

Additional Supplementary Material:

Integrative spatiotemporal map of nucleocytoplasmic transport

Barak Raveh^{*,1,2,3,4}, Roi Eliasian¹, Shaked Rashkovits¹, Daniel Russel^{2,3,4}, Ryo Hayama⁵, Samuel E. Sparks⁵, Digvijay Singh⁷, Roderick Lim⁹, Elizabeth Villa^{7,8}, Michael P. Rout^{*,5}, David Cowburn^{*,6}, Andrej Sali^{*,2,3,4}

¹ School of Computer Science and Engineering, Hebrew University of Jerusalem, Jerusalem, Israel.

² Quantitative Biosciences Institute, University of California, San Francisco, San Francisco, CA 94158, USA.

³ Department of Bioengineering and Therapeutic Sciences, University of California, San Francisco, San Francisco, CA 94158, USA.

⁴ Department of Pharmaceutical Chemistry, University of California, San Francisco, San Francisco, CA 94158, USA

⁵ Laboratory of Cellular and Structural Biology, The Rockefeller University, New York, NY 10065, USA

⁶ Departments of Biochemistry and Systems & Computational Biology, Albert Einstein College of Medicine, Bronx, NY 10461, USA

⁷ School of Biological Sciences, University of California San Diego, La Jolla, CA 92093, USA.

⁸ Howard Hughes Medical Institute, University of California San Diego, La Jolla, CA 92093, USA.

⁹ Biozentrum and the Swiss Nanoscience Institute, University of Basel, 4056 Basel, Switzerland.

*** Corresponding Authors**

Keywords: nucleocytoplasmic transport; nuclear pore complex; integrative modeling; spatiotemporal modeling; Brownian Dynamics simulations

Supplementary Tables

This document contains a subset of the supplementary tables. The additional supplementary tables will be updated in this document in the coming weeks.

Table of contents

Supplementary Tables	2
Table of contents	2
Supplementary Table S7. SANS measurements from Sparks et al. 2017.	3
Supplementary Table S8. Ensemble modeling data from SANS	4
Supplementary Table S9: Constructs used in LH and SS-RH SAXS	5
Supplementary Table S10: Parameters for various FG nups after model training	7

Supplementary Table S7A. SANS measurements from Sparks et al. 2017.

Sample	Debye analysis	P(r)	
	R_g (Å)	R_g (Å)	D_{max} (Å)
[^2H]-FSFG-K [0.6 mM] (8mg/ml)	36.2 ± 0.3	35.8 ± 1.1	127.0
[^2H]-FSFG-K [0.6 mM] Kap95 [0.25 mM]	39.6 ± 0.8	42.3 ± 2.7	147.0
[^2H]-FSFG-K [0.6 mM] Kap95 [0.5 mM]	41.8 ± 1.2	44.2 ± 1.7	143.5
[^2H]-FSFG-K [0.6 mM] NTF2 [0.3 mM]	41.2 ± 0.3	39.8 ± 1.9	142.8
[^2H]-FSFG-K [0.6 mM] NTF2 [0.6 mM]	45.6 ± 0.5	42.2 ± 2.2	151.0
[^2H]-FSFG-K [0.6 mM] NTF2 [1.2 mM]	47.9 ± 0.6	46.4 ± 2.9	168.0

Supplementary Table S8. Ensemble modeling data from SANS {Sparks et al.,}

Sample	χ^2	Selected ensemble R_g (Å)	Selected ensemble D_{max} (Å)	Selected ensemble end-to-end distance (Å) Ca(N)-Ca(C)
[2 H]-FSFG-K [0.6 mM] (8mg/ml)	0.20	35.5	87.4	73.7
[2 H]-FSFG-K [0.6 mM] Kap95 [0.25 mM]	0.26	46.9	129.1	118.0
[2 H]-FSFG-K [0.6 mM] Kap95 [0.5 mM]	0.38	47.0	137.4	135.0
[2 H]-FSFG-K [0.6 mM] NTF2 [0.3 mM]	0.17	38.5	119.0	119.0
[2 H]-FSFG-K [0.6 mM] NTF2 [0.6 mM]	0.15	40.7	117.6	115.3
[2 H]-FSFG-K [0.6 mM] NTF2 [1.2 mM]	0.27	42.0	141.0	128.1
FSFG-K [375 mM] + [2 H]-Kap95 [75 μ M]	0.99	46.6	120.9	108.3

Supplementary Table S9. Constructs used in LH and SS-RH SAXS.

