neXt-MP50 Challenge

translating the code of life

Executive Summary November 10, 2021

	Add the numbers
A. Papers submitted to the JPR 2021 HPP SI	**4**
B. Papers published in the JPR 2020 HPP SI	**4**
C. Papers relevant to the HPP published elsewhere 2020/2021	**33**
D. How many MPs (PE2–4) identified as PE1 since 2017	**540**
E. How many MPs (PE2–4) found in 2020 now listed in neXt-Prot as PE1	**240**
F. How many candidate MPs found in 2020, but not meeting Guidelines?	**58 (11 become PE1)**

Chromosome Number: 2

PIC Leaders: Lydie Lane (SIB/University of Geneva)

Part I: Missing Proteins: next-MP50 Challenge

Major lab members or partners contributing to the neXt-MP50 Challenge

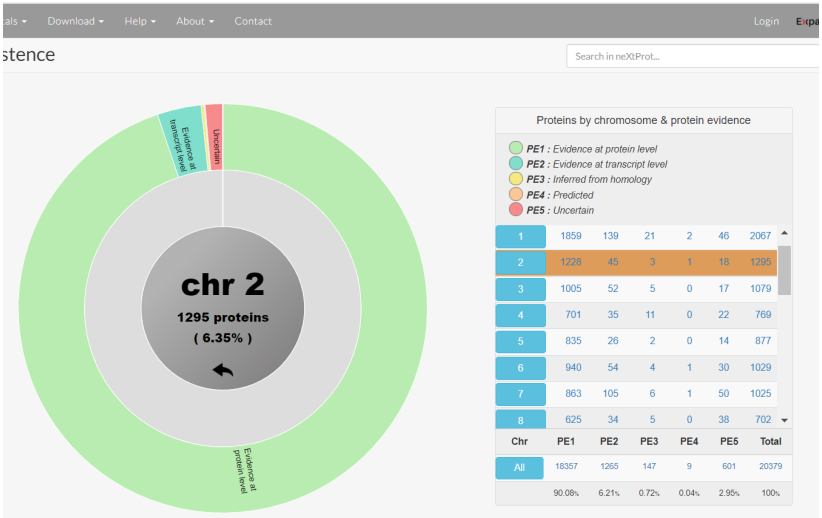
The neXtProt team

As we have been focusing on the neXt-CP50 challenge, we have not contributed to the neXt-MP50 challenge this year except from maintaining neXtProt

Status of the Chromosome "parts list" for your Chromosome:

(https://www.nextprot.org/about/protein-existence)

There are still 49 MPs (45 PE2, 3 PE3 and 1 PE4).



A) Titles and authors of papers submitted to the **2021** JPR SI or planned.

Omenn GS, Lane L, Overall CM, Paik YK, Cristea IM, Corrales FJ, Lindskog C, Bandeira N, Aebersold R, Moritz RL, Deutsch EW (**2021**) *Progress on Identifying and Characterizing the Human Proteome: 2021 Metrics from the HUPO Human Proteome Project*

B) Titles and authors of papers published in the **2020** JPR SI.

Vandenbrouck Y, Pineau C, Lane L. (**2020**) *The Functionally Unannotated Proteome of Human Male Tissues: A Shared Resource to Uncover New Protein Functions Associated with Reproductive Biology.* **J Proteome Res.** 19(12):4782-4794. doi: 10.1021/acs.jproteome.0c00516 PMID: 33064489

Omenn GS, Lane L, Overall CM, Cristea IM, Corrales FJ, Lindskog C, Paik YK, Van Eyk JE, Liu S, Pennington SR, Snyder MP, Baker MS, Bandeira N, Aebersold R, Moritz RL, Deutsch EW. (**2020**) *Research on the Human Proteome Reaches a Major Milestone: >90% of Predicted Human Proteins Now Credibly Detected, According to the HUPO Human Proteome Project.* **J Proteome Res**. 19(12):4735-4746. doi: 10.1021/acs.jproteome.0c00485. PMID: 32931287

C) Titles and authors of other HPP relevant papers submitted elsewhere in 2020/2021.

Adhikari S, Nice EC, Deutsch EW, Lane L, Omenn GS, Pennington SR, Paik YK, Overall CM, Corrales FJ, Cristea IM, Van Eyk JE, Uhlén M, Lindskog C, Chan DW, Bairoch A, Waddington JC, Justice JL, LaBaer J, Rodriguez H, He F, Kostrzewa M, Ping P, Gundry RL, Stewart P, Srivastava S, Srivastava S, Nogueira FCS, Domont GB, Vandenbrouck Y, Lam MPY, Wennersten S, Vizcaino JA, Wilkins M, Schwenk JM, Lundberg E, Bandeira N, Marko-Varga G, Weintraub ST, Pineau C, Kusebauch U, Moritz RL, Ahn SB, Palmblad M, Snyder MP, Aebersold R, Baker MS. (**2020**) *A high-stringency blueprint of the human proteome*. **Nat Commun**. 16;11(1):5301. doi: 10.1038/s41467-020-19045-9.

Zahn-Zabal M, Michel PA, Gateau A, Nikitin F, Schaeffer M, Audot E, Gaudet P, Duek PD, Teixeira D, Rech de Laval V, Samarasinghe K, Bairoch A, Lane L. (**2020**) *The neXtProt knowledgebase in 2020: data, tools and usability improvements*. **Nucleic Acids Res.** 48(D1):D328-D334.

Zahn-Zabal M, Lane L. (**2020**) *What will neXtProt help us achieve in 2020 and beyond?* **Expert Rev Proteomics** 17(2):95-98.

D) How many PE1-found MPs since HUPO-2017 has your chromosome group reported in papers?

In Carapito et al. 2017, we reported the validation of 12 PE2 proteins by SRM and IHC.
In Robin et al. 2018, we reported the validation of 1 PE2 protein (FRAT2) by reanalysing MS/MS data on 41 HeLa cell datasets.
In the two articles by Macron et al. 2018, we reported the validation of 14 PE2 proteins and 1 PE5 protein (SHISA8) by analysing CSF by MS/MS

We did not report any validation of MP in 2019 and 2020

E) How many PE1-found MPs since HUPO-2019 are now in NeXt-Prot as PE1 proteins? Please check each of your MPs that you reported in the JPR SI.

N/A

F) How many candidate MPs found, but not meeting the guidelines 3.0? (Please state number of peptides identified, their length, and biological replicates found in).

N/A

G) Any significant clinical or other successes re a MP that you wish us to consider highlighting in the report.

N/A

Chromosome Number: 3

PIC Leaders: Takeshi Kawamura (Associate professor, Isotope Science Center, The University of Tokyo)

Part I: Missing Proteins: neXt-MP50 Challenge

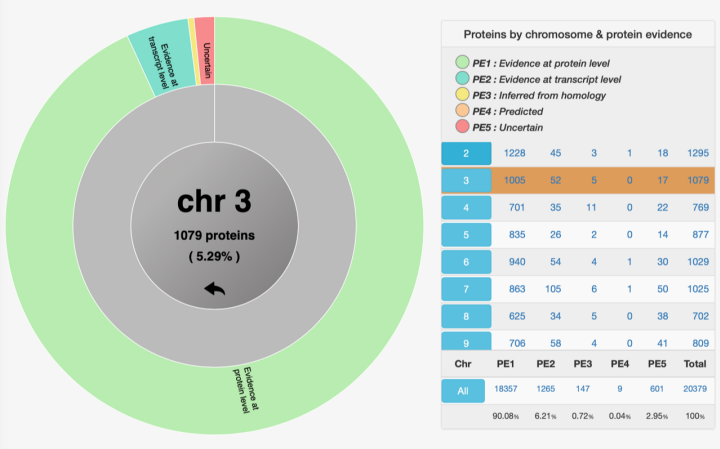
Major lab members or partners contributing to the neXt-MP50 Challenge

Kazuki Yamamoto (Assistant professor)
Yoko Chikaoka (Researcher)

Co-PI
Toshihide Nishimura (Professor, St. Marianna University School of Medicine)
Hiromasa Tojo (Professor, St. Marianna University School of Medicine)

Status of the Chromosome "parts list" for your Chromosome:

(https://www.nextprot.org/about/protein-existence)



Chr	PE1	PE2	PE3	PE4	PE5	Total
2	1228	45	3	1	18	1295
3	1005	52	5	0	17	1079
4	701	35	11	0	22	769
5	835	26	2	0	14	877
6	940	54	4	1	30	1029
7	863	105	6	1	50	1025
8	625	34	5	0	38	702
9	706	58	4	0	41	809
All	18357	1265	147	9	601	20379
	90.08%	6.21%	0.72%	0.04%	2.95%	100%

A) Titles and authors of papers submitted to the **2021** JPR SI. What is their publication status.
No

B) Titles and authors of papers published in the **2020** JPR SI.
No

C) Titles and authors of other HPP relevant papers submitted elsewhere in 2020/2021.

1. Activation of CpG-rich promoters mediated by MLL drives MOZ-rearranged leukemia. Ryo Miyamoto, Hiroshi Okuda, Akinori Kanai, Satoshi Takahashi, **Takeshi Kawamura**, Hirotaka Matsui, Toshio Kitamura, Issay Kitabayashi, Toshiya Inaba, Akihiko Yokoyama. ***Cell Rep***. 32, 108200, September 29, 2020.
2. The human melanoma proteome atlas-Defining the molecular pathology. Betancourt LH, Gil J, Kim Y, Doma V, Çakır U, Sanchez A, Murillo JR, Kuras M, Parada IP, Sugihara Y, Appelqvist R, Wieslander E, Welinder C, Velasquez E, de Almeida NP, Woldmar N, Marko-Varga M, Pawłowski K, Eriksson J, Szeitz B, Baldetorp B, Ingvar C, Olsson H, Lundgren L, Lindberg H, Oskolas H, Lee B, Berge E, Sjögren M, Eriksson C, Kim D, Kwon HJ, Knudsen B, Rezeli M, Hong R, Horvatovich P, Miliotis T, **Nishimura T**, Kato H, Steinfelder E, Oppermann M, Miller K, Florindi F, Zhou Q, Domont GB, Pizzatti L, Nogueira FCS, Horvath P, Szadai L, Tímár J, Kárpáti S, Szász AM, Malm J, Fenyö D, Ekedahl H, Németh IB, Marko-Varga G. ***Clin Transl Med***. 2021 Jul;11(7):e473.
3. The Human Melanoma Proteome Atlas-Complementing the melanoma transcriptome.Betancourt LH, Gil J, Sanchez A, Doma V, Kuras M, Murillo JR, Velasquez E, Çakır U, Kim Y, Sugihara Y, Parada IP, Szeitz B, Appelqvist R, Wieslander E, Welinder C, de Almeida NP, Woldmar N, Marko-Varga M, Eriksson J, Pawłowski K, Baldetorp B, Ingvar C, Olsson H, Lundgren L, Lindberg H, Oskolas H, Lee B, Berge E, Sjögren M, Eriksson C, Kim D, Kwon HJ, Knudsen B, Rezeli M, Malm J, Hong R, Horvath P, Szász AM, Tímár J, Kárpáti S, Horvatovich P, Miliotis T, **Nishimura T**, Kato H, Steinfelder E, Oppermann M, Miller K, Florindi F, Zhou Q, Domont GB, Pizzatti L, Nogueira FCS, Szadai L, Németh IB, Ekedahl H, Fenyö D, Marko-Varga G. ***Clin Transl Med***. 2021 Jul;11(7):e451.
4. Protein co-expression network-based profiles revealed from laser-microdissected cancerous cells of lung squamous-cell

carcinomas. **Nishimura T**, Fujii K, Nakamura H, Naruki S, Sakai H, Kimura H, Miyazawa T, Takagi M, Furuya N, Marko-Varga G, Kato H, Saji H. ***Sci Rep***. 2021 Oct 12;11(1):20209.

D) How many PE1-found MPs since HUPO-2017 has your chromosome group reported in papers?
No

E) How many PE1-found MPs since HUPO-2020 are now in neXt-Prot as PE1 proteins? Please check each of your MPs that you reported in the JPR SI.

In 2019 (8-22-2019), PE1 was 969, now it is 1,005. MPs(PE2,3,4) has decreased from 98 to 74. The total number of genes has increased from 1,067 to 1,079.

F) How many candidate MPs found, but not meeting the guidelines 3.0? (Please state number of peptides identified, their length, and biological replicates found in).

We haven't found it in chromosome 3, but We have found it in other chromosomes. A part of it has been partially registered in ProteomeXchange. We have not yet validated the results with synthetic peptides.

	acc. code	gene name(s)	Chr.	PE	Nr. of peptides identified	length	Biological Replicates	comment
1	NX_Q8IVE0	CROCCP3	1	5	2	8(overlap)	9	
2	NX_H7BZ55	CROCC2	2	5	3	9, 9, 19	1	
3	NX_Q7Z2Y8	GVINP1	11	2	4	8, 7, 8, 16	8	Two peptides were detected in one of the eight replicates, and only one peptide was detected in the others. A total of four sites were identified.
4	NX_P0DP75	MED14OS	X	5	3	15, 8, 26	2	Two peptides were detected in one of the four replicates, and only one peptide was detected in the others. A total of three sites were identified

G) Any significant clinical or other successes re a MP that you wish us to consider highlighting in the report.
No

Chromosome Number: 4

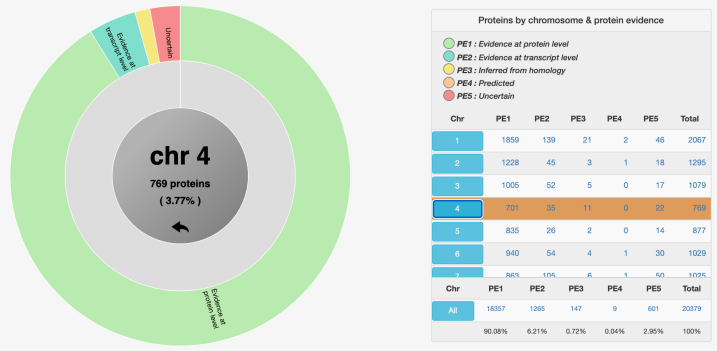
PIC Leaders:
Yu-Ju Chen, Chia-Li Han, Ting-Yi Sung, Sung-Liang Yu

Part I: Missing Proteins: neXt-MP50 Challenge

Major lab members or partners contributing to the neXt-MP50 Challenge
Yi-Ju Chen, Wen-Hsin Chang

Status of the Chromosome "parts list" for your Chromosome:

(https://www.nextprot.org/about/protein-existence)



A) Titles and authors of papers submitted to the **2021** JPR SI. What is their publication status.
NA

B) Titles and authors of papers published in the **2020** JPR SI.
"Mining Missing Proteins from Cancer Tissue Proteome" (*data mining ongoing*)

C) Titles and authors of other HPP relevant papers submitted elsewhere in 2020/2021.
Submission to International Journal of Molecular Sciences (IJMS)

D) How many PE1-found MPs since HUPO-2017 has your chromosome group reported in papers?
26 PE1-found MPs were reported in 2018. Based on our recent large-scale tissue proteomics profiling on non-smoking lung adenocarcinoma patients (Cell, 182, 226–244, 2020) and breast cancer patients (Manuscript in preparation), we identified 414 MPs (372 PE2 and 42 PE3), including 11 MPs in chromosome 4. The verification is under investigation according to guideline 3.0.

E) How many PE1-found MPs since HUPO-2020 are now in neXt-Prot as PE1 proteins? Please check each of your MPs that you reported in the JPR SI.
NA

F) How many candidate MPs found, but not meeting the guidelines 3.0? (Please state number of peptides identified, their length, and biological replicates found in).
NA

G) Any significant clinical or other successes re a MP that you wish us to consider highlighting in the report.

