

The minimum information required for reporting a molecular interaction experiment (MIMIx)

Sandra Orchard¹, Lukasz Salwinski², Samuel Kerrien¹, Luisa Montecchi-Palazzi¹, Matthias Oesterheld³, Volker Stümpflen³, Arnaud Ceol⁴, Andrew Chatr-aryamontri⁴, John Armstrong⁵, Peter Woollard⁵, John J. Salama⁶, Susan Moore^{6,19}, Jérôme Wojcik¹⁸, Gary D. Bader⁷, Marc Vidal⁸, Michael E. Cusick⁸, Mark Gerstein⁹, Anne-Claude Gavin¹⁰, Giulio Superti-Furga¹¹, Jack Greenblatt⁷, Joel Bader¹², Peter Uetz¹³, Mike Tyers¹⁴, Pierre Legrain¹⁵, Stan Fields¹⁶, Nicola Mulder¹⁷, Michael Gilson²⁰, Michael Niepmann²¹, Lyle Burgoon²², Javier De Las Rivas²³, Carlos Prieto²³, Victoria M. Perreau²⁴, Chris Hogue⁵, Hans-Werner Mewes³, Rolf Apweiler¹, Ioannis Xenarios¹⁸, David Eisenberg², Gianni Cesareni⁴ & Henning Hermjakob¹

Supplementary Materials

Note 1

Controlled Vocabulary Terms Relating to Interaction Detection Methods for Molecular Interactions

id: MI:0004

name: affinity chromatography technology

def: "This class of approaches is characterised by the use of affinity resins as tools to purify molecule of interest (baits) and their binding partners. The baits can be captured by a variety of high affinity ligands linked to a resin - for example, antibodies specific for the bait itself, antibodies for specific tags engineered to be expressed as part of the bait or other high affinity binders such as glutathione resins for GST fusion proteins, metal resins for histidine-tagged proteins." [PMID:7708014]

related_synonym: "Affinity purification" []

exact_synonym: "affinity chrom" []

is_a: MI:0091 ! chromatography technology

is_a: MI:0400 ! affinity technology

[Term]

id: MI:0006

name: anti bait coimmunoprecipitation

def: "A specific antibody for the protein of interest (bait) is available, this is used to generate a high affinity resin to capture the endogenous bait present in a sample." [PMID:7708014]

exact_synonym: "anti bait coip" []

is_a: MI:0019 ! coimmunoprecipitation

[Term]

id: MI:0007

name: anti tag coimmunoprecipitation

def: "A specific antibody for the protein of interest is not available, therefore the bait protein is expressed as a hybrid protein fused to a tag peptide/protein for which efficient and specific antibodies or a specific ligand are available." [PMID:7708014]

exact_synonym: "anti tag coip" []

is_a: MI:0019 ! coimmunoprecipitation

[Term]

id: MI:0008

name: array technology

def: "In this class of methodologies, the molecules to be tested are presented ordered in an array format (typically at high density) on planar supports. The characteristics and chemical nature of the planar support can vary. This format permits the simultaneous assay, in controlled conditions, of several thousand proteins/peptides/nucleic acids for different functions, for instance their ability to bind any given molecule." [PMID:14755292]

is_a: MI:0400 ! affinity technology

[Term]

id: MI:0009

name: bacterial display

def: "The protein of interest is presented on the outer membrane of Gram negative bacteria by expressing it as a fusion partner to peptide signals that direct heterologous proteins to the cell surface.

For instance, a single chain Fv (scFv) antibody fragment, consisting of the variable heavy and variable light domains from two separate anti-digoxin monoclonal antibodies, was displayed on the outer membrane of Escherichia coli by fusing it to an Lpp-OmpA. Similar systems have also been developed for gram positive bacteria. Fluorescence-activated cell sorting (FACS), is used to specifically select clones displaying a protein binding to scFv-producing cells." [PMID:10436088, PMID:8248129]

is_a: MI:0034 ! display technology

is_a: MI:0054 ! fluorescence-activated cell sorting

[Term]

id: MI:0010

name: beta galactosidase complementation

def: "Beta-galactosidase activity can be used to monitor the interaction of chimeric proteins. Pairs of inactive beta gal deletion mutants are capable of complementing to restore activity when fused to interacting protein partners. Critical to the success of this system is the choice of two poorly complementing mutant moieties, since strongly complementing mutants spontaneously assemble and produce functional beta-gal activity detectable in absence of any fused protein fragment."

[PMID:12042868, PMID:9237989]

exact_synonym: "beta galactosidase" []

is_a: MI:0228 ! cytoplasmic complementation assay

[Term]

id: MI:0011

name: beta lactamase complementation

def: "This strategy is based on a protein fragment complementation assay (PCA) of the enzyme TEM-1 beta-lactamase. The approach includes a simple colorimetric in vitro assays using the cephalosporin nitrocefin and assays in intact cells using the fluorescent substrate CCF2/AM. The combination of in vitro colorimetric and in vivo fluorescence assays of beta-lactamase in mammalian cells permits a variety of sensitive and high-throughput large-scale applications." [PMID:12042868]

exact_synonym: "beta lactamase" []

is_a: MI:0228 ! cytoplasmic complementation assay

[Term]

id: MI:0012

name: bioluminescence resonance energy transfer

def: "In this variation of the FRET assay the donor fluorophore is replaced by a luciferase (typically Renilla luciferase). In the presence of its substrate, the luciferase catalyses a bioluminescent reaction that excites the acceptor fluorophore through a resonance energy transfer mechanism. As with FRET the energy transfer occurs only if the protein fused to the luciferase and the one fused to the acceptor fluorophore are in close proximity (10-100 Angstrom)." [PMID:9874787, PMID for application instance:10725388]

related_synonym: "BRET" []

related_synonym: "LRET" []

exact_synonym: "bret" []

is_a: MI:0051 ! fluorescence technology

[Term]

id: MI:0013

name: biophysical

def: "The application of physical principles and methods to biological experiments." [PMID:14755292]

is_a: MI:0045 ! experimental interaction detection

[Term]

id: MI:0014

name: adenylate cyclase complementation

def: "Adenylate cyclase is encoded by the cyaA gene and contains a catalytic domain which can be proteolytically cleaved into two complementary fragments, T25 and T18, which remain associated in the presence of calmodulin in a fully active ternary complex. In the absence of calmodulin, the mixture of the two fragments does not exhibit detectable activity, suggesting that the two fragments do not associate. When expressed in an adenylate cyclase-deficient E. coli strain (E. coli lacks calmodulin or calmodulin-related proteins), the T25 and T18 fragments fused to putative interacting proteins are brought into close association which result in cAMP synthesis. The level of reconstructed adenylate cyclase can be estimated by monitoring the expression of a cAMP dependent reporter gene."

[PMID:9576956]

related_synonym: "bacterial two-hybrid" []

exact_synonym: "adenylate cyclase" []
is_a: MI:0228 ! cytoplasmic complementation assay

[Term]
id: MI:0016
name: circular dichroism
def: "Circular dichroism (CD) is observed when optically active molecules absorb left and right hand circularly polarized light slightly differently. Linearly polarized light can be viewed as a superposition of two components of circularly polarized light of equal amplitude and phase but opposite handedness. When this light passes through an optically active sample the two polarized components are absorbed differently. The difference in left and right handed absorbance $A(l) - A(r)$ is the signal registered in CD spectra. This signal displays distinct features corresponding to different secondary structures present in peptides, proteins and nucleic acids. The analysis of CD spectra can therefore yield valuable information about the secondary structure of biological macromolecules and the interactions among molecules that influence their structure." [PMID:11578931]
related_synonym: "CD" []
exact_synonym: "cd" []
is_a: MI:0013 ! biophysical

[Term]
id: MI:0017
name: classical fluorescence spectroscopy
def: "Proteins contain endogenous fluorophores such as tryptophan residue and heme or flavins groups. Protein folding and protein-protein interaction can be studied by monitoring changes in the tryptophan environment detected by changes in its intrinsic fluorescence. Changes in the fluorescence emission spectrum on complex formation can occur either due to a shift in the wavelength of maximum fluorescence emission or by a shift in fluorescence intensity caused by the mixing of two proteins. The interaction of two proteins causes a shift in the fluorescence emission spectrum relative to the sum of the individual fluorescence spectra, resulting in a difference spectrum $[F(\text{complex}) - 2 F(\text{sum})]$, which is a measurable effect of the interaction. Loss of fluorescence signal from a substrate can be used to measure protein cleavage." [PMID:7708014]
exact_synonym: "fluorescence spectr" []
is_a: MI:0051 ! fluorescence technology

[Term]
id: MI:0018
name: two hybrid
def: "The classical two-hybrid system is a method that uses transcriptional activity as a measure of protein-protein interaction. It relies on the modular nature of many site-specific transcriptional activators (GAL 4), which consist of a DNA-binding domain and a transcriptional activation domain. The DNA-binding domain serves to target the activator to the specific genes that will be expressed, and the activation domain contacts other proteins of the transcriptional machinery to enable transcription to occur. The two-hybrid system is based on the observation that the two domains of the activator need to be non-covalently brought together by the interaction of any two proteins. The application of this system requires the expression of two hybrid. Generally this assay is performed in yeast cell, but it can also be carried out in other organism." [PMID:10967325, PMID:12634794, PMID:1946372]
related_synonym: "2-hybrid" []
related_synonym: "2H" []
related_synonym: "2h" []
related_synonym: "classical two hybrid" []
related_synonym: "Gal4 transcription regeneration" []
related_synonym: "two-hybrid" []
related_synonym: "yeast two hybrid" []
exact_synonym: "2 hybrid" []
is_a: MI:0232 ! transcriptional complementation assay

[Term]
id: MI:0019
name: coimmunoprecipitation
def: "In this approach an antibody, specific for the protein of interest (bait) or any tag expressed within a fusion protein, is used to separate the bait from a protein mixture or a cell lysate and to capture its ligand simultaneously. The protein partners that bind to the bait protein retained by the resin can then be eluted and identified." [PMID:7708014]
related_synonym: "co-immunoprecipitation" []

related_synonym: "Co-IP" []
related_synonym: "CoIp" []
related_synonym: "immunoprecipitation" []
exact_synonym: "coip" []
is_a: MI:0004 ! affinity chromatography technology

[Term]
id: MI:0020
name: transmission electron microscopy
def: "During the treatment for microscope analysis a tissue section is incubated with high-specificity antibodies coupled to heavy metals (gold). Any tissue section can then be analysed by electron microscopy to localise the target proteins within the cell. This method supports very high resolution colocalisation of different molecules in a cell." [PMID:14755292]
exact_synonym: "tem" []
is_a: MI:0040 ! electron microscopy

[Term]
id: MI:0021
name: colocalization by fluorescent probes cloning
def: "Two proteins can be localised to cell compartments, in the same experiment, if they are expressed as chimeric proteins fused to distinct proteins fluorescing at different wavelengths (Green Fluorescent Protein and Red Fluorescent Protein for example). Using a confocal microscope the two proteins can be visualized in living cells and it can be determined whether they have the same subcellular location. Fluorescence microscopy of cells expressing a GFP fusion protein can also demonstrate dynamic processes such as its translocation from one subcellular compartment to another.\nOBSOLETE: use imaging technique (MI:0428) and specific probe as feature of each interacting protein."
[PMID:14755292]
exact_synonym: "coloc fluoresc probe" []
is_obsolete: true

[Term]
id: MI:0022
name: colocalization by immunostaining
def: "The subcellular location of a protein can be demonstrated by treating cells fixed on a microscope slide with an antibody specific for the protein of interest. A secondary antibody conjugated with a reactive enzyme (e.g. horseradish peroxidase) is then added. Following a washing step to remove the unbound secondary ligand, a chromogenic substrate (e.g. 3,3', 5,5' tetramethyl benzidine chromogen [TMB]) is converted to a soluble coloured product by the conjugated enzyme and can then be visualised by standard microscopic techniques.\nOBSOLETE since combination of Interaction Detection Method and Interaction Type. Consider using the Interaction Detection Method imaging techniques (MI:0428) coupled with Interaction Type colocalisation (MI:0401) and Participant detection immunostaining (MI:0402) instead." [PMID:14755292]
related_synonym: "Immunofluorescence Staining" []
related_synonym: "Immunostaining" []
exact_synonym: "coloc immunostaining" []
is_obsolete: true

[Term]
id: MI:0023
name: colocalization/visualisation technologies
def: "Techniques enabling the identification of the subcellular localisation of a protein or complex. Two different proteins show a similar distribution in the cell are said to co-localise. Obsolete since combination of Interaction Detection Method and Interaction Type.\nOBSOLETE. Consider using imaging techniques (MI:0428) as interaction detection method coupled with colocalisation (MI:0401) as interaction type and predetermined (MI:0396) as participant detection." [PMID:14755292]
exact_synonym: "coloc visual technol" []
is_obsolete: true