Abbreviation	Simple Structure	Sequence	#AA	MW (da)
(FSFG-K-274-397)-6His	M-Nsp1(274-397)-LEHHHHHH	MDNKTNTTPSFSGAKSDENKAGATSKPAFSFG AKPEEKDDNSSKPAFSFGAKSNEDKQDGTAKPAFSF GAKPAEKNNNETSKPAF SFGAKSDEKKDGDASKPAFSFGAKPDENKASATSKPA LEHHHHHH	133	14135
(FG-N)-6His	MGT-Nsp1(48-172)-SHMHHHHH H	MGTSAPNNTNNANSSITPAFGSNNTGNTAFGNSNPTS NVFGSNNSTTNTFGSNSAGTSLFGSSSAQQTKSNGT AGGNTFGSSSLFNNSTNSNTTKPAFGGLNFGGGNNT TPSSTGNANTSNNLFGATASHMHHHHHH	137	13649
(FG-N)-(FSFG-K)-6His	MGT-Nsp1(48-172)-ASATSKPA-Nsp1(284-397)-SHHHHHH	MGTSAPNNTNNANSSITPAFGSNNTGNTAFGNSNPTS NVFGSNNSTTNTFGSNSAGTSLFGSSSAQQTKSNGT AGGNTFGSSSLFNNSTNSNTTKPAFGGLNFGGGNNT TPSSTGNANTSNNLFGATAASATSKPAFSFGAKSDEN KAGATSKPAFSFGAKPEEKDDNSSKPAFSFGAKSNE DKQDGTAKPAFSFGAKPAEKNNNETSKPAFSFGAKSD EKKDGDASKPAFSFGAKPDENKASATSKPASHHHHH H	257	25955
(FG-N)-(FSFG-K)-Tet-6His	MGT-Nsp1(48-172)-ASATSKPA-Nsp1(284-397)-SHMGEYFTLQIRGRERFEMFRELNEALELKDAQAHMHHHHHH	MGTSAPNNTNNANSSITPAFGSNNTGNTAFGNSNPTS NVFGSNNSTTNTFGSNSAGTSLFGSSSAQQTKSNGT AGGNTFGSSSLFNNSTNSNTTKPAFGGLNFGGGNNT TPSSTGNANTSNNLFGATAASATSKPAFSFGAKSDEN KAGATSKPAFSFGAKPEEKDDNSSKPAFSFGAKSNE DKQDGTAKPAFSFGAKPAEKNNNETSKPAFSFGAKSD EKKDGDASKPAFSFGAKPDENKASATSKPASHMGEYF TLQIRGRERFEMFRELNEALELKDAQAHMHHHHHH	292	30235
2mer	M-Nsp1(346-397)-HHHHHH	MAKPAEKNNNETSKPAFSFGAKSDEKKDGDASKPAFS FGAKPDENKASATSKPAHHHHHH	60	6455
Nsp1FG	M-Nsp1(30-591)	MSTGAGAFGTGQSTFGFNNSAPNNTNNANSSITPA FGSNNTGNTAFGNSNPTSNVFGSNNSTTNT FGSNSAGTSLFGSSSAQQTKSNGTAGGNT FGSSSLFNNSTNSNTTKPAFGGLNFGGGNNTTPSSTG NANTSNNLFGATANANKPAFSFGATTNDDKKTEPDKP AFSFNSSVGNKTDQAPTTGFSFGSQLGGDKTVNEA AKPSLSFGSGSAGANPAGASQPEPTTNEPAKPAFSFG TATSDNKTNTTPSFSGAKSDENKAGATSKPAFSFG AKPEEKDDNSSKPAFSFGAKSNEDKQDGTAKPAFS FGAKPAEKNNNETSKPA FSFGAKSDEKKDGDASKPAFSFGAKPDENKASATSK PA FSFGAKPEEKDDNSSKPAFSFGAKSNEDKQDGTAK PA	571	58494.47

