INTERSPIA tutorial for using the main analysis interface

Configuring the analysis using user's data

- (1) Entering the list of genes or proteins
 - (1-1) Selecting the type of the input genes or proteins

User can select the type of the input genes or proteins among Ensembl protein identifier (Ensembl_PRO), Ensembl gene identifier (Ensembl), Gene ID (EntrezGene), Gene symbol, and UniProt accession (UniprotKB).

1. Input genes or proteins	
Ensembl_PRO (e.g. ENSP00000361930)	•
Ensembl_PRO (e.g. ENSP00000361930)	
Ensembl (e.g. ENSG00000166913) UniProtKB (e.g. P31946)	
EntrezGene (e.g. 7529)	
Gene symbol (e.g. YWHAB)	
Choose File No file chosen	

(1-2) Entering the list of genes or proteins manually

User can enter the list of genes or proteins in the text box as shown below. The format of genes or proteins need to match with the selected type above.

When user enters genes or proteins with the type of the Ensembl identifier, target species is automatically selected.

1	. Input genes or proteins 🖤	
	Ensembl_PRO (e.g. ENSP00000361930)	•
	EN SP00000263228	*
	EN SP00000263969	
	ENSP00000264122	
	ENSP00000264414	
	ENSP00000264552	
	ENSP00000265339	
	ENSP00000265428	
	ENSP00000266087	*
	Choose File No file chosen	

(1-3) Uploading a file with the list of genes or proteins

User can upload the file contained the list of genes or proteins.

When user uploads genes or proteins with the type of the Ensembl identifier, target species is automatically selected.

1. Input genes or proteins 🔍	
Ensembl_PRO (e.g. ENSP00000361930)	•
ENSP00000217173	
ENSP00000219548	
ENSP00000222402	
ENSP00000253023	
ENSP00000255040	
ENSP00000256474	
ENSP00000259089	
ENSP00000259939	-
Choose File Example input.txt	

- (2) Additional parameters
 - (2-1) Protein extension process

User can optional select whether the protein extension process based on the random walk with restart algorithm is used or not.

The default setting is "No" (do not use the protein extension process). It means that only the input genes or proteins are used in the analysis.

When user selects "Yes", the percentage value between (0.1% to 10%) must be selected. This value is used as a cutoff filtering top cutoff percentage of extended proteins to include this analysis.

2. Addition	al paramet	ters		2. Additiona	al paramet	ers	
2-1. Protein	n extensio	n proc	ess Ø	2-1. Protein	extensio	n proc	ess 🛛
No	•	1	%	Yes	•	1	%

(2-2) STRING confidence score cutoff

User can select the confidence cutoff score used in the STRING database.

"0.7" is the most widely used cutoff score.

When user selects a cutoff score, interactions with a confidence score greater than or equal to the

	2-2. STF	RING confidence score o	utoff 🛛
	0.7	Ŧ	
	0.2		
	0.3		
	0.4		
_	0.5		J
	0.6		
1a	0.7		
	0.8		
	0.9		

cutoff score are considered as "existing interactions". On the other hand, interactions with a

confidence score less than the cutoff are considered as "not existed interactions".

The STRING database contains direct (physical) as well as indirect (functional) interactions.

(3) Selecting a target species

User can select the target species for this analysis. The target species needs to match with the input genes or proteins.

The list of target species also provides the number of protein-protein interactions (PPIs) supported by our web server.

The numbers before and after the slash in parentheses represent the number of PPIs with the STRING confidence score greater than or equal to the user selected cutoff and the number of all PPIs in the STRING database respectively.

The number of PPIs is automatically updated as user selects a different STRING confidence score cutoff.

3. Select a target species	species	
Human (Homo_sapiens, 360,341 / 5,676,528 PPIs)	•	
Target species		
Armadillo (Dasypus_novemcinctus, 96,181 / 3,204,866 PPIs)		
Bat (Myotis_lucifugus, 208,768 / 6,980,614 PPIs)		
Cat (Felis_catus, 188,113 / 6,645,145 PPIs)		
Chimpanzee (Pan_troglodytes, 129,069 / 6,114,571 PPIs)		
Cow (Bos_taurus, 368,873 / 4,889,001 PPIs)		
Dog (Canis familiaris, 295,282 / 5,527,804 PPIs)		
Elephant (Loxodonta_africana, 203,580 / 5,879,013 PPIs)		
Ferret (Mustela putorius furo, 216,469 / 5,073,881 PPIs)		
Flying fox (Pteropus vampyrus, 126,222 / 5,788,640 PPIs)		
Galago (Otolemur garnettii, 366,960 / 6,206,508 PPIs)		

(4) Selecting other species for comparison

User can select less than 10 species to compare with the target species.

The list of species also provides the number of protein-protein interactions (PPIs) supported by our web server.