[Term]
id: MI:0024
name: confirmational text mining
def: "Text mining is used to support interactions which have been determined by other methods."
[PMID:14755292]
exact_synonym: "conformational tm" []

is_a: MI:0110 ! text mining

[Term]

id: MI:0025

name: copurification

def: "Approaches designed to separate cell components on the basis of their physicochemical properties. The observation that two or more proteins copurify in one or several conditions is taken as an indication that they form a molecular complex. OBSOLETE since too non-specific. Consider use of cosedimentation (MI:0027) or comigration in non denaturing gel electrophoresis (MI:0404) or affinity chromatography technologies (MI:0004) or molecular sieving (MI:0071) or for unspecific cases biochemical (MI:0401)." [PMID:14755292]

is_obsolete: true

[Term]

id: MI:0026

name: correlated mutations

def: "Pairs of multiple alignments of orthologous sequences are used to identify potential interacting partners as proteins that show covariation of their residue identities between different species. Proteins displaying inter-protein correlated mutations during evolution are likely to be interacting proteins due to co-adapted evolution of their protein interacting interfaces." [PMID:11933068]

is_a: MI:0101 ! sequence based prediction

is_a: MI:0660 ! feature prediction

[Term]

id: MI:0027

name: cosedimentation

def: "Separation of a protein mixture under the influence of artificial gravity." [PMID:14755292]

is_a: MI:0401 ! biochemical

[Term]

id: MI:0028

name: cosedimentation in solution

def: "The ultracentrifuge can be used to characterise and/or purify macromolecules in solution according to their mass and hydrodynamic properties. Sedimentation studies provide information about the molecular weight and shape of a molecule. It is also possible to measure the association state of the sample. Both the mass of a molecule and its shape, that influences the friction forces and diffusion that counterbalances gravity, determine the sedimentation speed." [PMID:10410796]

exact_synonym: "solution sedimentati" []

is_a: MI:0027 ! cosedimentation

[Term]

id: MI:0029

name: cosedimentation through density gradient

def: "Sedimentation through a density gradient measures the sedimentation rate of a mixture of proteins through either a glycerol or sucrose gradient. Two interacting proteins will sediment mostly as a complex at concentrations above the binding constant. By varying the concentration of one or both of the complex constituents and taking into account the dilution of the species during sedimentation, one can reasonably accurately estimate the binding constant." [PMID:10410796]

exact_synonym: "density sedimentatio" []

is_a: MI:0027 ! cosedimentation

[Term]

id: MI:0030

name: cross-linking study

def: "Analysis of complexes obtained by chemical treatments that promote the formation of covalent bonds among molecules in close proximity." [PMID:14755292]

exact_synonym: "crosslink" []

is_a: MI:0401 ! biochemical

[Term]

id: MI:0031

name: protein cross-linking with a bifunctional reagent

def: "A cross-linker is a bifunctional molecule having two reactive ends linked by a spacer, often containing a disulfide bond. Cross-linkers induce the formation of covalent bonds among proteins that

are neighbours. When a reducing agent is added the disulfide bridge is cleaved, the cross-linked pairs are released and can be identified. There are various classes of cross-linkers, the most common are those having photoreactive groups that become reactive fluorophores when activated by UV light thereby resulting in photolabeling the cross-linked moieties." [PMID:10679368, PMID:7708014]

related_synonym: "Label transfer techniques" []

related_synonym: "Photoaffinity labelling" []

exact_synonym: "protein crosslink" []

is_a: MI:0030 ! cross-linking study

[Term]

id: MI:0032

name: de novo protein sequencing by mass spectrometry

def: "The strategy to determine the complete amino acid sequence of a protein by mass spectrometry relies on the generation of a nested set of fragments differing by one amino acid. This permits to reveal the identity of the residue that has been removed at each degradation step by measuring the mass difference of fragments differing of one residue. Peptide fragments can be obtained by protease treatment combined with the fragmentation promoted by collision (or other methods) within a tandem mass spectrometer. This approach can be carried out with LC MS/MS (Liquid Chromatography Tandem Mass Spectrometry), nanoESI MS/MS (nanoElectrospray Ionisation tandem mass spectrometry), or FTMS (Fourier Transform mass spectrometry) instruments." [PMID:10984529]

exact_synonym: "ms protein sequence" []

is_a: MI:0093 ! protein sequence identification

is_a: MI:0427 ! mass spectrometry

is_a: MI:0659 ! experimental feature detection

[Term]

id: MI:0033

name: deletion analysis

def: "In this approach, once a molecule is demonstrated to participate in an interaction, several deletion derivatives are produced and tested in the binding assay to identify the minimal fragment (domain) that can still support the interaction." [PMID:14755292]

is_a: MI:0074 ! mutation analysis

[Term]

id: MI:0034

name: display technology

def: "All the methods that permit the physical linking of a protein/peptide to its coding sequence. As a consequence affinity purification of the displayed peptide results in the genetic enrichment of its coding sequence. By these technologies genes encoding a peptide with desired binding properties can be selected over an excess of up to 10¹² unrelated molecules." [PMID:14755292]

is_a: MI:0400 ! affinity technology

[Term]

id: MI:0035

name: docking

def: "Predicts the structure of a protein-protein complex from the unbound structures of its components. The initial approach in the majority of docking procedures is based largely on the 'rigid-body' assumption, whereby the proteins are treated as solid objects. Initial scoring of a complex is based on geometric fit or surface complementarity. This generally requires some knowledge of the binding site to limit the number of solutions." [PMID:9631301, PMID for application

instance:11478868]

is_a: MI:0105 ! structure based prediction

is_a: MI:0577 ! feature prediction from structure

[Term]

id: MI:0036

name: domain fusion

def: "The rosetta stone, or domain fusion procedure, is based on the assumption that proteins whose homologues in other organisms happen to be fused into a single protein chain are likely to interact or to be functionally related." [PMID:10573422]

related_synonym: "Rosetta Stone" []

is_a: MI:0058 ! genome based prediction

is_a: MI:0101 ! sequence based prediction

[Term]

id: MI:0037

name: domain profile pairs

def: "This approach uses a protein interaction network of a given organism to infer interaction in another organism using information about the interacting region. The regions or domains involved in interactions are clustered if they share sequence similarity and have common interacting partners. The resulting domain profiles are then used to screen the proteome of another organism and domain-domain interactions are inferred. Ultimately, an inferred protein interaction map is built in this second organism." [PMID:11473021]

is_a: MI:0046 ! experimental knowledge based

is_a: MI:0101 ! sequence based prediction

is_a: MI:0660 ! feature prediction

[Term]

id: MI:0038

name: dynamic light scattering

def: "In dynamic light scattering, particle diffusion in solution gives rise to fluctuations in the intensity of the scattered light on the microsecond scale. The hydrodynamic radius of the particles can be easily calculated." [PMID:9013660]

exact_synonym: "dls" []

is_a: MI:0067 ! light scattering

[Term]

id: MI:0040

name: electron microscopy

def: "Electron microscopy methods provide insights into the structure of biological macromolecules and their supramolecular assemblies. Resolution is on average around 10 Angstroms but can reach the atomic level when the samples analysed are 2D crystals. Different types of samples can be analysed by electron microscopy: crystals, single particles like viruses, macromolecular complexes or entire cells and tissue sections. Samples can be chemically fixed or vitrified by rapid freezing in liquid ethane, and then transferred into the electron microscope. Data collection consists of the recording of electron diffraction data (2D crystals) and images. Depending on the type of sample, different approaches are used to analyse and merge images and electron diffraction data." [PMID:11785754]

related_synonym: "Electron cryomicroscopy" []

related_synonym: "Electron crystallography" []

is_a: MI:0428 ! imaging techniques

[Term]

id: MI:0041

name: electron nuclear double resonance

def: "A combination of NMR and EPR. The lines in the EPR spectrum that are caused by coupling of an unpaired electron nearby nuclei change in intensity when these nuclei are excited at their NMR frequency." [PMID:11817959, PMID:11988476, PMID for application instance:12186859]

related_synonym: "ENDOR" []

exact_synonym: "endor" []

is_a: MI:0043 ! electron resonance

[Term]

id: MI:0042

name: electron paramagnetic resonance

def: "EPR (also called ESR, Electron Spin Resonance) spectroscopy is analogous to NMR, but is based on the excitation of unpaired electrons instead of nuclei. Unpaired (single) electrons are only found in radicals and some metal ions (paramagnetic species); the EPR spectrum provides information about the environment and mobility of the paramagnetic species. The magnetic interaction of two paramagnetic centres in a protein can be used to calculate the distance between them; this allows studies of the movements and interactions of protein segments. In proteins without any intrinsic unpaired electrons it is possible to attach a radical probe (spin label). Stable nitroxide radicals can be bound to amino acid residues, in analogy with fluorescent probes. In combination with site directed mutagenesis this method is used in particular to study structure and assembly of membrane proteins, by measuring with EPR whether an amino acid is in a polar or non polar environment." [PMID:11817959]

related_synonym: "EPR" []

related_synonym: "ESR" []

exact_synonym: "epr" []

is_a: MI:0043 ! electron resonance

[Term]
id: MI:0043
name: electron resonance
def: "A form of spectroscopy in which the absorption of microwave by a sample in a strong magnetic field is used to study atoms or molecules with unpaired electrons." [PMID:14755292]
is_a: MI:0013 ! biophysical
is_a: MI:0659 ! experimental feature detection

[Term]
id: MI:0045
name: experimental interaction detection
def: "Methods based on laboratory experiments." [PMID:14755292]
exact_synonym: "experimental interac" []
is_a: MI:0001 ! interaction detection method

[Term]
id: MI:0046
name: experimental knowledge based
def: "Predictive algorithms that rely on the information obtained by experimental results." [PMID:14755292]
exact_synonym: "experimental info" []
is_a: MI:0063 ! interaction prediction

[Term]
id: MI:0047
name: far western blotting
def: "Proteins are fractionated by PAGE (SDS-polyacrylamide gel electrophoresis), transferred to a nitrocellulose membrane and tested for the ability to bind to a protein, a peptide, or any other ligand. Cell lysates can also be fractionated before gel electrophoresis to increase the sensitivity of the method for detecting interactions with rare proteins. Denaturants are removed during the blotting procedure, which allows many proteins to recover (or partially recover) activity. However, if biological activity is not recoverable, the proteins can be fractionated by a non denaturing gel system. This variation of the method eliminates the problem of activity regeneration and allows the detection of binding when the presence of a protein complex is required for binding. The protein probe can be prepared by any one of several procedures, while fusion affinity tags greatly facilitate purification. Synthesis in *E. coli* with a GST fusion, epitope tag, or other affinity tag is most commonly used. The protein of interest can then be radioactively labelled, biotinylated, or used in the blotting procedure as an unlabeled probe that is detected by a specific antibody." [PMID:7708014]
related_synonym: "Affinity blotting" []
is_a: MI:0400 ! affinity technology

[Term]
id: MI:0048
name: filamentous phage display
def: "Filamentous phages (M13, f1, fd) have been extensively used to develop and implement the technology of phage display. Repertoires of relatively short peptides of random amino acid sequences or cDNA libraries have been constructed and searched successfully. Most experiments have taken advantage of the ability to assemble phages decorated with hybrid versions of the receptor protein pIII or of the major coat protein pVIII. Both systems allow the display of foreign peptides by fusion to the amino-terminus of the capsid protein but differ in the number of peptide copies that can be displayed on each phage particle. Display libraries of very diverse protein fragments have been constructed by fusing either genomic or cDNA fragments to gene III or gene VIII." [PMID:7682645]
exact_synonym: "filamentous phage" []
is_a: MI:0084 ! phage display

