**Supplementary Text 1: Details of ARepA’s case studies**

**Case Study 1: Human Prostate cancer**

* 1. **Retrieve data**
* Get all metadata (pkl files) downloaded for human (Homo sapiens) *a priori*.

Metadata query that searches for all datasets that are prostate cancer specific using the unpickle.py script in the source directory:

$ for i in \*.pkl; do python /arepa/src/unpickle.py < $i| grep “prostate cancer”; done

This query resulted in 10 human prostate cancer specific gene expression datasets as listed in Supplementary Table S1 as of 02/2013.

* Get these datasets from ARepA with activated Sleipnir option (default: “off” -> change into “on”) in order to obtain co-expression networks as “dat/dab” files.
  1. **Meta-analysis**

The script *casestudy1.py* can be executed in order to reproduce the case study results:

$ python casestudy1.py <path to pcl files (step 1)> casestudy1\_querygenes.txt

* Meta-analyze these single co-expression networks into a single prostate cancer specific functional network using unsupervised data integration averaging across normalized co-expression values (z-scores) using *Combiner* in Sleipnir with default setting.

$ Combiner -t dat -o <combined.dab> -n <data.dab>\*

Documentation: <http://libsleipnir.bitbucket.org/Combiner.html>

Reference: Huttenhower et al. Genome Res. 2009 Jun;19(6):1093-106

* Extract a high-confidence subgraph based on the probabilistic graph search algorithm. This starts from a user-defined set of query genes and identifies *k* additional neighbours in the network that are connected with high confidence to the original query genes. Here, we defined a query gene set of twelve genes from the NFκB signaling pathway in BioCarta (Supplementary Table 4) and a neighbourhood size of *k*=10 using *Dat2Graph* in Sleipnir with default setting.

$ Dat2Graph -i <data.dab> -t dat -q <genes.txt> -k 10

Documentation: <http://libsleipnir.bitbucket.org/Dat2Graph.html>

Reference: Huttenhower et al. Genome Res. 2009 Jun;19(6):1093-106, Myers et al. Genome Biology 2005, **6**:R114

* Result: High-confidence NFκB signalling network containing 22 genes in total as illustrated in Figure 2.

**Case Study 2: Mouse meta-analysis**

1. **Retrieve data**

* Get all metadata (pkl files) downloaded for mouse (Mus musculus) *a priori*.
* Metadata query that are germ-free and tissue specific using the unpickle.py script in the source directory:

$ for i in \*.pkl; do python /arepa/src/unpickle.py < $i| grep “germ-free” | grep “wild type” | grep “intestinal tract”; done

This query resulted in 6 case-control datasets from germ-free versus wild type mouse as listed in Supplementary Table 2 as of 02/2013.

* Get these 6 datasets from ARepA with default setting to obtain normalized and standardized expression datasets as “pcl” files.

1. **Meta-analysis**

The scripts *casestudy2\_computeDE.py, casestudy2\_computeMean.py,* and *casestudy2\_forestplpots.R* can be executed in order to reproduce the case study results.

* Compute log fold changes and confidence intervals (2.5% and 97.5%) for all genes between germ-free gut and wild type gut within each dataset using the R/limma package, resulting in ~3,600 differentially expressed genes (*casestudy2\_computeDE.py, casestudy2\_computeMean.py)*.

Reference: Smith et al. Springer 2005: 397-420

* Perform a meta-analysis using the R/metafor package on the six datasets by applying a random-effects model on the fold changes with default options, fitting the model with the restricted maximum-likelihood estimator (REML) (*casestudy2\_forestplpots.R)*. The false discovery rate was controlled by the Benjamini-Hochberg method.

Reference: Viechtbauer et al. 2010 Journal of Statistical Software 36(3): 1-48.

* Test all genes and their resulting meta-p-values for gene set enrichment (GSEA) in KEGG and BioCarta pathways.
* Result: 2 pathways that were significantly enriched for upregulated genes under germ-free conditions (using 1,000 permutations), while 15 pathways were enriched for downregulated genes as shown in Supplementary Table 3.

