9S	-7.5	-6.9	-6.8	-6.8	-6.5	-6.6	-6.5	-8.3	-8.0	-8.1	-7.6	-7.4	-8.7	-8.1	-8.1	-8.5
Skat_4X9S	-6.4	-7.0	-7.1	-6.7	-7.7	-6.3	-6.2	-8.1	-8.5	-9.2	-8.3	-7.6	-9.0	-8.5	-9.7	-9.5
SMg1_4X9S	-6.5	-6.9	-7.2	-7.1	-6.5	-6.9	-6.4	-7.3	-7.8	-7.7	-8.3	-7.5	-7.9	-8.4	-9.5	-7.6
SMg1_W9T	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
Sniv_2VEP	-6.2	-6.8	-7.4	-6.5	-7.8	-6.8	-6.7	-9.5	-9.6	-9.4	-8.7	-8.2	-8.6	-9.1	-9.9	-5.4
Scla_2VEP	-6.4	-7.7	-7.4	-7.2	-7.6	-7.5	-7.2	-8.5	-10.1	-10.0	-8.7	-7.9	-8.8	-9.1	-10.2	-6.9
Save_2VEP	-7.6	-6.7	-7.1	-7.2	-7.1	-6.5	-6.6	-7.8	-8.6	-10.0	-8.6	-7.5	-8.3	-8.5	-8.8	-5.3
Spur_2vEP	-6.8	-7.3	-6.9	-6.7	-8.0	-7.9	-7.7	-8.5	-9.5	-9.8	-8.1	-7.8	-8.7	-9.7	-10.0	-5.3
Scar_2Y89	-6.9	-6.8	-7.5	-7.1	-7.8	-7.3	-7.2	-8.3	-9.2	-8.3	-8.9	-8.4	-8.9	-9.3	-9.4	-5.3
Mtub_2Y88	-10.0	-7.8	-8.9	-5.4	-8.6	-7.3	-7.8	-9.5	-10.9	-10.3	-9.5	-9.0	-9.8	-9.8	-11.3	-10.1
Mtub_2Y89	-8.7	-8.6	-9.6	-9.4	-6.4	-5.9	-5.6	-7.1	-7.0	-7.5	-7.3	-6.8	-7.4	-8.4	-7.5	-8.1
Mtub_2Y85	-8.2	-7.9	-9.2	-7.4	-7.6	-7.5	-7.6	-8.4	-9.7	-9.5	-9.3	-7.8	-8.6	-8.6	-10.2	-9.8
Mtub_3ZS4	-10.0	-10.5	-11.4	-6.4	-8.2	-7.2	-7.0	-9.6	-10.2	-10.2	-9.9	-8.5	-9.3	-9.6	-10.9	-10.0
S34_3ZS4	-7.4	-8.0	-7.6	-6.4	-5.2	-5.4	-5.2	-6.1	-6.8	-6.4	-5.7	-6.4	-6.3	-6.3	-7.1	-7.4
Cdip_4AXK	-9.2	-7.8	-10.9	-7.2	-7.5	-7.6	-7.7	-8.9	-9.0	-9.8	-9.0	-8.3	-8.8	-9.2	-10.1	-9.5
Cjei_4AXK	-7.5	-6.9	-7.2	-6.5	-8.4	-8.0	-8.5	-8.7	-9.5	-9.6	-9.4	-8.8	-9.0	-9.5	-9.3	-8.1
Aaur	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
Aaur_4WD0	-8.2	-8.5	-8.1	-7.9	-5.7	-5.5	-5.9	-7.0	-7.5	-7.2	-7.1	-6.7	-7.1	-7.4	-8.4	-7.2
Acar_4X2R	-10.6	-9.3	-9.3	-7.2	-7.2	-6.8	-7.3	-7.5	-9.5	-9.8	-8.0	-7.8	-9.3	-8.9	-10.3	-9.6
Auro_4X2R	-9.9	-9.1	-9.8	-8.1	-7.4	-7.1	-7.4	-7.8	-9.2	-9.8	-8.3	-7.8	-9.3	-9.0	-9.9	-9.0
Afer_4WD0	-5.7	-5.8	-6.0	-5.7	-6.7	-6.2	-5.8	-7.6	-9.2	-8.8	-7.8	-7.4	-8.3	-8.4	-9.3	-6.7
Sent_5AHE	-9.1	-5.4	-6.4	-5.2	-8.0	-7.3	-7.4	-8.8	-10.7	-10.2	-8.7	-8.7	-9.6	-9.9	-10.9	-7.8

