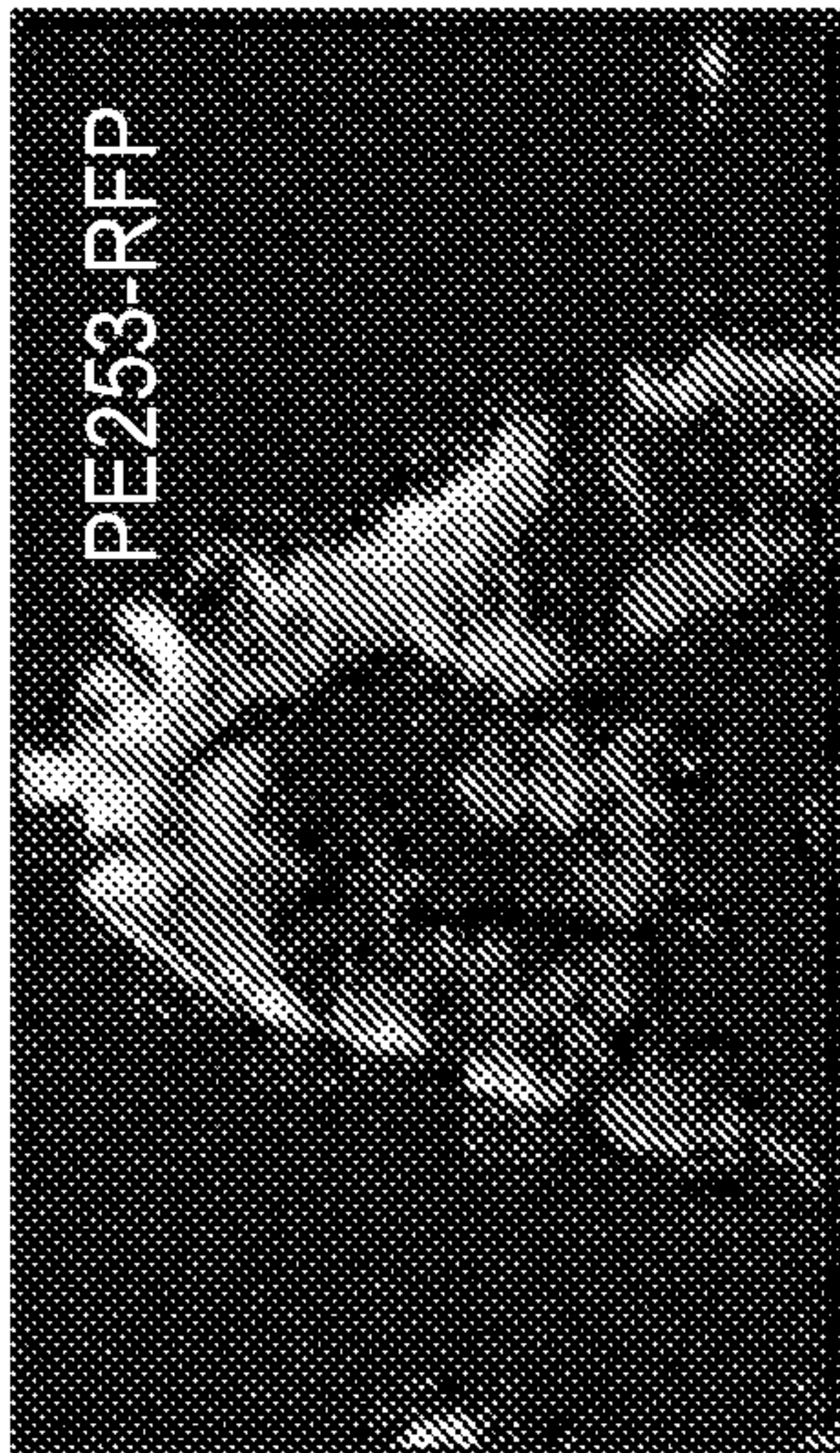
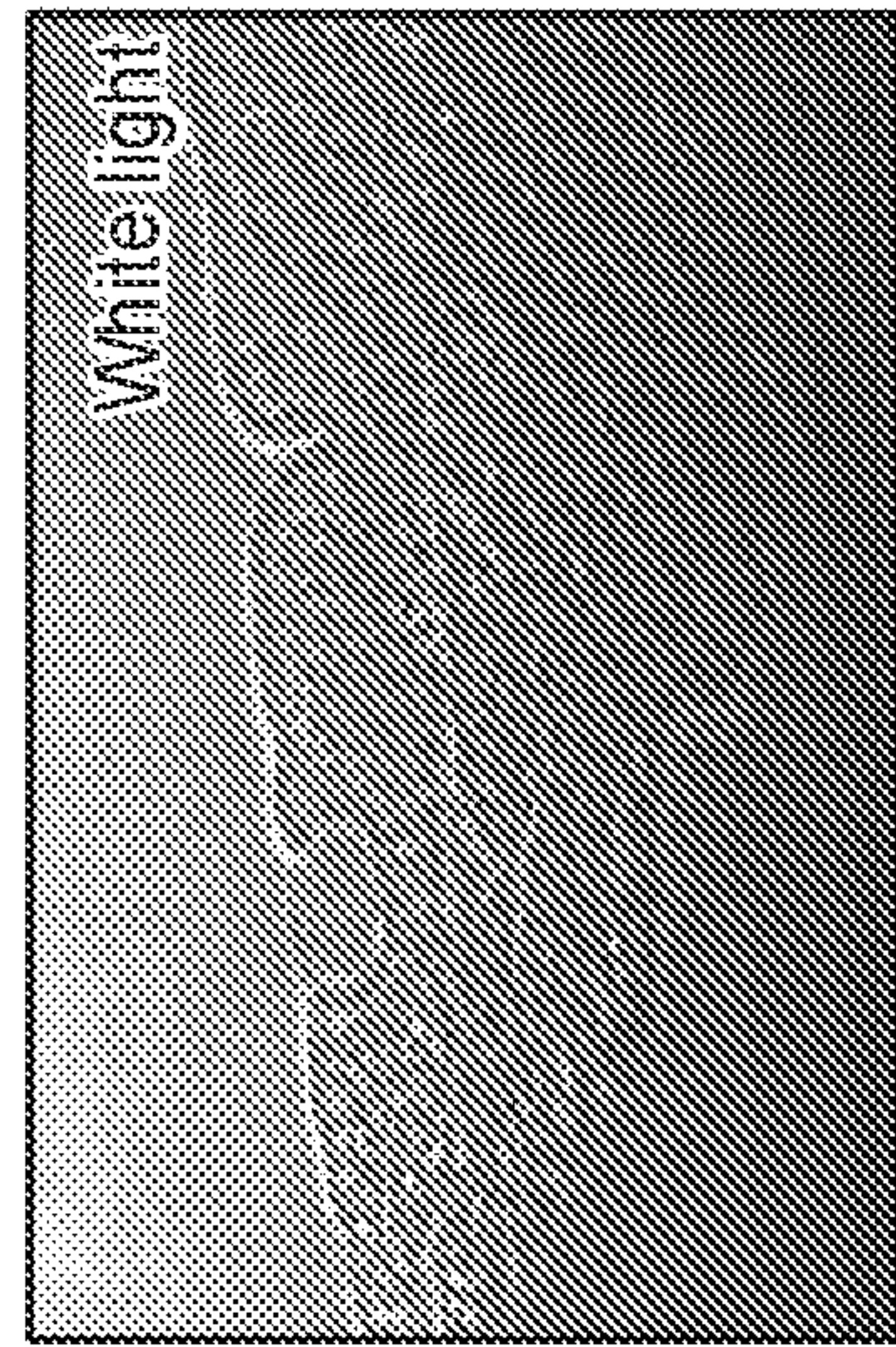
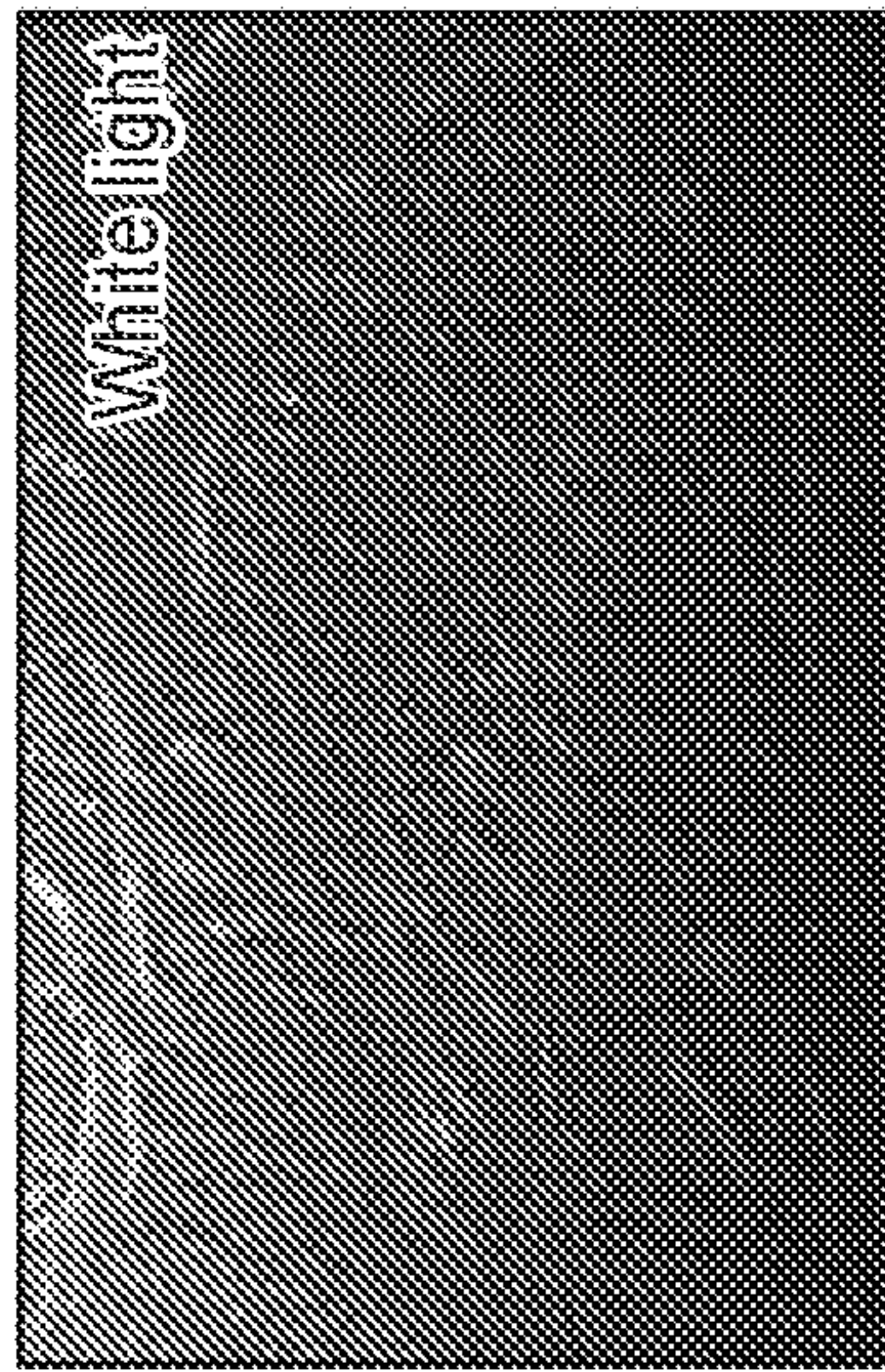
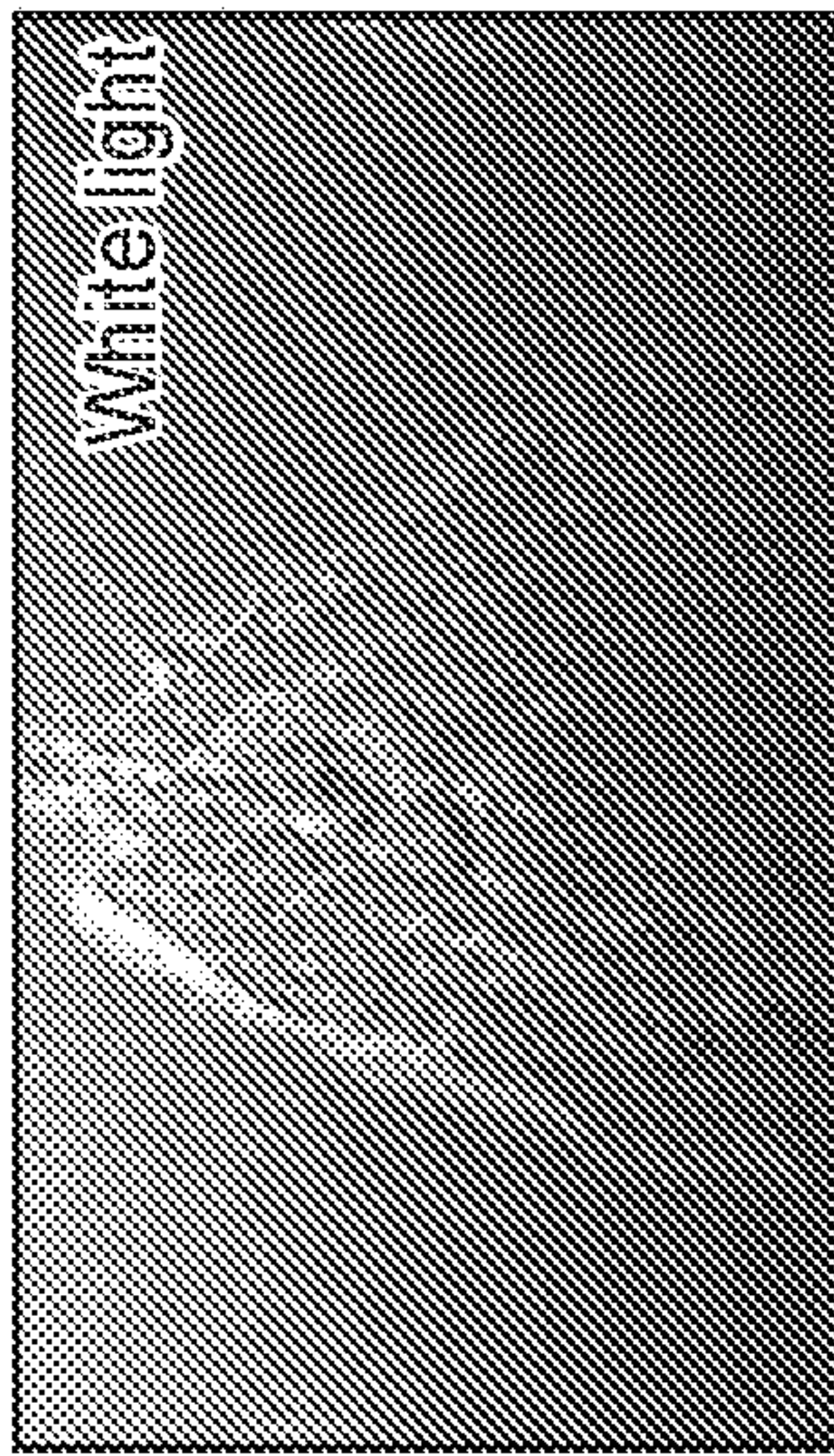
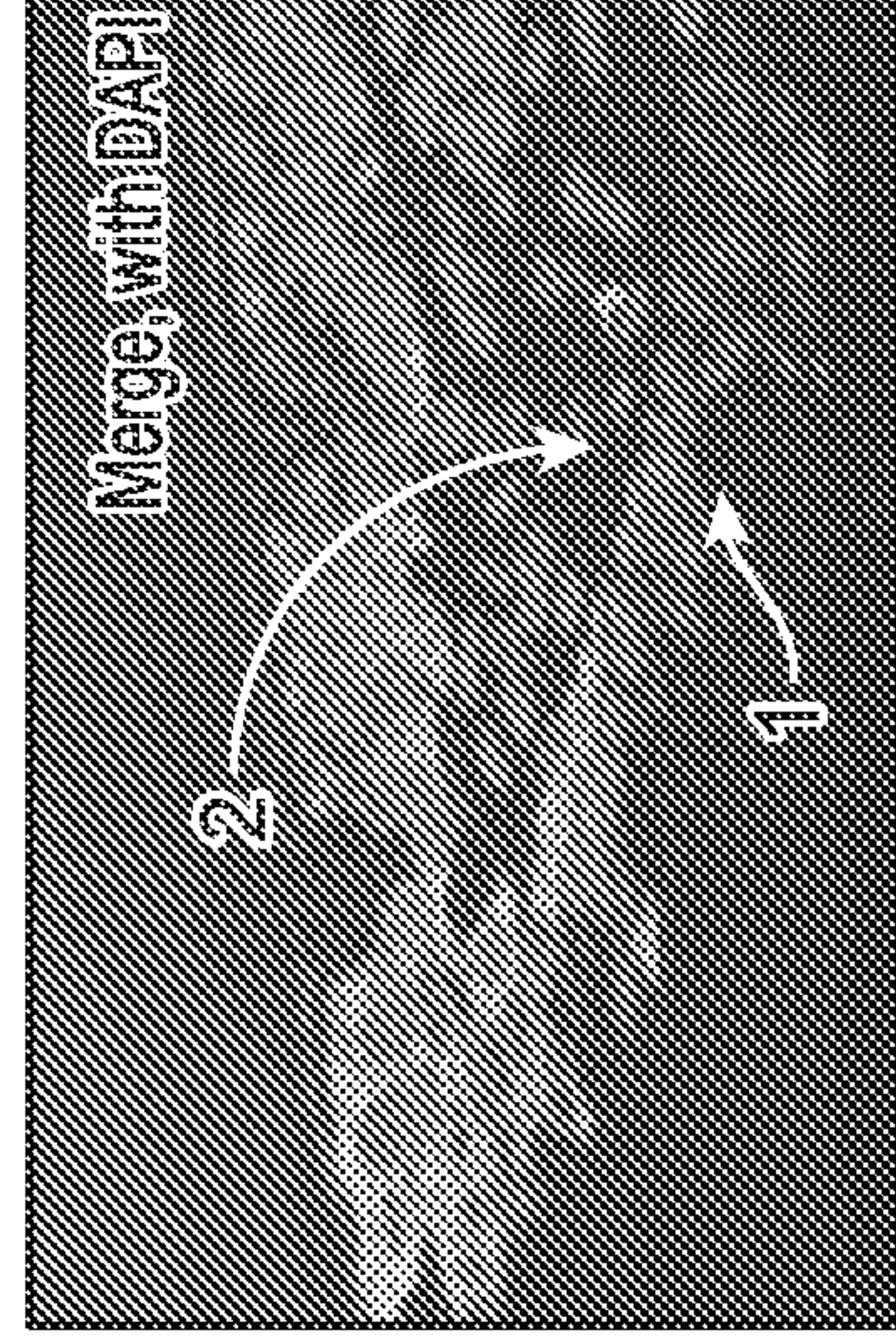
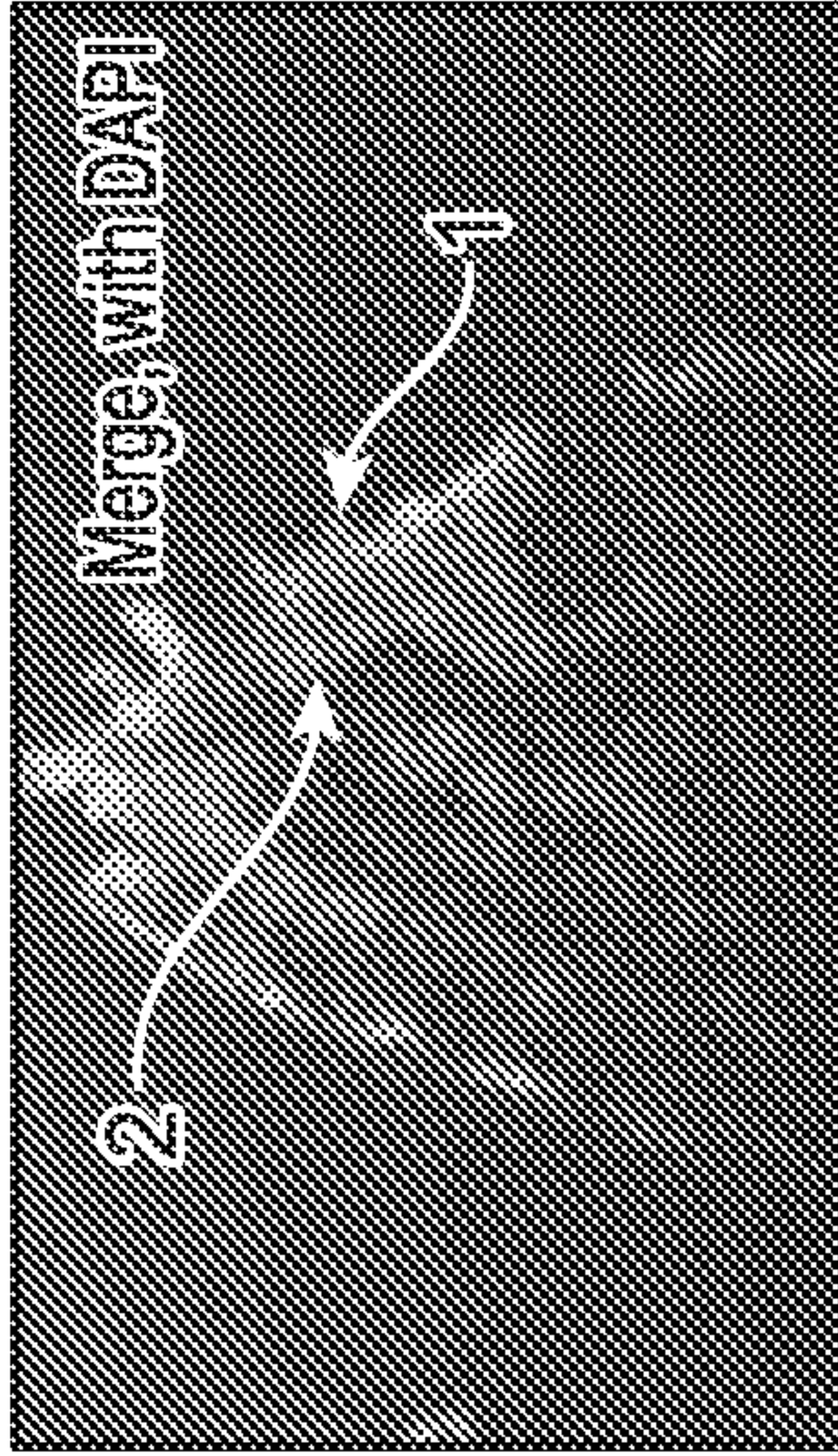