[Term]
id: MI:0049
name: filter binding
def: "A method in which separation depends upon the ability of one participant to bind to a filter or membrane which the other participants do not. Molecules interacting with the bound molecule will also be retain on the filter. For example, proteins expressed by different clones of an expression library are bound to a nitrocellulose membrane, by colony (bacterial library) or plaque (phage library) blotting. A labelled protein can then be used as a probe to identify clones expressing proteins that interact with the

probe. Interactions occur on the nitrocellulose filters. The method is highly general and therefore widely applicable. A variety of approaches can be used to label the ligand, alternatively the ligand can be detected by a specific antibody. " [PMID:7708014]

related_synonym: "Filter overlay assay" []

is_a: MI:0400 ! affinity technology

[Term]

id: MI:0050

name: flag tag coimmunoprecipitation

def: "The protein of interest is expressed as a fusion to the peptide DYKDDDDKV for which antibodies are commercially available. Sometimes multiple copies of the peptide are fused in tandem.\nOBSOLETE redundant term. Map to feature type: flag-tagged (MI:0518) and Interaction detection method: anti tag coimmunoprecipitation (MI:0007)." [PMID:14755292]

exact_synonym: "flag tag coip" []

is_obsolete: true

[Term]

id: MI:0051

name: fluorescence technology

def: "Techniques based upon the measurement of the emission of one or more photons by a molecule activated by the absorption of a quantum of electro-magnetic radiation. Typically the emission, which is characterised by a wavelength that is longer than the one of excitatory radiation, occurs within 10-8 seconds." [PMID:14755292]

exact_synonym: "fluorescence" []

is_a: MI:0013 ! biophysical

[Term]

id: MI:0052

name: fluorescence correlation spectroscopy

def: "FCS monitors the random motion of fluorescently labelled molecules inside a defined volume irradiated by a focused laser beam. These fluctuations provide information on the rate of diffusion or diffusion time of a particle and this is directly dependent on the particle mass. As a consequence, any increase in the mass of a biomolecule, e.g. as a result of an interaction with a second molecule, is readily detected as an increase in the diffusion time of the particle. From these results the concentration of the different molecules can be calculated as well as their binding constant." [PMID:10733953]

related_synonym: "FCS" []

exact_synonym: "fcs" []

is_a: MI:0051 ! fluorescence technology

[Term]

id: MI:0053

name: fluorescence polarization spectroscopy

def: "Because of the long lifetimes of excited fluorescent molecules (nanoseconds), fluorescence can be used to monitor the rotational motion of molecules, which occurs on this timescale. This is accomplished experimentally by excitation with plane-polarized light, followed by measurement of the emission at parallel and perpendicular planes. Since rotational correlation times depend on the size of the molecule, this method can be used to measure the binding of two proteins because the observed polarization increase when a larger complex is formed. A fluorescence anisotropy experiment is normally carried out with a protein bearing a covalently added fluorescent group, which increases both the observed fluorescence lifetime of the excited state and the intensity of the fluorescent signal. Residue modification can be assessed by addition of an antibody which binds to the modified residue and alters the molecular weight of the complex. A variation of this technique has been used to show interaction of a DNA binding protein with another protein. In this case the DNA rather than protein is fluorescently labelled." [PMID:12805227, PMID:7708014]

related_synonym: "Fluorescence anisotropy" []

related_synonym: "FPS" []

exact_synonym: "fps" []

is_a: MI:0051 ! fluorescence technology

[Term]

id: MI:0054

name: fluorescence-activated cell sorting

def: "Cells in suspension flow through a laser beam, the scattered light or emitted fluorescence is measured, filtered and converted to digital values. Cells can be sorted according to their properties.

Using flow cytometry, any fluorescent or light scattering experiment can be carried out on entire cells. With this instrument, interactions occurring either on cell surfaces or in any other subcellular location can be studied by using suitable fluorescent labels." [PMID:11988464]

related_synonym: "FACS" []

related_synonym: "Flow cytometry" []

exact_synonym: "facs" []

is_a: MI:0051 ! fluorescence technology

[Term]

id: MI:0055

name: fluorescent resonance energy transfer

def: "FRET is a quantum mechanical process involving the radiationless transfer of energy from a donor fluorophore to an appropriately positioned acceptor fluorophore. The fluorophores are genetically fused to the protein in analysis and cotransfected. Three basic conditions must be fulfilled for FRET to occur between a donor molecule and acceptor molecule. First, the donor emission spectrum must significantly overlap the absorption spectrum of the acceptor. Second, the distance between the donor and acceptor fluorophores must fall within the range 20 to 100 Angstrom. Third, the donor and acceptor fluorophores must be in favourable orientations." [PMID:11558993]

related_synonym: "FRET" []

related_synonym: "FRET analysis" []

related_synonym: "RET" []

exact_synonym: "fret" []

is_a: MI:0051 ! fluorescence technology

[Term]

id: MI:0057

name: gene neighbourhood

def: "Gene pairs that show a conserved topological neighbourhood in many prokaryotic genomes are considered by this approach to encode interacting or functionally related proteins. By measuring the physical distance of any given gene pair in different genomes, interacting partners are inferred."

[PMID:9787636]

is_a: MI:0058 ! genome based prediction

[Term]

id: MI:0058

name: genome based prediction

def: "Methods that require fully sequenced genomes either because they are based on the comparison of genome topology or on the identification of orthologous sequences in different genomes."

[PMID:14755292]

exact_synonym: "genome prediction" []

is_a: MI:0063 ! interaction prediction

[Term]

id: MI:0063

name: interaction prediction

def: "Computational methods to predict an interaction." [PMID:14755292]

related_synonym: "in silico methods" []

exact_synonym: "predicted interac" []

is_a: MI:0001 ! interaction detection method

[Term]

id: MI:0064

name: interologs mapping

def: "Protein interactions, experimentally detected in an organism, are extended to a second organism assuming that homologous proteins, in different organisms, maintain their interaction properties."

[PMID:11731503]

related_synonym: "Homology based interaction prediction" []

is_a: MI:0046 ! experimental knowledge based

is_a: MI:0101 ! sequence based prediction

[Term]

id: MI:0065

name: isothermal titration calorimetry

def: "Isothermal titration calorimetry (ITC) measures directly the energy associated with a chemical reaction triggered by the mixing of two components. A typical ITC experiment is carried out by the stepwise addition of one of the reactants (~10⁻⁶ L per injection) into the reaction cell (~1mL) containing the second reactant. The chemical reaction occurring after each injection either releases or absorbs heat (q_i) proportional to the amount of ligand that binds to the protein with a characteristic binding enthalpy (DH). As modern ITC instruments operate on the heat compensation principle, the instrumental response (measured signal) is the amount of power (microcalories per second) necessary to maintain constant the temperature difference between the reaction and the reference cells. Because the amount of uncomplexed protein available progressively decreases after each successive injection, the magnitude of the peaks becomes progressively smaller until complete saturation is achieved. The difference between the concentration of bound ligand in the ith and (i-1)th injections depends on the binding constant K_a and the total ligand injected. The calculations depend on the binding model (number of substrates). Analysis of the data yields DH and DG = -RTlnK_a. The entropy change is obtained by using the standard thermodynamic expression DG = DH-TDS." [PMID:11785756]

related_synonym: "ITC" []

exact_synonym: "itc" []

is_a: MI:0013 ! biophysical

[Term]

id: MI:0066

name: lambda phage display

def: "Morphologically classified as one of the siphoviridae, lambda is a temperate bacteriophage of E.coli, with a double-stranded DNA genome. It has an icosahedral head attached to a flexible helical tail. Both the tail protein pV and the head protein pD have been used for displaying (C or N terminally) foreign peptides on the viral capsid." [PMID:7682645]

exact_synonym: "lambda phage" []

is_a: MI:0084 ! phage display

[Term]

id: MI:0067

name: light scattering

def: "Dynamic and static laser light scattering probes the size, shape, and structure of biological macromolecules or of their assemblies. A beam is focused on an optically clear cylindrical cell containing the sample. Most of the light passes directly through the sample. A small portion of the light is scattered; the scattered light intensity containing information about the scattering particle is detected at an angle (typically in the range 15-180degrees) from the direction of the incident beam."

[PMID:9013660]

is_a: MI:0013 ! biophysical

[Term]

id: MI:0069

name: mass spectrometry studies of complexes

def: "Mass spectrometric approaches to the study of protein in complexes permits the identification of subunit stoichiometry and transient associations. By preserving complexes intact in the mass spectrometer, mass measurement can be used for monitoring changes in different experimental conditions, or to investigate how variations of collision energy affect their dissociation. Complexes can be transferred into the gas phase by a nanoflow ESI (Electrospray Ionisation) ionisation device. This is the method of choice for the investigation of the higher-order structure of biomolecules because it allows direct analysis of dilute aqueous solutions and the desolvation process is efficient and closer to the native-like solution environment. Mass measurements of intact macromolecular complexes is largely the domain of time of-flight (TOF) MS. This is mostly due to the high sensitivity and speed of TOF analysis, as well as the virtually unlimited m/z range. Quadrupole TOF (Q-TOF) type mass spectrometers combine a quadrupole mass filter with an orthogonal TOF analyser. These spectrometers provide supplementary structural information for ions isolated in the quadrupole and analysed in the TOF. This allows complexes well in excess of 60 kDa to be dissociated and consequently their subunit composition can be determined." [PMID:12504676, PMID for application instance:12057199]

exact_synonym: "ms of complexes" []

is_a: MI:0013 ! biophysical

[Term]

id: MI:0071

name: molecular sieving

def: "In sizing columns (gel filtration), the elution position of a protein or of a complex depends on its Stokes radius. Molecules with a radius that is smaller than the bead size are retained and retarded by

the interaction with the matrix. The observation that two proteins, loaded on a sieving column, elute in a fraction(s) corresponding to a MW that is larger than the MW of either protein may be taken as an indication that the two proteins interact. Furthermore this technique provides a conceptually simple method for evaluating the affinity of the interaction." [PMID:7708014]

related_synonym: "Gel Filtration" []

related_synonym: "Siezing column" []

related_synonym: "Size Exclusion Chromatography" []

is_a: MI:0013 ! biophysical

is_a: MI:0091 ! chromatography technology

[Term]

[Term]

id: MI:0073

name: mrna display

def: "This method relies on the covalent coupling of mRNA to the nascent polypeptide. The mRNA (natural or artificial) is first covalently linked to a short DNA linker carrying a puromycin moiety. The mRNA mixture is then translated in vitro. When the ribosome reaches the RNA-DNA junction the ribosome stalls and the puromycin moiety enters the peptidyltransferase site of the ribosome and forms a covalent linkage to the nascent polypeptide. As a result the protein and the mRNA are covalently joined and can be isolated from the ribosome and purified. In the current protocol, a cDNA strand is then synthesised to form a less sticky RNA-DNA hybrid and these complexes are finally used for affinity selection. As in most display approaches, several selections cycles (3-6) are sufficient to enrich for mRNAs encoding ligand proteins." [PMID:11551470]

is_a: MI:0034 ! display technology

[Term]

id: MI:0075

name: myc tag coimmunoprecipitation

def: "The protein of interest is expressed as a fusion to the peptide EUKLISEED (a fragment of the Myc oncogene protein) for which antibodies are commercially available. Sometimes multiple copies of the peptide are fused in tandem.\nOBSOLETE redundant term. Map to feature type: myc-tagged (MI:0522) and Interaction detection method: anti tag coimmunoprecipitation (MI:0007)."

[PMID:14755292]

exact_synonym: "myc tag coip" []

is_obsolete: true

[Term]

id: MI:0077

name: nuclear magnetic resonance

def: "NMR requires a small volume of concentrated protein solution that is placed in a strong magnetic field. Certain atomic nuclei, and in particular hydrogen, have a magnetic moment or spin; that is, they have an intrinsic magnetisation, like a bar magnet. The spin aligns along the strong magnetic field, but can be changed to a misaligned excited state in response to applied radio frequency (RF) pulses of electromagnetic radiation. When the excited hydrogen nuclei relax to their aligned state, they emit RF radiation, which can be measured and displayed as a spectrum. The nature of the emitted radiation depends on the environment of each hydrogen nucleus, and if one nucleus is excited, it will influence the absorption and emission of radiation by other nuclei that lie close to it. It is consequently possible, by an ingenious elaboration of the basic NMR technique known as two-dimensional NMR, to distinguish the signals from hydrogen nuclei in different amino acid residues and to identify and measure the small shifts in these signals that occur when these hydrogen nuclei lie close enough to interact: the size of such a shift reveals the distance between the interacting pair of hydrogen atoms. In this way NMR can give information about the distances between the parts of the protein molecule. NMR provides information about interacting atoms thereby permitting to obtain information about protein structure and protein-protein interaction." [PMID:12120505, PMID for application

instance:12062432]

related_synonym: "NMR" []

exact_synonym: "nmr" []

is_a: MI:0013 ! biophysical

is_a: MI:0659 ! experimental feature detection

[Term]

id: MI:0081

name: peptide array

def: "The peptide synthesis methods offer numerous opportunities to synthesise and subsequently screen large arrays of synthetic peptides on planar cellulose supports. Discrete spots are arranged as arrays on membrane sheets where each spot is individually accessed by manual or automated delivery of the appropriate reagent solutions. Over the past few years protein-protein recognition, peptide-metal ion interactions, peptide-nucleic acid binding, enzymatic modification of peptides experiments, have been explored using synthetic peptide arrays on planar support." [PMID:11167074]
is_a: MI:0008 ! array technology