1. **Chr4-DP1:** The protein expression of Chr4-DP1 is upregulated in tumor tissue of lung cancer patient. The mRNA expression levels were profiled in 22 lung cancer cell lines. Chr4-DP1-knockdown cancer cells were established, and the preliminary result showed that Chr4-DP1 slightly promotes cancer proliferation and colonization; no effect on ER stress markers' expression level.
2. **Chr4-DP2:** The protein expression of Chr4-DP2 is downregulated in tumor tissue of lung cancer patient and associated with better survival. The mRNA expression levels were profiled in 22 lung cancer cell lines; Chr4-DP2-overexpressing cells were established, and the preliminary result showed that Chr4-DP2 suppresses cancer proliferation, migration and invasion; RNA-seq was done for Chr4-DP2-overexpressing cells.
3. **Chr4-DP3:** The protein expression of Chr4-DP3 is upregulated in tumor tissue of lung cancer patient and associated with poor survival. mRNA expression levels were profiled in 22 lung cancer cell lines. Chr4-DP3-knockdown cancer cells were established and the bio-functions are testing.
4. **Chr4-DP4:** The protein expression of Chr4-DP4 is upregulated in tumor tissue of lung cancer patient and associated with poor survival. mRNA expression levels were profiled in 22 lung cancer cell lines; Chr4-DP4-knockdown cancer cells were established and the bio-functions are testing.
5. **Chr4-DP5:** The protein expression of Chr4-DP5 is upregulated in tumor tissue of lung cancer patient and associated with poor survival. mRNA expression levels were profiled in 22 lung cancer cell lines; Preparation of Chr4-DP5-knockdown cancer cells is going.
6. **Chr4-DP6:** The protein level of Chr4-DP6 is downregulated in tumor tissue of lung cancer patient. mRNA expression levels were profiled in 22 lung cancer cell lines; Preparation of Chr4-DP6-overexpressing cancer cells is ongoing.

Chromosome Number: 5

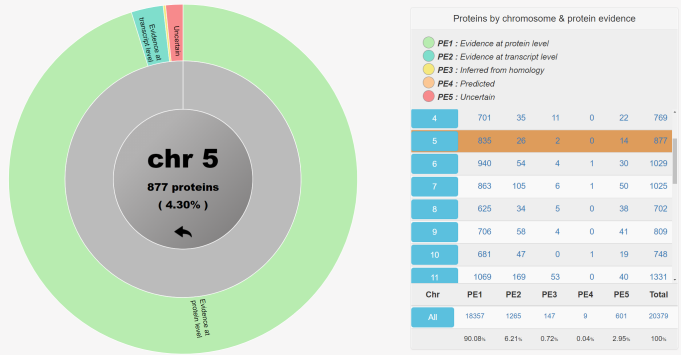
PIC Leaders: Peter Horvatovich

Part I: Missing Proteins: neXt-MP50 Challenge

Major lab members or partners contributing to the neXt-MP50 Challenge
in collaboration with Gyorgy-Marko Varga and Gilberto Domont

Status of the Chromosome "parts list" for your Chromosome:

(https://www.nextprot.org/about/protein-existence)



A) Titles and authors of papers submitted to the **2021** JPR SI. What is their publication status.
None

B) Titles and authors of papers published in the **2020** JPR SI.
None

C) Titles and authors of other HPP relevant papers submitted elsewhere in 2020/2021.

- Betancourt, Lazaro Hiram; Gil, Jeovanis; Sanchez, Aniel; Doma, Viktória; Kuras, Magdalena; Murillo, Jimmy Rodriguez; Velasquez, Erika; Çakır, Uğur; Kim, Yonghyo; Sugihara, Yutaka, The Human Melanoma Proteome Atlas—Complementing the melanoma transcriptome, Clinical and translational medicine 11 (7), e451
- Betancourt, Lazaro Hiram; Gil, Jeovanis; Kim, Yonghyo; Doma, Viktória; Çakır, Uğur; Sanchez, Aniel; Murillo, Jimmy Rodriguez; Kuras, Magdalena; Parada, Indira Pla; Sugihara, Yutaka, The human melanoma proteome atlas—Defining the molecular pathology, Clinical and translational medicine 11 (7), e473
- Gil, Jeovanis; Kim, Yonghyo; Szeitz, Beáta; Doma, Viktória; Çakır, Uğur; de Almeida, Natália Pinto; Hagemeijer, Yanick Paco; Guryev, Victor; Johansson, Jenny G; Sharma, Yogita, Proteogenomics Reveals how Metastatic Melanoma Modulates the Immune System to Allow Immune Evasion, bioRxiv
- Sanchez, Aniel; Kuras, Magdalena; Murillo, Jimmy Rodriguez; Pla, Indira; Pawlowski, Krzysztof; Szasz, A Marcell; Gil, Jeovanis; Nogueira, Fabio; Perez-Riverol, Yasset; Eriksson, Jonatan, Novel functional proteins coded by the human genome discovered in metastases of melanoma patients, Cell biology and toxicology 36 (3), 261-272
- Brandsma, Corry-Anke; Guryev, Victor; Timens, Wim; Ciconelle, Ana; Postma, Dirkje S; Bischoff, Rainer; Johansson, Maria; Ovchinnikova, Ekaterina S; Malm, Johan; Marko-Varga, Gyorgy, Integrated proteogenomic approach identifying a protein signature of COPD and a new splice variant of SORBS1, Thorax 75 (2), 180-183

D) How many PE1-found MPs since HUPO-2017 has your chromosome group reported in papers?
9

E) How many PE1-found MPs since HUPO-2020 are now in neXt-Prot as PE1 proteins? Please check each of your MPs that you reported in the JPR SI.
3

F) How many candidate MPs found, but not meeting the guidelines 3.0? (Please state number of peptides identified, their length, and biological replicates found in).
24 from these 11 become PE1

G) Any significant clinical or other successes re a MP that you wish us to consider highlighting in the report.
none

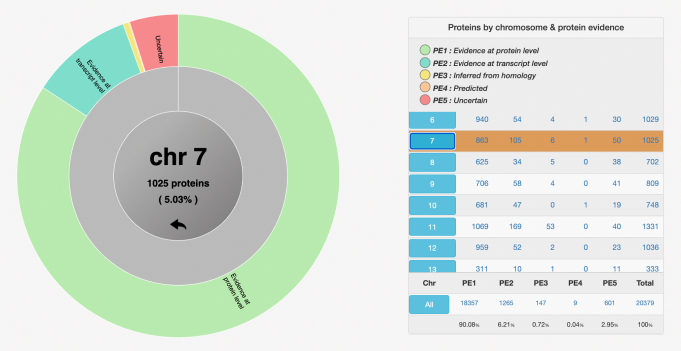
Chromosome Number 7

PIC Leader: Prof Ed Nice

Part I: Missing Proteins: neXt-MP50 Challenge

Major lab members or partners contributing to the neXt-MP50 Challenge
We continue to address the missing proteins, especially olfactory receptors, using a HPP/BD approach in conjunction with bioinformatics. Key labs involved are those of Prof Mark Baker (Macquarie) and Prof Ed Nice (Monash) with data compiled from all searchable publications. This approach also has the potential to find missing proteins for other chromosomes.

Status of the Chromosome "parts list" for your Chromosome:



Increase from 828 to 863: 35 NEW ENTRIES

A) Titles and authors of papers submitted to the **2021** JPR SI. What is their publication status.
Use of a Recombinant Biomarker Protein DDA Library Increases DIA Coverage of Low Abundance Plasma Proteins.
Ahn SB, Kamath KS, Mohamedali A, Noor Z, Wu JX, Pascovici D, Adhikari S, Cheruku HR, Guillemin GJ, McKay MJ, **Nice EC**, Baker MS.J Proteome Res. 2021 May 7;20(5):2374-2389.

B) Titles and authors of papers published in the **2020** JPR SI.
Research on the Human Proteome Reaches a Major Milestone: >90% of Predicted Human Proteins Now Credibly Detected, According to the HUPO Human Proteome Project.
Omenn GS, Lane L, Overall CM, Cristea IM, Corrales FJ, Lindskog C, Paik YK, Van Eyk JE, Liu S, Pennington SR, Snyder MP, Baker MS, Bandeira N, Aebersold R, Moritz RL, Deutsch EW.J Proteome Res. 2020 Dec 4;19(12):4735-4746. doi: 10.1021/acs.jproteome.0c00485.

C) Titles and authors of other HPP relevant papers submitted elsewhere in 2020/2021.
Proteomics, Personalized Medicine and Cancer.
Su M, Zhang Z, Zhou L, Han C, Huang C, **Nice EC.** Cancers (Basel). 2021 May 21;13(11):2512

The separation sciences, the front end to proteomics: An historical perspective.
Nice EC. Biomed Chromatogr. 2021 Jan;35(1):e4995. doi: 10.1002/bmc.4995.

Recent advances in autophagic machinery: a proteomic perspective.
Wu X, Luo L, Kong R, Song Y, Li Q, **Nice EC**, Wang K.Expert Rev Proteomics. 2020 Jul-Aug;17(7-8):561-579. doi: 10.1080/14789450.2020.1808464. Epub 2020 Aug 30.

The status of proteomics as we enter the 2020s: Towards personalised/precision medicine.
Nice EC. Anal Biochem. 2020 Jul 31:113840. doi: 10.1016/j.ab.2020.113840.

The omics revolution: beyond genomics. A meeting report.
Nice EC. Clin Proteomics. 2020 Jan 24;17:1. doi: 10.1186/s12014-020-9266-9

A high-stringency blueprint of the human proteome.
Adhikari S, **Nice EC**, Deutsch EW, Lane L, Omenn GS, Pennington SR, Paik YK, Overall CM, Corrales FJ, Cristea IM, Van Eyk JE, Uhlén M, Lindskog C, Chan DW, Bairoch A, Waddington JC, Justice JL, LaBaer J, Rodriguez H, He F, Kostrzewa M, Ping P, Gundry RL, Stewart P, Srivastava S, Srivastava S, Nogueira FCS, Domont GB, Vandenbrouck Y, Lam MPY, Wennersten S, Vizcaino JA, Wilkins M, Schwenk JM, Lundberg E, Bandeira N, Marko-Varga G, Weintraub ST, Pineau C, Kusebauch U, Moritz RL, Ahn SB, Palmblad M, Snyder MP, Aebersold R, Baker MS

D) How many PE1-found MPs since HUPO-2017 has your chromosome group reported in papers?
0

E) How many PE1-found MPs since HUPO-2020 are now in neXt-Prot as PE1 proteins? Please check each of your MPs that you reported in the JPR SI.
N/A

F) How many candidate MPs found, but not meeting the guidelines 3.0? (Please state number of peptides identified, their length, and biological replicates found in).
N/A

G) Any significant clinical or other successes re a MP that you wish us to consider highlighting in the report.
N/A

Chromosome Number: 9

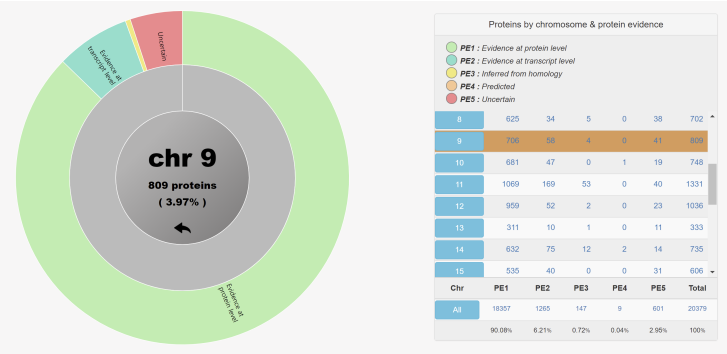
PIC Leaders: Je-Yoel Cho

Part I: Missing Proteins: neXt-MP50 Challenge

Major lab members or partners contributing to the neXt-MP50 Challenge
Soo-Youn Lee, Yong-In Kim, Dong Wook Kim, HuiSu Kim, Hyoung-Min Park, Jinwhan Eugene Lee

Status of the Chromosome "parts list" for your Chromosome:

(https://www.nextprot.org/about/protein-existence)



A) Titles and authors of papers submitted to the **2021** JPR SI. What is their publciation status.
Not submitted yet

B) Titles and authors of papers published in the **2020** JPR SI.
Not published yet

C) Titles and authors of other HPP relevant papers submitted elsewhere in 2020/2021.

Not submitted yet

D) How many PE1-found MPs since HUPO-2017 has your chromosome group reported in papers?

We found DEFB123 with two unique peptides in a testis, which collected from maturation arrest (MA) patient.

E) How many PE1-found MPs since HUPO-2020 are now in neXt-Prot as PE1 proteins? Please check each of your MPs that you reported in the JPR SI.

N/A

F) How many candidate MPs found, but not meeting the guidelines 3.0? (Please state number of peptides identified, their length, and biological replicates found in).

N/A

G) Any significant clinical or other successes re a MP that you wish us to consider highlighting in the report.

N/A

Chromosome Number: 10

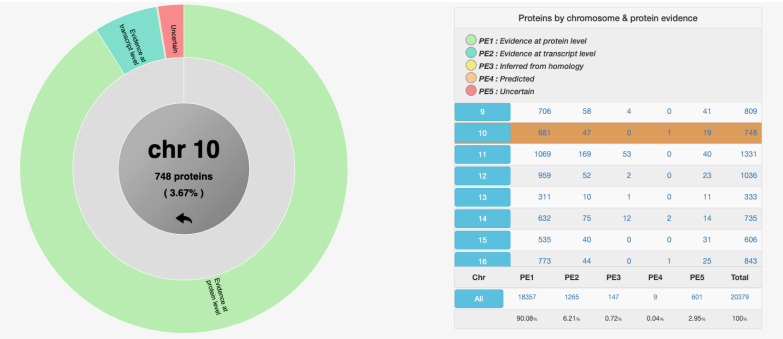
PIC Leaders: Joshua LaBaer, Jin Park

Part I: Missing Proteins: neXt-MP50 Challenge

Major lab members or partners contributing to the neXt-MP50 Challenge
Vel Murugan, Joe Miceli

Status of the Chromosome "parts list" for your Chromosome:

(https://www.nextprot.org/about/protein-existence)



Proteins by chromosome & protein evidence

PE1 : Evidence at protein level
PE2 : Evidence at transcript level
PE3 : Inferred from homology
PE4 : Predicted
PE5 : Uncertain

Chr	PE1	PE2	PE3	PE4	PE5	Total
9	706	58	4	0	41	809
10	681	47	0	1	19	748
11	1069	169	53	0	40	1331
12	959	52	2	0	23	1036
13	311	10	1	0	11	333
14	632	75	12	2	14	735
15	535	40	0	0	31	606
16	773	44	0	1	25	843
All	18357	1265	147	9	601	20379
	90.08%	6.21%	0.72%	0.04%	2.95%	100%

A) Titles and authors of papers submitted to the **2021** JPR SI. What is their publciation status.
NA

B) Titles and authors of papers published in the **2020** JPR SI.
NA

C) Titles and authors of other HPP relevant papers submitted elsewhere in 2020/2021.
NA

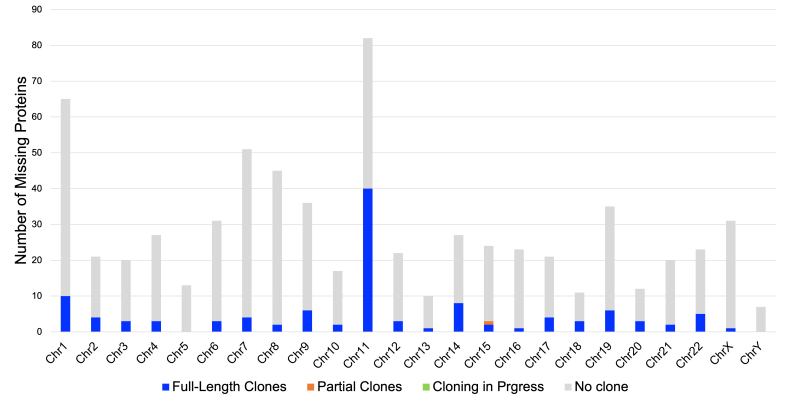
D) How many PE1-found MPs since HUPO-2017 has your chromosome group reported in papers?
0

E) How many PE1-found MPs since HUPO-2020 are now in neXt-Prot as PE1 proteins? Please check each of

your MPs that you reported in the JPR SI.
15

F) How many candidate MPs found, but not meeting the guidelines 3.0? (Please state number of peptides identified, their length, and biological replicates found in).
0

G) Any significant clinical or other successes re a MP that you wish us to consider highlighting in the report.
As a member of the 5-chromosome consortium of Chr 5, 10, 15, 16, and 19, we have been providing the IVTT-compatible plasmids for missing proteins to other members for IVTT-assisted SRM and continue to generate more plasmids. We have assembled a comprehensive and one of the world's largest collections of full-length Gateway plasmids representing 90% of all human protein-coding genes and are distributing the collection through our repository and distribution web portal DNASU (dnasu.org). Currently, we have full-length plasmids for 116 of 674 missing proteins (shown below), which is available to the entire C-HPP team.