Enzima	S13	S15	S14	S16	S10	S12	S9	S18	S5	S4	S8	S17	S7	S6	S11	S1
Ecoli_K12	-9.7	-9.7	-9.2	-6.1	-7.2	-6.6	-6.8	-8.6	-9.5	-9.1	-8.6	-8.2	-9.0	-8.6	-10.2	-9.9
Jden_4WUI	-8.3	-8.8	-8.2	-6.8	-6.2	-6.1	-6.0	-6.8	-7.4	-7.6	-7.5	-6.9	-7.6	-7.5	-7.7	-7.5
Ctra	-8.2	-8.0	-7.4	-7.2	-6.1	-5.5	-5.4	-6.5	-7.1	-7.1	-7.0	-6.2	-6.8	-6.8	-6.9	-7.2
SMg1_trpF	-7.2	-8.8	-8.2	-7.3	-6.2	-5.7	-5.7	-6.9	-6.6	-6.7	-7.3	-6.7	-7.6	-6.9	-7.5	-7.1
Aodo_4X2R	-8.5	-8.6	-8.8	-7.3	-7.1	-7.1	-7.1	-7.5	-9.7	-9.4	-7.8	-7.6	-9.6	-8.8	-10.1	-8.4

Enzima	S13	S15	S14	S16	S10	S12	S9	S18	S5	S4	S8	S17	S7	S6	S11	S1
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Molecular dynamics vs experimental data

PriA activity on S3 (PRA)

organism	Family	K_M	k_{cat}	$\frac{k_{cat}}{K_M}$	Pre MD	Pos MD	Reference
Afer	HisA	1.1 ± 0.2	0.05 ± 0.001	0.045	-10.1	-12.3	Noda-García L et al 2015
Ecoli	HisA	1.6	4.9	3.1	-9.9	-16	Henn-Sax et al. (2002)
Sent	HisA	17.0 ± 0.1	7.8 ± 2.4	4.5×10^5	-10.3	-20.1	Söderholm A et al (2015)
Aaur	PriB	2.1 ± 0.5	1.8 ± 0.2	0.9	-7.4		verduzco-castro 2016
Sipo	PriB	3.8 ± 0.2	0.82 ± 0.02	0.21	-8.2	-14.7	verduzco-castro 2016
SspC	PriB	11.4 ± 3.4	2.53 ± 0.74	0.22	-8.5	-12.7	verduzco-castro 2016
SMg1	PriB	13.2 ± 3.4	0.92 ± 0.19	0.069	-8	-15.2	verduzco-castro 2016
Ssvi	PriB	3.9 ± 0.89	0.69 ± 0.04	0.18	-8.2	-16.7	verduzco-castro 2016
Scoe	PriA	3.6 ± 0.7	1.3 ± 0.2	0.4	-8.4	-15	Noda-García et al (2010)
Sglob	PriA	4.2 ± 0.8	0.74 ± 0.03	0.18	-9.2	-16.7	verduzco-castro
Mtub 2Y85	priA	190.23	0.012 – 9.7				Due et al 2011
Mtub 3ZS4	priA	?	-9.9				Due et al 2011 (To be published)
Auro	priA	4.0 ± 0.9	0.2 ± 0.03	0.04	-9.2		Vazquez-Juarez (2016)
Cjei	PriA	2.3 ± 0.2	0.9 ± 0.08	0.39	-8.5		Noda-García et al (2013)
Cdip	subHisA	4.4 ± 0.5	2.6 ± 0.3	0.59	-9.2		Noda-García et al (2013)
SMg1 TrpF	TrpF3	-	-	-	-6.9	-9.6	verduzco-castro 2016
Jden	TrpF3	-	-	- -7.2	-9.4	16.8 ± 3.3	Verduzco-Castro E et al 2016
Acar	SubHisA	0.02					
Aodo	SubTrpF	-	-	-			

PriA activity on S7 (PROFAR) \$\$

organism	Family	K_M	k_{cat}	$\frac{k_{cat}}{K_M}$	Pre MD	Pos MD	Reference
Afer	HisA	-	-	-	-9.2	-9	Noda-García L et al. (2015)
Ecoli	HisA	-	-	-	-9	-11.1	Henn-Sax et al. (2002)
Sent	HisA	-	-	-	-9.6	-10.2	Söderholm A et al (2015)
Aaur	PriB	26.3 ± 6.3	0.37 ± 0.09	0.014	-7.1	-	verduzco-castro 2016
Sipo	PriB	60.8 ± 1.1	8.25 ± 0.4	0.14	-8	-8.5	verduzco-castro 2016
SspC	PriB	149.9 ± 29	1.4 ± 0.12	0.009	-8.5	-10.8	verduzco-castro 2016
SMg1	PriB	129.6 ± 34	0.29 ± 0.04	0.0022	-7.5	-11	verduzco-castro 2016
Ssvi	PriB	24.5 ± 4.0	1.6 ± 0.29	0.067	-8	-9.7	verduzco-castro 2016
Scoe	PriA	5.0 ± 0.08	3.4 ± 0.09	0.7	-8	-9.4	Noda-García et al (2010)
Sglob	PriA	11 ± 1.0	3.8 ± 0.2	0.34	-8.7	-9.4	verduzco-castro 2016
Mtub2Y85	priA	21	3.6	0.17	-8.6		Due et al 2011
Mtub3ZS4	priA				-9.3		Due et al 2011 (To be published)
Auro	priA	23 ± 6.5	0.5 ± 0.05	0.02	-9.3		Vazquez-Juarez (2016)
Cjei	PriA	5.1 ± 1.0	1.6 ± 0.16	0.31	-9		Noda-García et al (2013)
Cdip	subHisA	-	-	-	-8.8		Noda-García et al (2013)
SMg1 TrpF	TrpF3	8.4 ± 1.7	10.5 ± 2.4	1.25	-7.6	-9	verduzco-castro
Jden	TrpF3	16.8 ± 3.3	27 ± 1.6	1.6	-7.6	-7.7	verduzco-castro
Acar	SubHisA	Na	Na	0.02	Na	Na	Na
Aodo	SubTrpF	-	-	-	-	Na	Na