[Term]

id: MI:0084

name: phage display

def: "Peptide sequences or entire proteins can be displayed on phage capsids by fusion to coat proteins to generate a library of fusion phages each displaying a different peptide. Such a library can then be exploited to identify specific phages that display peptides that bind to any given bait molecule for instance an antibody. The selection is performed by a series of cycles of affinity purification known as panning. The bait protein, immobilized on a solid support (plastic, agarose, sepharose, magnetic beads and others) is soaked in the phage mixture and that phage that remains attached to the bait is amplified and carried through a further affinity purification step. Each cycle results in an approximately 1,000-fold enrichment of specific phage and after a few selection rounds (2-4), DNA sequencing of the tight-binding phage reveals only a small number of sequences. Phage display panning experiments can be carried out either on libraries of peptides of random amino acid sequence or on libraries of displaying natural peptides obtained by inserting cDNA fragments into the phage vector (cDNA libraries). Libraries have been assembled on several different phages (Fd, Lambda or T7)." [PMID:10975452, PMID:7708014]

is_a: MI:0034 ! display technology

[Term]

id: MI:0085

name: phylogenetic profile

def: "The phylogenetic profile of a protein stores information about the presence and the absence of that protein in a set of genomes. By clustering identical or similar profiles, proteins with similar functions and potentially interacting are identified." [PMID:10200254]

is_a: MI:0058 ! genome based prediction

[Term]

id: MI:0087

name: predictive text mining

def: "Methods based on natural language processing to detect possible interactions between proteins (direct physical interactions or indirect genetic interactions). This includes the detection of non ambiguous protein or gene names and analysis of the relation expressed in a sentence among them." [PMID:11791231]

exact_synonym: "predictive tm" []

is_a: MI:0110 ! text mining

[Term]

id: MI:0089

name: protein array

def: "The protein array technology allows the screening of biochemical activities or binding abilities of hundreds or thousands of protein samples in parallel. After synthesis and purification by high-throughput methodologies, the proteins are printed onto the chip by using an instrument (microarrayer) that is capable of spotting liquid samples in a reproducible manner onto a planar support. The ordered protein array can then be probed with labelled molecules to identify proteins that bind to the bait." [PMID:10976071, PMID:12067604]

is_a: MI:0008 ! array technology

[Term]

id: MI:0090

name: protein complementation assay

def: "The function of numerous proteins or ribonucleic particles (enzymes, transcription factors, and others) can be rationally dissected into two fragments that fold autonomously but cannot complement to reconstitute the complex function, unless they are located in close proximity. In a two hybrid experiment, restoration of the activity by complementation of the two fragments when expressed as

fusion with two polypeptides is taken as an evidence that the two polypeptides interact together."
[PMID:11495741]
related_synonym: "PCA" []
exact_synonym: "complementation" []
is_a: MI:0045 ! experimental interaction detection

[Term]
id: MI:0091
name: chromatography technology
def: "Used to separate and/or analyse complex mixtures. The components to be separated are distributed between two phases: a stationary phase (bed) and a mobile phase which percolates through the stationary bed. The nature of the two phases determines the separation criteria exploited by the column such as affinity, ionic charges, size or hydrophobicity of the molecules under analysis. Each type of column can be implemented with the mobile phase under atmospheric or high pressure condition. In this later case columns are designated as High Pressure Liquid Chromatography (HPLC)."
[PMID:14755292]
related_synonym: "column chromatography" []
exact_synonym: "chromatography" []
is_a: MI:0401 ! biochemical

[Term]
id: MI:0092
name: protein in situ array
def: "Protein In Situ Array is a method by which protein arrays are rapidly generated in one step directly from DNA, by cell-free protein expression and simultaneous in situ immobilisation at a surface. Individual genes or fragments are produce by PCR or RT-PCR depending on the source of genetic material using properly designed primers. The PISA is generated by cell-free protein synthesis using coupled transcription and translation to produce a double HexaHis-tagged protein, the reaction being carried out on a surface to which the protein adheres as soon as it is synthesised."
[PMID:11470888]
related_synonym: "PISA" []
exact_synonym: "pisa" []
is_a: MI:0008 ! array technology

[Term]
id: MI:0095
name: proteinchip(r) on a surface-enhanced laser desorption/ionization
def: "ProteinChip(r) Array technology is a surface-enhanced laser desorption/ionization (SELDI) approach (Ciphergen Biosystems Inc. Fremont, CA, USA) for sample fractionation accomplished by retentate chromatography. Retentate chromatography is performed on ProteinChip Arrays with varying chromatographic properties (e.g. anion exchange, cation exchange, metal affinity and reverse phase). By utilising arrays with differing surface chemistries in parallel and in series, a complex mixture of proteins, as from cells or body fluids, can be resolved into subsets of proteins with common properties. Specific analytes can also be examined by using preactivated arrays to which a bait molecule (such as an antibody or biotinylated DNA) is immobilized and a solution containing the binding partner(s) is presented to the array. This array-based immunoprecipitation or protein-binding experiment has been used with good success to study DNA-binding proteins, receptor-ligand interactions, and protein complexes. Any ligand retained on a SELDI chip can directly be identified by mass spectrometry."
[PMID:11827829]
related_synonym: "SELDI ProteinChip" []
exact_synonym: "seldi chip" []
is_a: MI:0008 ! array technology

[Term]
id: MI:0096
name: pull down
def: "A specific affinity chromatography method where a protein of interest (bait) is expressed as a fusion to an affinity tag (GST, HIS tag and others) and linked at high concentration to a support that has affinity for the tag. Purified proteins or cellular extracts are then adsorbed to the resin and the retained binding proteins are identified. Thus, in this approach, the protein that has affinity for the solid support (bait) is expressed and purified first, often in an heterologous system, and then challenged with a solution containing the candidate partner proteins. no antibodies to retain bait but other affinity stuffs." [PMID:14755292]
is_a: MI:0004 ! affinity chromatography technology

[Term]

id: MI:0097

name: reverse ras recruitment system

def: "In this complementation approach the bait can be any membrane protein (for example a receptor or a channel protein), the prey is cloned as a fusion protein of any cDNA from a library and the coding sequence of cytoplasmic RAS (cdc25 in yeast). If the bait and the prey interact, RAS is recruited close to the membrane and can activate cell growth. This procedure must take place in cells having a mutated RAS (Cdc25-2 yeast strain having a temperature sensitive mutation of RAS) to avoid constitutive growth activation." [PMID:11160938]

related_synonym: "reverse RRS" []

exact_synonym: "reverse rrs" []

is_a: MI:0228 ! cytoplasmic complementation assay

[Term]

id: MI:0098

name: ribosome display

def: "This method permits the coupling of phenotype to genotype via the formation of a non-covalent ternary complex between mRNAs and their encoded polypeptides while they are translated in an in vitro system. As a first step a cDNA library is constructed that encodes chimeric proteins in which the natural proteins or protein domains are fused to a C-terminal tether. As a consequence when the mRNA is translated in vitro the domain can fold while the tether is still in the ribosomal tunnel. Furthermore this chimeric mRNAs lack a stop codon, thus preventing release of the mRNA and the polypeptide from the ribosome. High concentrations of magnesium and low temperature further stabilise the ternary complex. Similarly to phage display, these complexes can be used directly to select for nucleic acids encoding proteins with desired properties." [PMID:11551470, PMID for application instance:12167034]

instance:12167034

is_a: MI:0034 ! display technology

[Term]

id: MI:0099

name: scintillation proximity assay

def: "SPA relies upon the fact that a beta particle emitted from a radioisotope decay can excite a fluorophore only when it is at a very short distance in water solution (few micrometers). The ligand is labelled with a radioactive atom and its potential partner is fixed to fluorophore containing beads, the emitted fluorescence proving their interaction can be measured in a scintillation counter. The scintillator measures only the amount of bound radiolabelled ligand. Competition experiment with cold competitor can be done to estimate the binding affinities (50% inhibitory concentration [IC50], cold ligand versus labelled ligand). Loss of signal can also be used to measure substrate cleavage by an enzyme, and labelled antibodies used to titrate the degree of modified residue present." [PMID:3866247]

related_synonym: "RIA Radio Immuno Assay" []

related_synonym: "SPA" []

exact_synonym: "spa" []

is_a: MI:0013 ! biophysical

[Term]

id: MI:0100

name: sequence based phylogenetic profile

def: "Multiple alignments of orthologous sequences in the same species and their corresponding phylogenetic trees are built. Every phylogenetic tree is computed as a matrix of distances between all possible protein pairs. The covariation of the distance matrices reveals interacting protein pairs." [PMID:11707606]

[PMID:11707606]

exact_synonym: "sequence phylogeny" []

is_a: MI:0058 ! genome based prediction

is_a: MI:0101 ! sequence based prediction

[Term]

id: MI:0101

name: sequence based prediction

def: "Computational methods based on evolutionary hypothesis, used as criteria to browse sequences and predict interacting pairs." [PMID:14755292]

exact_synonym: "predict from sequenc" []

is_a: MI:0063 ! interaction prediction

[Term]
id: MI:0103
name: southern blot
def: "A standard procedure to identify DNA fragments containing specific gene sequences. In this procedure i) a genome is fragmented using a restriction enzyme ii) the generated fragments are separated by electrophoresis iii) the fragments are transferred to a membrane iv) the membrane is incubated with a radio labelled probe that hybridises any complementary subsequence."
[PMID:14755292]
is_a: MI:0080 ! partial dna sequence identification by hybridization

[Term]
id: MI:0104
name: static light scattering
def: "In static light scattering, the average intensity of scattered light at multiple angles is measured. The data yield information on particle molecular weight, particle size and shape, and particle-particle interactions." [PMID:9013660]
exact_synonym: "sls" []
is_a: MI:0067 ! light scattering

[Term]
id: MI:0105
name: structure based prediction
def: "Methods based on 3D structure information." [PMID:14755292]
exact_synonym: "predict from struct" []
is_a: MI:0063 ! interaction prediction

[Term]
id: MI:0107
name: surface plasmon resonance
def: "This method measures formation of complex by monitoring changes in the resonance angle of light impinging on a gold surface as a result of changes in the refractive index of the surface. A ligand of interest (peptide or protein) is immobilized on a dextran polymer, and a solution of interacting protein is passed over it through a cell, with a gold wall coated with this polymer. Macromolecules that interact with the immobilized ligand are retained on the polymer surface, and alter the resonance angle of impinging light as a result of the change in refractive index brought about by the increased protein mass retained on the polymer surface. Since all proteins have the same refractive index and since there is a linear correlation between resonance angle shift and protein concentration near the surface, this allows one to measure changes in protein concentration at the surface as a consequence of protein interaction. Furthermore, this can be done in real time, allowing direct measurement of both the on rate and the off rate of complex formation." [PMID:11896282, PMID:12120258, PMID:16338355]
related_synonym: "BIAcore(r)" []
related_synonym: "Optical biosensor" []
exact_synonym: "spr" []
is_a: MI:0013 ! biophysical

[Term]
id: MI:0108
name: t7 phage display
def: "T7 is a double stranded DNA bacteriophage with a thin-walled icosahedral capsid, ~550 Angstrom in diameter, which is decorated by 415 copies of the capsid protein, the product of gene 10. gp10 can tolerate insertions at the carboxyterminus without losing its ability to be inserted into functional phage capsids. Both low density and high density display (albeit only with short peptides) can be achieved." [PMID:14755292]
comment: Reference not index in medline : Rosenberg, A., Griffin, K., Studier, W.S., McCormick, M., Berg, J., Novy, R., Mierendorf, R. in Novations, 1996, 6, 1.
exact_synonym: "t7 phage" []
is_a: MI:0084 ! phage display

[Term]
id: MI:0109
name: tap tag coimmunoprecipitation
def: "The TAP method involves the fusion of the TAP tag (encoding a calmodulin binding peptide, a TEV cleavage site, and the Staphylococcus aureus Protein A) to the target protein and the introduction

of the construct into the host cell or organism, maintaining the expression of the fusion protein at, or close to, its natural level. The fusion protein and associated components are recovered from cell extracts by affinity selection on an IgG matrix. After washing, the TEV protease is added to release the bound material. The eluate is incubated with calmodulin-coated beads in the presence of calcium. This second affinity step is required to remove the TEV protease as well as traces of contaminants remaining after the first affinity selection. After washing, the bound material is released with EGTA. This two steps purification steps ensures a highly selective complex purification of the tapped protein (first round of selection on the protein A, a high affinity tag) under mild condition (non denaturant pH or conditions required to remove the tag).\nOBSOLETE redundant term. Map to feature type: tap tagged (MI:0524) and as interaction detection method tandem affinity purification (MI:0676)."
[PMID:10504710]
exact_synonym: "tap tag coip" []
is_obsolete: true

[Term]
id: MI:0110
name: text mining
def: "Text mining methods can be used to predict or confirm interactions by automated processing of scientific literature. Co-occurrence in the same sentence of an abstract of gene products labels are analysed to evaluate whether it represents a valid evidence of an interaction." [PMID:14755292]
is_a: MI:0046 ! experimental knowledge based

[Term]
id: MI:0111
name: dihydrofolate reductase reconstruction
def: "The gene for DHFR is rationally dissected into two fragments called F[1,2] and F[3]. Two proteins or protein domains that are thought to bind to each other can then be fused to either of the two DHFR fragments. Reconstitution of enzyme activity can be monitored in vivo by cell survival in DHFR-negative cells grown in the absence of nucleotides. A fluorescence assay can also be carried out taking advantage of fMTX binding to reconstituted DHFR. The basis of this assay is that complementary fragments of DHFR, when expressed and reassembled in cells, will bind with high affinity (Kd 5 540 pM) to fMTX in a 1:1 complex. fMTX is retained in cells by this complex, whereas the unbound fMTX is actively and rapidly transported out of the cells. Survival depends only on the number of molecules of DHFR reassembled." [PMID:10318894]
exact_synonym: "dhfr reconstruction" []
is_a: MI:0228 ! cytoplasmic complementation assay

[Term]
id: MI:0112
name: ubiquitin reconstruction
def: "In this method the two proteins, whose interaction is under investigation, (in this case mostly membrane proteins) are fused to an terminal fragment (Nub) and to a C-terminal fragment of ubiquitin (Cub). The two fragments do not associate to form a functional ubiquitin unless the two fused membrane proteins form a complex. The association is monitored by an ingenious trick. The C-term fragment of ubiquitin is expressed as a fusion to a transcription factor that being linked to a membrane protein cannot perform its function unless it is released form the ubiquitin fusion by a specific protease. This achieved through the activity of UBP (an ubiquitin specific protease) that cleaves off the reporter protein only when a functional ubiquitin is reconstituted." [PMID:9560251]
exact_synonym: "ub reconstruction" []
is_a: MI:0228 ! cytoplasmic complementation assay

[Term]
id: MI:0114
name: x-ray crystallography
def: "Analysis of a diffraction pattern generated by a single crystal. X-rays have a wavelength, typically around 1 Angstrom (the diameter of a hydrogen atom). If a narrow parallel beam of X-rays is directed at a sample of a pure protein, most of the X-rays will pass straight through it. A small fraction, however, will be scattered by the atoms in the sample. If the sample is a well-ordered crystal, the scattered waves will reinforce one another at certain points and will appear as diffraction spots when the X-rays are recorded by a suitable detector. The position and intensity of each spot in the X-ray diffraction pattern contain information about the position and nature of the atoms in the crystal. The three-dimensional structure of a large molecule can be deduced from the electron-density map of its crystal. In recent years X-ray diffraction analysis has become increasingly automated, and now the slowest step is likely to be the production of suitable protein crystals. This requires high concentration

of very pure protein and empirical searching for the proper crystallisation conditions."