**Case Study 3: Bacterial data integration**

1. **Retrieve data**

* Get all gene expression data (GEO) and gene interaction data (IntAct, MPIDB, RegulonDB, Bacteriome and STRING, Table 2A) as of 02/2013 from ARepA with activated Sleipnir option (default: “off” -> change into “on”) in order to obtain co-expression networks as “dat/dab” files for *Bacillus subtilis, Escherichia coli* and *Pseudomonas aeruginosa*. Use UniRef90 as the output gene identifier for all bacteria.
  + - * GEO as of 02/2013, refer to Supplementary Table S2
      * Intact version 2013-01-30
      * MPIDB **Release 2009-11-18**
      * RegulonDB Release 8.1
      * Bacteriome as of 02/2013
      * STRING v9.0

1. **Meta-analysis**

The script *casestudy3.py* can be executed in order to reproduce the case study results; for each bacterium the script is executed individually, using the appropriate downloaded data files (pcl) and query genes (*genes\_secretion\_ecoli\_KOs.txt, genes\_secretion\_bacillus\_KOs.txt, genes\_secretion\_pseudomonas\_KOs.txt*):

$ python casestudy3.py <path to pcl files (step 1)> genes\_secretion\_ecoli\_KOs.txt

* Within each bacterium, integrate these data into a species-specific functional network using an unsupervised data integration averaging across normalized co-expression values (z-scores) using *Combiner* in Sleipnir with default setting.

$ Combiner -t dat -o <combined.dab> -n <data.dab>\*

Documentation: <http://libsleipnir.bitbucket.org/Combiner.html>

Reference: Huttenhower et al. Genome Res. 2009 Jun;19(6):1093-106

* Extract a high-confidence subgraph based on the probabilistic graph search algorithm. This query started from a user-defined set of genes of the sec and tat genes and genes from the Type I, Type II, Type III, Type V, and Type VI secretion systems (Supplementary Table 4B) and used a neighbourhood size of *k*=5 using *Dat2Graph* in Sleipnir with default setting.

$ Dat2Graph -i <data.dab> -t dat -q <genes.txt> -k 5

Documentation: <http://libsleipnir.bitbucket.org/Dat2Graph.html>

Reference: Huttenhower et al. Genome Res. 2009 Jun;19(6):1093-106, Myers et al. Genome Biology 2005, **6**:R114

* Result: 3 integrated functional networks as shown in Figure 4.

