

1-Exit of clones from the ORFeome

We have three human ORFeome

The CCSB 5.1 version (<http://horfdb.dfci.harvard.edu/hv5/index.php?page=orfsearch>). The clones are into pDON223 vector (Spectinomycin resistant) and don't have stop codon. Each clone is representing by a pool and not as unique clone. They are not sequenced. The clones are isolated by the platform and partially sequenced.

The CCSB 8.1 version (<http://horfdb.dfci.harvard.edu/index.php?page=orfsearch>)

The clones are into pDON223 vector (Spectinomycin resistant) and don't have stop codon. The clones are unique and sequenced.

It's necessary to prefer the version 8.1 so possible because the ORFeome is completely sequenced (it's not the case of the version 5.1). Approximately 20% of the clones of the version 5.1 contain error (mutations/deletion). There is less error into the version 8.1.

The Life Technologie ORFeome (http://orf.lifetechnologies.com/cgi-bin/ORF_Browser). The clones have a stop codon and are sequenced. They are cloned into pDon221 vector (Kanamycin resistant)

We have a Mouse ORFeome

It comes from Biovalley company. The clones are into pDON223 vector (Spectinomycin resistant). Some of them have a stop codon and others not.

We have a Yeast ORFeome

It comes from GE Healthcare Dharmacon (<http://dharmacon.gelifsciences.com/non-mammalian-cdna-and-orf/yeast-orf-collection/?productId=D886A3C9-EFDF-4EE4-B35D-E45C749C34BA>). The clones are cloned into a BG1805 vector (Ampiciline resistant)

Service : Clones are sequenced in the extremities and verified by digestion. According to their size, clones are not sequenced completely. The theoretical sequence, the experimental sequence, the alignment of both and the ORF sequence integrated into the pDON vector are supplied. The clone is delivered in DNA form coming from a miniprep. A part is archived into our freezer. It's fundamental to verify the isoform present into the ORFeome.

2-BP cloning reaction

The clone coming from BP reaction is completely sequenced. The theoretical sequence, the experimental sequence, the alignment of both are supplied. The clone is delivered in DNA form coming from a miniprep.

3-LR cloning reaction

The clone coming from LR reaction are tested by digestion. The sequence of the destination vectors are into the web

(http://www.igmm.cnrs.fr/spip.php?article326&equipe=COMMUN_psmgc&chemin_rub=89)

Service : The clone is tested by digestion and the gel photo is sent to the user. If the pDON is supplied for the user, we ask that the sequence of the ORF is certified. The ORF sequence integrated into the pDest vector are supplied and the clone is delivered in DNA form coming from a miniprep.