[PMID:14755292]

related_synonym: "X-ray" []

exact_synonym: "x-ray diffraction" []

is_a: MI:0013 ! biophysical

is_a: MI:0659 ! experimental feature detection

[Term]

id: MI:0115

name: yeast display

def: "The proteins are displayed on the surface of the yeast *S. cerevisiae* by fusion to signal sequences for protein secretion. This method is limited by the low efficiency of the yeast display system but can take full advantage of exploiting cell sorting methods (FACS) to isolate cells that display molecules with desired binding properties." [PMID:9181578]

is_a: MI:0034 ! display technology

is_a: MI:0054 ! fluorescence-activated cell sorting

[Term]

id: MI:0225

name: chromatin immunoprecipitation array

def: "The method combines a modified chromatin immunoprecipitation (ChIP) procedure, with DNA microarray analysis. Cells are fixed with formaldehyde, harvested, and disrupted by sonication. The DNA fragments cross-linked to a protein of interest are enriched by immunoprecipitation with a specific antibody. After reversal of the cross-links, the enriched DNA is amplified and labeled with a fluorescent dye (Cy5) by using a ligation-mediated polymerase chain reaction (LM-PCR). In parallel a sample of DNA that is not enriched by immunoprecipitation is subjected to LM-PCR in the presence of a different fluorophore (Cy3), and both immunoprecipitation (IP)-enriched and unenriched pools of labeled DNA were hybridized to a single DNA microarray containing a set of intergenic sequences. The ratio of the Cy5 to Cy3 fluorescence intensities measured at each DNA element in the microarray provided a measure of the extent of binding of the transcription factor to the corresponding genomic locus." [PMID:11125145, PMID:11206552]

exact_synonym: "chip-chip" []

is_a: MI:0008 ! array technology

is_a: MI:0402 ! chromatin immunoprecipitation assays

[Term]

id: MI:0226

name: ion exchange chromatography

def: "Stable complexes and their component proteins can be separated on the basis of their net charge by ion-exchange chromatography. If a protein has a net positive charge at pH 7, it will usually bind to a column of beads containing carboxylate groups, and can then be eluted by increasing the concentration of sodium chloride or another salt in the eluting buffer by competition of sodium ions with positively charged groups on the protein for binding to the column. Protein that have a low density of net positive charge will tend to emerge first, followed by those having a higher charge density. Positively charged complexes or proteins (cationic proteins) can be separated on negatively charged carboxymethyl-cellulose (CM-cellulose) columns. Conversely, negatively charged complexes or proteins (anionic proteins) can be separated by chromatography on positively charged diethylaminoethyl-cellulose (DEAE-cellulose) columns." [PMID:14755292]

related_synonym: "IEC" []

exact_synonym: "ion exchange chrom" []

is_a: MI:0091 ! chromatography technology

[Term]

id: MI:0227

name: reverse phase chromatography

def: "Reverse phase chromatography operates on the basis of hydrophilicity and lipophilicity. The stationary phase consists of silica based packings with n-alkyl chains covalently bound. For example, C-8 signifies an octyl chain and C-18 an octadecyl ligand in the matrix. The more hydrophobic the matrix on each ligand, the greater is the tendency of the column to retain hydrophobic moieties. Thus hydrophilic compounds elute more quickly than do hydrophobic compounds." [PMID:14755292]

exact_synonym: "reverse phase chrom" []

is_a: MI:0091 ! chromatography technology

[Term]

id: MI:0228
name: cytoplasmic complementation assay
def: "Protein complementation assay performed by dissecting a cytoplasmic protein activity and restoring it through the two hybrid proteins interaction." [PMID:14755292]
exact_synonym: "cytoplasmic compl" []
is_a: MI:0090 ! protein complementation assay

[Term]
id: MI:0229
name: green fluorescence protein complementation assay
def: "Protein-protein interaction trap based on fusions of bait and prey protein to two dissected fragment of GFP. The system implemented in E. coli, allow screening of partners, including membrane proteins and also detect transient interaction that are stabilized by the complemented GFP." [PMID:15631464]
exact_synonym: "gfp complementation" []
is_a: MI:0228 ! cytoplasmic complementation assay

[Term]
id: MI:0230
name: membrane bound complementation assay
def: "Protein complementation assay based on dissection of a membrane protein." [PMID:14755292]
exact_synonym: "membrane compl" []
is_a: MI:0090 ! protein complementation assay

[Term]
id: MI:0231
name: mammalian protein protein interaction trap
def: "The MAPPIT(mammalian protein-protein interaction Trap) is a screening method for protein-protein interaction in mammalian cells, based on the reconstitution of a membrane STAT (signal transducers and activators of transcription) receptor. The bait protein is fused to a STAT recruitment-deficient receptor and the prey protein to a functional STAT recruitment sites. In such a configuration, a given baitprey interaction restores a STAT-dependent responses leading to the expression of a reporter gene. This system, enable to demonstrate not only protein interaction but also modification-independent and tyrosine phosphorylation- dependent interactions." [PMID:12853652]
exact_synonym: "mappit" []
is_a: MI:0230 ! membrane bound complementation assay

[Term]
id: MI:0232
name: transcriptional complementation assay
def: "Protein complementation assay performed by dissecting a transcription factor activity (DNA binding domain and transcription activation domain) its restoration through the two hybrid proteins interaction that lead to a reporter gene expression." [PMID:14755292]
exact_synonym: "transcription compl" []
is_a: MI:0090 ! protein complementation assay

[Term]
id: MI:0254
name: genetic interference
def: "This term refers to methods that aim at interfering with the activity of a specific gene by altering the gene regulatory or coding sequences. This goal can be achieved either by a classical genetic approach (random mutagenesis followed by phenotype characterization and genetic mapping) or by a reverse genetics approach where a gene of interest is modified by directed mutagenesis." [PMID:14755292]
is_a: MI:0045 ! experimental interaction detection

[Term]
id: MI:0255
name: post transcriptional interference
def: "This term refers to methods designed to interfere with gene expression at post-transcriptional level rather than with the gene itself." [PMID:14755292]
exact_synonym: "expression interfer" []
is_a: MI:0045 ! experimental interaction detection

[Term]

id: MI:0256

name: rna interference

def: "RNA interference (RNAi) is a post-transcriptional gene silencing method reproducing a naturally occurring phenomena. RNAi is the process whereby double-stranded RNA (dsRNA) induces the sequence-specific degradation of homologous mRNA. RNAi or dsRNA-induced silencing phenomena are present in evolutionarily diverse organisms, e.g., nematodes, plants, fungi, and trypanosomes. The mechanisms by which RNAi works is initiated by a progressive cleavage of dsRNA into 21 to 23 nucleotide (nt) short interfering RNAs (siRNAs). These native siRNA duplexes are then incorporated into a protein complex called RNA-induced silencing complex (RISC). ATP-dependent unwinding of the siRNA duplex generates an active RISC complex. Guided by the antisense strand of siRNA, the active RISC complex recognizes and cleaves the corresponding mRNA." [PMID:12110901, PMID:12408823]

exact_synonym: "rnai" []

is_a: MI:0255 ! post transcriptional interference

[Term]

id: MI:0257

name: antisense rna

def: "This approach is based on the observation that expression of RNA that is complementary to a specific mRNA can decrease the synthesis of its gene product either by increasing the degradation of the targeted mRNA or by interfering with its translation." [PMID:1340158]

is_a: MI:0255 ! post transcriptional interference

[Term]

id: MI:0276

name: blue native page

def: "Blue native PAGE (BN-PAGE) permits a high-resolution separation of multi-protein complexes under native conditions. Blue native (BN)-PAGE is a charge shift method, in which the electrophoretic mobility of a complex is determined by the negative charge of the bound Coomassie dye and the size and shape of the complex. Coomassie does not act as a detergent and preserves the structure of complexes. Importantly, the resolution of BN-PAGE is much higher than that of other methods such as gel filtration or sucrose-gradient ultracentrifugation. Combined with other pre-purifications or dialysis steps this method permits the analysis of multi-protein complexes of whole cellular lysates by BN-PAGE." [PMID:14665681]

exact_synonym: "bn-page" []

is_a: MI:0404 ! comigration in non denaturing gel electrophoresis

[Term]

id: MI:0369

name: lex-a dimerization assay

def: "The method is based on the repression of a reporter gene activity by two LexA DNA binding domains with different binding specificities. LexA is a transcription factor with an N-terminal DNA binding/activation domain (DBAct) and a C-terminal dimerization domain. LexA dimerization is required to repress transcription efficiently. The discovery of LexA DNA binding domains that bind to different DNA sequence enabled the development of this system." [PMID:12446730]

related_synonym: "gallex" []

exact_synonym: "gallex" []

is_a: MI:0232 ! transcriptional complementation assay

[Term]

id: MI:0370

name: tox-r dimerization assay

def: "This assay allow identification of interactions in the inner membrane of E. coli. by using a chimeric construct ToxR-TM-MBP composed of the N-terminal DNA binding/transcriptional activation domain of ToxR (a dimerization dependant transcription factor) fused to a transmembrane domain of interest (TM) and a monomeric periplasmic anchor (the maltose binding protein). Association of the two TM results in the ToxR-mediated activation of a reporter gene such as CAT (chloroamphenicol acetyltransferase activity). The level of CAT expression indicates the strength of TM association. CAT expression can then be tested and quantify by measuring CAM resistance with disk diffusion assay or CAT activity assays on cell-free extracts." [PMID:9927659]

related_synonym: "toxcat" []

exact_synonym: "toxcat" []

is_a: MI:0230 ! membrane bound complementation assay

[Term]

id: MI:0397

name: two hybrid array

def: "Two-hybrid screening can be done in a colony array format, in which each colony expresses a defined pair of proteins. Because the particular protein pair expressed in each colony is defined by its position in the array, positive signals identify interacting proteins without further characterization, thus obviating the need for DNA purification and sequencing. The interrogation of a two-hybrid colony array usually involves a mating strategy in which every DNA binding domain hybrid (the bait) is tested against all activation domain hybrids (the preys) in a grid pattern. Arrays usually use full-length open reading frames." [PMID:11827624, PMID for application instance:10688190]

is_a: MI:0018 ! two hybrid

[Term]

id: MI:0398

name: two hybrid pooling approach

def: "In the pooling strategy the sets of bait and prey hybrid vectors are randomly mated. The positives double hybrid clones are sequenced to identify the interacting partners." [PMID:12634794, PMID for application instance:11283351]

exact_synonym: "two hybrid pooling" []

is_a: MI:0018 ! two hybrid

[Term]

id: MI:0399

name: two hybrid fragment pooling approach

def: "This two hybrid approach involves the screening of a large number of individual proteins against a comprehensive library of randomly generated fragment as prey. The usage of degenerated fragment allows identification of the minimal protein region required for the interaction. since multiple clones that encode overlapping regions of protein are often identified, the minimal domain for interaction may be readily apparent from the initial screen." [PMID:12634794, PMID for application instance:11196647]

exact_synonym: "2h fragment pooling" []

is_a: MI:0398 ! two hybrid pooling approach

[Term]

id: MI:0400

name: affinity technology

def: "Techniques which depend upon the strength of the interaction between two entities."