**MD5 checksum for Case Study 1**

753215e7d6fbd116b4b751f433033e87 GSE12348.dab

547f8b92a27abf7ee7e0fefbaea26a7d GSE14206.dab

de3b0b554e9fa7cda4c2d7e0cef1f44b GSE2443.dab

3f4220a6d8139d713ad99074eda1a683 GSE3868.dab

3963817e76c07e6c02659ad8197dbb53 GSE5377.dab

08904f7d70af612fe91d31ad5f733b1e GSE6956.dab

ae2b2c8168d238c361aa3cce7448e9a2 GSE7930.dab

fcdf73dcf42b3997f2b12467e3e4143e GSE8218.dab

b88dc8c8aaa786242c7d98104f866685 GSE8511.dab

0c5485652f3d39ee53c9873f6466516c GSE9347.dab

**MD5 checksum for Case Study 2**

575fa667f977f639fb50069d3e5a0792 GDS2163-GPL1261.pcl

150381b40ddd14addbb89d96a56a0efa GDS2968-GPL81.pcl

4ff3e873a2af0e6d8c9aa7e35bd7fdc4 GDS640-GPL81.pcl

0c20678d12ff95dcd0fe4d93b229af7b GSE13908.pcl

625b09bf6b5e80fbd0b88f0048ff90e7 GSE17438.pcl

9b7e4b6f0ea4d9646962f9d8b1250bd6 GSE22648.pcl

**MD5 checksum for Case Study 3**

*Bacillus subtilis:*

bbf0695a4ff2ae309922107f60d9f4e1 GSE1620.dab

b3b5d5ceceff8e6ad352c5d76a002649 GSE17435.dab

4840e395a2e62269d38c8fafde21e700 GSE17808.dab

da516613b5ab116a7ef47c34bbd277f0 GSE17809.dab

4a6ab0cd479b5f49395a6e74969b0863 GSE17884\_GPL3655.dab

5b1db24f4522a810296e1eb5792683f3 GSE2667.dab

8a28d01464336aff2236103b3aff6c8c GSE4670\_GPL3664.dab

3cb24da9c7661c1c09991923db5fa800 GSE4670\_GPL3665.dab

c95cc5c6305baf5b91b3206fc381a590 GSE4673\_GPL3645.dab

be446be42b537c7ebff9dee434b763d8 GSE4673\_GPL3655.dab

c429e420d9637d7f0fc013174c7be25a GSE4673\_GPL3663.dab

d8f8b8b5d579eb43231394a536fc92c4 GSE4673\_GPL3668.dab

d03231a60792bb213dcb81e8d206f474 GSE4673\_GPL3671.dab

4bdbcb37b67f158c9f5a9ce4d8fe7584 GSE5210\_GPL3950.dab

81e0861732eb9a093b27b7be17e0fad7 GSE6865.dab

d7392d2bf1134fdf52e28259d0396cfd IntAct\_taxid\_224308\_mapped.dab

07c39bf694266d0c81df9f4d09c827ee IntAct\_taxid\_224308\_pmid\_16796675\_mapped.dab

be32e7753e47936f507ecda87a8b9108 IntAct\_taxid\_224308\_pmid\_21630458\_mapped.dab

6ef2a3b6148dfb6c532d38452fa26877 MPIDB\_taxid\_1423\_mapped.dab

287ae47bb599d2abf247d27d98b9046a MPIDB\_taxid\_1423\_pmid\_18156271\_mapped.dab

5f9822a9489ecaba16130401931de1c4 PrositeDomine\_224308\_mapped.dab

89967dfbb1fd824c2da12eb32e46911a PrositeDomine\_655816\_mapped.dab

11dddea966bb18d4db674e8e2eaa05eb STRING\_taxid\_224308\_mode\_activation\_mapped.dab

202798abfb2ee9b8f6c92de508042986 STRING\_taxid\_224308\_mode\_binding\_mapped.dab

1afec781998537a533d8169b3e99fadd STRING\_taxid\_224308\_mode\_expression\_mapped.dab