Chromosome Number: 11

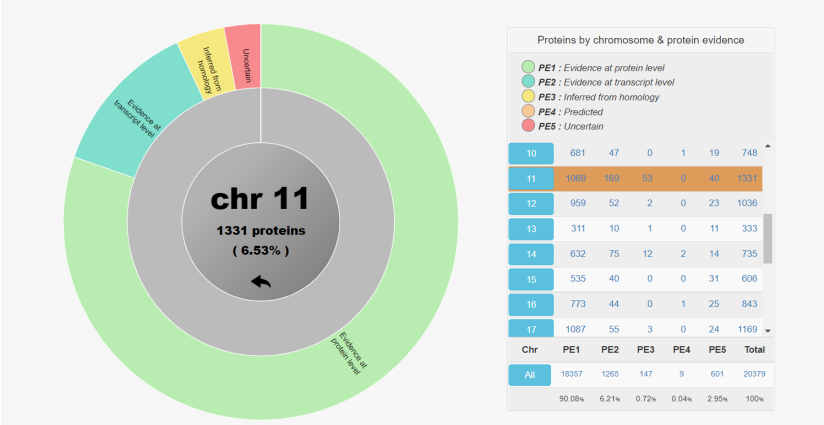
PIC Leaders: Jong Shin Yoo

Part I: Missing Proteins: neXt-MP50 Challenge

Major lab members or partners contributing to the neXt-MP50 Challenge

Status of the Chromosome "parts list" for your Chromosome:

(https://www.nextprot.org/about/protein-existence)



A) Titles and authors of papers submitted to the **2021** JPR SI. What is their publication status?
B) Titles and authors of papers published in the **2020** JPR SI.
C) Titles and authors of other HPP relevant papers submitted elsewhere in 2020/2021.

Chromosome-Centric Human Proteome Study of Chromosome 11 Team
Heeyoun Hwang ; Jin Young Kim ; Jong Shin Yoo,
Mass Spectrometry Letters, Volume 12 Issue 3 / Pages.60-65 / 2021 / 2233-4203(pISSN) / 2093-8950(eISSN)

A Proteotranscriptomic-Based Computational Drug-Repositioning Method for Alzheimer's Disease.
Lee SY, Song MY, Kim D, Park C, Park DK, Kim DG, Yoo JS, Kim YH.
Front Pharmacol. 2020 Jan 29;10:1653. doi: 10.3389/fphar.2019.01653. eCollection 2019.
PMID: 32063857

D) How many PE1-found MPs since HUPO-2017 has your chromosome group reported in papers?

E) How many PE1-found MPs since HUPO-2020 are now in neXt-Prot as PE1 proteins? Please check each of your MPs that you reported in the JPR SI.

F) How many candidate MPs found, but not meeting the guidelines 3.0? (Please state number of peptides identified, their length, and biological replicates found in).

G) Any significant clinical or other successes re a MP that you wish us to consider highlighting in the report.

Chromosome Number: 12

PIC Leaders: Ravi Sirdeshmukh (partners: Hari PS, Manoj K Gupta, Mahesh Kulkarni, Srikanth Rapole)
Yuju Chen, Taiwan; Terence Poon, Hong Kong; Radislaw Sobota, Singapore
[This Report is mainly based on the work from Ravi Sirdeshmukh Group, India]

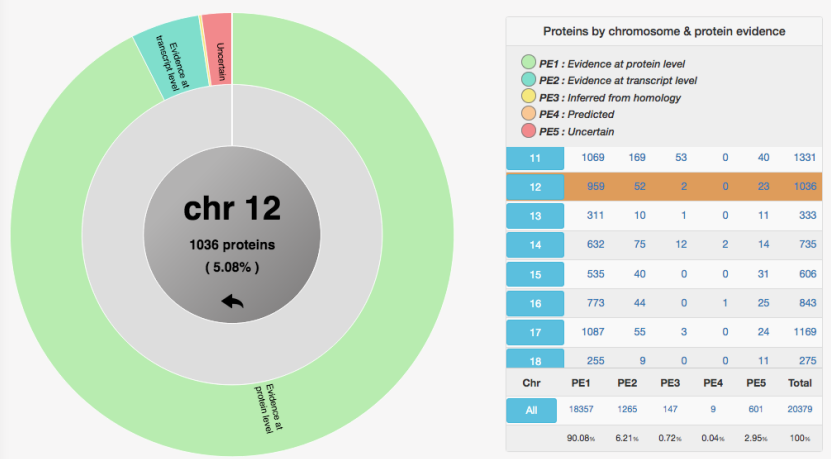
Part I: Missing Proteins: neXt-MP50 Challenge

Major lab members or partners contributing to the neXt-MP50 Challenge

RS group : (present) Hari PS, Manoj K Gupta

Status of the Chromosome "parts list" for your Chromosome:

(https://www.nextprot.org/about/protein-existence)



NeXtProt Version	PE1	Missing Proteins (MP)			PE5 (*)	Total entries/genes
		PE2 (*)	PE3 (*)	PE4		
Feb 18, 2021	959/960	52 (14)	2 (1)	0	23 (2)	1036/1037
Jul 17, 2020	954/955	54	3	0	23	1034/1035

* In the Feb 2021 version, there are 14 PE2, 1 PE3 and 3 PE5 genes that were identified in our study (JPR SI 2014)

New PE1 between 2020-2021

960-955= 5 (1 of them were also identified in our study) NX_A6NMB9

 # From total 89 MPs reported first from our study (JPR SI, 2014), 72 are now considered as protein level evidence as per NeXtProt Feb 2021

A) Titles and authors of papers submitted to the **2021** JPR SI. What is their publication status.
Nil

B) Titles and authors of papers published in the **2020** JPR SI. Nil

C) Titles and authors of other HPP relevant papers submitted elsewhere in 2020/2021.
Mole Cellu Prot (MCP) (2021) under revision

Proteogenomic analysis of breast cancer transcriptomic and proteomic data, using *de novo* transcript assembly: Genome-wide identification of novel peptides and potential clinical implications.
P. S. Hari[1], Lavanya Balakrishnan[1], Chaithanya Kotyada[1], Arivusudar Everad John[1], Nameeta Shah[1#] and Ravi Sirdeshmukh[1, 2, 3#].
[This manuscript is related to development of a proteogenomics pipeline and analysis of CPTAC dataset for novel peptides]

More manuscripts using the same approach are being submitted (About GBM related alternative splice forms , novel ORFs --- , from across multiple chromosomes)

D) How many PE1-found MPs since HUPO-2017 has your chromosome group reported in papers?

No new MPs reported after 2014 . Number reported 89 , 72 of them have entered
At PE1 level in next-Prot.

E) How many PE1-found MPs since HUPO-2020 are now in neXt-Prot as PE1 proteins? Please check each of your MPs that you reported in the JPR SI.

New PE1 between 2020-2021
960-955= 5 (1 of them NX_A6NMB9 was reported first time in our 2014 study

F) How many candidate MPs found, but not meeting the guidelines 3.0? (Please state number of peptides identified, their length, and biological replicates found in). Nil

-

G) Any significant clinical or other successes re an MP that you wish us to consider highlighting in the report. To be explored

Chromosome Number: 13

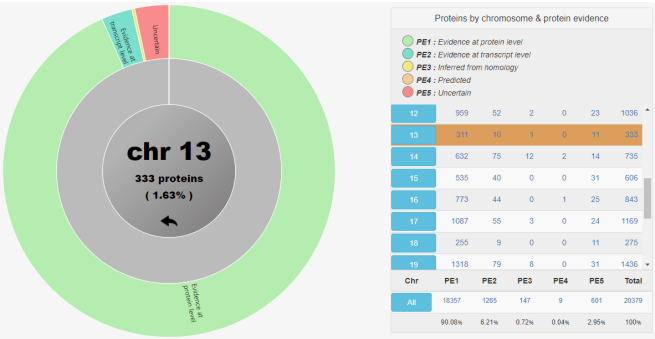
PIC Leaders:
Young-Ki Paik

Part I: Missing Proteins: next-MP50 Challenge

Major lab members or partners contributing to the neXt-MP50 Challenge

Keun-Na (YPRC, Yosei University, Korea)
Ju-Wan Kim (YPRC, Yosei University, Korea)
Jin-Young Cho (YPRC, Yosei University, Korea)
Chae-Yeon Kim (YPRC, Yosei University, Korea)
Jun-Young Park (YPRC, Yosei University, Korea)

Status of the Chromosome "parts list" for your Chromosome:
(https://www.nextprot.org/about/protein-existence)



A) Titles and authors of papers submitted to the **2021** JPR SI or planned.
- Ju-Wan Kim, Keun-Na, Jin-Young Cho and Young-Ki Paik. An Improved detection strategy of MPs by using sample fractionation and multiple data search (underway))

B) Titles and authors of papers published in the **2021** JPR SI.
- None

C) Titles and authors of other HPP relevant papers submitted elsewhere in 2020/2021.
- Na K, Kim M, Kim CY, Lim JS, Cho JY, Shin H, Lee HJ, Kang BJ, Han DH, Kim H, Baik JH, Swiatek-de Lange M, Karl J, Paik YK. Potential Regulatory Role of Human-Carboxylesterase-1 Glycosylation in Liver Cancer Cell Growth. J Proteome Res. 2020 Dec 4;19(12):4867-4883. doi: 10.1021/acs.jproteome.0c00787. Epub 2020 Nov 18. PMID: 33206527.

D) How many PE1-found MPs since HUPO-2017 has your chromosome group reported in papers?
- 0 protein (see F entry below)

E) How many PE1-found MPs since HUPO-2020 (2020-01-17) are now in NeXt-Prot as PE1 proteins? Please check each of your MPs that you reported in the JPR SI.
- According to the current version of neXtProt DB (2021-02-15), all of 7 target proteins became PE1. We don't want to claim them as candidate MPs because they were one-hit wonder in our studies (see F entry below).

F) How many candidate MPs found, but not meeting the guidelines 3.0? (Please state number of peptides identified, their length, and biological replicates found in).

neXtProt Acc.	Guideline 3.0	PE (2021.2.15)	Comment
NX_Q9BYX7	Not satisfied, One-hit wonder	5	One of two peptides (SSVEKSYELPDGQVITIGNER) has a part of known variants of PE1
NX_Q8NGC7	Not satisfied, One-hit wonder	2	

G) Any significant clinical or other successes re a MP that you wish us to consider highlighting in the report.
- None

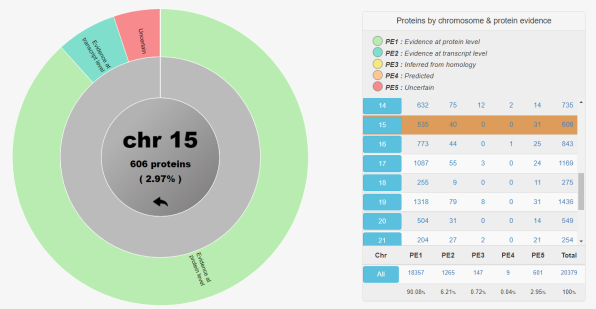
Chromosome Number: 15

PIC Gilberto B Domont
Co-Chair Fabio CS Nogueira

Part I: Missing Proteins: neXt-MP50 Challenge

Major lab members or partners contributing to the neXt-MP50 Challenge
Lund University: Lazaro Betancourt, Aniel Sanchez, Jeovanis Gil, Gyorgy Marko-Varga
Federal University of Rio de Janeiro: Jessica Guedes, Natalia P Almeida, Patricia Sosa, Maurício Quinões,

Status of the Chromosome "parts list" for your Chromosome:



A) Titles and authors of papers submitted to the 2021 JPR SI. What is their publciation status.
NONE

B) Titles and authors of papers published in the 2020 JPR SI.

NONE
C) Titles and authors of other HPP relevant papers submitted elsewhere in 2020/2021.
Novel Functional Proteins Coded by the Human Genome Discovered in Metastases of Melanoma Patients
Aniel Sanchez, *et al.*
Cell. Biol.Toxicol. (2020) 36: 261-272
9 MPs identified already in neXtProt + 24 for validation

The Human Melanoma Proteome Atlas – Complementing the Melanome Transcriptome
Lazaro H Betancourt et al.
Clin Transl Med. (2021) 11: e451 doi: 10.1002/ctm2.451
Reports 26 "new missing proteins": 3 MPs that match HPP Guideline 3.0. plus 19 also identified as transcripts in melanoma tumor samples, plus 4 candidates.

The Human Melanoma Proteome Atlas – Defining the Molecular Pathology
Lazaro H Betancourt et al.
Clin Transl Med. (2021) 11: e473 . doi: 10.1002/ctm2.473.

D) How many PE1-found MPs since HUPO-2017 has your chromosome group reported in papers?
12 (twelve)

E) How many PE1-found MPs since HUPO-2020 are now in neXt-Prot as PE1 proteins? Please check each of your MPs that you reported in the JPR SI.

6 (six)
F) How many candidate MPs found, but not meeting the guidelines 3.0? (Please state number of peptides identified, their length, and biological replicates found in).
47 candidates.

Chromosome Number: 16

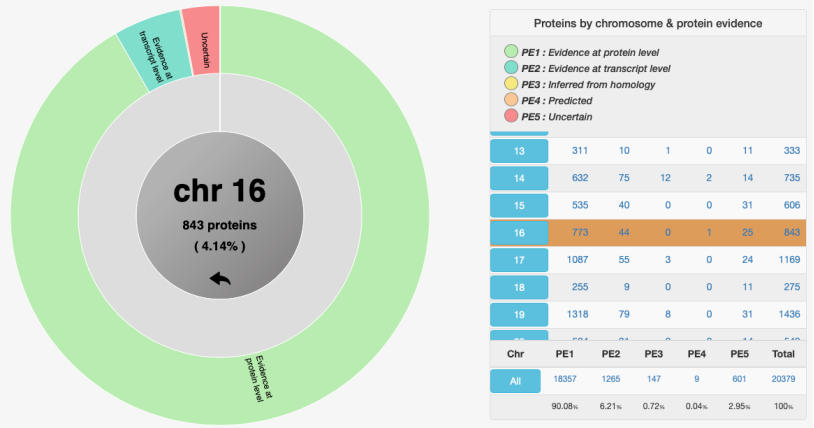
PIC Leaders:

Part I: Missing Proteins: next-MP50 Challenge

Major lab members or partners contributing to the neXt-MP50 Challenge
CNB-CSIC (FJ Corrales, A Paradela), UCM (Concha Gil), INIBIC (F. Blanco, C Ruiz), CIB-CSIC (I Casal), CBMSO-CSIC (A Marina), CNIO (J. Muñoz), CNIC (J Vázquez), IIBB (J Abian, M Carrscal), PCB (E Oliveira), IRB (M Vilaseca), CIMA (V. Segura), CRG (E Sabido), VHIO (F Canals), UV (MM Sánchez del Pino), CIC-USAL (M Fuentes), CIC bioGUNE (F Elortza), UPV (JM Arizmendi), Navarrabiomed (J Fernández, E Santamaría), IJC (C de la Torre), HNP (ME González Barderas), FJD (G Álvarez Llamas), IACS (I Orera), IPBLN-CSIC (J Sancho).