[PMID:14755292]

exact_synonym: "affinity techniques" []

is_a: MI:0401 ! biochemical

[Term]

id: MI:0401

name: biochemical

def: "The application of chemical principles and methods to biological experiments."

[PMID:14755292]

is_a: MI:0045 ! experimental interaction detection

[Term]

id: MI:0402

name: chromatin immunoprecipitation assays

def: "Chromatin immunoprecipitation (ChIP) is a powerful approach that allows one to define the interaction of factors with specific chromosomal sites in living cells. ChIP involves treating cells or tissue briefly with formaldehyde to crosslink proteins to DNA. An antibody against a protein suspected of binding a given cis-element is then used to immunoprecipitate chromatin fragments. Polymerase chain reaction analysis of the immunoprecipitate with primers flanking the cis-element reveals whether a specific DNA sequence is recovered in an immune-specific manner and therefore whether the protein contacted the site in living cells." [PMID:12054902]

exact_synonym: "ch-ip" []

is_a: MI:0400 ! affinity technology

[Term]

id: MI:0404
name: comigration in non denaturing gel electrophoresis
def: "comigration by non denaturing gel electrophoresis." [PMID:14755292]
exact_synonym: "comig non denat gel" []
is_a: MI:0807 ! comigration in gel electrophoresis

[Term]
id: MI:0405
name: competition binding
def: "Competitive binding experiments measure equilibrium binding of a single concentration of ligand at various concentrations of an unlabeled competitor. Analysis of these data gives the affinity of the receptor for the competitor." [PMID:14755292]
is_a: MI:0400 ! affinity technology

[Term]
id: MI:0406
name: deacetylase assay
def: "Measures the catalysis of the hydrolysis of an acetyl group or groups from a substrate molecule." [PMID:14755292]
is_a: MI:0415 ! enzymatic study

[Term]
id: MI:0409
name: dna footprinting
def: "Experimental method used to identify the region of a nucleic acid involved in an interaction with a protein. One sample of a radiolabeled nucleic acid of known sequence is submitted to partial digestion. A second sample is incubated with its interacting partner and then is submitted to the same partial digestion. The two samples are then analyzed in parallel by electrophoresis on a denaturing acrylamide gel. After autoradiography the identification of the bands that correspond to fragments missing from the lane loaded with the second sample reveals the region of the nucleic acid that is protected from nuclease digestion upon binding." [PMID:14755292]
is_a: MI:0417 ! footprinting

[Term]
id: MI:0410
name: electron tomography
def: "Electron tomography is a general method for three-dimensional (3D) reconstruction of single, transparent objects from a series of projection images (i.e. from a tilt series) recorded with a transmission electron microscope. It offers the opportunity to obtain 3D information on structural cellular arrangements with a high resolution." [PMID:12160704]
exact_synonym: "sem" []
is_a: MI:0040 ! electron microscopy

[Term]
id: MI:0411
name: enzyme linked immunosorbent assay
def: "Following non-covalent binding of a purified primary ligand to a solid phase, a blocking reagent is added to prevent any non-specific binding. A specific antigen is then allowed to bind to the primary ligand. Unbound antigen is removed by washing and a secondary antibody conjugated to an enzyme (e.g. horseradish peroxidase) is added. Following a washing step to remove unbound secondary ligand, the extent to which a chromogenic substrate (e.g. 3,3', 5,5' tetramethyl benzidine chromogen [TMB]) is converted to a soluble coloured product by the conjugated enzyme in a given time is determined by spectrophotometry using a standard microplate absorbance reader. A similar type of approach can be utilized to detect enzymatic activities. The substrate, attached to a solid phase is incubated in the presence of the enzyme and the enzymatic modification is monitored by an antibody that is specific for the modified substrate (for instance a phosphorylated protein)." [PMID:11906746]
related_synonym: "ELISA" []
exact_synonym: "elisa" []
is_a: MI:0400 ! affinity technology
is_a: MI:0421 ! identification by antibody

[Term]
id: MI:0412
name: electrophoretic mobility supershift assay

def: "The EMSA supershift is a EMSA experiment carried out using a third lane loaded with the radiolabeled nucleic acid, a protein mixture and an antibody for a specific protein. If an extra retardation is observed, this is due to the formation of a larger complex including the antibody. By this approach, at least one protein of the complex is directly identified." [PMID:12169687]
exact_synonym: "emsa supershift" []
is_a: MI:0413 ! electrophoretic mobility shift assay

[Term]
id: MI:0413
name: electrophoretic mobility shift assay
def: "This method proves the interaction between a nucleic acid and a protein partner. On the same electrophoresis gel 1 lane is loaded with a radiolabeled nucleic acid of known sequence, a second lane is loaded with the same nucleic acid together with a purified protein (or a protein mixture). After the electrophoresis run and autoradiography by comparing the nucleic acid migration in the two lanes the retardation of the nucleic acid due to its interaction with a protein can be easily observed." [PMID:12169687]
related_synonym: "band shift" []
related_synonym: "Gel retardation assay" []
exact_synonym: "emsa" []
is_a: MI:0400 ! affinity technology

[Term]
id: MI:0415
name: enzymatic study
def: "Participants are enzyme or substrate in a biochemical reaction." [PMID:14755292]
is_a: MI:0401 ! biochemical

[Term]
id: MI:0416
name: fluorescence microscopy
def: "Fluorescent microscopy uses a high intensity light to illuminate the sample. This light excites fluorescence species in the sample, which then emit light of a longer wavelength. A fluorescent microscope also produces a magnified image of the sample, but the image is based on the second light source -- the light emanating from the fluorescent species -- rather than from the light originally used to illuminate, and excite, the sample." [PMID:14755292]
exact_synonym: "fluorescence imaging" []
is_a: MI:0428 ! imaging techniques

[Term]
id: MI:0417
name: footprinting
def: "footprinting analysis is used to identify regions of molecules directly involved in binding other macromolecules and therefore protected from the effects of degradative enzymes." [PMID:14755292]
is_a: MI:0401 ! biochemical

[Term]
id: MI:0419
name: gtpase assay
def: "Measures the catalysis of the reaction: $GTP + H_2O = GDP + phosphate$." [PMID:14755292]
related_synonym: "gtp hydrolysis" []
related_synonym: "GTPase" []
is_a: MI:0879 ! nucleoside triphosphatase assay

[Term]
id: MI:0420
name: kinase homogeneous time resolved fluorescence
def: "Measures quenching of the nonradiative energy transfer between fluorescent long-lifetime lanthanide chelates and different acceptors. Relies on a fluorescence energy donor and acceptor being added from close proximity on the phosphorylated substrate due to the action of the kinase." [PMID:14987100]
related_synonym: "homogeneous time-resolved fluorescence" []
related_synonym: "kinase HTRF" []
exact_synonym: "kinase htrf" []
is_a: MI:0424 ! protein kinase assay

is_a: MI:0510 ! homogeneous time resolved fluorescence

[Term]

id: MI:0423

name: in-gel kinase assay

def: "Substrate protein radio-labelled by kinase transferring an isotope of phosphate from the nucleotide. Substrate isolated by gel electrophoresis and radio-labelling confirmed by autoradiography." [PMID:14755292]

exact_synonym: "in gel kinase assay" []

is_a: MI:0424 ! protein kinase assay

[Term]

id: MI:0424

name: protein kinase assay

def: "Catalysis of the transfer of a phosphate group, usually from ATP, to a protein substrate." [PMID:14755292]

is_a: MI:0415 ! enzymatic study

[Term]

id: MI:0425

name: kinase scintillation proximity assay

def: "Relies on the radiolabelling of a peptide substrate immobilized on a scintillant coated SPA-bead. The kinase transfers a phosphate isotope from the nucleotide to the substrate." [PMID:14755292]

exact_synonym: "kinase spa" []

is_a: MI:0099 ! scintillation proximity assay

is_a: MI:0424 ! protein kinase assay

[Term]

id: MI:0426

name: light microscopy

def: "Light visible microscopy uses environmental light to illuminate the sample and produce a magnified image of the sample." [PMID:14755292]

is_a: MI:0428 ! imaging techniques

[Term]

id: MI:0428

name: imaging techniques

def: "Methods that provide images of molecules at various resolution depending on the technology used." [PMID:14755292]

related_synonym: "microscopy" []

is_a: MI:0045 ! experimental interaction detection

[Term]

id: MI:0430

name: nucleic acid uv cross-linking assay

def: "Radiolabelled nucleic acids with a known sequence are incubated with purified proteins or reproducible protein mixture (HPLC eluate fractions) and then irradiated with UV. The complex are treated with an enzyme to remove the unbound nucleic acid (Rnase A or Dnase I for example), washed and then analyzed by electrophoresis. The eventual complexes are identified by autoradiography. The proteins involved in the complex can be recognized by specific antibodies or by retrieving the original protein mixture and carrying further analysis on it. The affinity of the binding can be estimated by adding a non-labelled nucleic acid as competitor before the UV irradiation." [PMID:14755292]

exact_synonym: "nucl ac uv crosslink" []

is_a: MI:0030 ! cross-linking study

[Term]

id: MI:0432

name: one hybrid

def: "Protein-DNA complementation assay where a single promoter act as bait and is screened against a library of prey transcription factors." [PMID:10589421]

related_synonym: "one-hybrid" []

related_synonym: "yeast one hybrid" []

related_synonym: "yeast one-hybrid" []

is_a: MI:0232 ! transcriptional complementation assay

[Term]
id: MI:0434
name: phosphatase assay
def: "Measures the catalysis of the reaction: a phosphoprotein + H₂O = a protein + phosphate."
[PMID:14755292]
is_a: MI:0415 ! enzymatic study

[Term]
id: MI:0435
name: protease assay
def: "Measures the enzymatic hydrolysis of a peptide bond within a peptide or protein substrate."
[PMID:14755292]
is_a: MI:0415 ! enzymatic study

[Term]
id: MI:0437
name: protein tri hybrid
def: "Two hybrid assay performed with a third protein component is co-transfected into a recombinant yeast strain together with a bait and a prey construct. Negative control show that the interaction between the bait and the prey do not occur when the third protein is not co-transfected."
[PMID:12052864, PMID:12761205, PMID:12935900]
related_synonym: "bridge assay" []
related_synonym: "trihybrid" []
is_a: MI:0018 ! two hybrid
is_a: MI:0588 ! 3 hybrid method

[Term]
id: MI:0438
name: rna tri hybrid
def: "In vivo reconstruction of specific RNA-proteins interactions. The DNA binding and transcription activator domains of GAL4 are brought together via the interaction of recombinant RNA. The first hybrid protein contains the DNA binding domain of GAL4 fused to RevM10 (a mutated RNA binding protein of HIV-1 that binds specifically to the Rev responsive element RRE of the env gene). A recombinant RNA contains the RRE sequence and a target RNA sequence X. The second hybrid protein contains the activation domain of GAL4 fused to protein Y tested for its ability to bind the target RNA X. If this interaction occurs the three hybrid reconstructs GAL4 and the transcription of a reporter gene is activated." [PMID:12162957, PMID:8972875]
related_synonym: "Three hybrid system" []
is_a: MI:0232 ! transcriptional complementation assay
is_a: MI:0588 ! 3 hybrid method

[Term]
id: MI:0439
name: random spore analysis
def: "A technique used to detect genetic interactions between 2 (or more) genes in a sporulating organism by scoring a large population of haploid spores for a phenotype and correlating the phenotype with the presence of single vs double (multiple) mutations. A diploid heterozygous organism harbouring mutations in two (or more) genes is induced to sporulate. Resulting spores are meiotic segregants that are haploid and are either wild type or mutant at each locus. Spores are scored for a phenotype, such as loss of viability." [PMID:14755292]
related_synonym: "random-spore analysis" []
related_synonym: "RSA" []
related_synonym: "spore germination" []
exact_synonym: "rsa" []
is_a: MI:0254 ! genetic interference

[Term]
id: MI:0440
name: saturation binding
def: "Saturation binding experiments measure specific ligand binding at equilibrium at various concentrations of the ligand. Analysis of these data can determine receptor number and affinity."
[PMID:14755292]
is_a: MI:0400 ! affinity technology

[Term]
id: MI:0441
name: synthetic genetic analysis
def: "Identification of genetic interactions by generation of an organism harbouring mutations in 2 or more genes and scoring for a phenotype, such as loss of viability, that is not observed for any of the mutations in isolation." [PMID:14755292]
related_synonym: "SGA" []
exact_synonym: "sga" []
is_a: MI:0254 ! genetic interference

[Term]
id: MI:0508
name: deacetylase radiometric assay
def: "Measures the release of radiolabelled acetic acid from pre-labeled histone." [PMID:14755292]
exact_synonym: "radiolabeled acetate" []
is_a: MI:0406 ! deacetylase assay