\$\$

Con actividad de FolE i.e activa para el compuesto V Adams et al (2014) Genome size vs Total antismash cluster coloured by order

Non darwinian trayectories

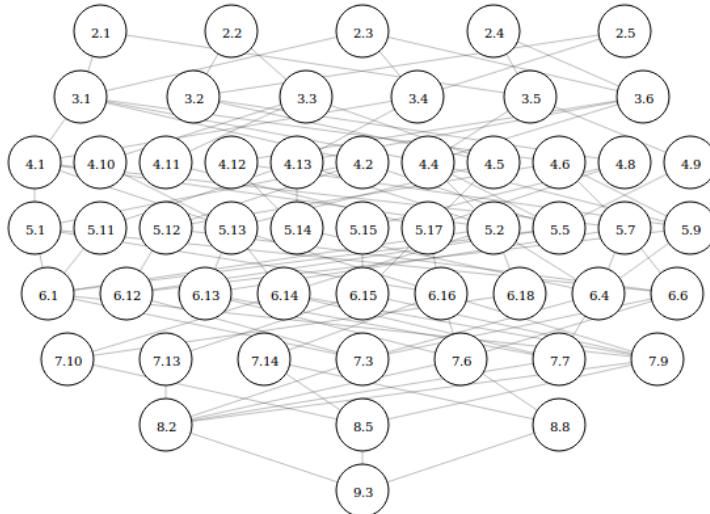


Figure 13: Non darwinian trayectories

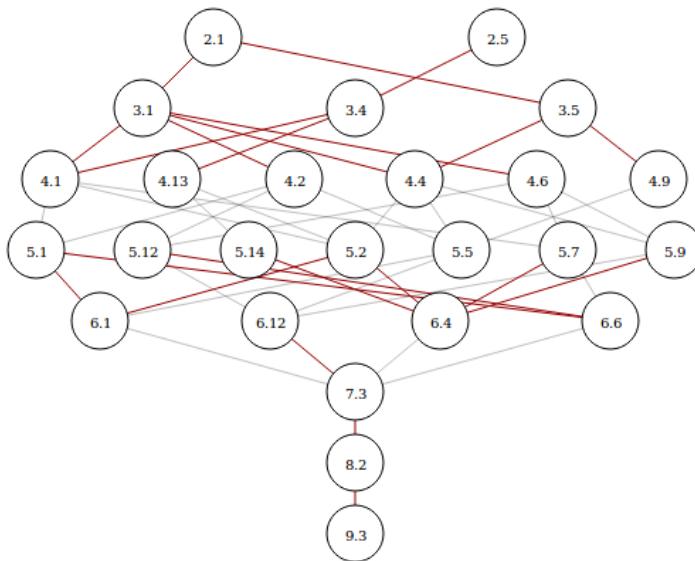


Figure 14: Positive increments on PRA

HisF Arabinosa TrpC TrpD acido antranilico PRPP gliceroles de Ana checar resistencias

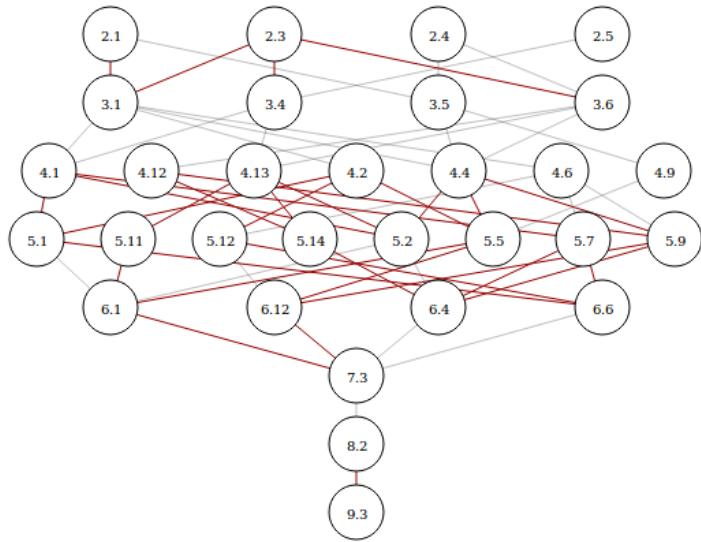


Figure 15: Positive increments on ProFAR



Figure 16: gel

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#sessionInfo()
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