Status of the Chromosome "parts list" for your Chromosome:

(https://www.nextprot.org/about/protein-existence)



A) Titles and authors of papers submitted to the **2021** JPR SI or planned.
B) Titles and authors of papers published in the **2020** JPR SI.
UPEFinder: A Bioinformatic Tool for the Study of Uncharacterized Proteins Based on Gene Expression Correlation and the PageRank Algorithm
José González-Gomariz, Guillermo Serrano, Carlos M. Tilve-Álvarez, Fernando J. Corrales, Elizabeth Guruceaga*, and Victor Segura*
Journal of Proteome Research 2020, 19, 12, 4795-4807 (Article)
Publication Date (Web):November 6, 2020

Smelling the Dark Proteome: Functional Characterization of PITH Domain-Containing Protein 1 (C1orf128) in Olfactory Metabolism
Mercedes Lachén-Montes, Naroa Mendizuri, Karina Ausín, Alberto Pérez-Mediavilla, Mikel Azkargorta, Ibon Iloro, Felix Elortza, Hiroyuki Kondo, Izumi Ohigashi, Isidre Ferrer, Rafael de la Torre, Patricia Robledo, Joaquín Fernández-Irigoyen, and Enrique Santamaría*
Journal of Proteome Research 2020, 19, 12, 4826-4843 (Article)ACS AuthorChoice
Publication Date (Web):November 13, 2020

C) Titles and authors of other HPP relevant papers submitted elsewhere in 2020/2021.

These are papers more related to B/D initiatives that we coordinate re Liver, Infectious and Rheumatic-autoimmune. Feel free to consider them or not since they are not directly related to C-HPP.

A Proteomic Approach for Systematic Mapping of Substrates of Human Deubiquitinating Enzymes.

Ramirez J, Prieto G, Olazabal-Herrero A, Borràs E, Fernandez-Vigo E, Alduntzin U, Osinalde N, Beaskoetxea J, Lectez B, Aloria K, Rodriguez JA, Paradela A, Sabidó E, Muñoz J, **Corrales F**, Arizmendi JM, Mayor U.Int J Mol Sci. 2021 May 3;22(9):4851. doi: 10.3390/ijms22094851.PMID: 34063716

Management practices for placenta accreta spectrum patients: a Latin American hospital survey.
Nieto-Calvache AJ, Palacios-Jaraquemada JM, Hidalgo A, Vergara-Galliadi LM, Cortés Charry R, Aguilera Daga LR, Verástegui Goyzueta R, Osanan G, Fernandez J, **Corrales F**, Mereci W, Yuen-Chon V, Guevara E, Zúñiga LA, Girón, Turcios FE, Muñoz H, Perez AM, Meade P, Basanta N, Pineda JP.J Matern Fetal Neonatal Med. 2021 Apr 11:1-8. doi: 10.1080/14767058.2021.1906858. Online ahead of print.PMID: 33843408

Long-Term Liver Expression of an Apolipoprotein A-I Mimetic Peptide Attenuates Interferon-Alpha-Induced Inflammation and Promotes Antiviral Activity.
Fernandez-Sendin M, Di Trani CA, Bella A, Vasquez M, Ardaiz N, Gomar C, Arrizabalaga L, Ciordia S, **Corrales FJ**, Aranda F, Berraondo P.Front Immunol. 2021 Feb 23;11:620283. doi: 10.3389/fimmu.2020.620283. eCollection 2020.PMID: 33708194

Quantitative proteomics-based analyses performed on pre-eclampsia samples in the 2004-2020 period: a systematic review.
Navajas R, **Corrales F**, Paradela A.Clin Proteomics. 2021 Jan 26;18(1):6. doi: 10.1186/s12014-021-09313-1.PMID: 33499801

UPEFinder: A Bioinformatic Tool for the Study of Uncharacterized Proteins Based on Gene Expression Correlation and the PageRank Algorithm.
González-Gomariz J, Serrano G, Tilve-Álvarez CM, **Corrales FJ**, Guruceaga E, Segura V.J Proteome Res. 2020 Dec 4;19(12):4795-4807. doi: 10.1021/acs.jproteome.0c00364. Epub 2020 Nov 6.PMID: 33155801

Why Does COVID-19 Affect Patients with Spinal Cord Injury Milder? A Case-Control Study: Results from Two Observational Cohorts.
Calvo E, Corbacho-Alonso N, Sastre-Oliva T, Nuñez E, Baena-Galan P, Hernandez-Fernandez G, Rodriguez-Cola M, Jimenez-Velasco I, **Corrales FJ**, Gambarrutta-Malfati C, Gutierrez-Henares F, Lopez-Dolado E, Gil-Agudo A, Vazquez J, Mourino-Alvarez L, Barderas MG.J Pers Med. 2020 Oct 21;10(4):182. doi: 10.3390/jpm10040182.PMID: 33096722

Proteomics Insights Into the Molecular Basis of SARS-CoV-2 Infection: What We Can Learn From the Human Olfactory Axis.
Lachén-Montes M, **Corrales FJ**, Fernández-Irigoyen J, Santamaría E.Front Microbiol. 2020 Sep 22;11:2101. doi: 10.3389/fmicb.2020.02101. eCollection 2020.PMID: 33071996

A high-stringency blueprint of the human proteome.
Adhikari S, Nice EC, Deutsch EW, Lane L, Omenn GS, Pennington SR, Paik YK, Overall CM, **Corrales FJ**, Cristea IM, Van Eyk JE, Uhlén M, Lindskog C, Chan DW, Bairoch A, Waddington JC, Justice JL, LaBaer J, Rodriguez H, He F, Kostrzewa M, Ping P, Gundry RL, Stewart P, Srivastava S, Srivastava S, Nogueira FCS, Domont GB, Vandenbrouck Y, Lam MPY, Wennersten S, Vizcaino JA, Wilkins M, Schwenk JM, Lundberg E, Bandeira N, Marko-Varga G, Weintraub ST, Pineau C, Kusebauch U, Moritz RL, Ahn SB, Palmblad M, Snyder MP, Aebersold R, Baker MS.Nat Commun. 2020 Oct 16;11(1):5301. doi: 10.1038/s41467-020-19045-9.PMID: 33067450

Digging deeper into bile proteome.
Ciordia S, Alvarez-Sola G, Rullán M, Urman JM, Ávila MA, **Corrales FJ.**J Proteomics. 2021 Jan 6;230:103984. doi: 10.1016/j.jprot.2020.103984. Epub 2020 Sep 12.PMID: 32932008

Research on the Human Proteome Reaches a Major Milestone: >90% of Predicted Human Proteins Now Credibly Detected, According to the HUPO Human Proteome Project.
Omenn GS, Lane L, Overall CM, Cristea IM, **Corrales FJ**, Lindskog C, Paik YK, Van Eyk JE, Liu S, Pennington SR, Snyder MP, Baker MS, Bandeira N, Aebersold R, Moritz RL, Deutsch EW.J Proteome Res. 2020 Dec 4;19(12):4735-4746. doi: 10.1021/acs.jproteome.0c00485. Epub 2020 Oct 19.PMID: 32931287

Pilot Multi-Omic Analysis of Human Bile from Benign and Malignant Biliary Strictures: A Machine-Learning Approach.

Urman JM, Herranz JM, Uriarte I, Rullán M, Oyón D, González B, Fernandez-Urién I, Carrascosa J, Bolado F, Zabalza L, Arechederra M, Alvarez-Sola G, Colyn L, Latasa MU, Puchades-Carrasco L, Pineda-Lucena A, Iraburu MJ, Iruarrizaga-Lejarreta M, Alonso C, Sangro B, Purroy A, Gil I, Carmona L, Cubero FJ, Martínez-Chantar ML, Banales JM, Romero MR, Macias RIR, Monte MJ, Marín JJG, Vila JJ, **Corrales FJ**, Berasain C, Fernández-Barrena MG, Avila MA. Cancers (Basel). 2020 Jun 21;12(6):1644. doi: 10.3390/cancers12061644.PMID: 32575903

The COVID-19 MS Coalition-accelerating diagnostics, prognostics, and treatment.
Struwe W, Emmott E, Bailey M, Sharon M, Sinz A, **Corrales FJ**, Thalassinos K, Braybrook J, Mills C, Barran P; COVID-19 MS Coalition.Lancet. 2020 Jun 6;395(10239):1761-1762. doi: 10.1016/S0140-6736(20)31211-3. Epub 2020 May 27.PMID: 32473097

Analytical techniques for multiplex analysis of protein biomarkers.
Van Gool A, **Corrales F**, Čolović M, Krstić D, Oliver-Martos B, Martínez-Cáceres E, Jakasa I, Gajski G, Brun V, Kyriacou K, Burzynska-Pedziwiatr I, Wozniak LA, Nierkens S, Pascual García C, Katrlik J, Bojic-Trbojevic Z, Vacek J, Llorente A, Antohe F, Suica V, Suarez G, t'Kindt R, Martin P, Penque D, Martins IL, Bodoki E, Iacob BC, Aydindogan E, Timur S, Allinson J, Sutton C, Luider T, Wittfooth S, Sammar M.Expert Rev Proteomics. 2020 Apr;17(4):257-273. doi: 10.1080/14789450.2020.1763174. Epub 2020 May 19.PMID: 32427033

Mesenchymal Stem Cell-Derived Extracellular Vesicle Isolation and Their Protein Cargo Characterization.
Morente-López M, Fafián-Labora JA, Carrera M, de Toro FJ, **Gil C**, Mateos J, Arufe MC.Methods Mol Biol. 2021;2259:3-12. doi: 10.1007/978-1-0716-1178-4_1.PMID: 33687705

Vultures from different trophic guilds show distinct oral pathogenic yeast signatures and co-occurrence networks.
Pitarch A, **Gil C**, Blanco G.Sci Total Environ. 2020 Jun 25;723:138166. doi: 10.1016/j.scitotenv.2020.138166. Epub 2020 Mar 23.PMID: 32224410

Multiomics Substrates of Resistance to Emerging Pathogens? Transcriptome and Proteome Profile of a Vancomycin-Resistant *Enterococcus faecalis* Clinical Strain.
Pinto L, Torres C, **Gil C**, Santos HM, Capelo JL, Borges V, Gomes JP, Silva C, Vieira L, Poeta P, Igrejas G.OMICS. 2020 Feb;24(2):81-95. doi: 10.1089/omi.2019.0164.PMID: 32073998

Nucleic Acid Programmable Protein Arrays (NAPPA) for the Discovery of Autoantibodies in Osteoarthritis.
Lourido L, Camacho-Encina M, **Blanco FJ**, **Ruiz-Romero C.**Methods Mol Biol. 2021;2344:181-190. doi: 10.1007/978-1-0716-1562-1_13.PMID: 34115360

A clinical model including protein biomarkers predicts radiographic knee osteoarthritis: a prospective study using data from the Osteoarthritis Initiative.
Lourido L, Balboa-Barreiro V, **Ruiz-Romero C**, Rego-Pérez I, Camacho-Encina M, Paz-González R, Calamia V, Oreiro N, Nilsson P, **Blanco FJ.**Osteoarthritis Cartilage. 2021 Apr 30:S1063-4584(21)00707-X. doi: 10.1016/j.joca.2021.04.011. Online ahead of print.PMID: 33933586

Serum Proteomic Profiling in Rheumatoid Arthritis by Antibody Suspension Bead Arrays.
Lourido L, Paz-González R, **Ruiz-Romero C**, Nilsson P, **Blanco FJ.**Methods Mol Biol. 2021;2259:143-151. doi: 10.1007/978-1-0716-1178-4_8.PMID: 33687712

Identification of a distinct lipidomic profile in the osteoarthritic synovial membrane by mass spectrometry imaging.
Rocha B, Cillero-Pastor B, **Ruiz-Romero C**, Paine MRL, Cañete JD, Heeren RMA, **Blanco FJ.**Osteoarthritis Cartilage. 2021 May;29(5):750-761. doi: 10.1016/j.joca.2020.12.025. Epub 2021 Feb 11.PMID: 33582239

Association of serum anti-centromere protein F antibodies with clinical response to infliximab in patients with rheumatoid arthritis: A prospective study.
Lourido L, **Ruiz-Romero C**, Picchi F, Diz-Rosales N, Vilaboa-Galán S, Fernández-López C, Tasende JAP, Pérez-Pampín E, Regueiro C, Mera-Varela A, Gonzalez A, Hambardzumyan K, Saevarsdottir S, Nilsson P, **Blanco FJ.**Semin Arthritis Rheum. 2020 Oct;50(5):1101-1108. doi: 10.1016/j.semarthrit.2020.06.010. Epub 2020 Jun 30.PMID: 32920323

Integrative Metabolic Pathway Analysis Reveals Novel Therapeutic Targets in Osteoarthritis.

Rocha B, Cillero-Pastor B, Eijkel G, Calamia V, Fernandez-Puente P, Paine MRL, **Ruiz-Romero C**, Heeren RMA, **Blanco FJ.**Mol Cell Proteomics. 2020 Apr;19(4):574-588. doi: 10.1074/mcp.RA119.001821. Epub 2020 Jan 24.PMID: 31980557

D) How many PE1-found MPs since HUPO-2017 has your chromosome group reported in papers?
Datasets from human studies have been carefully analyzed for MPs and uPE proteins but no reliable hit was found.

E) How many PE1-found MPs since HUPO-2019 are now in NeXt-Prot as PE1 proteins? Please check each of your MPs that you reported in the JPR SI.

F) How many candidate MPs found, but not meeting the guidelines 3.0? (Please state number of peptides identified, their length, and biological replicates found in).

G) Any significant clinical or other successes re a MP that you wish us to consider highlighting in the report.

Chromosome Number: 17

PIC Leaders: Gil Omenn & Mike Snyder

Part I: Missing Proteins: neXt-MP50 Challenge

Major lab members or partners contributing to the neXt-MP50 Challenge

Status of the Chromosome "parts list" for your Chromosome:

(https://www.nextprot.org/about/protein-existence)

A) Titles and authors of papers submitted to the **2021** JPR SI. What is their publication status?
JPR, other than SI:
Huang X, Zhang C, Pearce R, **Omenn GS**, Zhang Y. Identifying the zoonotic origin of SARS-CoV-2 by modeling the binding affinity between the spike receptor-binding domain and host ACE2. J Proteome Res. 2020 Dec 4;19(12):4844-4856. PMID: 33175551; PMCID: PMC7770890.
Zhang C, Zheng W, Cheng M, **Omenn GS**, Freddolino PL, Zhang Y. Functions of essential genes and a scale-free protein interaction network revealed by structure-based function and interaction prediction for a minimal genome. J Proteome Res. 2021 Feb 5;20(2):1178-1189. PMID: 33393786; PMCID: PMC7867644.

B) Titles and authors of papers published in the **2020** JPR SI.
Omenn GS, Lane L, Overall CM, Cristea IM, Corrales FJ, Lindskog C, Paik YK, Van Eyk JE, Liu S, Pennington SR, Snyder MP, Baker MS, Bandeira N, Aebersold R, Moritz RL, Deutsch EW. Research on the Human Proteome Reaches a Major Milestone: >90% of Predicted Human Proteins Now Credibly Detected, according to the HUPO Human Proteome Project. J Proteome Res. 2020 Dec 4;19(12):4735-4746. PMID: 32931287; PMCID: PMC7718309.