FIG. 1

Anti-PE: 1/1,000
AbII: 1/100



AGEL

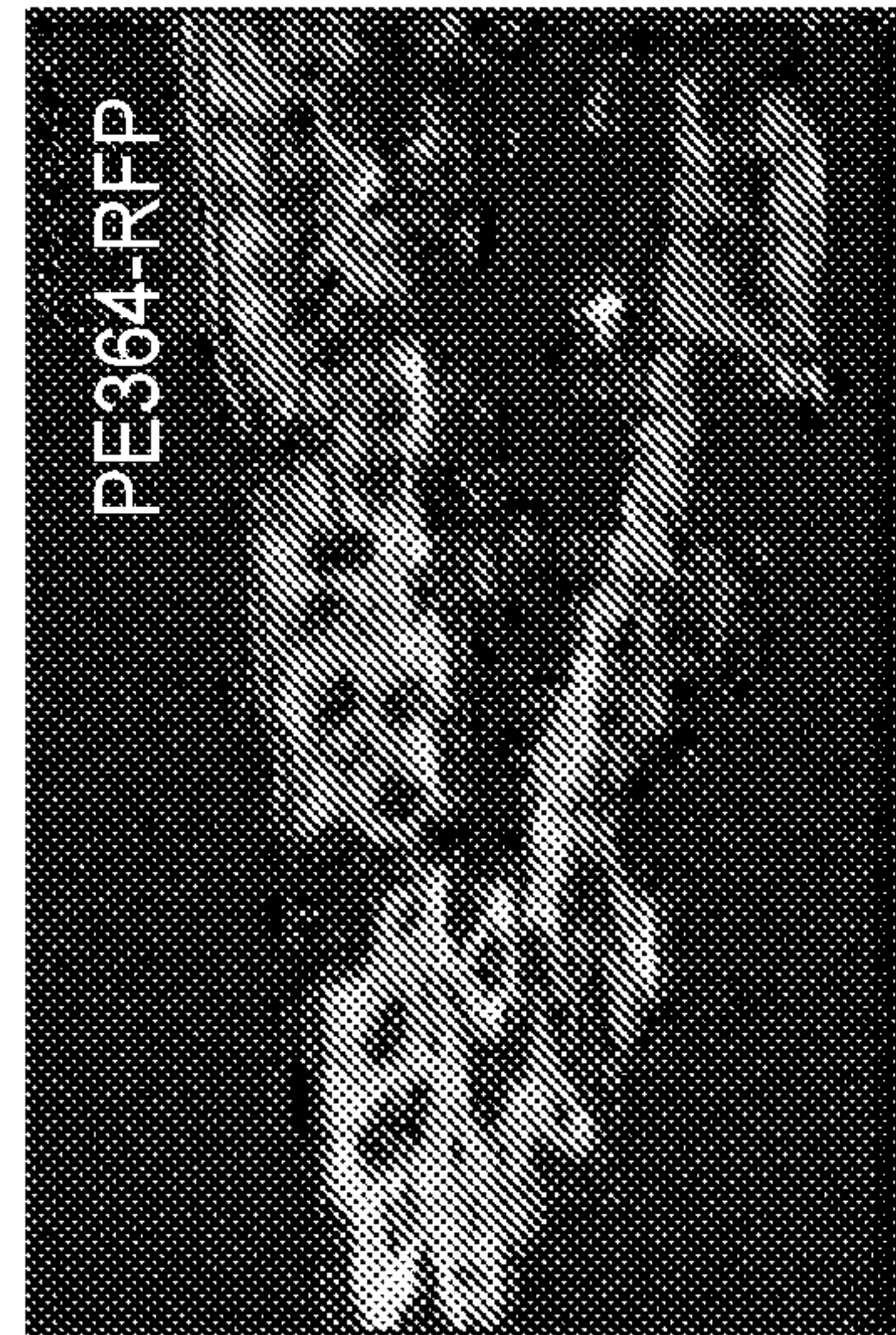
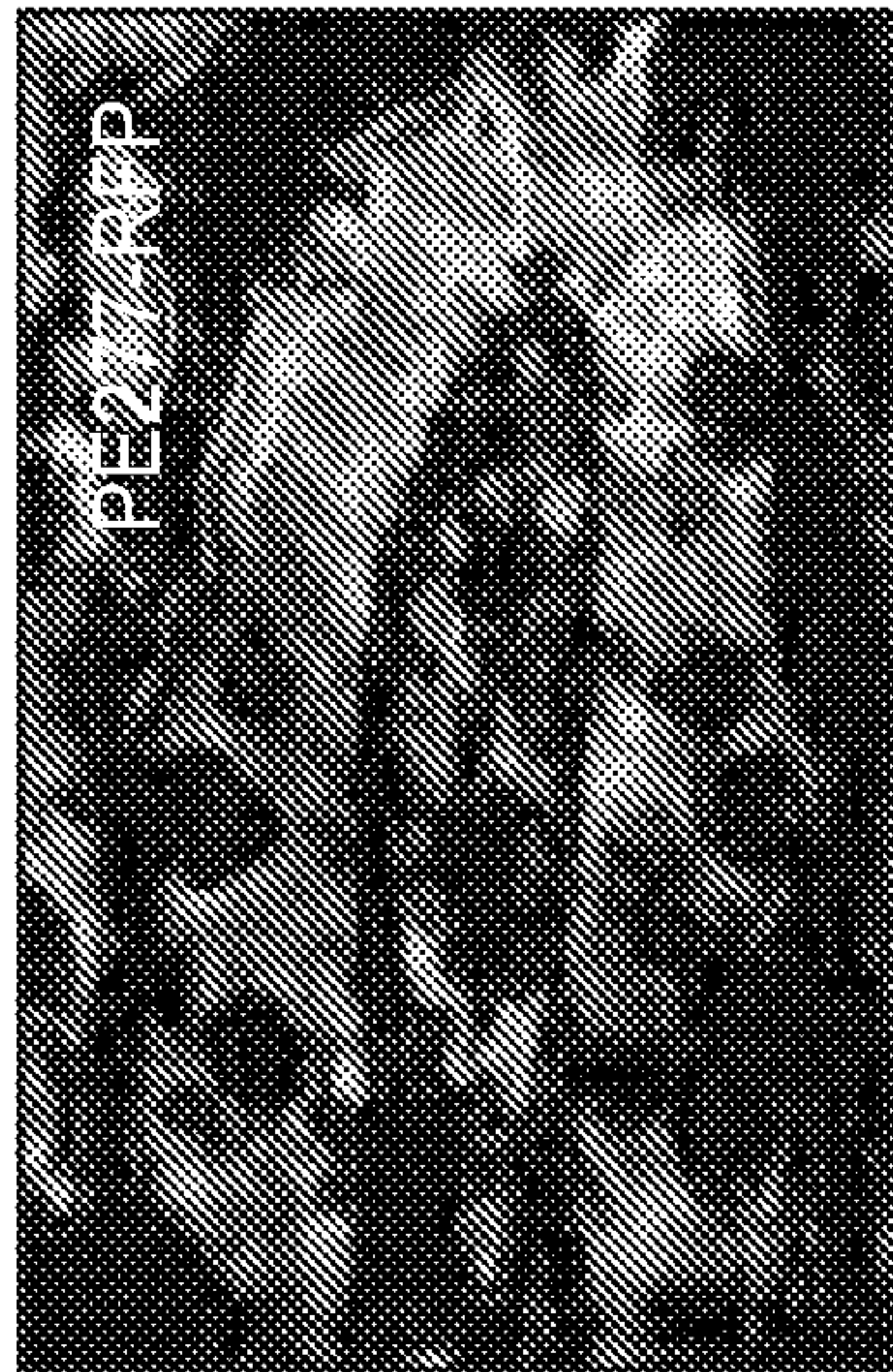