[Term]
id: MI:0509
name: phosphatase homogeneous time resolved fluorescence
def: "Measures quenching of the nonradiative energy transfer between fluorescent long-lifetime lanthanide chelates and different acceptors. Relies on a fluorescence energy donor and acceptor being removed from close proximity on the phosphorylated substrate due to the action of the phosphatase." [PMID:14987100]
related_synonym: "homogeneous time-resolved fluorescence" []
related_synonym: "phosphatase HTRF" []
exact_synonym: "phosphatase htrf" []
is_a: MI:0434 ! phosphatase assay
is_a: MI:0510 ! homogeneous time resolved fluorescence

[Term]
id: MI:0510
name: homogeneous time resolved fluorescence
def: "Methods based on the exceptionally long fluorescence lifetime characteristics of certain fluorophores, which allows the elimination of the effects of background fluorescence. Uses nonradiative energy transfer or quenching between fluorescent lanthanide chelates and different acceptors to measure reaction rates." [PMID:14987100]
related_synonym: "homogeneous time-resolved fluorescence" []
exact_synonym: "htrf" []
is_a: MI:0051 ! fluorescence technology

[Term]
id: MI:0511
name: protease homogeneous time resolved fluorescence
def: "Measures quenching of the nonradiative energy transfer between fluorescent long-lifetime lanthanide chelates and different acceptors. Fluorescence donor and acceptor are on the same peptide molecule and separated by the action of the protease." [PMID:14987100]
related_synonym: "Protease HTRF" []
exact_synonym: "protease htrf" []
is_a: MI:0435 ! protease assay
is_a: MI:0510 ! homogeneous time resolved fluorescence

[Term]
id: MI:0512
name: zymography
def: "Samples run on a gelatine containing gels under non-reducing condition, gels then incubated under conditions in which the enzyme is active. Gels are stained with coomasie and gelatine-free regions of the gel taken as a measure of enzyme activity." [PMID:2071592]
is_a: MI:0435 ! protease assay

[Term]
id: MI:0513
name: collagen film assay

def: "Measures the amount of radiolabel released into the medium when enzyme is added onto a film of isotope-labelled collagen." [PMID:6247938]
is_a: MI:0435 ! protease assay

[Term]
id: MI:0514
name: in gel phosphatase assay
def: "Substrate protein pre-radiolabelled either synthetically or through the action of a kinase transferring an isotope of phosphate from a nucleotide. Substrate then exposed to phosphate under assay conditions. Substrate isolated by gel electrophoresis and loss of radiolabelling confirmed by autoradiography." [PMID:14755292]
exact_synonym: "in gel phosphatase" []
is_a: MI:0434 ! phosphatase assay

[Term]
id: MI:0515
name: methyltransferase assay
def: "Measures the catalysis of the transfer of a methyl group to an acceptor molecule." [PMID:14755292]
exact_synonym: "methyltransferase as" []
is_a: MI:0415 ! enzymatic study

[Term]
id: MI:0516
name: methyltransferase radiometric assay
def: "Measures the transfer of a radiolabelled methyl group of a donor, for example S-adenosyl-L-methionine (SAM) to a carboxyl group of an acceptor." [PMID:14755292]
exact_synonym: "radiolabeled methyl" []
is_a: MI:0515 ! methyltransferase assay

[Term]
id: MI:0588
name: 3 hybrid method
def: "Group of method based on complementation assay where a third participant is shown to be necessary for the binding of a given bait prey pair." [PMID:14755292]
is_a: MI:0090 ! protein complementation assay

[Term]
id: MI:0602
name: chemical footprinting
def: "Sites of sequence-specific DNA-protein interaction are identified by altered reactivity of a chemical probe to DNA bound by a protein compared to the same nucleotide sequence in naked DNA. Nucleotides in close contact with the binding protein are protected from modification or cleavage by the probe. In certain case DNA-protein interactions can be indicated by enhanced modification or cleavage by the probe at particular nucleotides. When these probes are administered to intact cells, the pattern of protection from the probes identifies the location of DNA-protein interactions in vivo." [PMID:8238889]
exact_synonym: "chemical footprint" []
is_a: MI:0409 ! dna footprinting

[Term]
id: MI:0603
name: dimethylsulphate footprinting
def: "Dimethylsulphate (DMS) is the most commonly used chemical to study DNA-protein interactions. DMS induces methylation of guanine residues so DNA interaction with protein binding to AT rich sequences or to the phosphate backbone may be not detected by DMS footprinting. However as DMS diffuses across membrane it can also be used for in vivo footprinting. The experiment involves the treatment with DMS of two DNA samples with identical sequence, one protein bound and the other naked. The two samples are treated with piperidine to induce chemical cleavage of the DMS modified guanine residues followed by digestion with restriction enzymes. Once labelled the samples are run in parallel on a gel to visualize the pattern of nested fragments sharing a common end generated by restriction enzyme(or PCR primer extension) and a variable end guanine dependent. The missing bands of the protein bound sample correspond to the guanine residues protected from modification by an interaction." [PMID:8238889]

exact_synonym: "dms footprinting" []
is_a: MI:0602 ! chemical footprinting

[Term]
id: MI:0604
name: potassium permanganate footprinting
def: "Potassium permanganate bind to single-stranded pyrimidine residues, it is commonly used to detect promoters opening regions in vivo. KMnO4 treatment of cells, followed by treatment with piperidine, followed by either PCR and/or acrylamide gel electrophoresis allows detection of interaction between transcription factor and the DNA sequence under their control." [PMID:8238889]
exact_synonym: "k-mn-04 footprinting" []
is_a: MI:0602 ! chemical footprinting

[Term]
id: MI:0605
name: enzymatic footprinting
def: "Sites of sequence-specific DNA-protein interaction are identified by altered reactivity of an enzymatic probe to DNA bound by a protein compared to the same nucleotide sequence in naked DNA. Nucleotides in close contact with the binding protein are protected from cleavage by the enzyme. When these enzymes are administrated to intact cells, the pattern of protection from the probes identifies the location of DNA-protein interactions in vivo." [PMID:8238889]
exact_synonym: "enzymatic footprint" []
is_a: MI:0409 ! dna footprinting

[Term]
id: MI:0606
name: DNase I footprinting
def: "Deoxyribonuclease I (DNase I) do not have high specificity for given sequences or residues, thus footprinting with DNase I permits the exact delineation of the protein-DNA binding site. Moreover DNase I, can be used for in vivo footprinting by treating intact cells with permeabilising drugs. In this latter case DNase I in vivo footprinting allow studies of the chromatin structure in genomic DNA." [PMID:8238889]
exact_synonym: "dnase 1 footprinting" []
is_a: MI:0605 ! enzymatic footprinting

[Term]
id: MI:0655
name: lambda repressor two hybrid
def: "A protein of interest (the bait) is fused to the full-length bacteriophage lambda repressor protein (lambdacl, 237 amino acids), containing the amino terminal DNA-binding domain and the carboxylterminal dimerization domain. The corresponding target (prey) protein is fused to the N-terminal domain of the alfa-subunit of RNA polymerase (248 amino acids). The bait is tethered to the lambda operator sequence upstream of the reporter promoter through the DNA-binding domain of lambdacl. When the bait and prey interact, they recruit and stabilize the binding of RNA polymerase at the promoter and activate the transcription of the HIS3 reporter gene. Due to the tendency of both the lambda repressor protein and the N-terminal domain of the alfa-subunit of RNA polymerase to dimerize, this system might not be optimal for the analysis of proteins that self-associate unless their interaction with other protein partners depends on the oligomerization." [PMID:15792953]
related_synonym: "BacterioMatch" []
exact_synonym: "lambda two hybrid" []
is_a: MI:0232 ! transcriptional complementation assay

[Term]
id: MI:0657
name: systematic evolution of ligands by exponential enrichment
def: "RNA and cDNA constructs with variable central sequences and a constant flanking region are collected in a complex library. The library is then screened to select either specific binding partners of a bait molecule (generally a protein) or particular enzymatic activities of the nucleic acid molecules themselves. The selected nucleic acids are amplified using the constant flanking regions to increase their abundance. Cycles of selection-amplification can be repeated to increase the specificity of the targets that, at the end, are individually identified by sequencing." [PMID:11539574]
related_synonym: "in vitro evolution of nucleic acids" []
exact_synonym: "selex" []
is_a: MI:0400 ! affinity technology

[Term]
id: MI:0663
name: confocal microscopy
def: "A confocal is a standard epifluorescence microscope with improvement essentially coming from the rejection of out-of-focus light interference. Confocal imaging system achieves this by two strategies: a) by illuminating a single point of the specimen at any one time with a focused beam, so that illumination intensity drops off rapidly and b) by the use of blocking a pinhole aperture in a conjugate focal plane to the specimen so that light emitted away from the point in the specimen being illuminated is blocked from reaching the detector. Only the light from the single point illuminated of the specimen passing through the image pinhole is detected by a photodetector. Usually a computer is used to control the sequential scanning of the sample and to assemble the image for display onto a video screen." [PMID:14755292]
is_a: MI:0428 ! imaging techniques

[Term]
id: MI:0676
name: tandem affinity purification
def: "Tandem affinity purification allows rapid purification under native conditions of complexes, even when expressed at their natural level. Prior knowledge of complex composition or function is not required. The TAP method requires fusion of the a multiple tag, either N- or C-terminally, to the target (or bait) protein of interest. The multiple tag allows two steps purification steps ensuring a highly selective complex purification." [PMID:11403571]
related_synonym: "TAP" []
exact_synonym: "tap" []
is_a: MI:0004 ! affinity chromatography technology

[Term]
id: MI:0678
name: antibody array
def: "A microarray consisting of antibodies spotted on a solid support in appropriate orientation is incubated with a biological sample (or antigen). Some proteins are captured by the antibodies in the array. Protein of forming complexes on the array are identified according to their prior labelling (tag, ELISA, biotin and others)." [PMID:12454649]
related_synonym: "antigen capture assay" []
related_synonym: "sandwich immunoassay" []
is_a: MI:0008 ! array technology

[Term]
id: MI:0695
name: sandwich immunoassay
def: "Antibody array where proteins retained by the arrayed antibodies are identified using a detector antibody. The detector antibody is either modified with a directly detectable label (enzyme, fluorescent molecule, isotope, etc.), or it is biotinylated for detection after subsequent probing with labeled streptavidin." [PMID:12454649]
is_a: MI:0678 ! antibody array

[Term]
id: MI:0696
name: polymerase assay
def: "Measures the catalysis of the transfer of a free nucleotidyl group to a nucleic acid chain." [PMID:14755292]
related_synonym: "nucleotidyltransferase assay" []
is_a: MI:0415 ! enzymatic study

[Term]
id: MI:0697
name: dna directed dna polymerase assay
def: "Measures the catalysis of the reaction: deoxynucleoside triphosphate + DNA(n) = diphosphate + DNA(n+1); the synthesis of DNA from deoxyribonucleotide triphosphates in the presence of a DNA template or primer." [PMID:14755292]
exact_synonym: "dna dna pol assay" []
is_a: MI:0696 ! polymerase assay

[Term]

id: MI:0698

name: dna directed rna polymerase assay

def: "Measures the catalysis of the reaction: nucleoside triphosphate + RNA(n) = diphosphate + RNA(n+1). Utilizes a DNA template, i.e. the catalysis of DNA-template-directed extension of the 3'-end of an RNA strand by one nucleotide at a time. Can initiate a chain 'de novo'." [PMID:14755292]

exact_synonym: "dna rna pol assay" []

is_a: MI:0696 ! polymerase assay

[Term]

id: MI:0699

name: rna directed dna polymerase assay

def: "Measures the catalysis of the reaction: deoxynucleoside triphosphate + DNA(n) = diphosphate + DNA(n+1). Catalyzes RNA-template-directed extension of the 3'- end of a DNA strand by one deoxynucleotide at a time." [PMID:14755292]

exact_synonym: "rna dna pol assay" []

is_a: MI:0696 ! polymerase assay

[Term]

id: MI:0700

name: rna directed rna polymerase assay

def: "Measures the catalysis of the reaction: nucleoside triphosphate + RNA (n) = diphosphate + RNA (n+1); uses an RNA template." [PMID:14755292]

exact_synonym: "rna rna pol assay" []

is_a: MI:0696 ! polymerase assay

[Term]

id: MI:0726

name: reverse two hybrid

def: "Yeast strains are generated in which expression of DB-X/AD-Y or DBPX hybrid proteins is toxic under particular conditions (negative selection). Under these conditions, dissociation of an interaction should provide a selective advantage thereby facilitating detection: a few growing yeast colonies in which DB-X/AD-Y (or DBPX/binding site) fail to interact should be identified among many nongrowing colonies containing interacting DB-X/AD-Y or DBPX/binding site." [PMID:8816797]

is_a: MI:0232 ! transcriptional complementation assay

[Term]

id: MI:0727

name: lexa b52 complementation

def: "Yeast two-hybrid system using Escherichia coli LexA amino acids 1-202 as the DNA-binding domain (BD), E. coli B42 acidic sequence as the activation domain (AD), and two reporters, lacZ and LEU2, each containing upstream LexA binding elements." [PMID:14613974]

related_synonym: "LexA B52 transcription complementation" []

exact_synonym: "lexa b52 complement" []

is_a: MI:0018 ! two hybrid

[Term]

id: MI:0728

name: gal4 vp16 complementation

def: "A chimeric protein consisting of the GAL4 DNA-binding domain (aa 1-147 of GAL4) and a transcriptional activation domain from the herpes simplex virus protein VP16 (either aa 411-490 or aa 411-455) can specifically activate transcription of a reporter gene located downstream of GAL4 DNA binding sites and the E1B minimal promoter. Similarly, two chimeric proteins, one encoding a chimeric GAL4 protein and the other encoding a chimeric VP16 protein, can activate the reporter gene, if the domains fused to the GAL4 and VP16 sequences can complex with appropriate conformation. However, if the domains fused to the GAL4 and VP16 sequences do not interact specifically to form a + complex that reconstitutes GAL4 function, the reporter gene cannot be activated.\n"