C) Titles and authors of other HPP relevant papers submitted elsewhere in 2020/2021.
Adhikari S, Nice EC, Deutsch EW, Lane L, **Omenn GS**, Pennington SR, Paik YK, Overall CM, Corrales FJ, Cristea IM, Van Eyk JE, Uhlén M, Lindskog C, Chan DW, Bairoch A, Waddington JC, Justice JL, LaBaer J, Rodriguez H, He F, Kostrzewa M, Ping P, Gundry RL, Stewart P, Srivastava S, Srivastava S, Nogueira FCS, Domont GB, Vandenbrouck Y, Lam MPY, Wennersten S, Vizcaino JA, Wilkins M, Schwenk JM, Lundberg E, Bandeira N, Marko-Varga G, Weintraub ST, Pineau C, Kusebauch U, Moritz RL, Ahn SB, Palmblad M, Snyder MP, Aebersold R, Baker MS. A high-stringency blueprint of the human proteome. Nat Commun. 2020 Oct 16;11(1):5301. PMID: 33067450; PMCID: PMC7568584.

Mann SP, Treit PV, Geyer PE, **Omenn GS**, Mann M. Ethical principles, constraints and opportunities in clinical proteomics. Mol Cell Proteomics. 2021 Jan 14;20:100046. Epub ahead of print. PMID: 33453411; PMCID: PMC7950205.

Omenn GS. Reflections on the HUPO Human Proteome Project, the flagship project of the Human Proteome Organization, at 10 years. Mol Cell Proteomics. 2021 Feb 25:100062. Epub ahead of print. PMID: 33640492.
Li HD, Yang C, Zhang Z, Yang M, Wu FX, **Omenn GS**, Wang J. IsoResolve: Predicting splice isoform functions by integrating gene and isoform-level features with domain adaptation. Bioinformatics. 2020 Sep 23:btaa829. doi: 10.1093/bioinformatics/btaa829. PMID: 32966552.

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Huang X, Pearce R, **Omenn GS**, Zhang Y. Computationally assessing the potency of 13 guanidinobenzoyl- or aminidinobenzoyl-containing drugs to inhibit TMPRSS2 for COVID-19 treatment. International J Molecular Science, 2021 Jun30; 11:7060; PMID:34209110

Satpathy S, Krug K, Jean Beltran PM, Savage SR, Petralia F, Kumar-Sinha C, Dou Y, Reva B, Kane MH, Avanessian SC, Vasaikar SV, Krek A, Lei JT, Jaehnig EJ, Omelchenko T, Geffen Y, Bergstrom EJ, Stathias V, Christianson KE, Heiman DI, Cieslik MP, Cao S, Song X, Ji J, Liu W, Li K, Wen B, Li Y, Gümüş ZH, Selvan ME, Soundararajan R, Visal TH, Raso MG, Parra ER, Babur Ö, Vats P, Anand S, Schraink T, Cornwell M, Rodrigues FM, Zhu H, Mo CK, Zhang Y, da Veiga Leprevost F, Huang C, Chinnaiyan AM, Wyczalkowski MA, **Omenn GS**, Newton CJ, Schurer S, Ruggles KV, Fenyö D, Jewell SD, Thiagarajan M, Mesri M, Rodriguez H, Mani SA, Udeshi ND, Getz G, Suh J, Li QK, Hostetter G, Paik PK, Dhanasekaran SM, Govindan R, Ding L, Robles AI, Clauser KR, Nesvizhskii AI, Wang P, Carr SA, Zhang B, Mani DR, Gillette MA; Clinical Proteomic Tumor Analysis Consortium. A proteogenomic portrait of lung squamous cell carcinoma. Cell. 2021 Aug 5;184(16):4348-4371.e40. doi: 10.1016/j.cell.2021.07.016.PMID: 34358469

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D) How many PE1-found MPs since HUPO-2017 has your chromosome group reported in papers?

E) How many PE1-found MPs since HUPO-2020 are now in neXt-Prot as PE1 proteins? Please check each of your MPs that you reported in the JPR SI.

F) How many candidate MPs found, but not meeting the guidelines 3.0? (Please state number of peptides identified, their length, and biological replicates found in).

G) Any significant clinical or other successes re a MP that you wish us to consider highlighting in the report.

Chromosome Number: 18

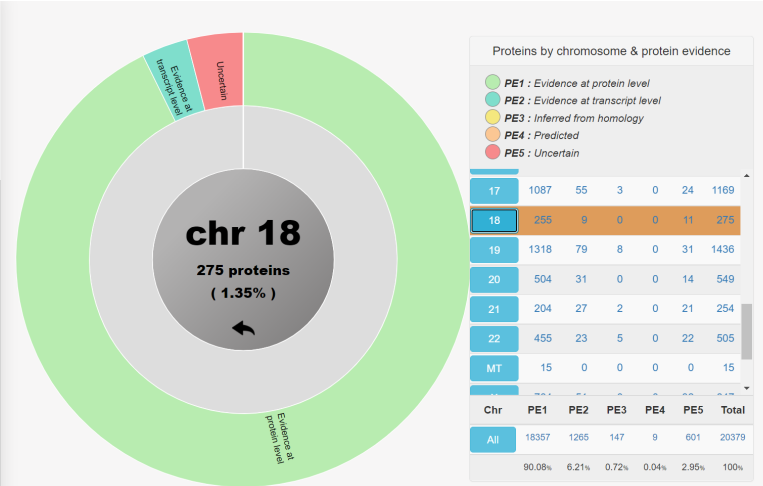
PIC Leaders:

Alexander Archakov
Elena Ponomarenko (bioinformatics), Andrey Lisitsa (standardization)
Part I: Missing Proteins: next-MP50 Challenge

Major lab members or partners contributing to the neXt-MP50 Challenge
Katerina V. Poverennaya, Ekaterina V. Ilgisonis, Olga I. Kiseleva, Sergey P. Radko, Mikhail V. Gorshkov, Stanislav N. Naryzhny, Elena S. Zorina, Svetlana S. Novikova, Arthur T. Kopylov, Olga V. Tikhonova, Eugene N. Nikolaev, Victor G. Zgoda

Status of the Chromosome "parts list" for your Chromosome:

(https://www.nextprot.org/about/protein-existence)



A) Titles and authors of papers submitted to the **2021** JPR SI or planned.

1) **Number of detected proteins as the function of the sensitivity of proteomic technology in human liver cells:** Archakov A.I., Ilgisonis E.V., Lisitsa A.V., Ponomarenko E.A., Vavilov N.E., Zgoda V.G.
2) **Evolution of protein functional annotation: text mining study** Ekaterina V. Ilgisonis, Pavel Pogodin, Olga I. Kiseleva, Elena A. Ponomarenko

B) Titles and authors of papers published in the **2020** JPR SI.

Proteomic Analysis of Chr 18 Proteins Using 2D Fractionation
Nikita E. Vavilov, Victor G. Zgoda, Olga V. Tikhonova, Tatiana E. Farafonova, Natalya A. Shushkova, Svetlana E. Novikova, Konstantin N. Yarygin, Sergey P. Radko, Ekaterina V. Ilgisonis, Elena A. Ponomarenko, Andrey V. Lisitsa, and Alexander I. Archakov

C) Titles and authors of other HPP relevant papers submitted elsewhere in 2020/2021.

Savosina P., Karasev D., Veselovsky A., Miroshnichenko Yu., Sobolev B., Functional and structural features of proteins associated with alternative splicing, International Journal of Biological Macromolecules, 2020, 147, 513-520

Radko S., Ptitsyn K., Novikova S., Kiseleva Y., Moysa A., Kurbatov L., Mannanova M., Zgoda V., Ponomarenko E., Lisitsa A., Archakov A., Evaluation of Aptamers as Affnity Reagents for an Enhancement of SRM-Based Detection of Low-Abundance Proteins in Blood Plasma, Biomedicines, 2020, 8(5), 133

Kiseleva O., Zgoda V., Naryzhny S., Poverennaya E., Empowering Shotgun Mass Spectrometry with 2DE: A HepG2 Study, International Journal of Molecular Sciences, 2020, 21(11), 3813

Poverennaya E., Kiseleva O., Romanova A., Pyatnitskiy M., Predicting Functions of Uncharacterized Human Proteins: From Canonical to Proteoforms, Genes, 2020, 1(6), 677

D) How many PE1-found MPs since HUPO-2017 has your chromosome group reported in papers?
No PE1-found MPs since HUPO-2017 were detected by chr18 team.

E) How many PE1-found MPs since HUPO-2019 are now in NeXt-Prot as PE1 proteins? Please check each of your MPs that you reported in the JPR SI.

Since HUPO-2019 two (B2RU33 and Q9BXX2) MPs were found at PE1. We confirm.

F) How many candidate MPs found, but not meeting the guidelines 3.0? (Please state number of peptides identified, their length, and biological replicates found in).
1 protein, 1 peptide, 1 replicate.

UniProt AC	Gene name	PE	Peptide	Sample
Q9BXX2	ANKRD30B	PE2	GPEPPNPFSERVYTEK	Prostate biopsy

G) Any significant clinical or other successes re a MP that you wish us to consider highlighting in the report.

We're developing and modifying the experimental protocol of missing protein isolation using gene editing. We are improving experimental protocol, based on 2D fractionating, because it showed promising results in HepG2 protein profiling (Vavilov N.E., Zgoda V.G., Tikhonova O.V., Farafonova T.E., Shushkova N.A., Novikova S.E., Yarygin K.N., Radko S.P., Ilgisonis E.V., Ponomarenko E.A., Lisitsa A.V., Archakov A.I., Proteomic Analysis of Chr 18 Proteins Using 2D Fractionation, Journal of Proteome Research, 2020, 19(12), 4901–4906).

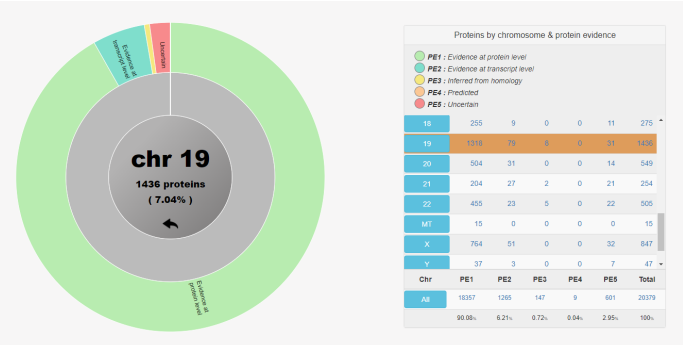
Chromosome Number: 19

PIC Leaders: Sergio Encarnación-Guevara

Part I: Missing Proteins: neXt-MP50 Challenge

Major lab members or partners contributing to the neXt-MP50 Challenge
Orlando Morales-Tarré, Magdalena Hernández-Ortiz, Ramiro Alonso, María del Carmen Vargas-Lagunas, Nilda del Carmen Sánchez, Gloria Angelina Herrera Quiterio, Victor Emmanuel Osio Becerro. Proteomics Laboratory at Center for Genomic Sciences UNAM.
Jeovanis Gil-Valdes. Lund University, Department of Clinical Sciences
Julio Collado-Vides, Program of Computational Biology at Center for Genomic Sciences UNAM.
Alejandro García Carranca, National Cancer Institute
Osbaldo Resendis Antonio, National Institute of Genomic Medicine.
Guadalupe Ayala, National Institute of Public Health

Status of the Chromosome "parts list" for your Chromosome:



A) Titles and authors of papers submitted to the **2021** JPR SI. What is their publication status.
None

B) Titles and authors of papers published in the **2020** JPR SI.
None

C) Titles and authors of other HPP relevant papers submitted elsewhere in 2020/2021.

Gómez-Caudillo L, Ortega-Lozano AJ, Martínez-Batallar ÁG, Rosas-Vargas H, Minauro-Sanmiguel F, Encarnación-Guevara S. Principal component analysis on LC-MS/MS and 2DE-MALDI-TOF in glioblastoma cell lines reveals that mitochondria act as organelle sensors of the metabolic state in glioblastoma. Oncol Rep. 2020 Aug;44(2):661-673. doi: 10.3892/or.2020.7625. Epub 2020 May 27.

Sanchez A, Kuras M, Murillo JR, Pla I, Pawlowski K, Szasz AM, Gil J, et al., Novel functional proteins coded by the human genome discovered in metastases of melanoma patients. Cell Biol Toxicol. 2020 Jun;36(3):261-272. doi: 10.1007/s10565-019-09494-4. Epub 2019 Oct 10.

D) How many PE1-found MPs since HUPO-2017 has your chromosome group reported in papers?
Three

E) How many PE1-found MPs since HUPO-2020 are now in neXt-Prot as PE1 proteins? Please check each of your MPs that you reported in the JPR SI.
One

F) How many candidate MPs found, but not meeting the guidelines 3.0? (Please state number of peptides identified, their length, and biological replicates found in).

We are still working on the validation through the use of synthetic peptides of the 4 proteins listed in the following table, these proteins were identified in the indicated cell lines, under conditions of inhibition of sirtuin 1 by compound EX527.

ID	Gene name	Peptide	Sample Identified
Q8IVC4	ZNF584	KDALVLHQR	SiHa EX527
		VIQHQDTHSEGKPR	SiHa EX527
P52742	ZNF135	IHTGEKPYKCTQCGR	CaLo CTRL
		IHTGEKPYECNQCGR	SiHa EX527
Q8WXB4	ZNF606	IHTGEKPYKCNQCER	SiHa EX527
		SALTKHER	SiHa EX527
		THTGEKPYR	HaCaT CTRL
Q9Y2G7	ZFP30	EKPYECGECGKAFR	SiHa EX527
		IKSCGLEEQESPHEVCFR	SiHa EX527

G) Any significant clinical or other successes re a MP that you wish us to consider highlighting in the report.

Currently, we are using chemical inhibition and siRNA silencing of epigenetic enzymes such as lysine deacetylases (KDACs), DNA histones and methyltransferases in different cancer models, looking for the adequate expression conditions of MPs, our current emphasis is the analysis of proteins belonging to cell membranes and secretomes (exosomes). The next stage will involve a large-scale proteomic analysis to identify and validate the presence of MPs using synthetic peptides. Some of these proteins will be selected to study their function in cancer cells.

Chromosome Number: X

PIC Leaders:
Yasushi Ishihama (PI)
Tadashi Yamamoto (co-PI)

Part I: Missing Proteins: neXt-MP50 Challenge

Major lab members or partners contributing to the neXt-MP50 Challenge

- **TEAM A:** Yasushi Ishihama, Norie Araki, Susumu Goto, Shin Kawano, Masaki Matsumoto, Shujiro Okuda, Naoyuki Sugiyama, Akiyasu C Yoshizawa, Mio Iwasaki, Tsuyoshi Tabata, Yoshinori Yamanouchi, Yuki Moriya, Daiki Kobayashi, Atsushi Hatano, Tomoyo Takami
- **Team B:** Tadashi Yamamoto, Yoshitoshi Hirao, Tomohiro Uchimoto, Keiko Yamamoto, Yanagita Kengo

Status of the Chromosome "parts list" for your Chromosome:

(https://www.nextprot.org/about/protein-existence)

A) Titles and authors of papers submitted to the **2021** JPR SI. What is their publciation status.
None

B) Titles and authors of papers published in the **2020** JPR SI.
None

C) Titles and authors of other HPP relevant papers submitted elsewhere in 2020/2021.