FIG. 2

Anti-PE: 1/1,000
AbII: 1/100



FIG. 2A

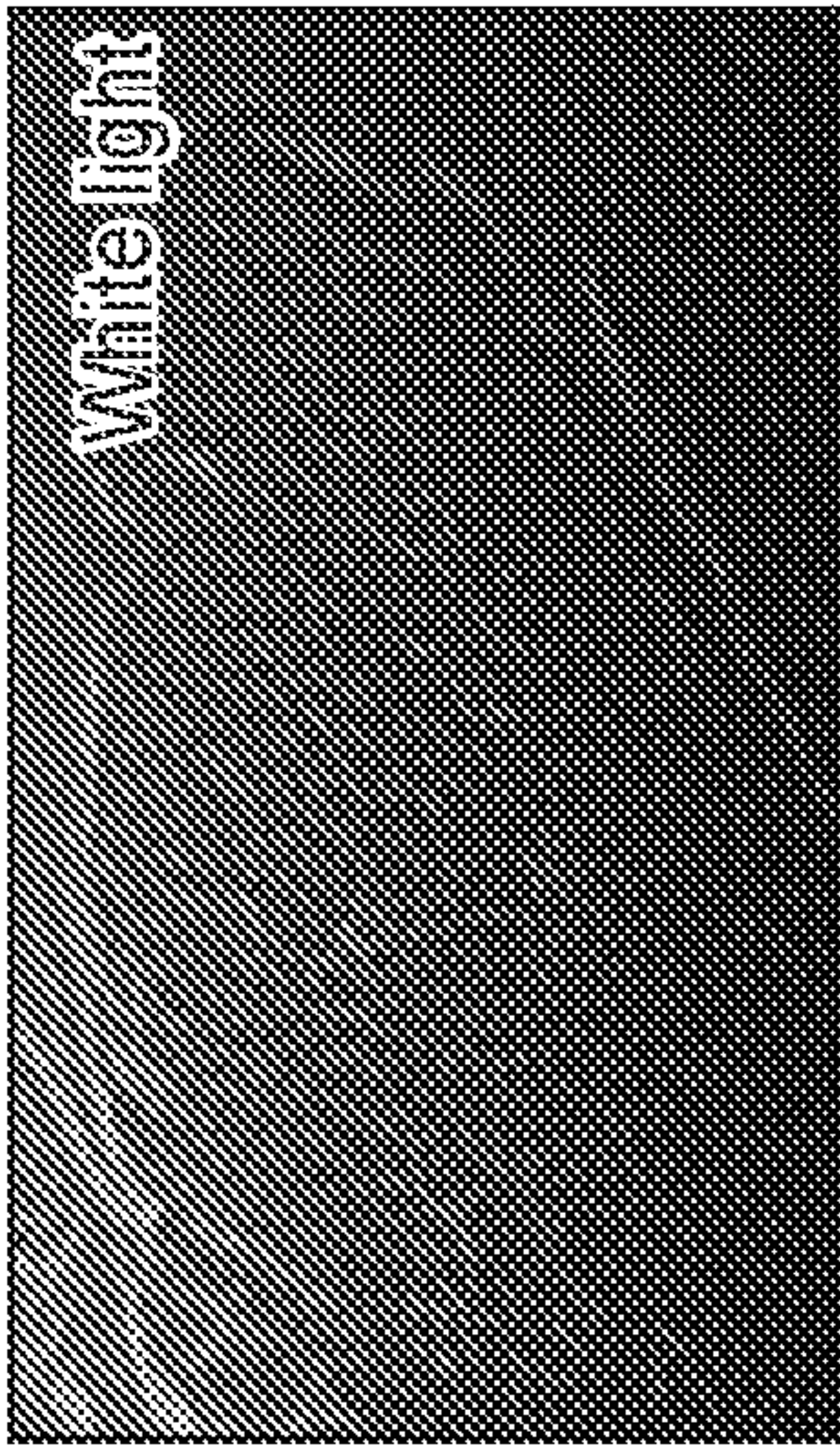


FIG. 2B

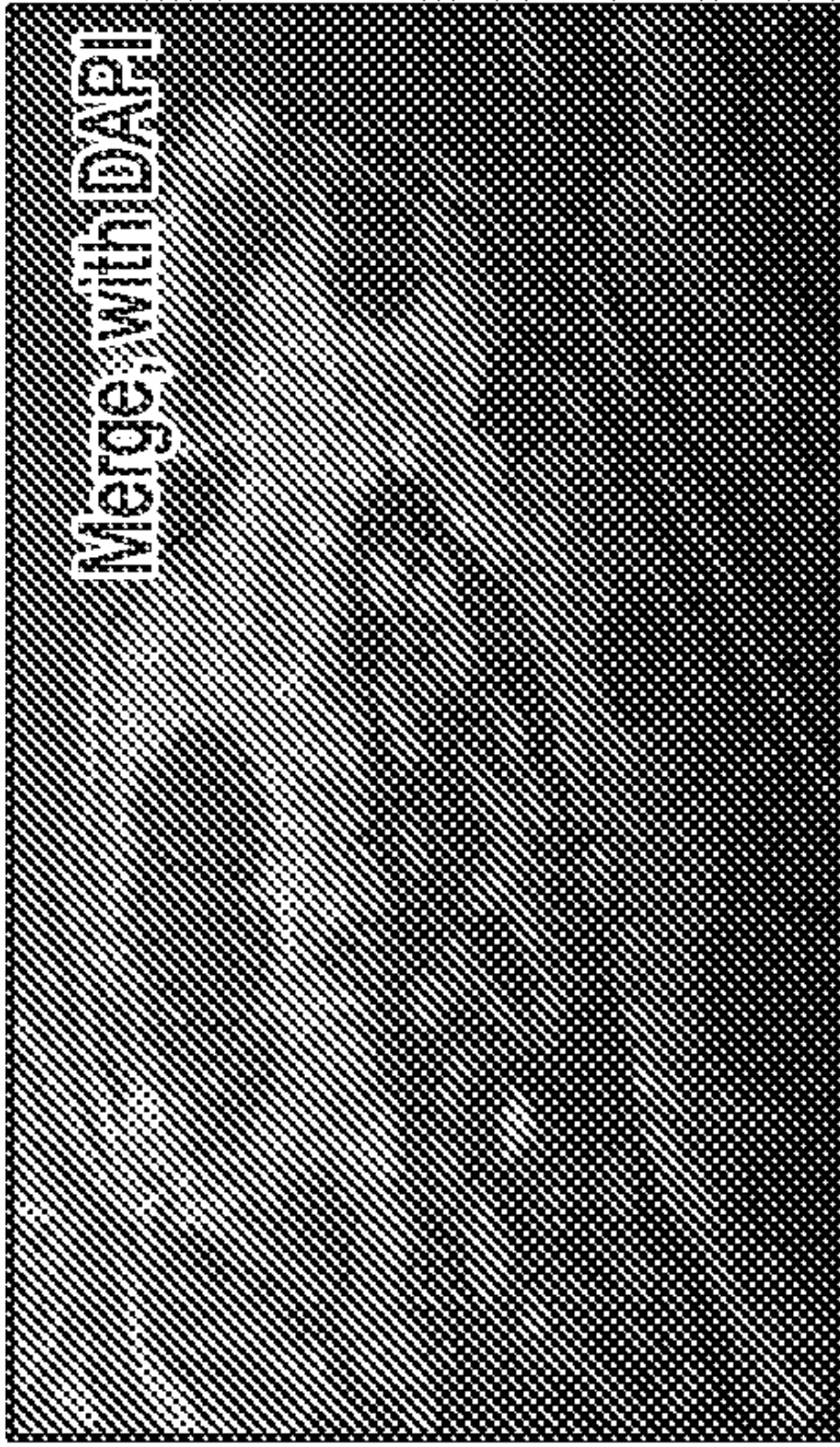


FIG. 2C

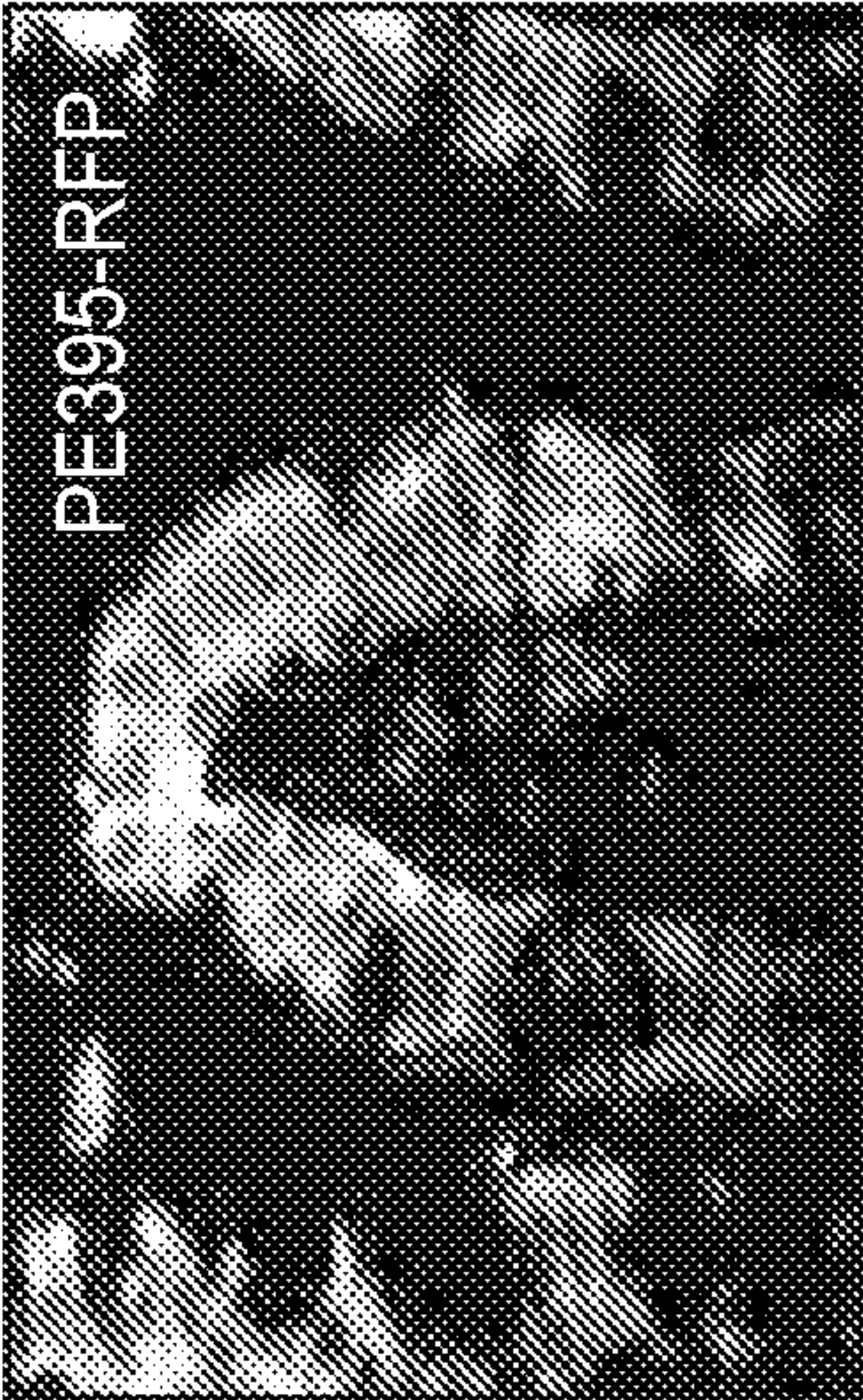


FIG. 2D



FIG. 2E



FIG. 2F