89ba34b1fd709b557e462d324f8f35a8 STRING\_taxid\_224308\_mode\_ptmod\_mapped.dab

*Pseudomonas aeruginosa:*

f60e62236f2d46ecf63c70e284dc882e GDS1910-GPL84.dab

5cd2215656cdcbf92b5af6ef8c3fdc55 GDS2111-GPL84.dab

7e671ca973c7fd0fae3135186597066b GDS2317-GPL84.dab

ee967ef6aa4b84d7404ffd25ecbb1f60 GDS2377-GPL84.dab

40583ed4babeb6c6ecc447398cda56fc GDS2502-GPL84.dab

1f0a869796060fbf77aa491d079b17a0 GDS2869-GPL84.dab

cb2cfc9454c73a15287abc0c6d8efef7 GDS2870-GPL84.dab

51ef56834afc5f0dbbf51ce651ae49c2 GDS2893-GPL84.dab

09a9be513800ff85f9b014e8ccd87a09 GDS3174-GPL84.dab

f68c4052cc528a4332a20fcaf62542aa GDS3251-GPL84.dab

f8053bf1b8c2d29dc6c10e6865303cff GDS3562-GPL84.dab

1ec76be13c6c9b3888949f81d81d140a GSE10030.dab

1521f1cbdaa6b91d3531d5853f79a72a GSE10304.dab

74a9722597cb2beaf8fc7ce574924988 GSE10362.dab

3b72660832c3a17787a4c59584299f28 GSE10604.dab

87307e18edb7f9354a3934be40f89a7a GSE12207.dab

6d913d5a8ecfea419c076b85750373db GSE12738.dab

302f2e4e8b47e6506c6b2206aeb5b00d GSE13871.dab

6a6b46eb7af537c8455fc613cb0409e9 GSE14253.dab

a4b55b70447d399a021869f5dcbe3c31 GSE15697.dab

7fc5f8654ef8919f9c4cc4773c0ef477 GSE17111.dab

768dcd5baec4cac86efa7cd47e2429ca GSE17116.dab

f2768979fbc4350906e2beb14e2f2785 GSE17147.dab

a0d7851029e1c4b981f6b742b1932232 GSE17179.dab

9f13fc7caa2e2c0ad0b8f3600ae047c8 GSE17296.dab

cc688f9caac7770753eae612dacda92d GSE21508.dab

e2831c4dc3527372f41e9604d92f3162 GSE21704.dab

7fd2fa57b27c396e8a2c87fb3edb64f5 GSE21966.dab

f6922ffd175e3b1231d4c7fdbddc9cdf GSE22164.dab

0bc4e500ee64c6a662e54aaf1a18ced4 GSE22665.dab

cb6c95c86c7cf25a8d14f71f07aefa32 GSE22999.dab

19c93f3510ac9a1e230451f3fef2eb1d GSE23007.dab

c3a1fa95f42cc08d1cb098d744fc3ef3 GSE23367.dab

0e36c7a256758c549ae890be4fc1b6b9 GSE24784.dab

0c502996edf0862012578b076939d71f GSE25128.dab

baa011dc9744d34fd987623a11a456fc GSE25129.dab

bb4ab638cea2a25d806e6b1a983db569 GSE25130.dab

5fb35b12811cec35afe06df60fe8bbdf GSE25481.dab

ae7ac91e6fcaeea4c2abf82e784a44d3 GSE25595.dab

65971b583cc0748ba6f52ccddb93919c GSE25945.dab

07f6f2d7704f087a998ac5e34192cd03 GSE26142.dab

f41609261793acb5670ba1edef2e37b2 GSE27674.dab

cbc6e4483db2289d12a3bce33e102945 GSE28719.dab

bad03da3c380c3a1e64dbe14c53d57a9 GSE28953.dab

2be586279c3c30d76444fdfb48c9293c GSE29665.dab

81355350ad60fd6f4ce3d8cb5da3c5a8 GSE31227.dab

a2b661891552a88e9d4935a582c4212c GSE33188.dab

dfdb270d9dad1be976678b0e61f6cf58 GSE33275.dab

bec752b3205b09d91083d3a505fc1e77 GSE34141.dab

2dd08a51065302d55b26f983c619e233 GSE34762.dab

5b695d1e1f794c6e3af9b43794eb6612 GSE35286.dab

7a0b7a87d4b91d6d504e22318d43727e GSE36647.dab

3696cc776b06761bf98833d2ab810e52 GSE6122.dab

8a96c3b677f85b1784ec0f8cf4414ca6 GSE7968.dab

6f7745dd0b6b503e4618a51df13172b5 GSE8083.dab

3fb94d7f615bd6f12cd7fe6e202e8014 GSE8408.dab