1. Combinatorial analysis of translation dynamics reveals eIF2 dependence of translation initiation at near-cognate codons. Ichihara K, Matsumoto A, Nishida H, Kito Y, Shimizu H, Shichino Y, Iwasaki S, Imami K, Ishihama Y, Nakayama KI. Nucleic Acids Res. 2021 Jul 21;49(13):7298-7317. doi: 10.1093/nar/gkab549.
2. Exploring the landscape of ectodomain shedding by quantitative protein terminomics. Tsumagari K, Chang CH, Ishihama Y. iScience. 2021 Mar 2;24(4):102259. doi: 10.1016/j.isci.2021.102259. eCollection 2021 Apr 23.
3. Aberrant splicing isoforms detected by full-length transcriptome sequencing as transcripts of potential neoantigens in non-small cell lung cancer. Oka M, Xu L, Suzuki T, Yoshikawa T, Sakamoto H, Uemura H, Yoshizawa AC, Suzuki Y, Nakatsura T, Ishihama Y, Suzuki A, Seki M. Genome Biol. 2021 Jan 4;22(1):9. doi: 10.1186/s13059-020-02240-8.
4. Isolation of Acetylated and Unmodified Protein N-Terminal Peptides by Strong Cation Exchange Chromatographic Separation of TrypN-Digested Peptides. Chang CH, Chang HY, Rappsilber J, Ishihama Y. Mol Cell Proteomics. 2020 Nov 24;20:100003. doi: 10.1074/mcp.TIR120.002148.
5. Sequence-Specific Model for Predicting Peptide Collision Cross Section Values in Proteomic Ion Mobility Spectrometry. Chang CH, Yeung D, Spicer V, Ogata K, Krokhin O, Ishihama Y. J Proteome Res. 2021 Jun 16. doi: 10.1021/acs.jproteome.1c00185.
6. The jPOST Repository as a Public Data Repository for Shotgun Proteomics. Watanabe Y, Yoshizawa AC, Ishihama Y, Okuda S. Methods Mol Biol. 2021;2259:309-322. doi: 10.1007/978-1-0716-1178-4_20.
7. The ProteomeXchange consortium in 2020: enabling 'big data' approaches in proteomics. Deutsch EW, Bandeira N, Sharma V, Perez-Riverol Y, Carver JJ, Kundu DJ, García-Seisdedos D, Jarnuczak AF, Hewapathirana S, Pullman BS, Wertz J, Sun Z, Kawano S, Okuda S, Watanabe Y, Hermjakob H, MacLean B, MacCoss MJ, Zhu Y, Ishihama Y, Vizcaíno JA. Nucleic Acids Res. 2020 Jan 8;48(D1):D1145-D1152. doi: 10.1093/nar/gkz984.

D) How many PE1-found MPs since HUPO-2017 has your chromosome group reported in papers?
29 Proteins out of 41 MPs, we found in GPM, are now PE=1 in NeXtProt 2021.

E) How many PE1-found MPs since HUPO-2020 are now in neXt-Prot as PE1 proteins? Please check each of

your MPs that you reported in the JPR SI.
9

F) How many candidate MPs found, but not meeting the guidelines 3.0? (Please state number of peptides identified, their length, and biological replicates found in).
NX_Q9NS67-1 3 pepts 13,16,10, n=1
NX_Q9H354-1 1 pept 16, n=1
NX_Q9BZK3-1 2 pepts 14, 15, n=1
NX_Q9BZ68-1 1 pept 20, n=1
NX_Q96NG8-1 1 pept 10, n=1
NX_Q92928-1 1 pept 19, n=1
NX_Q8IZP2-1 2 pepts 14, 12, n=1
NX_Q8IX06-1 6 pepts 11, 12, 11, 18, 11, 10, n=1
NX_Q6ZN08-1 1 pept 10, n=1
NX_Q58FF6-1 3 pepts 15, 21, 20, n=1
NX_P59074-1 1 pept 27, n=1
NX_I3L273-1 1 pept 12, n=1
NX_C9JQL5-1 1 pept 25, n=1
NX_A0A0J9YWL9-1 6 pepts 24, 8, 14, 15, 21, 20, n=1

Chromosome Number: Y

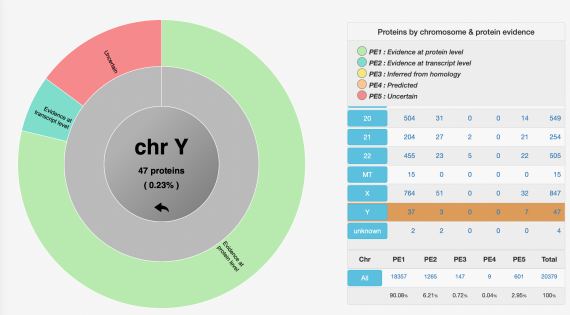
PIC Leaders: Ghasem Hosseini Salekdeh

Part I: Missing Proteins: neXt-MP50 Challenge

Major lab members or partners contributing to the neXt-MP50 Challenge

Status of the Chromosome "parts list" for your Chromosome:

(https://www.nextprot.org/about/protein-existence)



A) Titles and authors of papers submitted to the **2021** JPR SI. What is their publication status?

B) Titles and authors of papers published in the **2020** JPR SI.

Alikhani M, Karamzadeh R, Rahimi P, Adib S, Baharvand H, Salekdeh GH. Human Proteome Project and Human Pluripotent Stem Cells: Odd Bedfellows or a Perfect Match?. Journal of Proteome Research. 2020 Oct 30;19(12):4747-53.

C) Titles and authors of other HPP relevant papers submitted elsewhere in 2020/2021.

Heydari R, Jangravi Z, Maleknia S, Seresht-Ahmadi M, Bahari Z, Salekdeh GH, Meyfour A. New insights into the human Y chromosome and men's susceptibility to disease. Experimental Cell Research (under review)

Dehkordi SN, Khani F, Hassani SN, Baharvand H, Soleimanpour-Lichaei HR, Salekdeh GH. The Contribution of Y Chromosome Genes to Spontaneous Differentiation of Human Embryonic Stem Cells into Embryoid Bodies In Vitro. Cell Journal. 2021 Apr;23(1):40.

D) How many PE1-found MPs since HUPO-2017 has your chromosome group reported in papers?

E) How many PE1-found MPs since HUPO-2020 are now in neXt-Prot as PE1 proteins? Please check each of your MPs that you reported in the JPR SI.

F) How many candidate MPs found, but not meeting the guidelines 3.0? (Please state number of peptides identified, their length, and biological replicates found in).

G) Any significant clinical or other successes re a MP that you wish us to consider highlighting in the report.

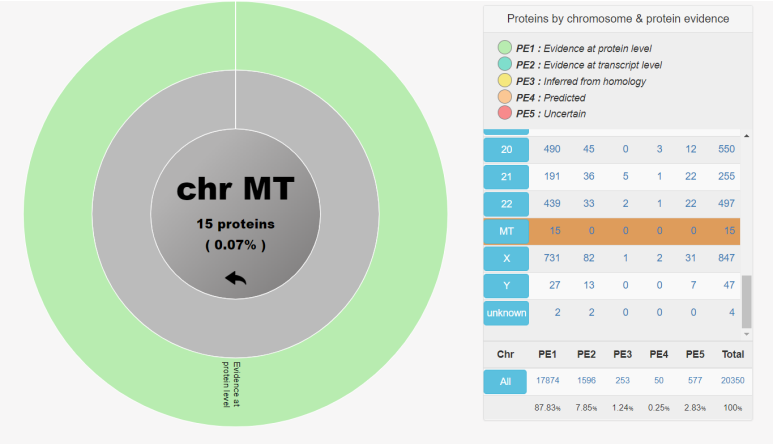
Chromosome Number: Mt

PIC Leaders: Andrea Urbani
MT-Mitochondria

Part I: Missing Proteins: neXt-MP50 Challenge

Major lab members or partners contributing to the neXt-MP50 Challenge
Andrea Urbani, Paola Roncada, Mauro Fasano, Alberio T, Pieroni L, Ronci M, Brioschi M, Cunsolo V, Foti S, Giusti L, Greco V, Lucacchini A, Soggiu A, Zilocchi M, Iavarone F, Persichilli S, Castagnola M, Ciregia F, Sickmann A, Babu M.

Status of the Chromosome "parts list" for your Chromosome:



A) Titles and authors of papers submitted to the **2021** JPR SI. What is their publciation status.
none

B) Titles and authors of papers published in the **2020** JPR SI.
none

C) Titles and authors of other HPP relevant papers submitted elsewhere in 2020/2021.
Aly KA, Moutaoufik MT, Phanse S, Zhang Q, Babu M. From fuzziness to precision medicine: on the rapidly evolving proteomics with implications in mitochondrial connectivity to rare human disease. iScience. 2021 Jan 6;24(2):102030. doi: 10.1016/j.isci.2020.102030.

Zilocchi M, Broderick K, Phanse S, Aly KA, Babu M. Mitochondria under the spotlight: On the implications of mitochondrial dysfunction and its connectivity to neuropsychiatric disorders. Comput Struct Biotechnol J. 2020 Sep 14;18:2535-2546. doi: 10.1016/j.csbj.2020.09.008.

Ferri A, Garcia-Roves PM, Pieroni L. Editorial: Mitochondrial Proteomics: Understanding Mitochondria Function and Dysfunction Through the Characterization of Their Proteome. Front Cell Dev Biol. 2020 Dec 10;8:608753. doi: 10.3389/fcell.2020.608753. eCollection 2020.

Pittalà MGG, Reina S, Cubisino S, Cucina A, Formicola B, Cunsolo V, Foti S, Saletti R, Messina A. Post-Translational Modification Analysis of VDAC1 in ALS-SOD1 Model Cells Reveals Specific Asparagine and Glutamine Deamidation Antioxidants (Basel). 2020 Dec 2;9(12):1218. doi: 10.3390/antiox9121218.

Reina S, Pittalà M, Guarino F, Messina A, De Pinto V, Foti S, Saletti R. Cysteine Oxidations in Mitochondrial Membrane Proteins: The Case of VDAC Isoforms in Mammals. Front Cell Dev Biol . 2020 Jun 4;8:397. doi: 10.3389/fcell.2020.00397. eCollection 2020.

Pittalà MGG, Saletti R, Reina S, Cunsolo V, De Pinto V, Foti S. A High Resolution Mass Spectrometry Study Reveals the Potential of Disulfide Formation in Human Mitochondrial Voltage-Dependent Anion Selective Channel Isoforms (hVDACs). Int J Mol Sci. 2020 Feb 21;21(4):1468. doi: 10.3390/ijms21041468.

Marini F, Carregari VC, Greco V, Ronci M, Iavarone F, Persichilli S, Castagnola M, Urbani A, Pieroni L. Exploring the HeLa Dark Mitochondrial Proteome. Front Cell Dev Biol. 2020 Mar 5;8:137. doi: 10.3389/fcell.2020.00137. PMID: 32195257; PMCID: PMC7066081.

Zilocchi M, Colugnat I, Lualdi M, Meduri M, Marini F, Corasolla Carregari V, Moutaoufik MT, Phanse S, Pieroni L, Babu M, Garavaglia B, Fasano M, Alberio T. Exploring the Impact of PARK2 Mutations on the Total and Mitochondrial Proteome of Human Skin Fibroblasts. Front Cell Dev Biol. 2020 Jun 11;8:423. doi: 10.3389/fcell.2020.00423. PMID: 32596240; PMCID: PMC7300190.

Zilocchi M, Moutaoufik MT, Jessulat M, Phanse S, Aly KA, Babu M. Misconnecting the dots: altered mitochondrial protein-protein interactions and their role in neurodegenerative disorders.
Expert Rev Proteomics. 2020 Feb;17(2):119-136. doi: 10.1080/14789450.2020.1723419. Epub 2020 Feb 6. PMID: 31986926

Other achievements:

"The Mitochondrial Human Proteome Project (mt-HPP) virtual day: a Mitochondrial Chromosome-centric Human Proteome Project Initiative"- July 13th 2021 Chair: Andrea Urbani, co-chair: Paola Roncada; Organizer: Viviana Greco (https://www.hupo.org/event-4407536)

Special Issue "Mitochondrial Proteomics in Neuroscience and Neurodegenerative Diseases", Int J Mol Sci. 2021. Guest editors: Viviana Greco and Luisa Pieroni.

D) How many PE1-found MPs since HUPO-2017 has your chromosome group reported in papers?
3

E) How many PE1-found MPs since HUPO-2020 are now in neXt-Prot as PE1 proteins? Please check each of your MPs that you reported in the JPR SI.
3

F) How many candidate MPs found, but not meeting the guidelines 3.0? (Please state number of peptides identified, their length, and biological replicates found in).

Protein name	Accession (Gene name)	Identified in	Chr #	Matching proteotypic Peptides	RNA cell category (HPA)	Main location (HPA)
RAS p21 protein activator 4B	NX_C9J798, RASA4B	U-2 OS, HepG2, SH-SY5Y	7	ELSGGAEAGTVPTSPGK, VVQQEEGWFR, DITGSSDPYCIVK, VSINNTGLLGSYHPGVFR, AHLGALLSALSR	Cell line enhanced (TPM U-2 OS = 16.6, TPM SH-SY5Y = 14.3)	Localized to the Cell Junctions (uncertain) In addition localized to

						the Vesicles (uncertain)
RAS p21 protein activator 4	NX_O43374, RASA4	U-2 OS, HepG2, SH-SY5Y	7	ELSGGAEAGTVPTSPGK, VVQQEEGWFR, DITGSSDPYCIVK, VSINNTGLLGSYHPGVFR, AHLGALLSALSR	Cell line enhanced (TPM U-2 OS = 23.3, TPM SH-SY5Y = 34.2)	Localized to the Vesicles (uncertain)
60S acidic ribosomal protein P0-like	NX_Q8NHW5, RPLP0P6	Hek293	2	AFLADPSAFVAAAPVAADTTAAPAAAAAPAK, FLADPSAFVAAAPVAADTTAAPAAAAAPAK	N/A	N/A
Putative keratin-87 protein	NX_A6NCN2, KRT87P	HepG2	12	KSDLEANVEALTQEIDFLR, SDLEANVEALTQEIDFLR, KSDLEANVEALTQEIDFLRR, SDLEANVEALTQEIDFLRR, LEANVEALTQEIDFLR, LASELNHVQEVLEGYK	N/A	N/A
PR domain zinc finger protein 13	NX_Q9H4Q3, PRDM13	HeLa	6	LDSGTLPPAVAAAGGTGGGGSGGSGAGKPK, AAGGTGGGGSGGSGAGKPK, VAAAGGTGGGGSGGSGAGKPK	Not detected	Not available
Protein CXorf40B	NX_Q96DE9, CXorf40B	SH-SY5Y	X	LGMTPAQIQALLR, YLTVISNPR, WLLEPIPR	Expressed in all (TPM SH-SY5Y = 19.4)	Not available

G) Any significant clinical or other successes re a MP that you wish us to consider highlighting in the report.

none

neXt-CP50 Challenge

translating the code of life

HUMAN PROTEOME ORGANIZATION

Chromosome Number: 2

Part II: uPE1 Proteins (Dark Proteins): next-CP50 Challenge

Major lab members or partners contributing to the neXt-CP50 Challenge:

Paula Duek (SIB/University of Geneva)
Amos Bairoch (SIB/University of Geneva)
Monique Zahn-Zabal (SIB)
Camille Mary (University of Geneva) *[left in September 2020]*

A) Please list the neXt-CP50 Challenge Proteins that your team is characterising and briefly describe your teams progress made to date including any publications or planned papers in 2021.

C12orf73:
Zhang S, Reljić B, Liang C, Kerouanton B, Francisco JC, Peh JH, Mary C, Jagannathan NS, Olexiouk V, Tang C, Fidelito G, Nama S, Cheng RK, Wee CL, Wang LC, Duek Roggli P, Sampath P, Lane L, Petretto E, Sobota RM, Jesuthasan S, Tucker-Kellogg L, Reversade B, Menschaert G, Sun L, Stroud DA, Ho L. (**2020**) *Mitochondrial peptide BRAWNIN is essential for vertebrate respiratory complex III assembly*. **Nat Commun**. 11(1):1312.