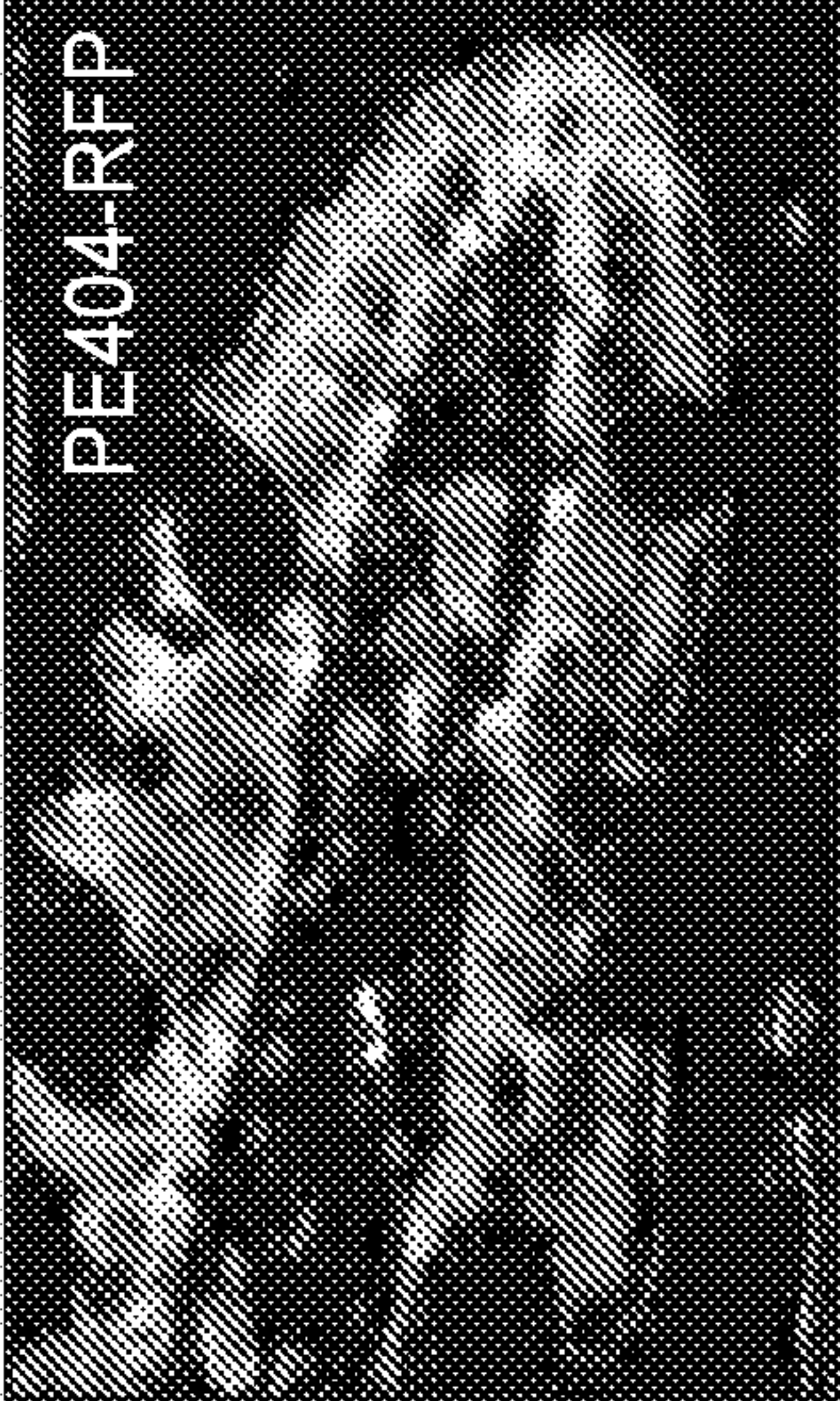


FIG. 2G

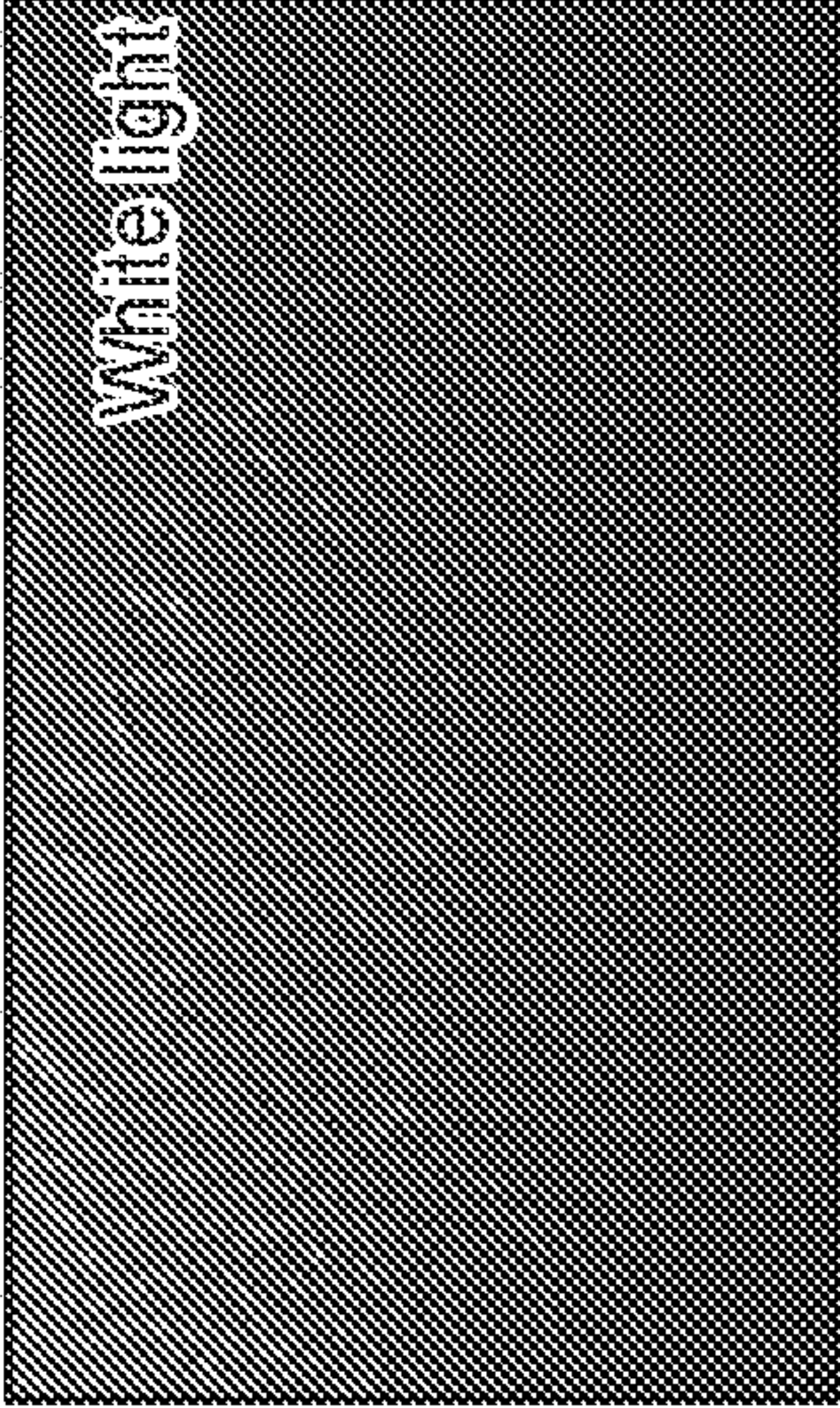


FIG. 2H

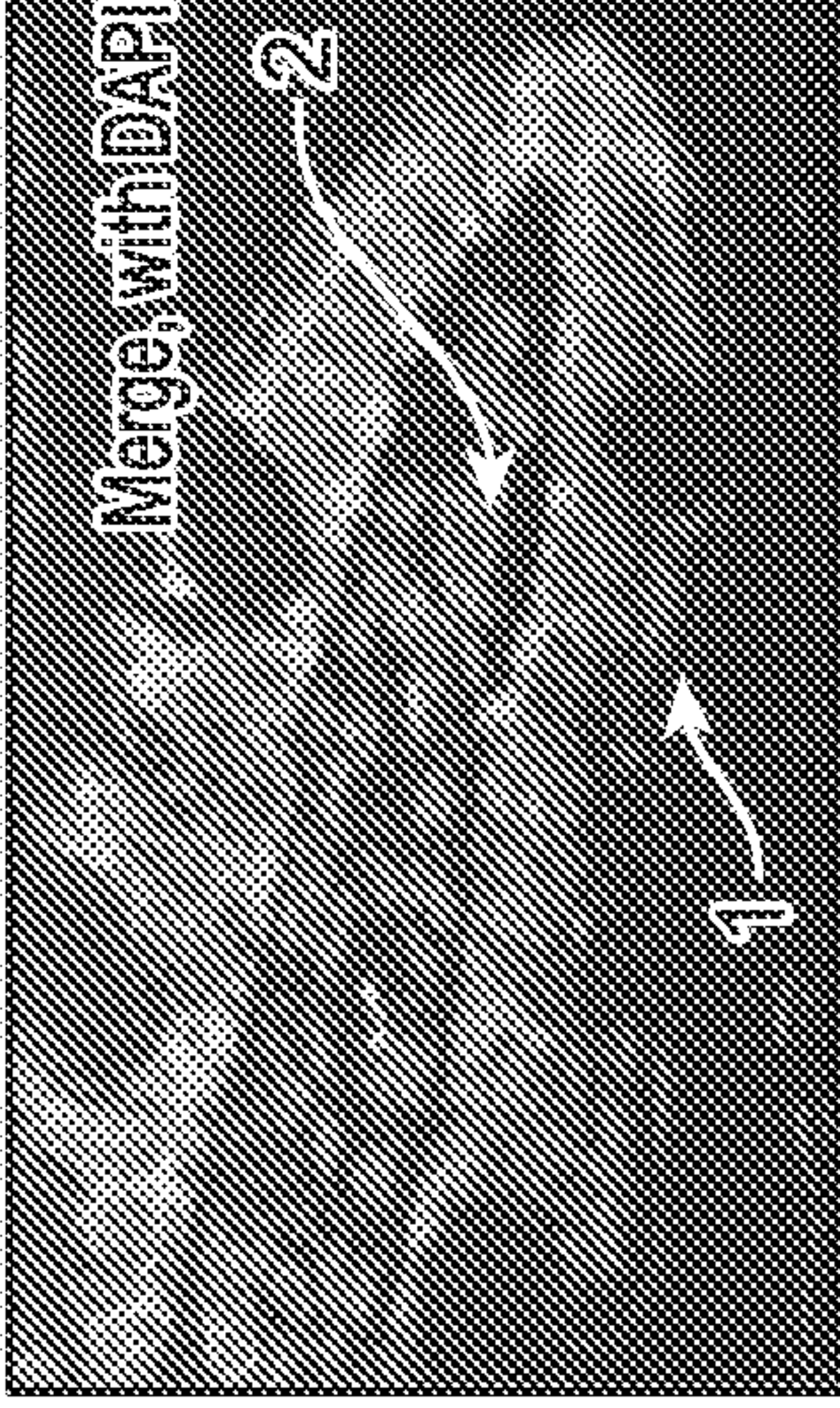


FIG. 2I

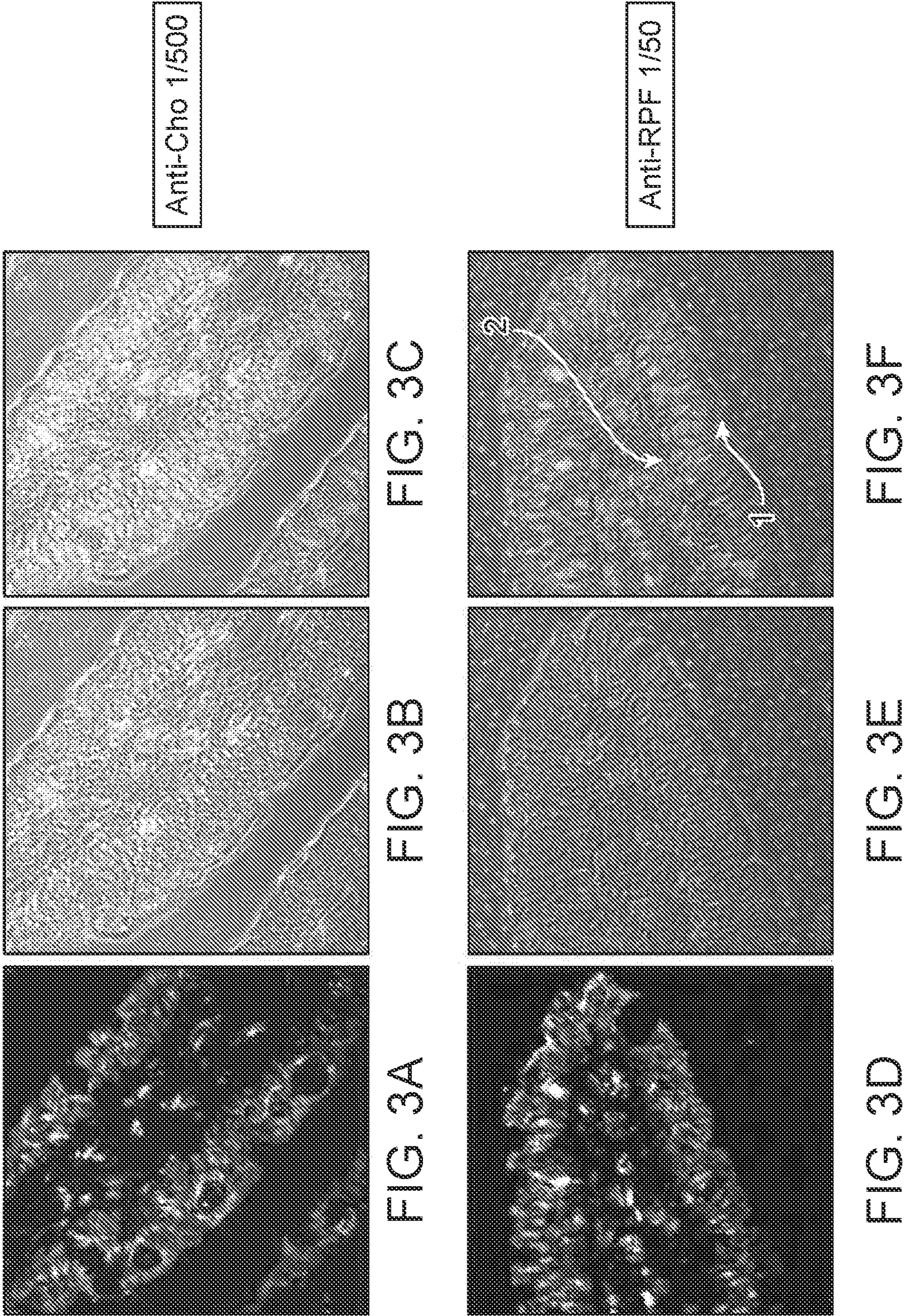


FIG. 3

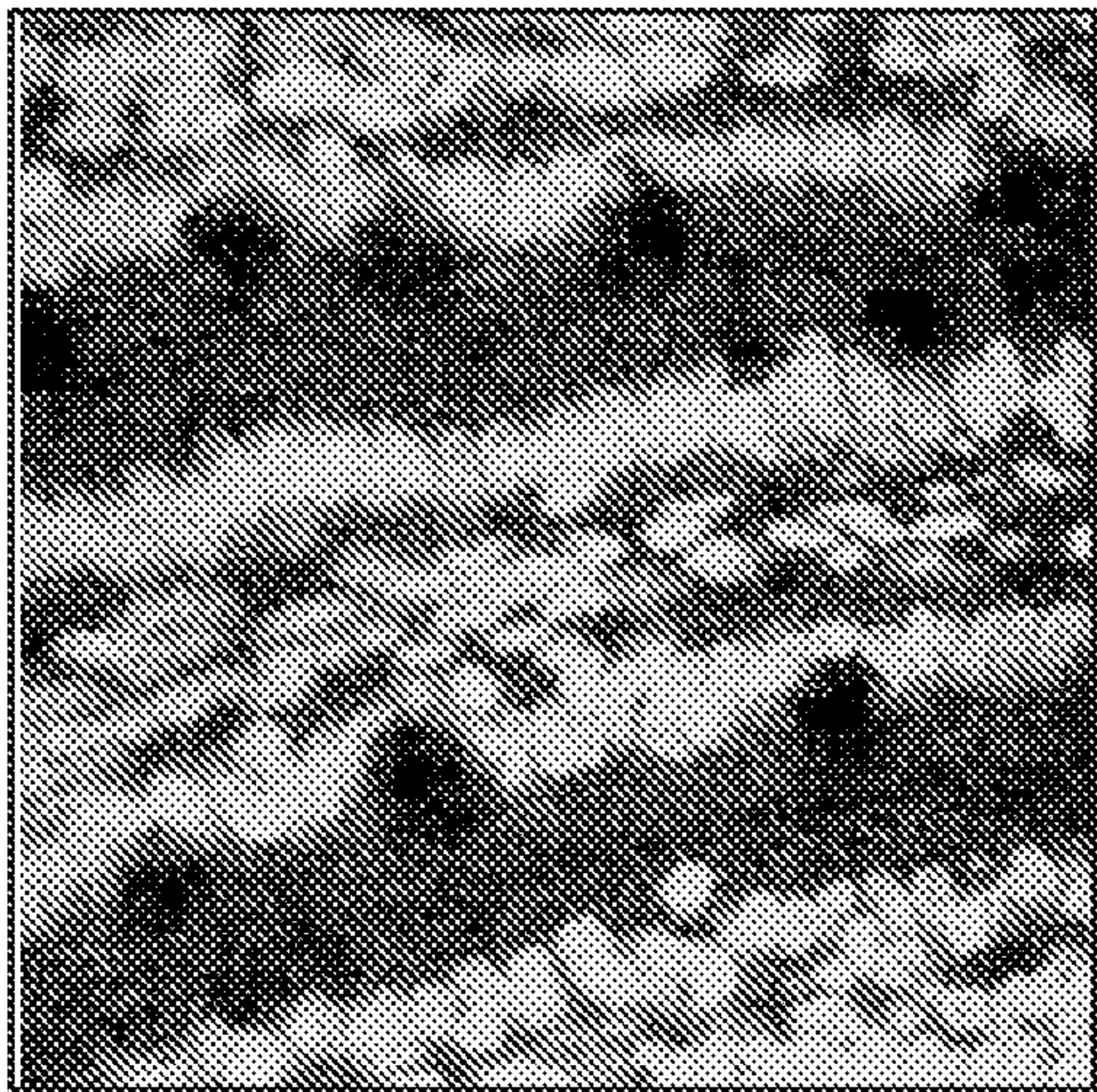