cd28df46e2b0e36a3b8e6b4c18a56c19 GSE9255.dab

77ed9c087ebaf4601eb2efb896b11c51 GSE9592.dab

1d2279b335e8440f21190656fb5c758f GSE9621.dab

286c4206b4a7bb41d6a2e5fe0fad4c9a GSE9657.dab

2a9385c37ae53d07c9fb32a7bff6a46e GSE9926.dab

6315e60b2cd90a99894535c20febcf92 GSE9989.dab

2d08e3f2965c96a9949b413eea8d4bde GSE9991.dab

4f6664071cb9c026764b3be697c8a63f IntAct\_taxid\_208964.dab

1985885ce2655c686749628438c6fcc8 MPIDB\_taxid\_287.dab

*Escherichia coli:*

84da0d582f616861624b85f523efd21d GDS100\_GPL18.dab

0009b3e9086e0ade7c296cba00c451bc GDS1804\_GPL3500.dab

01a9d33558a5b69405f8ef836f61098a GDS1827\_GPL199.dab

1bca33359063318972b0b45c4c28be1e GDS1848\_GPL189.dab

1894a20ab291e5bc9ef3c3789f799b27 GDS1963\_GPL1436.dab

635165b5642007fabcce91a82347a293 GDS2427\_GPL3154.dab

516187436e0af11bcb05589e73048e50 GDS2578\_GPL3503.dab

669791a0831b432d6b006b021c768270 GDS2579\_GPL3503.dab

bd051f697272f99aa0705ee6b60b089d GDS2580\_GPL3503.dab

64d9dddcf60b3a6d7c716a12cb8b85b8 GDS2581\_GPL3503.dab

2a651dc28c9fea4a1322b86503677839 GDS2582\_GPL3503.dab

25c8e0e0b0d31633b330b5d398effa83 GDS2583\_GPL3503.dab

a0c402b03dae466888d7b4366251972e GDS2584\_GPL3503.dab

116ca8bc896c2f5770a2ad000392b328 GDS2585\_GPL3503.dab

d8db38e84d7d7efb6d86428edd94ccfb GDS2586\_GPL3503.dab

135ae44cbdaf183a4222e9186257f922 GDS2587\_GPL3503.dab

8c226d00d0a0ae7f70e36d1247fb45a0 GDS2588\_GPL3503.dab

72f8d423b60599216858d10fd7e743c2 GDS2589\_GPL3503.dab

a1747319aa35933025c99c6c6eaf6f23 GDS2590\_GPL3503.dab

9c838ebd92d32461d4a5ad7e90270e62 GDS2591\_GPL3503.dab

b2ffc424af03477ea91b810bee116955 GDS2592\_GPL3503.dab

b7cee01dd37712015befb9bee51166ee GDS2593\_GPL3503.dab

5dc1ea4cba179e16dc726068340b3a9b GDS2594\_GPL3503.dab

d4ceead0d0a620c87bbc699ec3d04663 GDS2595\_GPL3503.dab

ba0f9b9b72d99ec3eb84bbd77d43bf8e GDS2596\_GPL3503.dab

8a9131268affee713884cf562d38ff0a GDS2597\_GPL3503.dab

901dc012e7be85f97f360004b6dd61f7 GDS2598\_GPL3503.dab

0a1c967005570053bff827b263754464 GDS2599\_GPL3503.dab

af9478605baf02e71d16a0b4a8958223 GDS2600\_GPL3503.dab

85e7d642a0bc8dbc14bf554b338772f8 GDS2753\_GPL3154.dab

04591df7de0f9797d6d8e76e054c32da GDS2825\_GPL189.dab

f6d973f333be8d7f02ac2cdcd8a9a714 GDS2879\_GPL3154.dab

2d5bcc735594247b5ffe311f5fc93d0d GDS3123\_GPL199.dab

302ee98b43068b08e77e0f872d8cb6fc GDS3421\_GPL3154.dab

b9887e13150a093eb236b3723e37afa4 GDS3597\_GPL3154.dab

b02c9747569edb72f9146109c403a807 GDS95\_GPL16.dab

4a242a66806607253f6e8ed89f27142a GDS96\_GPL17.dab

0a8c8c995476ebdf463b2dafe111b5cb GDS99\_GPL17.dab

b4f6e569f9113b5c3a99c5cd694db107 GSE10116.dab

3af2237a999b10e7fb54a38be54a3f0d GSE10158.dab

a891833b72fc1b16edb471eb457ff3bc GSE10319.dab

0b1ec90f4ac54c564f35c20575f88351 GSE10345.dab

5d4b0ab39318e359535b1518e8b5e6dc GSE10440.dab