Abbreviation	Simple Structure	Sequence	#AA	MW (da)
		FSFGAKPAEKNNNETSKPAFSFGAKSDEKKDGDASK PAFSFGAKSDEKKDSDSSKPAFSFGTKSNEKKDSGS SKPA FSFGAKPDEKKNDEVSKPAFSFGAKANEEKESDESK SA FSFGSKPTGKEEGDGAKAAISFGAKPEEQKSSDTSK PA FTFGLEHHHHHH		
GLFG	M-Nup100(318-44)SNLEHHHHH	MSLFGKANTFSNSASGGLFGQNNQQQGSGLFGQNS QTSGSSGLFGQNNQKQPNTFTQSNTGIGLFGQNNNQ QQQSTGLFGAKPAGTTGSLFGGNSSTQP NSLFGTTNVPTSNTQSQQGNLFGATKLTNSNLEHHHHH HH	138	14348
Nup100FG				
Nup49	M-Nup49-(1-269)	MFGLNKASSTPAGGLFGQASGASTGNANTGFSFGGTQTGQNTGPSTGG LFGAKPAGSTGGGLGASFGQQQQSQTNAFGGSATGGGLFGNKPNNNTA NTGGGLFGANSNSNSGSLFGSNNAQTSRGLFGNNNTNNINSSSGMNN ASAGLFGSKPAGGTSLFGNTSTSSAPAQNQGMFGAKPAGTSLFGNNAG NTTTGGGLFGSKPTGATSLFGSSNNNNNNNNNNNIMSASGGLFGNQQQ QLQQQFQMOCALQNLSQLPITPMTRISELLEHHHHHHH	277	27421.20
Nup1FG	M-Nup1-(352-1076)-LEHHHHHHH	MKATSSAGAVFKSSVEMGKTDKSTKTAEAPTLSFNFSQKANKTKAVDN TVPSTTLFNFGGKSDTVTSASQPFKFGKTSEKSENHTESDAPPKSTAP IFSFGKQEENGDEGDENEPRKRRLPVSEDNTKPLFDGKTGDQKE TKKGESEKDASGKPSFVFGASDKQAEGLPLFTFGKKADVTSNIDSSAQ FTFGKAATAKETHTKPSETPATIVKKPTFTFGQSTSENKISEGSAKPT FSFSKSEERKSSPISNEAAKPSFSFPGKPDVQAPTDDKTLKPTFSF TEPAQKDSSVVSEPKKPSFTFASSKTSQPKPLFSFGKSDAAKEPPGSN TSFSFTKPPANETDKRPTPPSFTFGGSTTNNTTTTSTKPSFSFGAPES MKSTASTAAANTEKLSNGFSFTKFNNHKEKSNSPTSFFDGSASTPIPI VLGKPTDATGNTTSSKSAFSFGTANTNGTNASANSTSFSFNAPATGNGT TTTSNTSGTNIAGTFNVGKPDQSIASGNTNGAGSAFGFSSSGTAATGA ASNQSSFNFGNNGAGGLNPFTSATSSSTNANAGLFNKPPSTNAQNVNVP SAFNFTGNNSTPGGGSVFNMNGNTNANTVFAGSNNQPHQSQTSPFNTN SSFTPSTVPNINFSGLNGGITNTATNALRPSDIFGANAASGSNSNVTN PSSIFGGAGVPPTTSFGQPQSAPNQMGMTNNGMSMGGMANRARIAR MRHSKRLEHHHHHHH	734	76081.56

Supplementary Table S10. Parameters for various FG nups following model training

Nsp1

Nup	subregion	Residue range	Self-k [kcal/mol/A]	self-range	Kap-k [kcal/mol/A ²]	Kap-range [A]	Non-specific k	Non-specific range	Backbone-k (k_2) [kcal/mol/A ²]	backbone-tau
Nsp1	N	1-180	1.47	6.00	2.64	5.5	0.08	5.00	0.0075	
	C	181-550	1.32	6.00	2.64	5.5	0.01	5.00	0.0075	
	s	551-636	1.28	6.00	-	-	0.01	5.00	0.0075	
Nup100		2-610	1.47	6.00	2.64	5.5	0.08	5.00	0.0075	
	s	551-800	1.28	6.00	-	-	0.01	5.00	0.0075	
Nup116		1-751	1.47	6.00	2.64	5.5	0.08	5.00	0.0075	
	s	751-950	1.28	6.00	-	-	0.01	5.00	0.0075	
Nup159		442-881	1.32	6.00	2.64	5.5	0.07	5.00	0.0075	
	s	882-1116	1.28	6.00	-	-	0.01	5.00	0.0075	
Nup49		1-240	1.47	6.00	2.64	5.5	0.08	5.00	0.0075	
	s	241-269	1.28	6.00	-	-	0.01	5.00	0.0075	
Nup57		1-200	1.47	6.00	2.64	5.5	0.08	5.00	0.0075	
	s	201-286	1.28	6.00	-	-	0.01	5.00	0.0075	
Nup145 N		1-200	1.47	6.00	2.64	5.5	0.08	5.00	0.0075	
	s	201-250	1.28	6.00	-	-	0.01	5.00	0.0075	
Nup1	s	201-325	1.28	6.00	-	-	0.01	5.00	0.0075	
	m	326-797	1.32	6.00	5.98	5.5	0.01	5.00	0.0075	
	c	798-1076	1.47	6.00	5.98	5.5	0.08	5.00	0.0075	
Nup60		399-539	1.32	6.00	5.98	5.5	0.01	5.00	0.0075	
	s	301-398	1.28	6.00	-	-	0.01	5.00	0.0075	