FIG. 4C

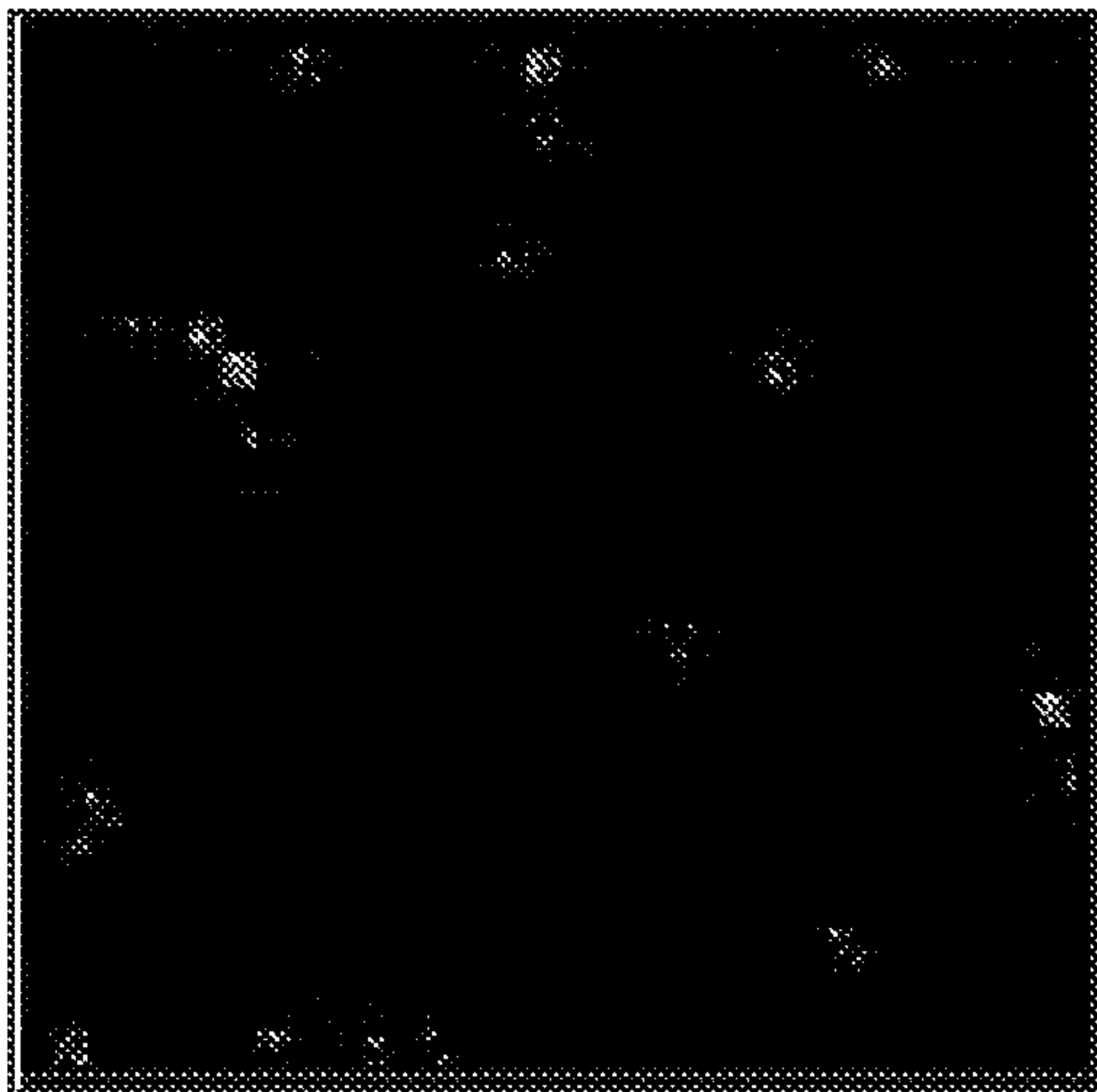


FIG. 4B

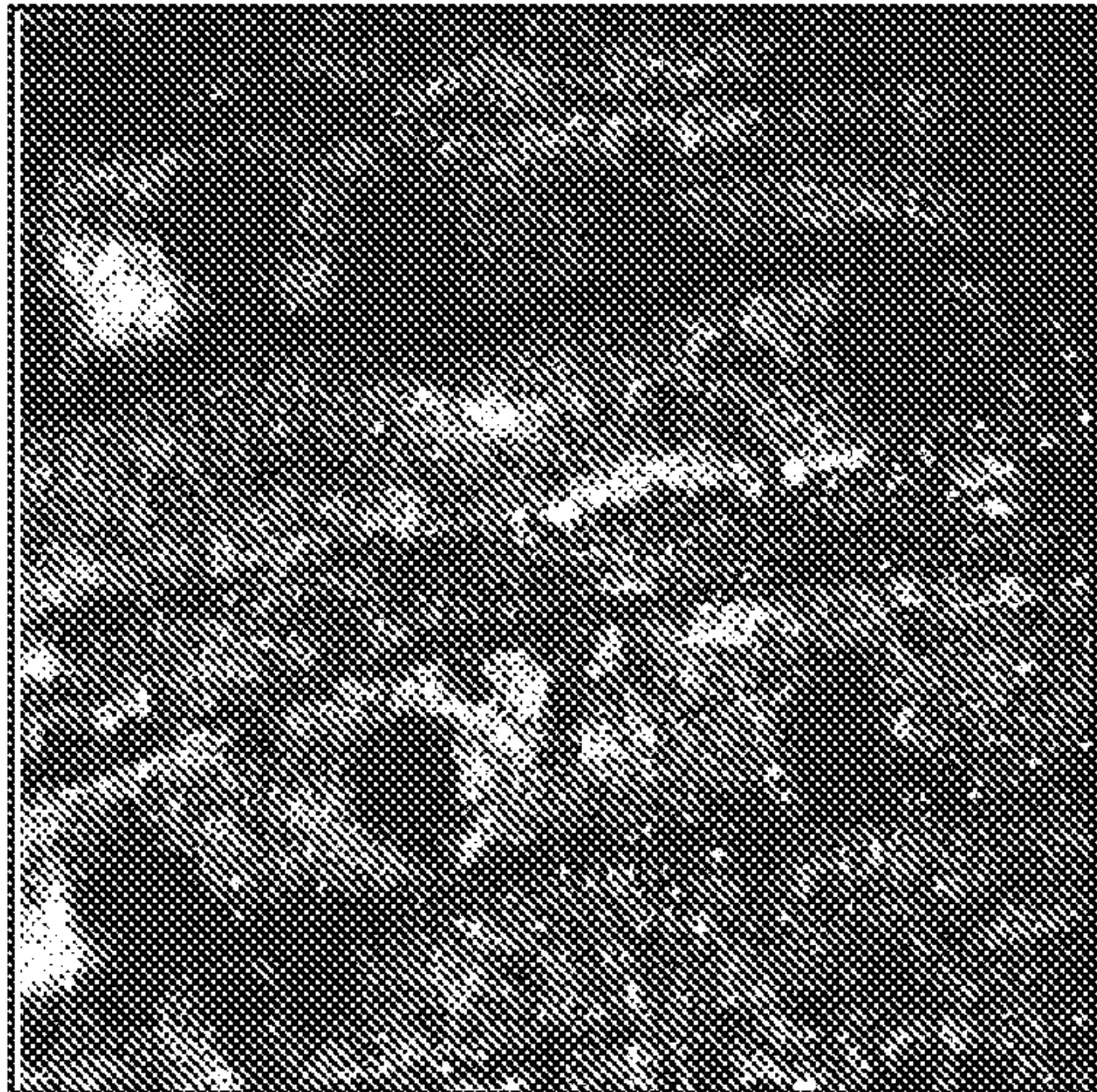


FIG. 4A

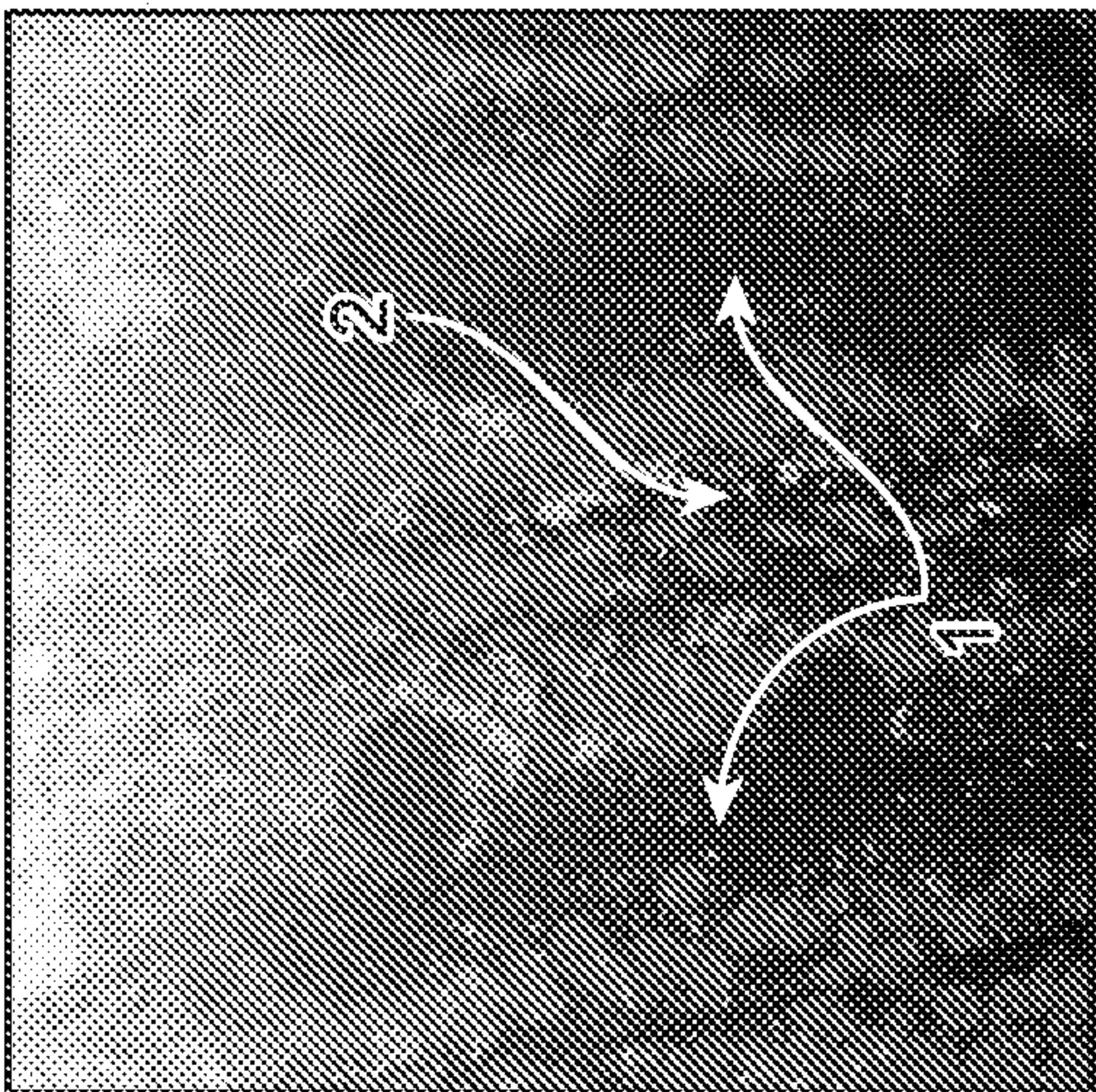


FIG. 4E

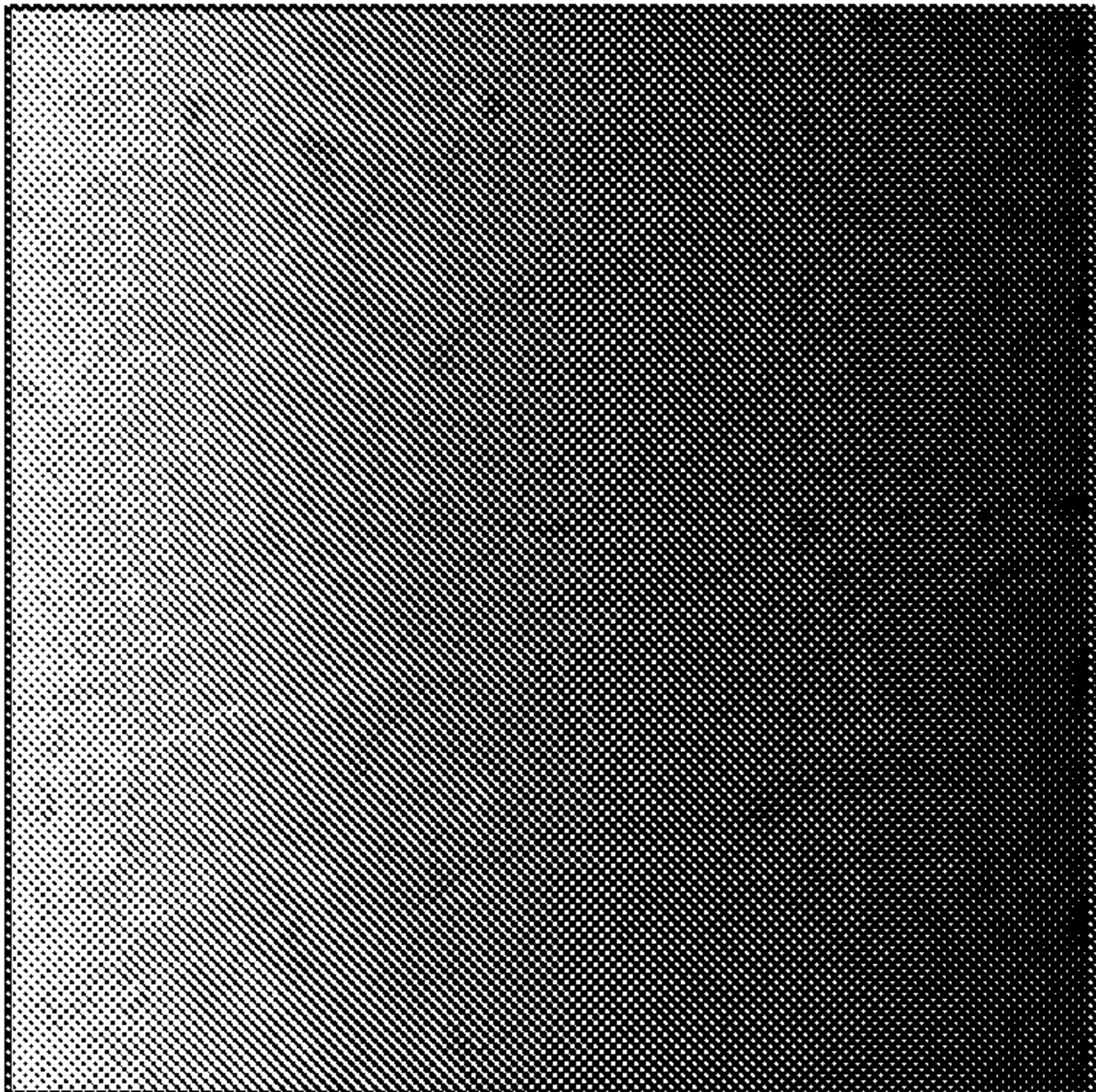


FIG. 4D

FIG. 4