User can click and drag a mouse, or use a mouse click holding down the shift key to highlight multiple species adjacent to each other, or highlight multiple interspersed species by mouse click holding down the ctrl key.

The highlighted species need to be moved to the below panel by clicking the "v" button.

If user selects the wrong species in the selected box, user can remove them using the " Λ " button.

4. Select other species to compare (less than 10 species)	 Select other species to compare (less than 10 species)
Available	Available
Armadia (Claspype, novemoricutu, 64 181 / 32,04 68 PPlo). Bat Milvols, Lovidous, 2007, 687 (-693) 014 PPls). Call (Arein, Latina, 183, 113 / 2016, 143 (-PPls). Cave (Bos, Lauras, 308, 113 / 405, 104 (-PPls). Cave (Bos, Lauras, 308, 113 / 408, 0001 PPls). Diog (Cans. 5-millon, 260, 267 / 408, 0001 PPls). Elephant (Loxodorda, africana, 203, 880 / 587, 304 PPls). Floring, Scott (Peropos, vampyrus, 128, 222 / 57, 886 APPls). Floring, Scott (Peropos, vampyrus, 128, 222 / 57, 886 APPls). Selected	Armadiio (Daspus, hovemoricus, 96, 1817 / 3.204, 666 PPIs) Bit (Ayola, Uculquay, 203, 706 / 980,014 PPIs) Dog (Caries, Jamiliaria, 265,282 / 5.827, 904 PPIs) Dog (Caries, Jamiliaria, 265,282 / 5.827, 904 PPIs) Elephant (Locotonta, africana, 205,300 / 5.079,013 PPIs) Ferret (Mustela, Johanna, 176, 272 / 178,846 DPIs) Phing, fact (Primoras, ximproy, 184,272 / 178,846 DPIs) Gladago (Oblemur, garreta, 305,800 / 1820,1627 BPIs) Gladago (Oblemur, garreta, 306,800 / 2205,108 PPIs) Gladago (Oblemur, 216,226 / 186,865 PPIs) Gladago (Oblemur, 216,226 / 186,865 PPIs) Gladago (Oblemur, 216,226 / 186,865 PPIs) Conta (Contin, goint), 216,226 / 186,865 PPIs) Conta (Contin, goint), 216,226 / 186,865 PPIs) Conta (Contin, goint), 216,226 / 186,865 PPIs) Conta (Contin, goint), 216,226 / 186,965 PPIs) Selected Selected
Click!	Cart (Folia_catus 188,113/6.645,145.9Pic) Champanzes (Fan_10020405,123,005/6,114,5/11976) Cow (Box_taurus, 388,873 / 4,889,001 PPils)

[Selecting other species to compare]



[Unselecting the chosen species]

(5) Clicking the "Submit" button to start the analysis

If user clicks this button, the results of ID mapping are showing in a separate window.

When clicking the "Continue" button in a separate window as shown below, the analysis begins using input genes or proteins and chosen parameters.

D mapping result					
Total	proteins	Failed prote	ins Succ	ess proteins	
1	4	0		14	
Ensembl_PRO	UniProtKB	Gene symbol	EntrezGene	Ensembl	
ENSCAFP00000001523	Q28350	MYC	403924	N/A	
	F1PW15	MYC	N/A	ENSCAFG0000001086	
ENSCAFP0000000978	E2RSB0	MAPK12	N/A	ENSCAFG0000000685	
ENSCAFP0000005256	E2QWW0	CLSPN	482481	ENSCAFG0000003539	
ENSCAFP0000008437	E2R962	MKKS	485771	ENSCAFG0000005651	
ENSCAFP00000013625	F1PTG8	LOC100856570	N/A	ENSCAFG00000030829	
ENSCAFP00000013943	E2RK98	FARS2	488204	ENSCAFG0000009483	
ENSCAFP00000020178	F1PXC1	USP28	489400	ENSCAFG00000013688	
ENSCAFP00000023887	E2QZS2	MAX	609969	ENSCAFG00000016212	
ENSCAFP00000025704	E2RFR2	USP43	N/A	ENSCAFG00000017439	
ENSCAFP00000026142	F1PG86	BBS4	N/A	ENSCAFG00000017717	
ENSCAFP00000027048	E2RPS0	USP14	480182	ENSCAFG0000018309	
ENSCAFP00000036357	J9NS82	HACL1	N/A	ENSCAFG0000005949	
ENSCAFP00000036914	J9NUS5	N/A	N/A	ENSCAFG0000009235	
ENSCAFP00000037171	J9NVH9	BBS10	N/A	ENSCAFG00000031800	

(6) An example data of the INTERSPIA analysis

When clicking the "Load an example" button, parameters of the analysis using example data are

automatically filled.



(7) Refreshing the analysis page

When the "Clear" button is clicked, all data are cleared and chosen parameter values are reset to default values.