C21orf91:
Reiche L, Göttle P, Lane L, Duek P, Park M, Azim K, Schütte J, Manousi A, Schira-Heinen J, Küry P. (**2021**) *C21orf91 Regulates Oligodendroglial Precursor Cell Fate-A Switch in the Glial Lineage?* **Front Cell Neurosci**. 15:653075. doi: 10.3389/fncel.2021.653075. PMID: 33796011

We had to close our lab in September 2020 due to lack of funding but we will continue to assist teams by providing in silico tools and expertise to characterize uPE1s.

Duek P, Mary C, Zahn-Zabal M, Bairoch A, Lane L (**2021**) *Functionathon: a manual data mining workflow to generate functional hypotheses for uncharacterized human proteins and its application by undergraduate students*, **Database** (Oxford), baab046. doi: 10.1093/database/baab046. PMID: 34318869.

B) Your opinion on the extension of next-CP50 (2018-2021).
Due to the nature of protein characterization work, which requires a lot more extra efforts in combination of in vitro, in vivo and in silico approaches, and COVID-19, we consider that the 3 year pilot phase needs to be extended. For instance, we can extend this to 2023 instead of 2021.

If you agree with this suggestion, please respond by checking “Yes” below. If not, you can add a few comments as you wish.

Yes (X) or/and Your comments: *indeed, experimental characterization work takes a lot of time!*

C) So far, we have received only two manuscripts on Dark Protein studies. We are wondering when we expect to see your 1st submission to the journal (JPR or any). Would it be possible for you to expect approximate date?
See above

D) Any suggestions on the Dark Protein Studies? e.g., resources, reagents, cell lines, ab, …others.
We have created new pages in neXtProt to host functional predictions on uPE1 proteins. They are regularly populated with new information from the community. Don’t hesitate to contact us and contribute!
https://www.nextprot.org/about/functional-proteome-project.

Chromosome Number: 3

Part I: uPE1 Proteins (Dark Proteins): neXt-CP50 Challenge:

Major lab members or partners contributing to the neXt-CP50 Challenge:

Lab member
Kazuki Yamamoto (Assistant professor)
Yoko Chikaoka (Researcher)

Partner
Toshie Takahashi (The University of Tokyo)

A) Please list the neXt-CP50 Challenge Proteins that your team is characterising and briefly describe your teams progress made to date including any publications or planned papers in 2020.

Acc. code	Protein name	Gene name
NX_Q9Y2S6	Translation machinery-associated protein 7	TMA7
NX_Q504Y3	Zinc finger CW-type PWWP domain protein 2	ZCWPW2
NX_Q12894	Interferon-related developmental regulator 2	IFRD2

There is no progress in our team.

TMA7 was reported this year as a biomarker of acute myeloid leukemia by RT-PCR (Int J Mol Sci. 2021 Apr 27;22(9):4575.)

ZCWPW2 was reported last year as one of five genes in the genomic region duplicated by irradiation of primary fibroblast(J Mol Med (Berl). 2020 Aug;98(8):1107-1123.).
Immunostaining data is available in Human protein atlas.
https://www.proteinatlas.org/ENSG00000206559-ZCWPW2/tissue
We have tried to detect ZCWPW2 in the system irradiating iPS cells, but so far no protein has been detected.

There is no recent report on IFRD2.

B) We are wondering when we expect to see your 1st submission to the journal (JPR or any). Would it be possible for you to state the approximate date?

We are planning to analyze the mechanism of ZCWPW2 under irradiation, but the timing of publication is uncertain.

C) Any suggestions on the Dark Protein Studies? e.g., resources, reagents, cell lines, ab, …others.

Chromosome Number: 4

Part I: uPE1 Proteins (Dark Proteins): neXt-CP50 Challenge:

Major lab members or partners contributing to the neXt-CP50 Challenge:
Yu-Wen Liao. Wen-Hsin Chang, Sung-Liang Yu

A) Please list the neXt-CP50 Challenge Proteins that your team is characterising and briefly describe your teams progress made to date including any publications or planned papers in 2020.

Functional characterization of missing protein, Ch4-DP1, in lung adenocarcinoma: Ch4-DP1 serves as a tumor suppressor

The information about the two uPE1 proteins selected for functional analysis, which were identified by our own from chromosome 4, were further compared with the expression data from paired tumor and adjacent normal tissues from 96 lung adenocarcinoma (LUAD) patients. In 96 lung adenocarcinoma patients, the expression level of CH4-DP1 was down-regulated in tumor tissues compared with adjacent normal tissues, particularly in late-stage patients (early vs late stage, $P < 0.05$), (N0 vs N1-2, $P = 0.0845$). In addition, CH4-DP1 was highly expressed in TP53 mutant patients. $P = 0.0034$.

To investigate the role of CH4-DP1 in lung cancer, we analyzed the clinical correlation of CH4-DP1 based on The Cancer Genome Atlas (TCGA) dataset in a cohort of 1,144 LUAD patients. We found that higher CH4-DP1 expression is associated with the favorable overall survival of lung adenocarcinoma patients (long rank p = 3.5e-09). Next, to further characterize the functional role of CH4-DP1, we established stable cell lines with CH4-DP1 overexpression. The gene expression profiles revealed 294 differentially expressed genes between CH4-DP1 overexpression and vector control cells by RNA sequencing. Pathway enrichment analysis shows the most significantly ranking pathways in cell adhesion and extracellular matrix remodelling. Several genes such as MMP-A, MMP-B and MMP-C were downregulated and therefore affecting cellular functions such as collagen mediated cell adhesion and motility. In addition, decreased LAMA had effect on cell proliferation, migration and invasion.

Given several invasion/migration pathways enriched in CH4-DP1-overexpressed transfectants and CH4-DP1 associated with better patients' outcome, we suggest that CH4-DP1 might be a metastasis suppressor. The cell mobility was performed by wound healing assay. The results showed that stably expressed CH4-DP1 in CL1-5, PC9 and H1650 cells could inhibit the cell migration ability 20%, 36%, 30%, respectively (mean ± SD, n = 3). The cell proliferation was performed by MTT assay. Overexpression of CH4-DP1 decreased cell proliferation. For the anchorage-dependent colony formation, control group cells and stable overexpressed CH4-DP1 cells were incubated for 2 weeks, then stained with methylene blue. We observed 40% decrease of growth-suppressive effect on CH4-DP1 overexpression cells.

Taken together, CH4-DP1 expression is associated with favorable survival, inhibition of cell migration and colony formation in vitro and expression analysis also indicates CH4-DP1 significantly alters invasion/migration related pathways. These preliminary results implied that CH4-DP1 acts as a tumor suppressor in lung cancer.

B) We are wondering when we expect to see your 1st submission to the journal (JPR or any). Would it be possible for you to state the approximate date?
Due to the challenge of protein functional characterization and the COVID-19 outbreak, we hope that the 3-year pilot phase can be further extended to another 1-2 year.

C) Any suggestions on the Dark Protein Studies? e.g., resources, reagents, cell lines, ab, …others.
NA

Chromosome Number: 9

Part I: uPE1 Proteins (Dark Proteins): neXt-CP50 Challenge:

Major lab members or partners contributing to the neXt-CP50 Challenge:
Dong Wook Kim, HuiSu Kim, Hyoung-Min Park, Yong-In Kim, Jinwhan Eugene Lee

A) Please list the neXt-CP50 Challenge Proteins that your team is characterising and briefly describe your teams progress made to date including any publications or planned papers in 2020.
We are trying to reveal biological function of MPs using human cell line models that express MPs and IP-MS. This strategy is not only useful for neXt-CP50 uPE1 functional characterization project, but also next-MP50 MPs identification and validation project. Two MPs (FOXD4, ARID3C) have been turned out its subcellular localization and binding partner proteins via our IP-MS strategy.

B) We are wondering when we expect to see your 1st submission to the journal (JPR or any). Would it be possible for you to state the approximate date?
2Q 2022 expected

C) Any suggestions on the Dark Protein Studies? e.g., resources, reagents, cell lines, ab, …others.
N/A

Chromosome Number: 10

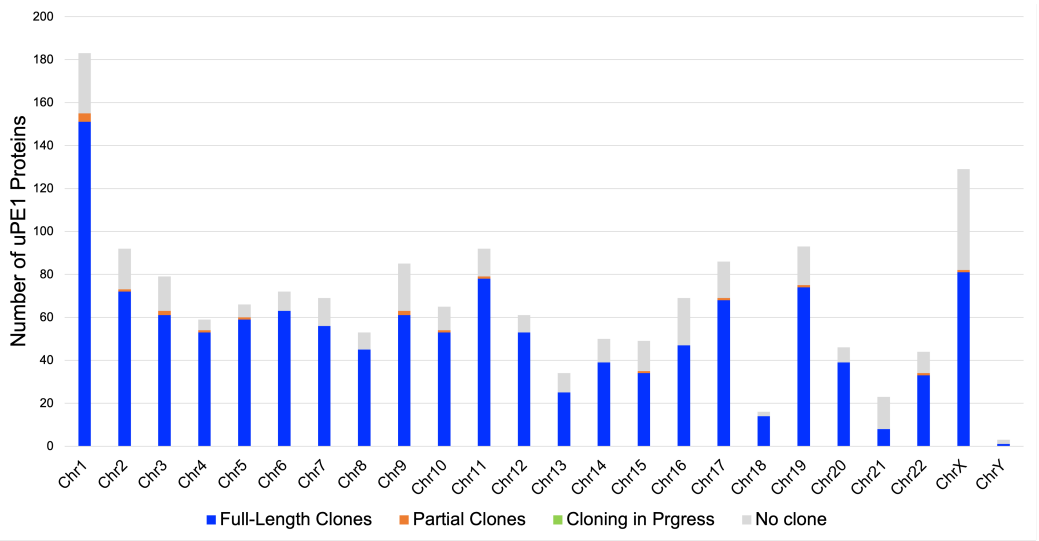
Part I: uPE1 Proteins (Dark Proteins): neXt-CP50 Challenge:

Major lab members or partners contributing to the neXt-CP50 Challenge:
Dustin Grief, Yining Zhang

A) Please list the neXt-CP50 Challenge Proteins that your team is characterising and briefly describe your teams progress made to date including any publications or planned papers in 2020.

We performed genome-wide CRISPR-based function genomics screen to identify mutations that can promote cancer progression, especially invasion, in breast epithelial cells expressing different mutant p53 proteins. From the *in vitro* cell-based screens, a few hundred hits were identified for 2 different p53 mutants, and we are currently down-selecting the top candidates, including several uPE1 proteins, for individual validation. In addition, we performed *in vivo* mouse-based CRISPR screen and identified tumor-initiating mutations.

We are also producing more full-length plasmid clones for uPE1 proteins for functional studies, and the current clone coverage is shown below. Currently, we have full-length plasmids for around 80% of 1,618 uPE1 proteins (shown below), which are available to the entire C-HPP team.



B) We are wondering when we expect to see your 1st submission to the journal (JPR or any). Would it be possible for you to state the approximate date?
We are aiming to submit the manuscript describing the CRISPR screening results in 2022.

C) Any suggestions on the Dark Protein Studies? e.g., resources, reagents, cell lines, ab, …others.
As mentioned above, we have a full-length plasmid collection for the majority of dark proteins in multiple vectors, which can be applied to many types of experiments for functional characterization of the Dark Proteins. IVTT-produced proteins (GST-tagged) can be used for targeted MS or antibody validation, and the Lenti-based plasmid can be used for cell-based assays or screening. All these are available to the C-HPP team via our web portal DNASU.org, and we are always open to collaboration.

Chromosome Number: 11

Part I: uPE1 Proteins (Dark Proteins): neXt-CP50 Challenge:

Major lab members or partners contributing to the neXt-CP50 Challenge:

A) Please list the neXt-CP50 Challenge Proteins that your team is characterising and briefly describe your teams progress made to date including any publications or planned papers in 2020.

Bioinformatic Prediction of Gene Ontology Terms of Uncharacterized Proteins from Chromosome 11.
Hwang H, Im JE, Yang Y, Kim H, Kwon KH, Kim YH, Kim JY, Yoo JS.
J Proteome Res. 2020 Dec 4;19(12):4907-4912. doi: 10.1021/acs.jproteome.0c00482. Epub 2020 Oct 22. PMID: 33089979

Flashlight into Function Unannotated C11orf52 Using Affinity Purification Mass Spectrometry
Authors: Yeji Yang, Heeyoun Hwang, Ji Eun Im, Kyungha Lee, Seong Hee Bhoo, Jong Shin Yoo, Yun-Hee Kim, Jin Young Kim
J Proteome Res. 2021 Nov 5. doi: 10.1021/acs.jproteome.1c00540. Online ahead of print.PMID: 34739247

To protect your IP you may wish to not disclose the protein ID. In this case please use this abbreviation to designate your target uPE1: Ch-DPx (Ch add number for Ch team, For Chr X, Y and Mt, = XDP1, YDP1 or MtDP1; DP, dark protein; x 1,2,3 etc)

Ch11-DP: CCDC90B (NX_Q9GZT6), SMAP (NX_O00193), and C11orf52 (NX_Q96A22)

Examples:
ATXN8 (Chr 13), CCDC70 (Chr 13)…
▶13DP1 for ATXN8, 13DP2 for CCDC70 and so on.

B) We are wondering when we expect to see your 1st submission to the journal (JPR or any). Would it be possible for you to state the approximate date?

Chromosome Number: Chromosome 12

Part I: uPE1 Proteins (Dark Proteins): neXt-CP50 Challenge:

Major lab members or partners contributing to the neXt-CP50 Challenge:
Ravi Sirdeshmukh (partners: Hari PS, Manoj K Gupta, Mahesh Kulkarni, Srikanth Rapole)
Yuju Chen, Taiwan ; Terence Poon, Hong Kong; Radislaw Sobota, Singapore

[This Report is mainly based on the work from Ravi Sirdeshmukh Group, India; but the work under progress envisages participation of other members in future] See under 'B' below.

A) Please list the neXt-CP50 Challenge Proteins that your team is characterising and briefly describe your teams progress made to date including any publications or planned papers in 2020.

To protect your IP you may wish to not disclose the protein ID. In this case please use this abbreviation to designate your target uPE1: Ch-DPx (Ch add number for Ch team, For Chr X, Y and Mt, = XDP1, YDP1 or MtDP1; DP, dark protein; x 1,2,3 etc)

Examples:
ATXN8 (Chr 13), CCDC70 (Chr 13)…
▶13DP1 for ATXN8, 13DP2 for CCDC70 and so on.

B) We are wondering when we expect to see your 1st submission to the journal (JPR or any). Would it be possible for you to state the approximate date?

We have not selected any MP for a detailed study as yet.

However, we have now identified many novel peptides across multiple chromosomes, in breast cancer

and glioblastoma through Proteogenomics analysis. The novel peptides map to alternative splicing events of known proteins, novel ORF, fusion proteins across multiple chromosomes.

This will form the base for future explorations/directions under C-HPP. We plan to take up key candidates for clinical validations, biochemical, structural and functional characterization.

This will also open opportunity for other members of our Team to participate in the effort on select candidates with a specific and targeted deep study work plan.

Chromosome Number: 13

Part I: uPE1 Proteins (Dark Proteins): next-CP50 Challenge

Major lab members or partners contributing to the neXt-CP50 Challenge:

Keun-Na (YPRC, Yosei University, Korea)
Ju-Wan Kim (YPRC, Yosei University, Korea)
Jin-Young Cho (YPRC, Yosei University, Korea)
Chae-Yeon Kim (YPRC, Yosei University, Korea)
Jun-Young Park (YPRC, Yosei University, Korea)
Lydie Lane (SIB/University of Geneva)
Paula Duek (SIB/University of Geneva)

A) Please list the neXt-CP50 Challenge Proteins that your team is characterising and briefly describe your teams progress made to date including any publications or planned papers in 2021.