Supplementary Table S11. Sequences of FG Nups domains used in Extended Data Fig. 1**NUP100****>NUP100 YKL068W SGDID:S000001551**

```
1    50 MFGNNRPMFGGSNLSFGSNTSSFGGQQSQQPNSLFGNSNNNNNSTSNNAQ
51   100 SGFGGFTSAAGSNSNSLFGNNNTQNNGAFGQSMGATQNSPFGSLNSSNAS
101  150 NGNTFGGSSSMGSFGGNTNNAFNNSNSTNSPFGFNKPNTGGTLFGSQNN
151  200 NSAGTSSLFGGQSTSTTGTFTGNTGSSFGTGLNGNGSNIIFAGNNSQSNTT
201  250 GSLFGNQSSAFGTNNQQGSLFGQQSQNTNNAFGNQNLGGSSFGSKPVG
251  300 SGSLFGQSNNTLGNTTNNRNLFGQMNSSNQSSNSGLFGQNSMNSSTQG
301  350 VFGQNNNQMQINGNNNNSLFGKANTFSNSASGGLFGQNNQQQGSLFGQN
351  400 SQTSGSSGLFGQNNQKQPNTFTQSN TGIGLFGQNNNQQQSTGLFGAKPA
401  450 GTTGS LFGGNSSTQPNLSLFGTTNVPTSNTQSQQGNSLFGATKLTNMPFGG
451  500 NPTANQSGSGNSLFGTKPASTTGS LFGNNTASTTV PSTNGLFGNNANNST
501  550 STTNTGLFGAKPDSQSKPALGGGLFGNSNSNSSTIGQNKPVFGGTTQNTG
551  600 LFGATGTNSSAVGSTGKLFGQNNNTLNVGTQNVPPVNNTTQNALLGTTAV
601  650 PSLQQAPVTNEQLFSKISIPNSITNPVKATT SKVNADMKRNSSLTSAYRL
651  700 APKPLFAPSSNGDAKFQKWGKTLERSDRGSSTSNSITDPESSYLNSNDLL
701  750 FDPDRRYLKHLVIKNNKNLNVINHNDDEASKVKLVFTTESASKDDQASS
751  800 SIAASKLTEKAHSPQTDLKDDHDESTPDPQSKSPNGSTSIPMIENEKISS
801  850 KVPGLLSNDVTFFKNYYISPSIETLGNKSLIELRKINNLVIGHRNYGKV
851  900 EFLEPVDLLNTPLDTLCGD LVTFGPKSCSIYENC SIKPEKGEGINVRCRV
901  950 TLYSCFPIDKETRKPIKNITHPLLKRSIAKLKENPVYKFESYDPVTGTYS
951  960 YTIDHPVLTHHHHHH
```

NUP1 FG Domain**>NUP1 YOR098C SGDID:S000005624**

```
1    50 MSSNTSSVMSSPRVEKRSFSSTLKSFFTNPNNKKRPSSKKVFSSNLSYANH
51   100 LEESDVEDTLHVNKRRKRVSGTSQHSDSLTQNNNNAPIIIYGTENTERPPL
101  150 LPILPIQRLRLLEKQQRVRNMRELGLIQSTEFPSITSSVILGSQSKSDEG
151  200 GSYLCTSSSTPSPIKNGSCTRQLAGKSGEDTNVGLPILKSLKNRSNRKR FH
201  250 SQSKGTVWSANFEYDLSEYDAIQKKDNKDKEGNAGGDQKTSENRNNIKSS
251  300 ISNGNLATGPNLTSEIEDLRADINSNRLSNPQKNLLLKGPASTVAKTAPI
301  350 QESFVPNSERSGTPTLKKNIEPKKD KESIVLPTVGFDFIKDNETPSKKTS
351  400 PKATSSAGAVFKSSVEMGKTDKSTKTAEAPTLSFNFSQKANKTKAVDNTV
401  450 PSTTLFNFGGKSDTVTSASQPFKFGKTSEKSENHTESDAPPKSTAPIFSF
```

```

451 500 GKQEENGDEGDDENEPKRKRRLPVSEDNTNKPFLDFGKTGDQKETKKGES
501 550 EKDASGKPSFVFGASDKQAEGTPLFTFGKKADVTSNIDSSAQFTFGKAAT
551 600 AKETHTKPSETPATIVKKPTFTFGQSTSENKISEGSAKPTFSFSKSEEER
601 650 KSSPISNEAAKPSFSFPGKPVVDVQAPTDDKTLKPTFSFTEPAQKDSSVVS
651 700 EPKKPSFTFASSKTSQPKPLFSFGKSDAAKEPPGSNTSFSFTKPPANETD
701 750 KRPTPPSFTFGGSTTNNTTTTSTKPSFSFGAPESMKSTASTAAANTEKLS
751 800 NGFSFTKFNHNKEKSNSPTSFFDGSASSTPIPVLGKPTDATGNTTSKSAF
801 850 SFGTANTNGTNASANSTSFNFNAPATGNGTTTTSTNTSGTNIAGTFNVGKP
851 900 DQSIASGNTNGAGSAFGFSSSGTAATGAASNQSSFNFGNNGAGGLNPFTS
901 950 ATSSTNANAGLFNKPPSTNAQNVNVPSAFNFTGNNSTPGGGSVFNMNGNT
951 1000 NANTVFAGSNNQPHQSQTPSFNTNSSFTPTVFNINFSGLNGGITNTATN
1001 1050 ALRPSDIFGANAASGSNSNVTNPSSIFGGAGGVPTTSFGQPQSAPNQMG
1051 1076 GTNNGMSMGGGVMANRKIARMRHSKRHHHHHH