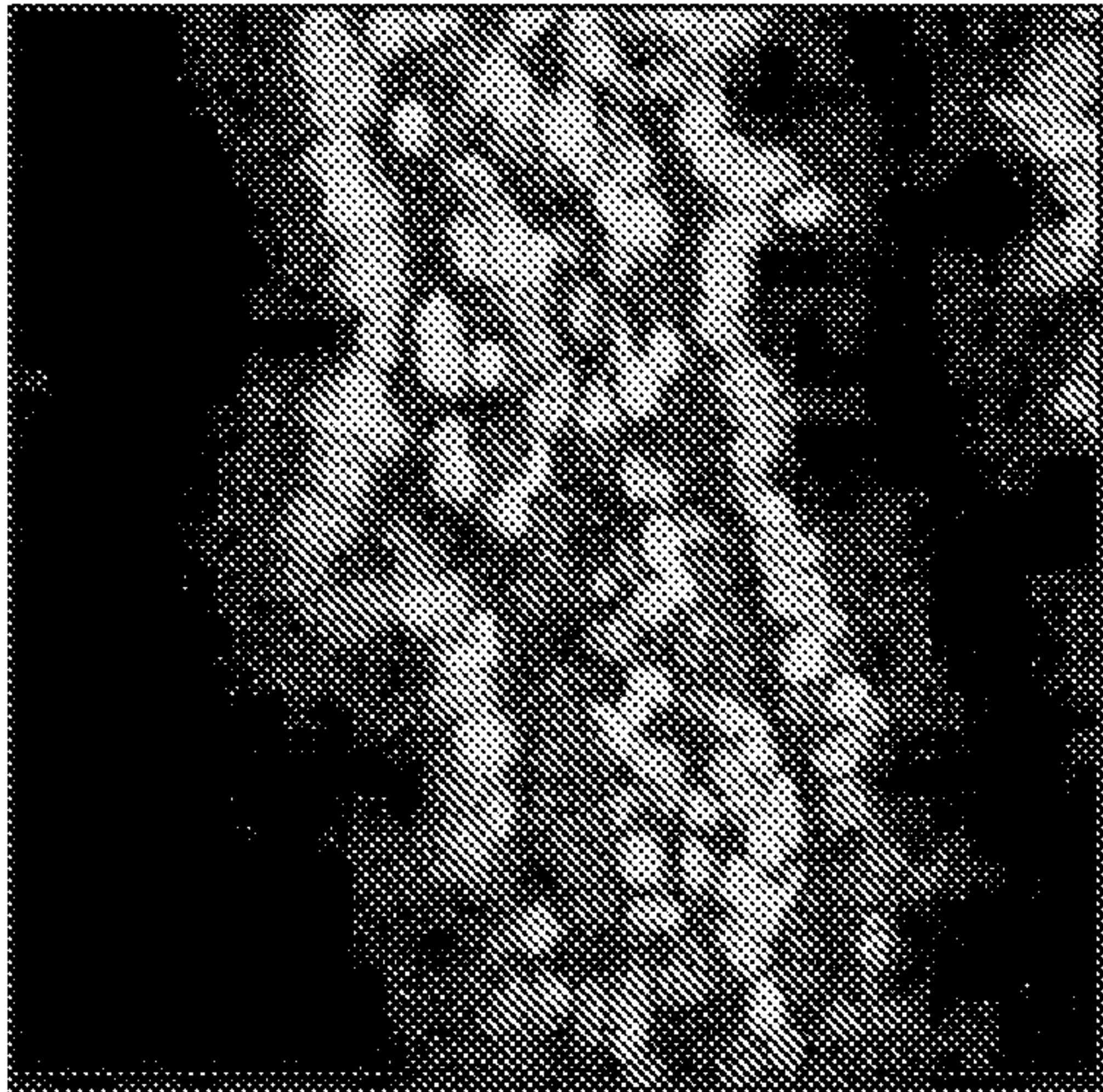


FIG. 5C

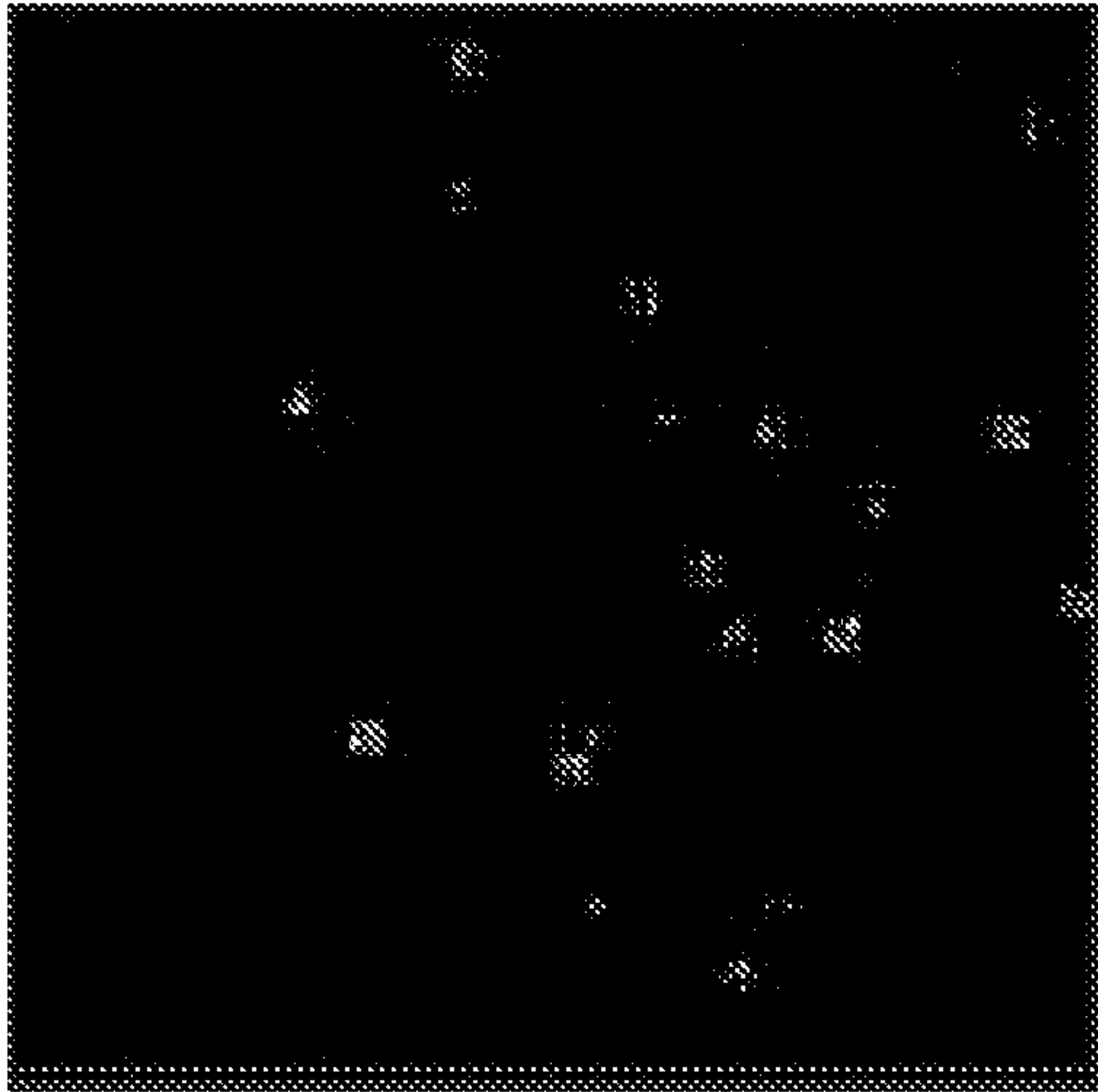


FIG. 5B

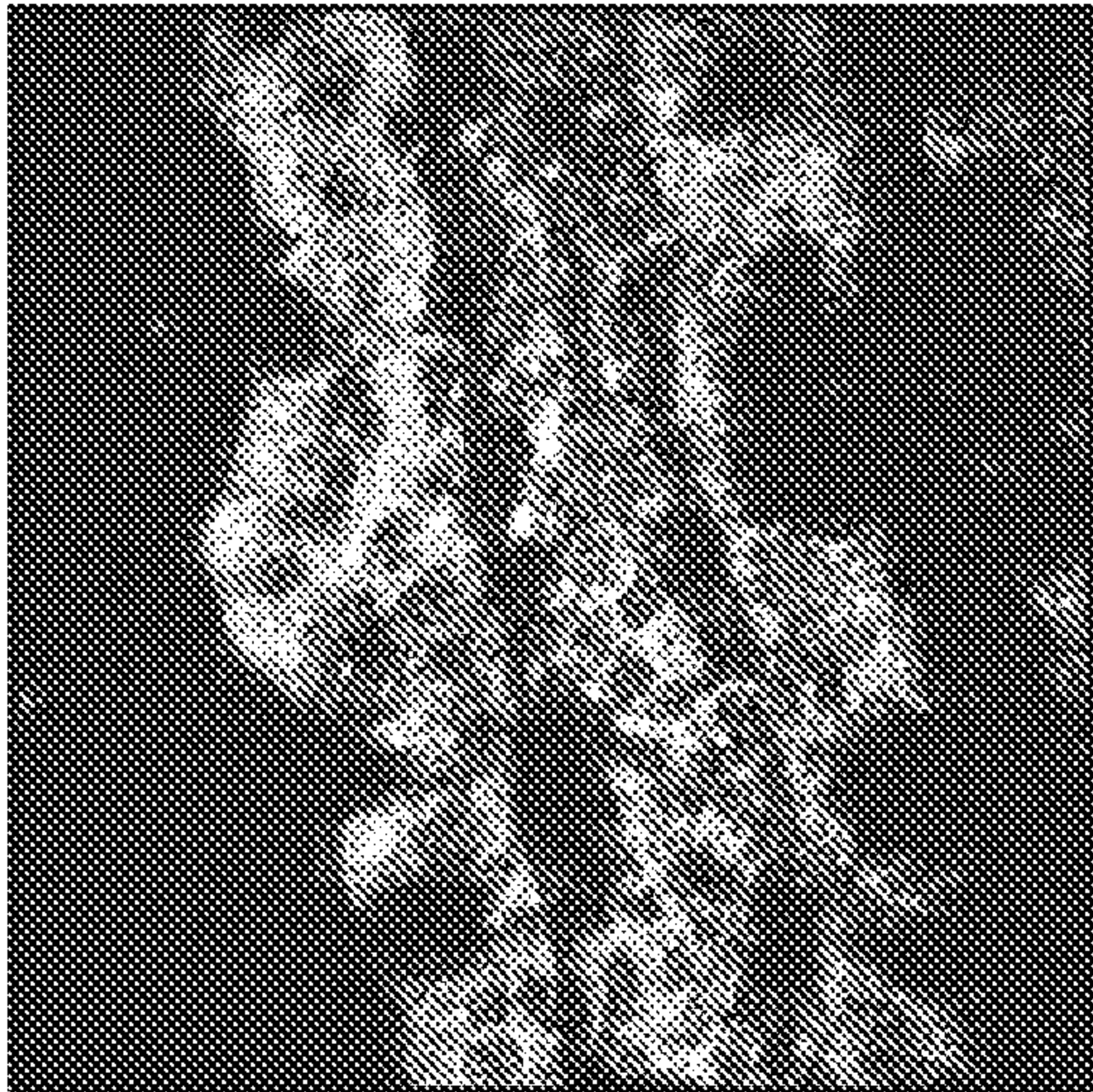


FIG. 5A

FIG. 5

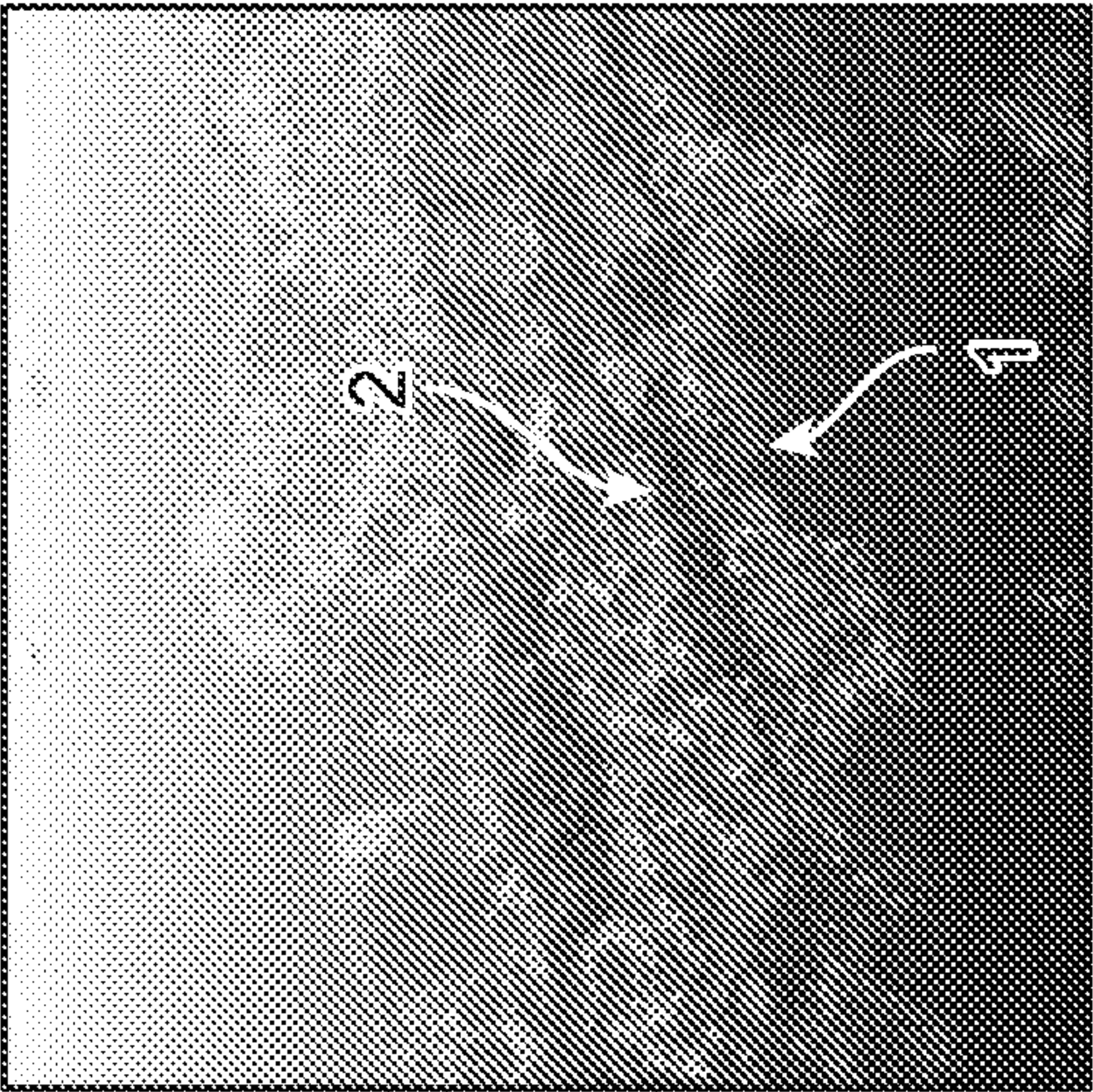


FIG. 5E

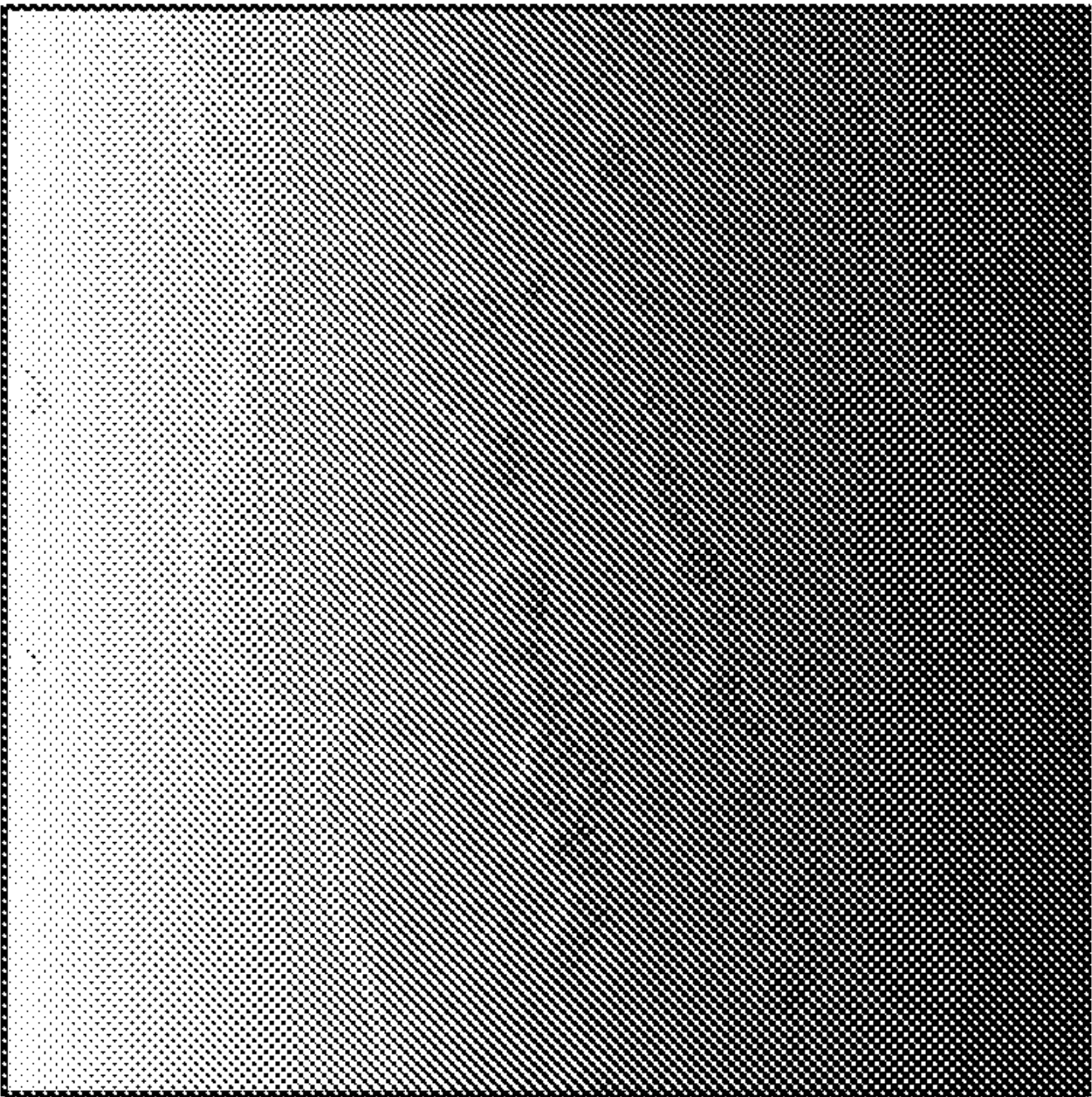