Chromosome	Nickname	Status
1	1DP1	CRISPR/cas9 mutant was successfully constructed for in vivo study in model animals C. elegans.
1	1DP2	Cancelled due to lower alignment score model animals C. elegans.
3	3DP1	Failure on the CRISPR/cas9 mutant construction due to lethal phenotype in model animals C. elegans.
4	4DP1	Cancelled due to publication by others during preparation of mutant construct.
4	4DP2	Lower priority
9	9DP1	Failure on the CRISPR/cas9 mutant construction due to sterile phenotype in model animals C. elegans.
10	10DP1	Cancelled due to lower priority
10	10DP2	CRISPR/cas9 mutant was successfully constructed for in vivo study in model animals C. elegans.
15	15DP1	Failure on the CRISPR/cas9 mutant construction due to lethal phenotype in model animals C. elegans.
17	17DP1	Cancelled due to lower priority
19	19DP1	CRISPR/cas9 mutant was successfully constructed for in vivo study in model animals C. elegans.
22	22DP1	Cancelled due to publication by others during preparation of mutant construct.
X	XDP1	Cancelled due to lower alignment score to model animals C. elegans.

B) Your opinion on the extension of next-CP50 (2018-2021).

Due to the nature of protein characterization work, which requires a lot more extra efforts in combination of in

vitro, in vivo and in silico approaches, and COVID-19, we consider that the 3 year pilot phase needs to be extended. For instance, we can extend this to 2023 instead of 2021.

If you agree with this suggestion, please respond by checking “Yes” below. If not, you can add a few comments as you wish.

C) So far, we have received only two manuscripts on Dark Protein studies. We are wondering when we expect to see your 1st submission to the journal (JPR or any). Would it be possible for you to expect approximate date?
-not yet this point.

Chromosome Number: 18

Part I: uPE1 Proteins (Dark Proteins): next-CP50 Challenge

Major lab members or partners contributing to the neXt-CP50 Challenge:

Ekaterina V. Ilgisonis, Katerina V.Poverennaya, Mikhail A.Pyatnitskii, Elena A. Ponomarenko

A) Please list the neXt-CP50 Challenge Proteins that your team is characterising and briefly describe your teams progress made to date including any publications or planned papers in 2020.
B2RU33 (chr18)
Q32NC0 (chr18)
Q68D86 (chr18)
Q9P2G3 (chr18)

B) Your opinion on the extension of next-CP50 (2018-2021).
Due to the nature of protein characterization work, which requires a lot more extra efforts in combination of in vitro, in vivo and in silico approaches, and COVID-19, we consider that the 3 year pilot phase needs to be extended. For instance, we can extend this to 2023 instead of 2021.

If you agree with this suggestion, please respond by checking “Yes” below. If not, you can add a few comments as you wish.
Yes ( ) or/and Your comments: Yes

C) So far, we have received only two manuscripts on Dark Protein studies. We are wondering when we expect to see your 1st submission to the journal (JPR or any). Would it be possible for you to expect approximate date?
We’re are going to submit 2 manuscripts by 30/06/2021

D) Any suggestions on the Dark Protein Studies? e.g., resources, reagents, cell lines, ab, …others.
We combined all the computational methods
A lot of protein function prediction methods were implemented, so we are going to perform the analysis using all of them for uncharacterized chromosome 18 proteins.

Chromosome Number: 19

Part I: uPE1 Proteins (Dark Proteins): neXt-CP50 Challenge:

Major lab members or partners contributing to the neXt-CP50 Challenge:

Nohemi Salinas Jazmín-School of Medicine, UNAM
Orlando Morales-Tarré, Emmanuel Osio Becerro, Gloria Angelina Herrera Quiterio, Magdalena Hernández-Ortiz, María del Carmen Vargas-Lagunas-Proteomics laboratory at Center for Genomic Sciences UNAM.
Jeovanis Gil-Valdes. Lund University, Department of Clinical Sciences
Julio Collado-Vides, Program of Computational Biology at Center for Genomic Sciences UNAM.
Emmanuel Salazar Bustamante-Universidad Autónoma del Estado de Morelos
Osbaldo Resendis Antonio, National Institute of Genomic Medicine.

Alejandro García Carranca, National Cancer Institute

A) Please list the neXt-CP50 Challenge Proteins that your team is characterising and briefly describe your teams progress made to date including any publications or planned papers in 2020.

uPE1s under research: 19DP1, 19DP2, 19DP3 and 19DP4

• We observed a differential expression of four proteins with no assigned function (19DP1, 19DP2, 19DP3 and 19DP4), measured by RT-PCR in cervical cancer (CC) cells, suggesting a potentially relevant role in this type of cancer. Using the Gene Expression Profiling Interactive Analysis (GEPIA) database, we analyzed the expression of the 19DP1, 19DP3 and, 19DP4 genes in different types of cancer. A differential expression of 19DP1, 19DP3 and, 19DP4 in normal tissues with respect to tumors were detected, as well as a significant difference in the survival of cancer patients with high and low expression of these proteins, which suggests that these proteins may be relevant in a tumour setting.

• 19DP1 mRNA is expressed without significant changes in SiHa (HPV 16 infected) and HeLa (HPV18) cervical cancer tumor cells compared to C33a (HPV negative) cervical cancer cells and HaCat non-tumor cells. While the 19DP1 protein has a differential expression in SiHa tumor cells increased, compared to C33a, HeLa and HaCaT. The absence of 19DP1 increases cell proliferation and migration in HaCaT cells.

Also, we are currently conducting immunohistochemistry experiments on cancerous breast, ovarian and cervical tissues in which, according to Protein Atlas, a very high expression of the mRNA of this gene is observed.

- One of the first evidences that we had regarding the role of the 19DP1 protein, was its apparent relationship with viral proteins of HPV18 and HPV16, therefore we found that we carried out cell-based screening of the target gene in cervical cancer cells.

Knockout (KO) was generated by the CRISPR/Cas9 system without any off-target effect detected. Western blot results showed successful validation of the 19DP1 knockout in the cervical cancer lines (HeLa, SiHa, C33A) and HaCaT cells.

• 19DP2 mRNA is significantly overexpressed in C33a cells and less expressed in SiHa and HeLa cancer lines (infected with HPV) and HaCaT control cells. Different independent proteomic analyzes stored in the EXPRESSION ATLAS EMBL-EBI database report overexpression of the 19DP2 protein in the A549 (lung adenocarcinoma), HeLa (cervical cancer) and SKOV3 (ovarian cancer) cell lines, which was corroborated by western blot in our laboratory. In addition, according to its amino acid sequence, and the *in silico* analysis carried out in our group for the prediction of the structure of 19DP2 using several available programs 19DP2 could have up to 3 transmembrane steps.

• 19DP3 protein is significantly overexpressed in SiHa cells with respect to C33a, HeLa and HaCaT, which suggests a relevant role in this type of CCU. With cisplatin (1uM) treatment, 19DP3 increases its expression in HaCaT, C33a and Hela cells, while in Siha cells it decreases, which suggests being involved in the response mechanism to treatment in a different way depending on the cellular context.

The 19DP4 protein was detected more expressed in HeLa and SiHa cells than in HaCaT and C33a cells. Treatment of these cells with Cisplatin (1uM) reduces its expression of the 19DP4 protein, so it could be related to the resistance of these cells to the drug.

B) We are wondering when we expect to see your 1st submission to the journal (JPR or any). Would it be possible for you to state the approximate date?

Very recently we have started laboratory work, we hope to have in the next 12 months solid experimental evidence of the function of at least 19DP1 and important advances in 19DP2. At that time, we will evaluate the possibility of publishing our results. For now, we are concluding a review of the family of proteins to which 19DP2 belongs, which we will send for publication shortly.

Chromosome Number:X

Part I: uPE1 Proteins (Dark Proteins): neXt-CP50 Challenge:

Major lab members or partners contributing to the neXt-CP50 Challenge:

PI: Yasushi Ishihama (Kyoto University)

(Members: jPOST team)

Masaki Matsumoto (Niigata University)
Shujiro Okuda (Niigata University)
Shin Kawano (Database Center for Life Science, DBCLS))
Susumu Goto (Database Center for Life Science, DBCLS))
Norie Araki (Kumamoto University)

(Partners)

Taiwan ICPC-Chr 4 team (PI: Prof Yu-Ju Chen)

A) Please list the neXt-CP50 Challenge Proteins that your team is characterising and briefly describe your teams progress made to date including any publications or planned papers in 2020.

Our jPOST team is in charge of validating the existence by SRM assay for the candidate proteins both in Chr-X and 4 selected by the Taiwan Chr-4 team based on their results from large-scale proteome measurements. Currently, 22 and 19 uPE1 protein candidates were selected for further analysis.

Table 41 uPE1 candidates for further validation

Chr 4	P78312	Q0P651	Q56VL3	Q5BJH2	Q5M9N0	Q68CR1
	Q6NW29	Q6ZU35	Q6ZUS6	Q86YA3	Q8IUW5	Q8N1A6
	Q8N8J7	Q8NEC7	Q8WVX3	Q96EY4	Q96QK8	Q9C0D6
	Q9C0F1	Q9P2B7	Q9ULE4	Q9Y605		
Chr X	A2AJT9	A6NJG2	A6ZKI3	O14668	Q14656	Q5JSJ4
	Q5U3C3	Q6P1M9	Q6ZTR5	Q7Z309	Q8N9E0	Q8NFB2
	Q8TB03	Q96A49	Q9BVG4	Q9BWD3	Q9H5V9	Q9HAI6
	Q9Y4X0					

In addition, 4 proteins such as sp|Q6IC83|CV042_HUMAN, sp|Q7Z570|Z804A_HUMAN, sp|Q8IVF6|AN18A_HUMAN and sp|Q8IZA3|H1FOO_HUMAN in human iPS cells will be analysed.

B) We are wondering when we expect to see your 1st submission to the journal (JPR or any). Would it be possible for you to state the approximate date?
Currently we do not have any clear deadline.

C) Any suggestions on the Dark Protein Studies? e.g., resources, reagents, cell lines, ab, …others.

Chromosome Number: Mt

Part I: uPE1 Proteins (Dark Proteins): neXt-CP50 Challenge:

Major lab members or partners contributing to the neXt-CP50 Challenge:

Andrea Urbani, Paola Roncada, Mauro Fasano, Alberio T, Pieroni L, Ronci M, Brioschi M, Cunsolo V, Giusti L, Greco V, Lucacchini A, Soggiu A, Zilocchi M, Marini F, Carregari VC, Iavarone F, Persichilli S, Castagnola M, Ciregia F, Sickmann A, Babu M.

A) Please list the neXt-CP50 Challenge Proteins that your team is characterising and briefly describe your teams progress made to date including any publications or planned papers in 2020.

TABLE 1 | List of mt-dark proteins identified in HeLa dataset.

Accession	Description	Mito evidence IMPI	Mito localization IMPI	HPA cell localization	Enzyme	Exp. fraction	Gravy score
NX_O60941-1	Dystrobrevin beta	Predicted	Unknown	Mitochondria (A)	Try; Glu-C	F1, F2	−0.55
NX_Q3SXM5-1	Inactive hydroxysteroid dehydrogenase-like protein 1	Known	OM	Intracellular, Membrane (P)	Chym; Try	F2	+0.15
NX_Q4VC31-1	Coiled-coil domain-containing protein 58	Known	IMS	Nucleoli, Mitochondria (A)	Try; Chym; Glu-C	F2	−0.60
NX_Q56VL3-1	OCIA domain-containing protein 2	Known	Unknown	Mitochondria (E)	Try	F2	−0.26
NX_Q8IYQ7-1	Threonine synthase-like 1	Known	Matrix	Nuclear bodies (A), Mitochondria (A), Cytosol (A)	Try	F2	−0.13
NX_Q8NFV4-1	Protein ABHD11	Known	Matrix	Mitochondria (S)	Try; Chym	F2	−0.09
NX_Q96EX1-1	Small integral membrane protein 12	Predicted	Unknown	Mitochondria (A)	Try	F2	−0.53
NX_Q96C01-1	Protein FAM136A	Known	IMS	Mitochondria (A)	Try; Glu-C	F2	−0.43
NX_Q96ER9-1	Coiled-coil domain-containing protein 51	Known	Matrix	Nucleosome (S), Mitochondria (S) Centrosome (A)	Try; Glu-C	F2	−0.38
NX_P56378-1	6.8 kDa mitochondrial proteolipid	Known	IM	Mitochondria (S),Nucleoli (S)	Try	F1,F2	−0.02
NX_Q9GZT6-1	Coiled-coil domain-containing protein 90B	Known	Matrix	Mitochondria (E)	Try; Glu-C	F2	−0.55
NX_A8MTT3-1	Protein CEBPZOS	Known	IMS	Nucleoplasm (A)	Try; Glu-C	F2	−0.27
NX_Q9H4I3-1	TraB domain-containing protein	Known	OM	Nucleus (A), Mitochondria (A)	Try	F2	−0.21
NX_Q9UFN0-1	Protein NipSnap homolog 3A	Known	Matrix	not available	Try; Glu-C	F2	−0.37
NX_Q6P1 × 6-1	UPF0598 protein C8orf82	Known	Matrix	Nucleus (A)	Try; Chym	F2	−0.23
NX_Q8N2U0-1	Transmembrane protein 256	Predicted	Unknown	Vesicles (A)	Try	F2	+0.46
NX_Q8WVI0-1	Small integral membrane protein 4	Predicted	Unknown	Nucleoplasm (A), Mitochondria (A)	Try	F2	−0.54
NX_Q8WW59-1	SPRY domain-containing protein 4	Known	Matrix	Nucleoplasm (A)	Try	F1,F2	−0.07
NX_Q96BQ5-1	Coiled-coil domain-containing protein 127	Known	OM/IMS	Nucleus (S), Nucleoli (S)	Try; Chym	F1,F2	−0.72
NX_Q96DB5-1	Regulator of microtubule dynamics protein 1	Known	OM/IMS	Centrosomes (S),Actin filaments (S)	Try	F1,F2	−0.37
NX_Q96KF7-1	Small integral membrane protein 8	Known	OM	Vesicles (A)	Try; Chym	F2	−0.55
NX_Q9NU23-1	LYR motif-containing protein 2	Known	matrix	Cytosol (A)	Try	F2	−0.70

Accession, neXtprot accession number of uPE1 mt-dark protein; Description, extended protein name; MitoEvidenceIMPI. Mitochondrial protein classification; MitoLocalization IMPI, protein localization within mitochondria according to IMPI; HPA cell localization, main protein location Approved, Predicted, Enhanced or Supported in Human Protein Atlas project; Enzyme, protease used to generate the identified proteolytic peptides; Experimental Fraction, experimental sample in which the corresponding protein has been retrieved; Gravy Score, Grand Average of Hydropathy, measure the hydrophobicity of a specific protein.

See for reference: Exploring the HeLa Dark Mitochondrial Proteome.
Marini F, Carregari VC, Greco V, Ronci M, Iavarone F, Persichilli S, Castagnola M, Urbani A, Pieroni L.
Front Cell Dev Biol. 2020 Mar 5;8:137. doi: 10.3389/fcell.2020.00137. eCollection 2020. PMID: 32195257

A) We are wondering when we expect to see your 1st submission to the journal (JPR or any). Would it be possible for you to state the approximate date?

The current pandemic crisis is limiting our investigations in this field, most probably we will not submit any contribution.

B) Any suggestions on the Dark Protein Studies? e.g., resources, reagents, cell lines, ab, …others.

1) Development of a joint NDA under the HUPO leadership for data sharing before publication of collected experimental and in silico evidences (eg. MS data, protein-protein interaction matrices, pQTR/eQTR , etc. etc.)
2) Lobbying for funding.