[PMID:1387709]

related_synonym: "karyoplasmic interaction ion strategy" []

related_synonym: "KISS" []

related_synonym: "mammalian two hybrid" []

exact_synonym: "gal4 vp16 complement" []

is_a: MI:0018 ! two hybrid

[Term]

id: MI:0729

name: luminescence based mammalian interactome mapping

def: "This strategy uses Renilla luciferase enzyme (RL) fused to proteins of interest, which are then coexpressed with individual Flag-tagged partners in mammalian cells. Their interactions were determined by performing an RL enzymatic assay on immunoprecipitates using an antibody against Flag." [PMID:15761153]

exact_synonym: "lumier" []

is_a: MI:0004 ! affinity chromatography technology

[Term]

id: MI:0807

name: comigration in gel electrophoresis

def: "The interaction of two molecules is determine by their very close proximity or the overlap of their relative bands in a gel." [PMID:14755292]

exact_synonym: "comigration in gel" []

is_a: MI:0401 ! biochemical

[Term]

id: MI:0808

name: comigration in sds page

def: "Method allowing the detection of strong interactions between two molecules by their very close proximity or the overlap of their relative bands in a denaturing SDS gel." [PMID:14755292, PMID:16732283]

exact_synonym: "comigration in sds" []

is_a: MI:0807 ! comigration in gel electrophoresis

[Term]

id: MI:0809

name: bimolecular fluorescence complementation

def: "The bimolecular fluorescence complementation (BiFC) is an assay for determination of the locations of protein interactions in living cells. This approach is based on complementation between two non fluorescent fragments of the yellow fluorescent protein (YFP) when they are brought together by interactions between proteins fused to each fragment." [PMID:11983170]

exact_synonym: "bifc" []

is_a: MI:0051 ! fluorescence technology

is_a: MI:0090 ! protein complementation assay

[Term]

id: MI:0813

name: proximity enzyme linked immunosorbent assay

def: "Method allowing efficient and precise interaction detection, along with extensive repertoires of specific binding reagents. It is based on proximity a ligation mechanism that enables sensitive high-capacity protein measurements by converting the detection of specific proteins to the analysis of DNA sequences. Proximity probes containing oligonucleotide extensions are designed to bind pairwise to target proteins and to form amplifiable tag sequences by ligation when brought in proximity." [PMID:15155907]

related_synonym: "pELISA" []

exact_synonym: "p elisa" []

is_a: MI:0400 ! affinity technology

is_a: MI:0421 ! identification by antibody

[Term]

id: MI:0814

name: protease accessibility laddering

def: "In protease accessibility laddering (PAL) tagged proteins are purified on magnetic beads in their natively folded state. While attached to the beads, proteins are probed with proteases. Proteolytic fragments are eluted and detected by immunoblotting with antibodies against the tag (e.g., Protein A, GFP, and 6xHis)." [PMID:16615907]

related_synonym: "pal" []

exact_synonym: "protease access" []

is_a: MI:0417 ! footprinting

[Term]

id: MI:0824
name: x-ray powder diffraction
def: "Analysis of a diffraction pattern generated by an isotropic sample composed of many randomly oriented crystals." [PMID:14755292]
related_synonym: "X-ray" []
exact_synonym: "x-ray powder diffrac" []
is_a: MI:0114 ! x-ray crystallography

[Term]
id: MI:0825
name: x-ray fiber diffraction
def: "Analysis of diffraction pattern of a partially ordered sample composed of fibers oriented parallel to each other." [PMID:14755292]
related_synonym: "X-ray" []
exact_synonym: "x-ray fiber diffrac" []
is_a: MI:0114 ! x-ray crystallography

[Term]
id: MI:0826
name: x ray scattering
def: "Method where the internal structure of a sample is derived from the intensity distribution of the scattered monochromatic X-ray beam at very low scattering angles." [PMID:14755292]
exact_synonym: "saxs" []
is_a: MI:0067 ! light scattering

[Term]
id: MI:0827
name: x-ray tomography
def: "X-ray Tomography is a branch of X-ray microscopy. A series of projection images are used to calculate a three dimensional reconstruction of an object. The technique has found many applications in materials science and later in biology and biomedical research. In terms of the latter, the National Center for X-ray Tomography (NCXT) is one of the principle developers of this technology, in particular for imaging whole, hydrated cells." [PMID:14755292]
is_a: MI:0428 ! imaging techniques

[Term]
id: MI:0841
name: phosphotransfer assay
def: "Measures the rate of a phosphate transfer between two proteins " [PMID:14755292]
exact_synonym: "phosphotransfer assa" []
is_a: MI:0415 ! enzymatic study

[Term]
id: MI:0858
name: immunodepleted coimmunoprecipitation
def: "Method involving consecutive CoIps on the same sample, a control one where an interaction is detected, and other CoIPs where the sample is previously treated with a specific antibody that precipitate a candidate interactor and lead to an interaction supression or a change in composition od a complex." [PMID:17081976]
related_synonym: "immunodepleted coip" []
exact_synonym: "immunodepletion" []
is_a: MI:0019 ! coimmunoprecipitation

[Term]
id: MI:0859
name: intermolecular force
def: "An optical tweezer is a scientific instrument that uses a focused laser beam to provide an attractive or repulsive force, depending on the index mismatch (typically on the order of piconewtons) to physically hold and move microscopic dielectric objects (i.e.highly resistants to the flow of an electric current). This instrument allow to measure the forces generated between interacting molecules - either at the level of just single interacting pair of molecules or at the level of larger molecular assemblies. " [PMID:17023539, PMID:17081984]
related_synonym: "force measurement" []
related_synonym: "molecular force measurement" []

related_synonym: "optical tweezer" []
related_synonym: "single molecule force measurement" []
related_synonym: "surface adhesion force measurement" []
is_a: MI:0013 ! biophysical

[Term]

id: MI:0872

name: atomic force microscopy

def: "The atomic force microscope (AFM) is a very high-resolution type of scanning probe microscope, with demonstrated resolution of fractions of a nanometer, more than 1000 times better than the optical diffraction limit. The AFM was invented by Binnig, Quate and Gerber in 1986, and is one of the foremost tools for imaging, measuring and manipulating matter at the nanoscale. The term 'microscope' in the name is actually a misnomer because it implies looking, while in fact the information is gathered by feeling out the surface with a mechanical

feeler. \nhttp://en.wikipedia.org/wiki/Atomic_force_microscope" [PMID:17502105]

related_synonym: "AFM" []

exact_synonym: "atomic force microsc" []

is_a: MI:0428 ! imaging techniques

[Term]

id: MI:0879

name: nucleoside triphosphatase assay

def: "Measures the catalysis of the hydrolysis of a nucleoside triphosphate into a nucleoside diphosphate plus phosphate." [PMID:14755292]

exact_synonym: "triphosphatase ass" []

is_a: MI:0415 ! enzymatic study

[Term]

id: MI:0880

name: atpase assay

def: "Measures the catalysis of the hydrolysis of ATP+ H2O = ADP + phosphate." [PMID:14755292]

is_a: MI:0879 ! nucleoside triphosphatase assay

Note2 Completed checklist for Croze et al¹⁹

- ∞ A submission should contain the essential administrative information:
 - contact email: ed_croze@berlex.com
 - publication title: Receptor for Activated C-Kinase (RACK-1), a WD Motif-Containing Protein, Specifically Associates with the Human Type I IFN Receptor
 - first author: croze
 - publication identifier: PMID 11046044
- ∞ Experiment: 1
Each experimental setup should be described separately, with the following parameters:
 - Host system: Yeast (TaxID:4932)
Interaction detection method: two hybrid (MI:0018).
 - Participant Identification method: nucleotide sequence (MI:0078).
- ∞ Interaction:
 - Participant list:
The list of all molecules participating in the interaction. The list can contain one to many elements. Each molecule should be characterised by:
 - ♣ Database: UniProt (MI:0486)
 - ♣ Accession number from that database. P48551
 - ♣ Version number. 1
 - ♣ Name. IFN- α R β L
 - ♣ The species of origin for the molecule. Identified by NCBI tax id. – not required, accession number given
Biological role: neutral component (MI:497)
 - ♣ Experimental role: bait (MI:0496)
 - ♣ Note –sequence is deletion mutant 281-515

 - ♣ Database: UniProt (MI:0486)
 - ♣ Accession number from that database. P63244
 - ♣ Version number. 3
 - ♣ Name. RACK-1
 - ♣ The species of origin for the molecule. Identified by NCBI tax id. – not required, accession number given
Biological role: neutral component (MI:497)
 - ♣ Experimental role: prey (MI:0498)
Note –protein sequence 90-316 identified by nucleotide sequencing

- ∞ Experiment: 2
 - Each experimental setup should be described separately, with the following parameters:
 - Host system: in vitro
 - Interaction detection method: pull down (MI:0096)
 - Participant Identification method: western blot (MI:0113).
 - ∞ Interaction:
 - Participant list:
 - The list of all molecules participating in the interaction. The list can contain one to many elements. Each molecule should be characterised by:
 - ♣ Database: UniProt (MI:0486)
 - ♣ Accession number from that database. P48551
 - ♣ Version number. 1
 - ♣ Name. IFN- α R β L
 - ♣ The species of origin for the molecule. Identified by NCBI tax id. – not required, accession number given
 - Biological role: neutral component (MI:497)
 - ♣ Experimental role: bait (MI:0496)
 - ♣ Note –sequence is deletion mutant 299-345

 - ♣ Database: UniProt (MI:0486)
 - ♣ Accession number from that database. P63244
 - ♣ Version number. 3
 - ♣ Name. RACK-1
 - ♣ The species of origin for the molecule. Identified by NCBI tax id. – not required, accession number given
 - Biological role: neutral component (MI:497)
 - ♣ Experimental role: prey (MI:0498)
- ∞ Experiment: 3
 - Each experimental setup should be described separately, with the following parameters:
 - Host system: Human (TaxID: 9606) – daudi cells
 - Interaction detection method: anti bait coimmunoprecipitation (MI:0006)
 - Participant Identification method: western blot (MI:0113).
- ∞ Interaction:
 - Participant list:
 - The list of all molecules participating in the interaction. The list can contain one to many elements. Each molecule should be characterised by:
 - ♣ Database: UniProt (MI:0486)
 - ♣ Accession number from that database. P48551
 - ♣ Version number. 1
 - ♣ Name. IFN- α R β L
 - ♣ The species of origin for the molecule. Identified by NCBI tax id. – not required, accession number given
 - Biological role: neutral component (MI:497)
 - ♣ Experimental role: bait (MI:0496)

- ♣ Database: UniProt (MI:0486)
- ♣ Accession number from that database. P63244
- ♣ Version number. 3
- ♣ Name. RACK-1
- ♣ The species of origin for the molecule. Identified by NCBI tax id. – not required, accession number given
- Biological role: neutral component (MI:497)
- ♣ Experimental role: prey (MI:0498)

∞ Experiment: 4

Each experimental setup should be described separately, with the following parameters:

- Host system: Yeast (TaxID:4932)
Interaction detection method: two hybrid (MI:0018).
- Participant Identification method: predetermined participant (MI:0396).
Note – directed two hybrid screen.

∞ Interaction:

- Participant list:

The list of all molecules participating in the interaction. The list can contain one to many elements. Each molecule should be characterised by:

- ♣ Database: UniProt (MI:0486)
- ♣ Accession number from that database. P48551
- ♣ Version number. 1
- ♣ Name. IFN- α R β L
- ♣ The species of origin for the molecule. Identified by NCBI tax id. – not required, accession number given
- Biological role: neutral component (MI:497)
- ♣ Experimental role: bait (MI:0496)
- ♣ Note – sequence is deletion mutant 281-515

- ♣ Database: UniProt (MI:0486)
- ♣ Accession number from that database. P63244
- ♣ Version number. 3
- ♣ Name. RACK-1
- ♣ The species of origin for the molecule. Identified by NCBI tax id. – not required, accession number given
- Biological role: neutral component (MI:497)
- ♣ Experimental role: prey (MI:0498)
- Note – sequence is deletion mutant 90-316