ffb9b3db7b25601248c906a0cc43ffb9 GSE11052.dab

c975ad2d13522fa30d28d95333a6e2f8 GSE11303.dab

ddef5ffb9d1cf381c99a8c7abd630858 GSE11779.dab

2e213e641b8057417297474f77f4a87c GSE11927.dab

48f2d1fd1d7e2bb47db46e6f411fc10a GSE11932.dab

3413590250b7dd68d4c37030161823ce GSE12006.dab

e8a5ec92e076b926ff935556ccf08852 GSE12190.dab

7628958c0faeb71033ea1364cacd4223 GSE12554.dab

7c8209b34e090a72f29c94351f3c6d6f GSE12701.dab

7784281d34d361639d0983823a2c8a91 GSE12750.dab

619fde48409e59e31ccb672e168e6796 GSE12797.dab

0d6b9a71c709918e54c6dab5be756290 GSE12831.dab

0875498ab0400c1bda8bc32017261714 GSE13444.dab

2292ebb972a006cd682c55293114307d GSE13562.dab

5a7fbb3cb1f6e26b2908679d996892f5 GSE13589.dab

6628eec9650427965f7daa75fe2dabf7 GSE13902.dab

f89cb36f2dd4b56732e4860599c94da8 GSE1421.dab

89c33fb960228a38cd8375235bbbeb38 GSE14472.dab

ae5669da08a78471a15fa430a689882a GSE15050.dab

753476125f1cdccf1b7935b4006fa089 GSE15059.dab

fdca5bd199efcf534aefdd1707c677f8 GSE15404.dab

6465dc22dbd791a3f7976c10030244cf GSE15405.dab

55678bbaf48bd25b54eed7806ecd52f3 GSE15406.dab

c2c2c3652f1b62c55ce34c65160bce55 GSE15500.dab

26f213c4e65cc4714a1e7c134b8870c4 GSE1730.dab

882e4ea0861d79c32eaa1a4130b58add GSE1735.dab

cdb1ef2dbe6dd5b82159bd79c4f96e33 GSE17465.dab

0d833e94e6fa6b450f35cb7693596a42 GSE17526.dab

ca6a71e02e73a8622c389e9bd7899033 GSE17584.dab

13dd718402c82b2348710f4ff7cba3e4 GSE1780.dab

b8651a9364aa1f2d5c43c470635279f9 GSE18118.dab

03b911e371a8b8e84c57094b5fb30de7 GSE18362.dab

f6f781501318efc0dbe677f646ae8805 GSE18623.dab

1ebf2317c64e18df36afd967adda9408 GSE18852.dab

efa7fbe4f9692ac3f815d6c1937cfbac GSE18972.dab

5f7f6fafb4ba949a175b3b0523005bbb GSE19655.dab

44b872b3174c43fd286bba35f7ae0f6e GSE19842.dab

0d638193bff601fab0cafa69c0c278ee GSE19879.dab

f6e00e1ebe6ef134153350a442ded146 GSE19953.dab

70ba1bc75edd85c3fb542a8968c4a7ae GSE20380.dab

4a32ce0cf56b627cbc74a7b1be13fe24 GSE21551.dab

5a900a1d8cefc6cab5dfe423a7a39b59 GSE21652.dab

699a40c0b77926674a3501610f58d661 GSE21820.dab

d0234af375b7d790271c5273b8b6cad0 GSE21893.dab

3dcd0f86233606c94be5fe467d413ecd GSE22057.dab

ba1c9dfb42b6131622dcbafd9881708f GSE23417.dab

a9dbe9b742d35a52822c4133c35d080e GSE25745.dab

daa0600be31feb8d069b8d596c5485fc GSE25982.dab

e1ffa1b8d6c7462655324145333e14d2 GSE2697.dab

173f273c81eb49306da1032a6f525e7d GSE28144.dab

fb77726c2d874b92962a6ffb117c4320 GSE28193.dab

2e93f1485f3183a3a2a1c178324f12cd GSE28412.dab

565c2abb93717c94db57d34e8f8e9482 GSE28795.dab

3e0202224f62e14d0c71ef3d052b1f94 GSE29076.dab

933e58d418ce832e62f074ecf8d5de3c GSE29439.dab

2656d18881cda5e4438b9d43a4a1324e GSE29440.dab

3b87db95233c887039a3fc52d074fdd8 GSE29486.dab

820da5fe0506299369d8d1d0d2e6dcf0 GSE29803.dab

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