FIG. 5D



FIG. 6C

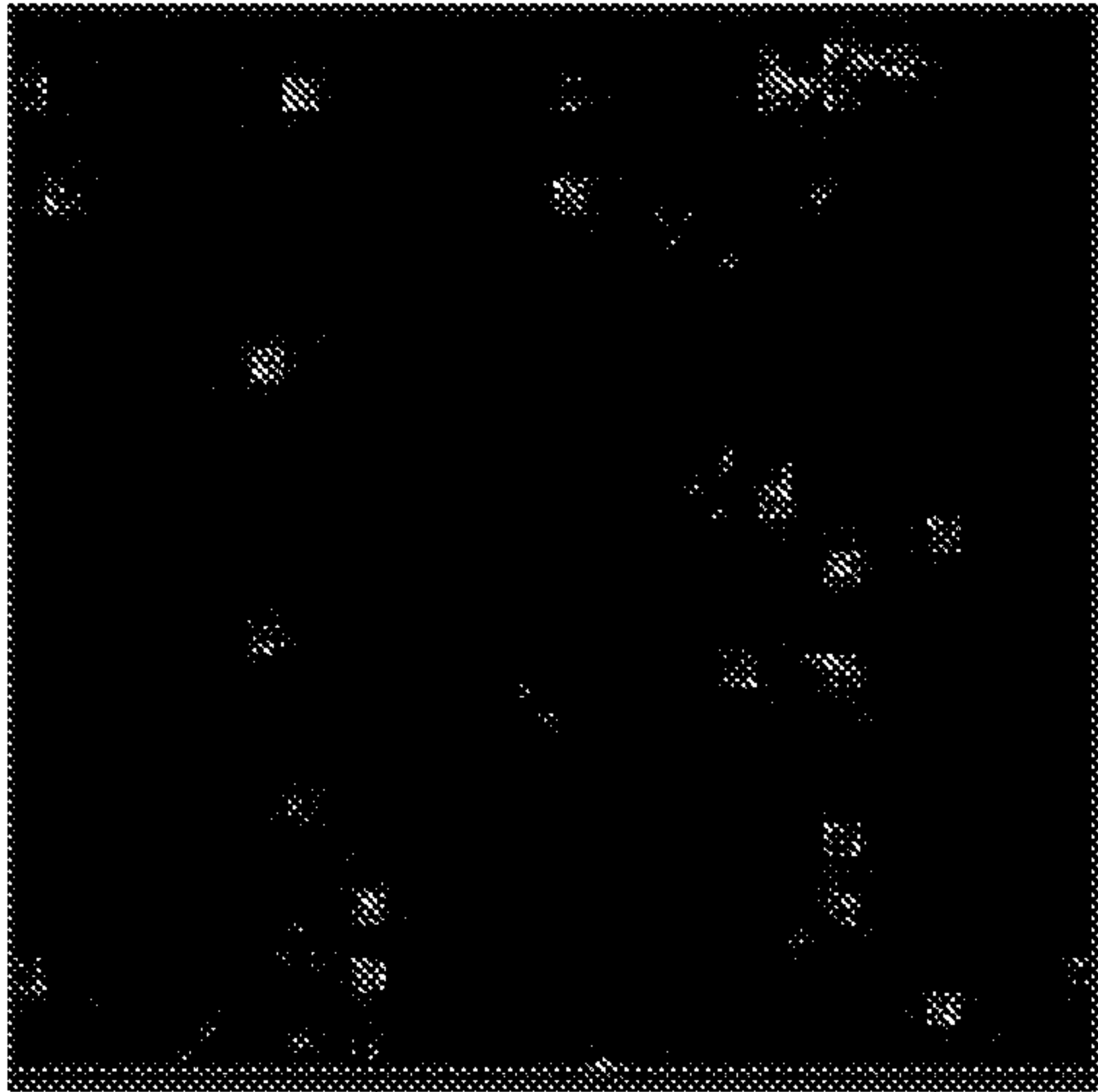


FIG. 6B

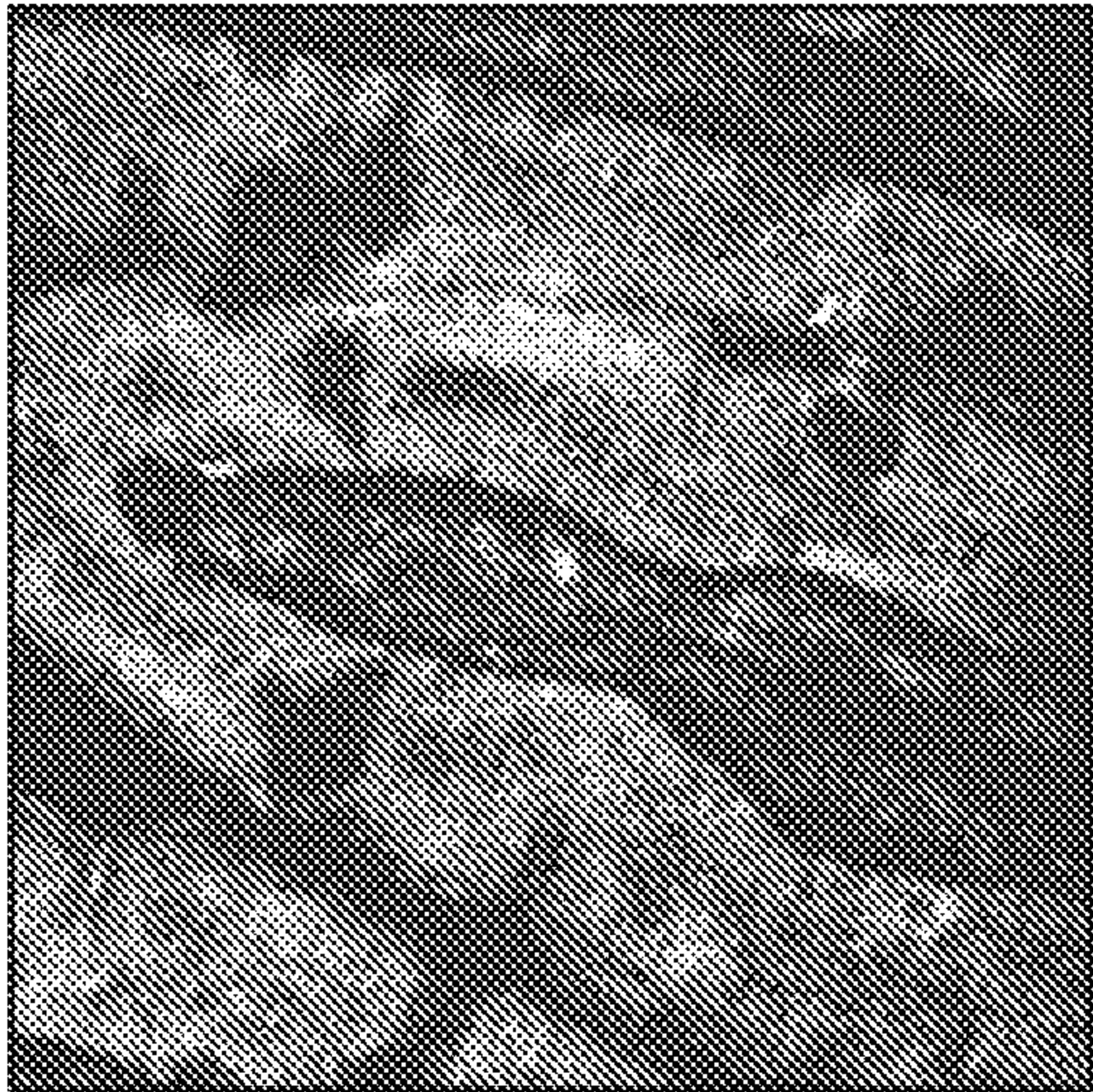


FIG. 6A

FIG. 6

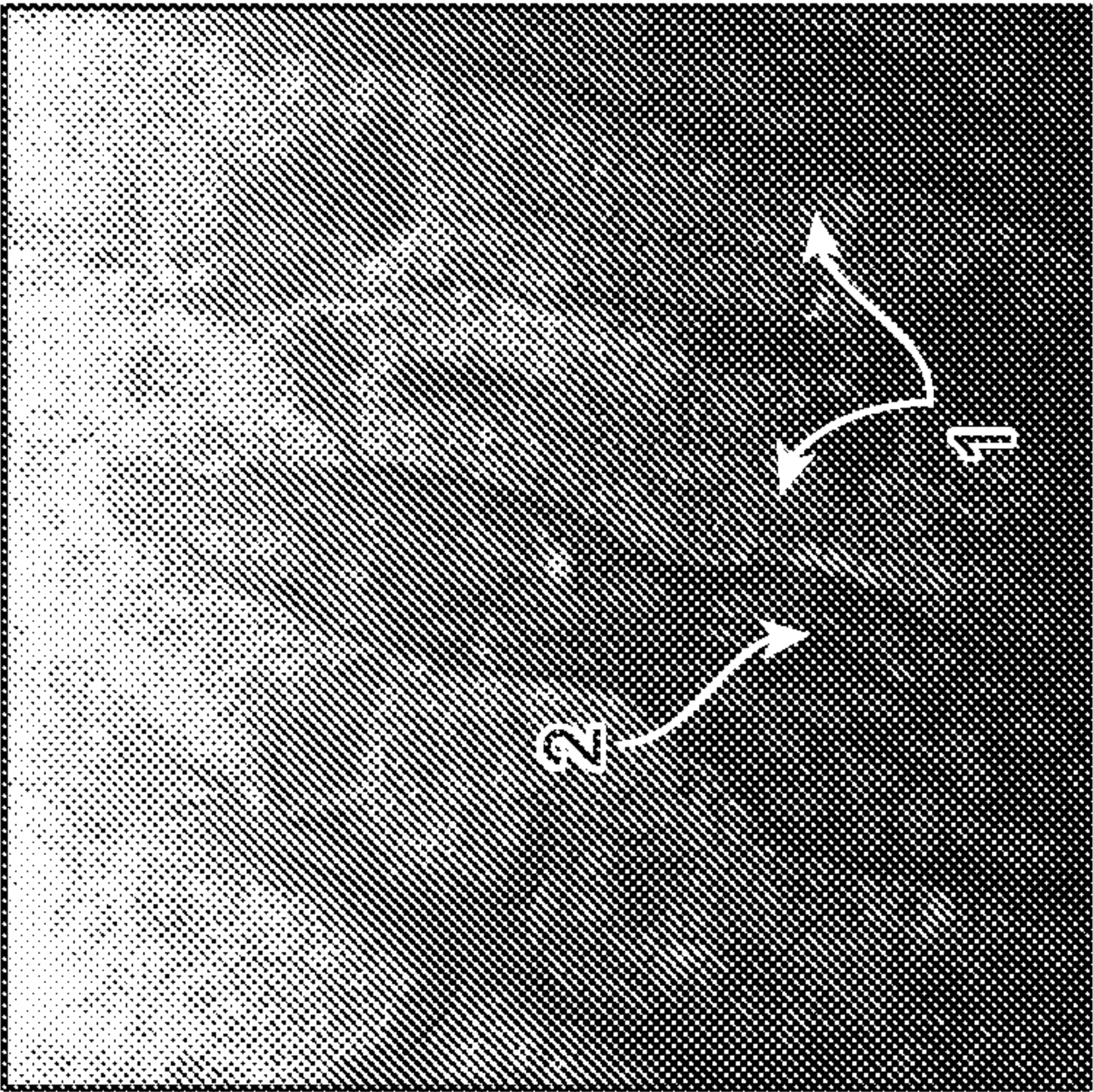


FIG. 6E

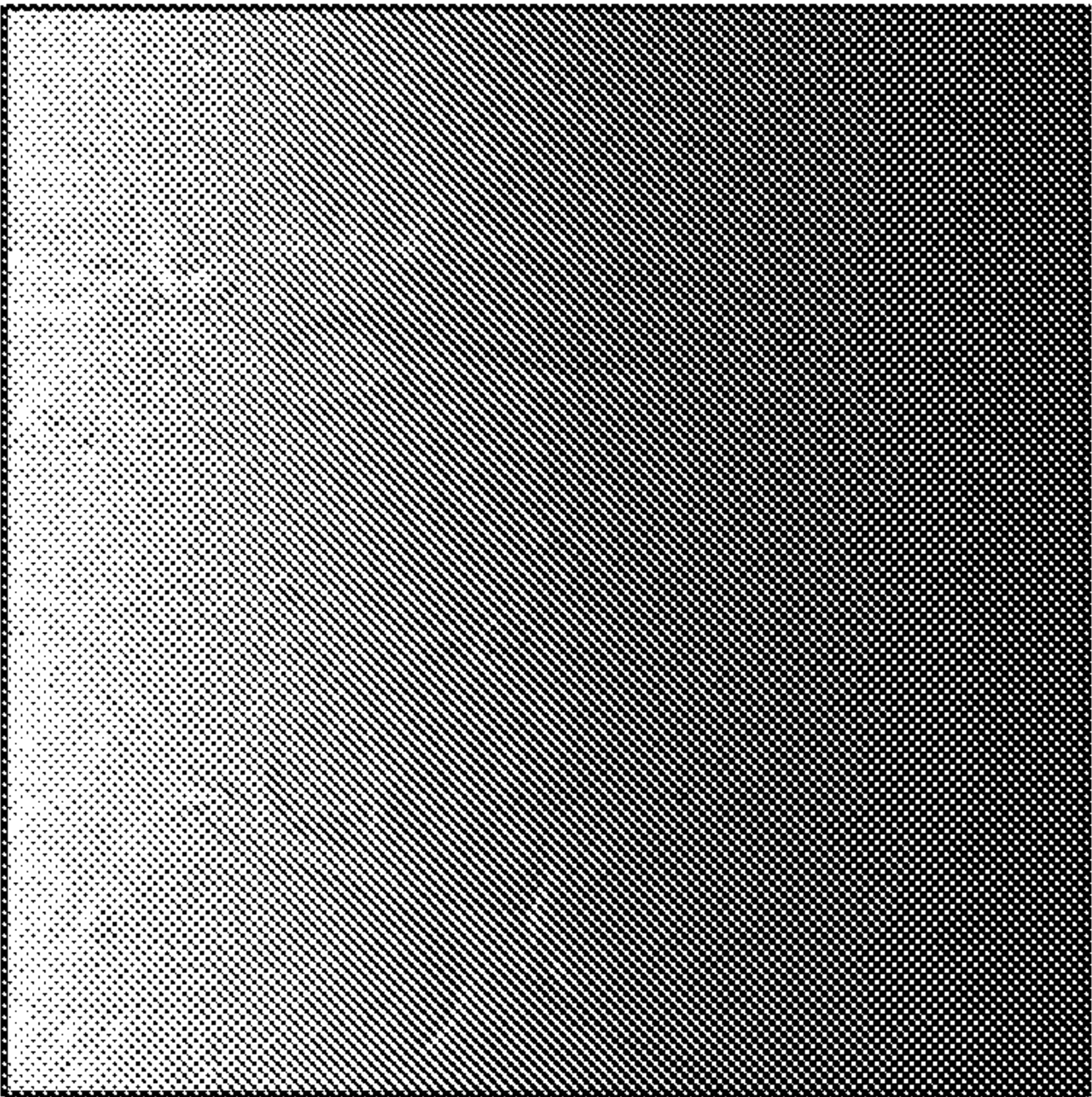