```

NSP49 FG domain

NUP49 YGL172W SGDID:S000003140

```

1 50 MFGLNKASSTPAGGLFGQASGASTGNANTGFSFGGTQTGQNTGPSTGGLF
51 100 GAKPAGSTGGLGASFGQQQQQSQTNAFGGSATTGGGLFGNKPNNANTGG
101 150 GLFGANSNSNSGSLFGSNNNAQTSRGLFGNNNTNNINNSSSGMNNASAGLF
151 200 GSKPAGGTSLFGNTSTSSAPAQNQGMFGAKPAGTSLFGNNAGNTTTGGGL
201 250 FGSKPTGATSLFGSSNNNNNNNNNSNNIMSASGGLFGNQQQQLQQQPQMOC
251 277 ALQNLSQLPITPMTRISELLGHHHHHH

```

NSP1 FG

>NSP1 YJL041W SGDID:S000003577

```

1 50 MNFNTPPQQNKTPFSFGTANNNSNTTNQNSSTGAGAFGTGQSTFGFNNSAP
51 100 NNTNNANSSITPAFGSNNTGNTAFGNSNPTSNVFGSNNSTTNTFGSNSAG
101 150 TSLFGSSSAQQTKSNGTAGGNTFGSSSLFNNSTNSNTTKPAFGGLNFGGG
151 200 NNTTPSSTGNANTSNNLFGATANANKPAFSFGATTNDDKKTEPDKPAFSF
201 250 NSSVGNKTDQAPTTGFSFGSQLGGNKTVNEAAKPSLSFGSGSAGANPAG
251 300 ASQPEPTTNEPAKPALSFGTATSDNKTNTTTPSFSGAKSDENKAGATSK
301 350 PAFSFGAKPEEKKDDNSSKPAFSFGAKSNEDKQDGTAKPAFSFGAKPAEK
351 400 NNNETSKPAFSFGAKSDEKKDGDASKPAFSFGAKPDENKASATSKPAFSF
401 450 GAKPEEKKDDNSSKPAFSFGAKSNEDKQDGTAKPAFSFGAKPAEKNNNET
451 500 SKPAFSFGAKSDEKKDGDASKPAFSFGAKSDEKKDSDSSKPAFSFGTKSN

```

501 550 EKKDSGSSKPAFSFGAKPDEKKNDEVSKPAFSFGAKANEEKESDESKSAF
551 600 SFGSKPTGKEEGDGAKAAISFGAKPEEQKSSDTSKPAFTFGAQKDNEKKT
601 650 EESSTGKSTADVKSDDLKLNKPVSLDNKTLDDLVTWKWTNQLTE
651 700 SASHFEQYTKKINSWDQVLVKGGEQISQLYSDAVMAEHSQNKIDQSLQYI
701 750 ERQQDELENFLDNFETKTEALLSDVVSTSSGAAANNNDQKRQQAYKTAQT
751 800 LDENLNSLSSNLSSLIVEINNVSNTFNKTTNIDINNEDENIQLIKILNSH
801 824 FDALRSLDDNSTSLEKQINSIKKHHHHHH