FIG. 6D



FIG. 7C

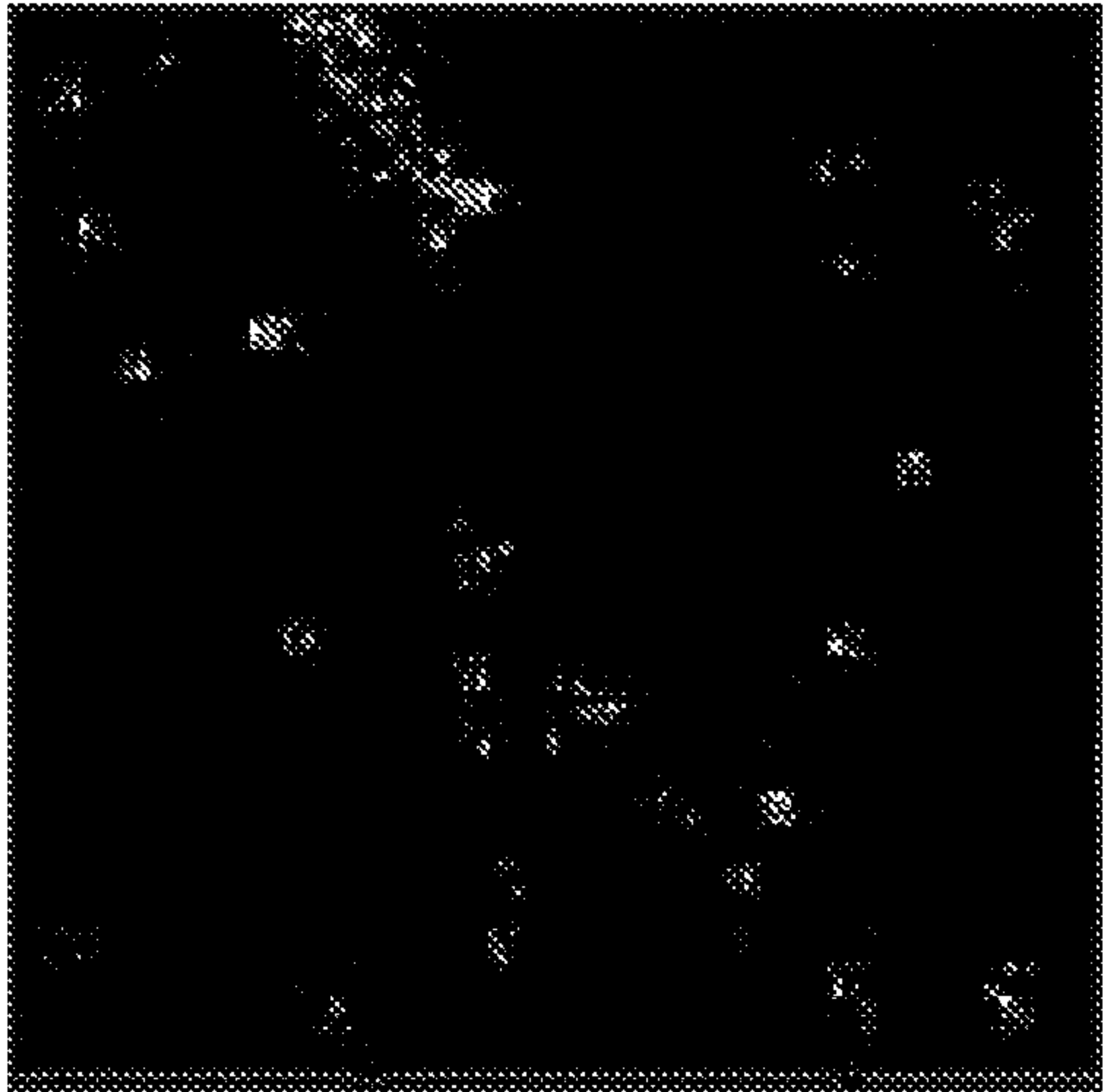


FIG. 7B

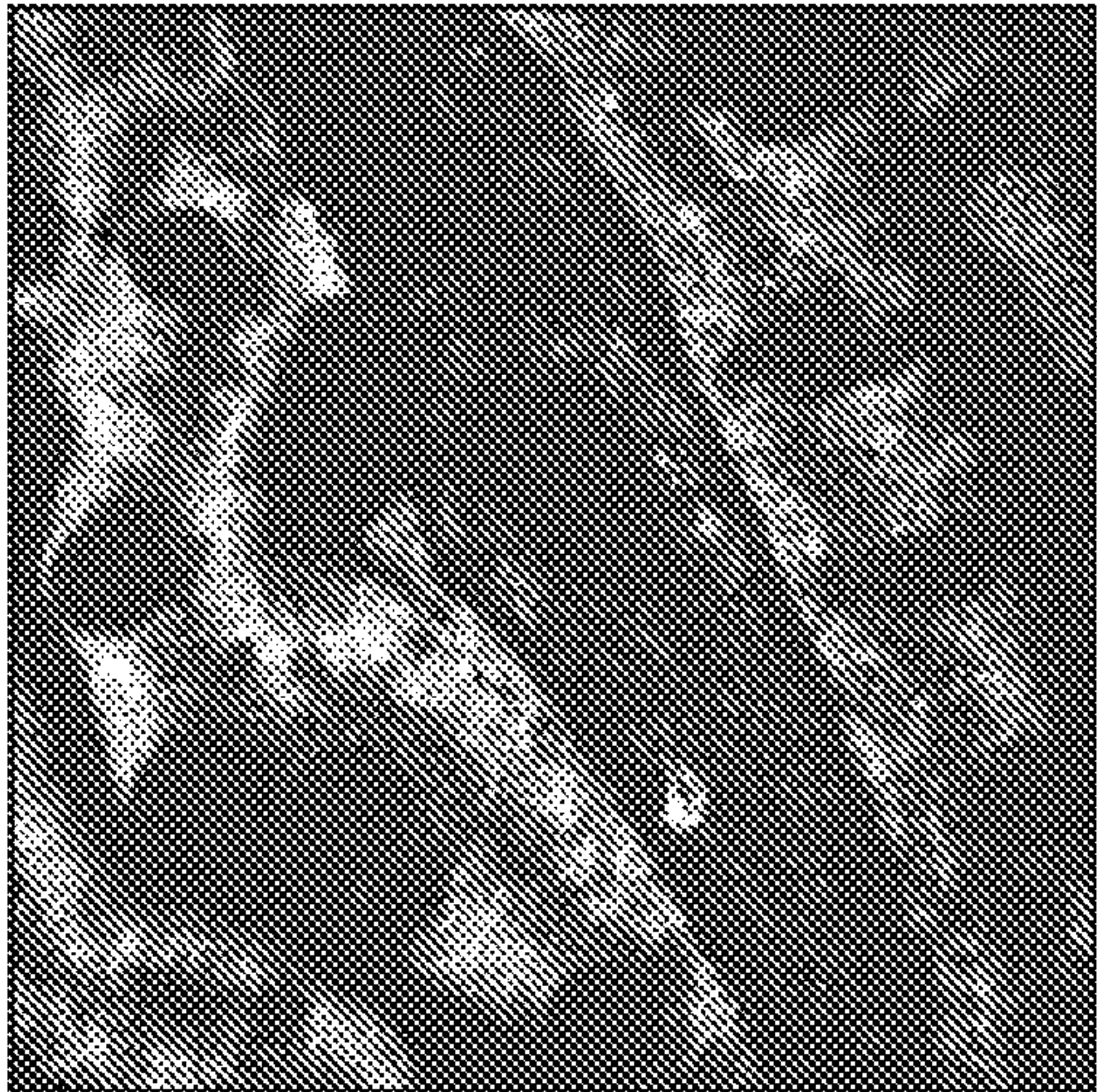


FIG. 7A

FIG. 7

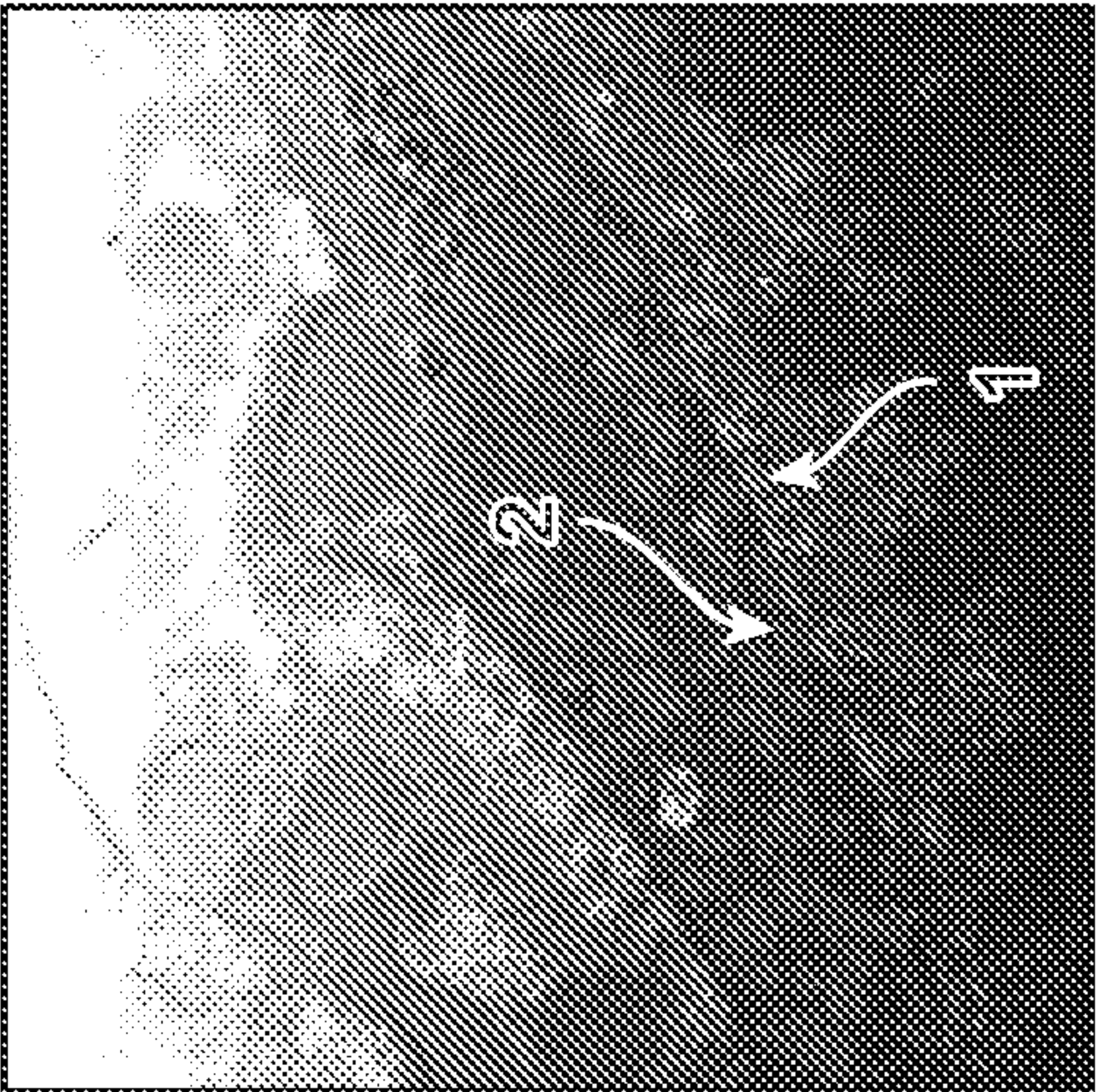


FIG. 7E

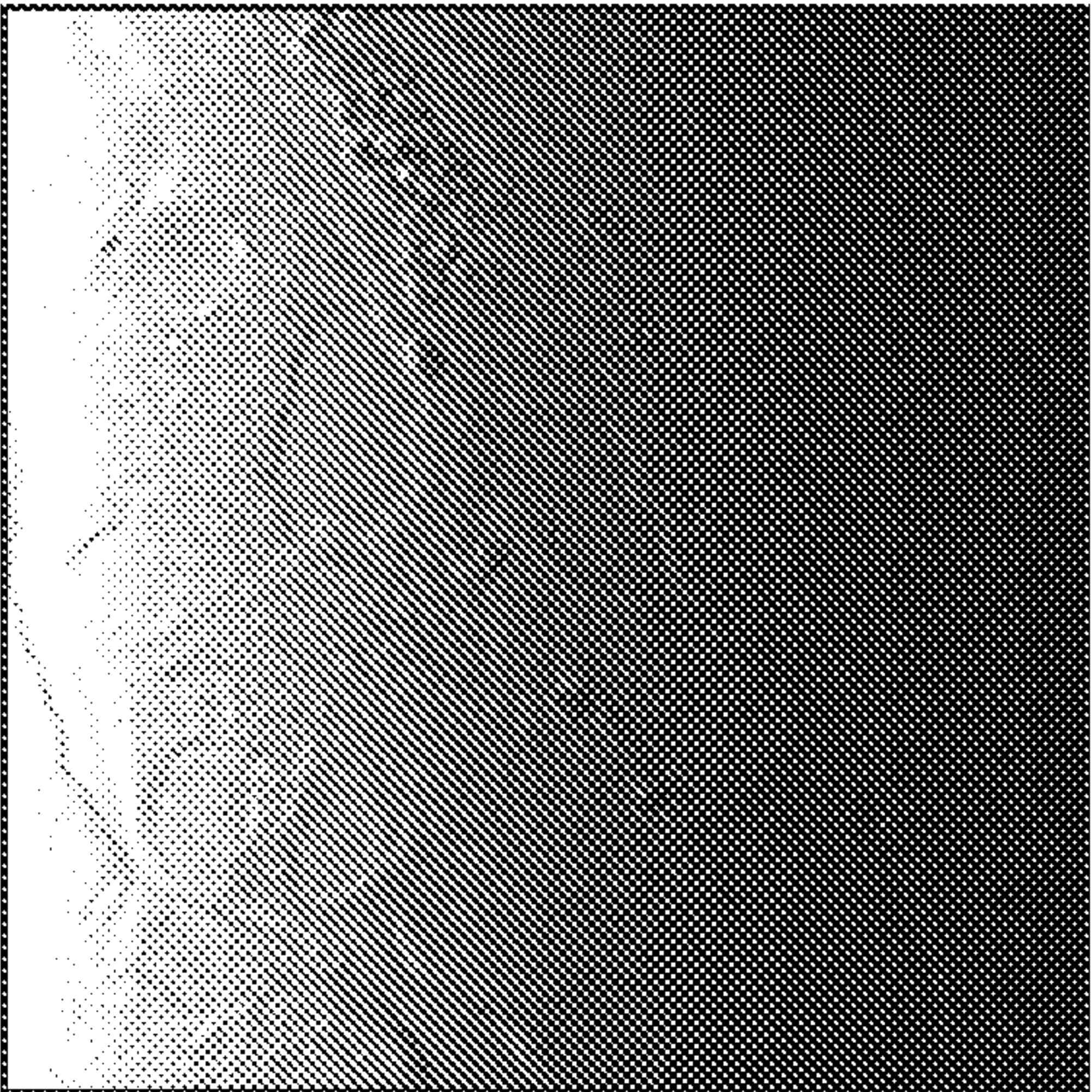


FIG. 7D

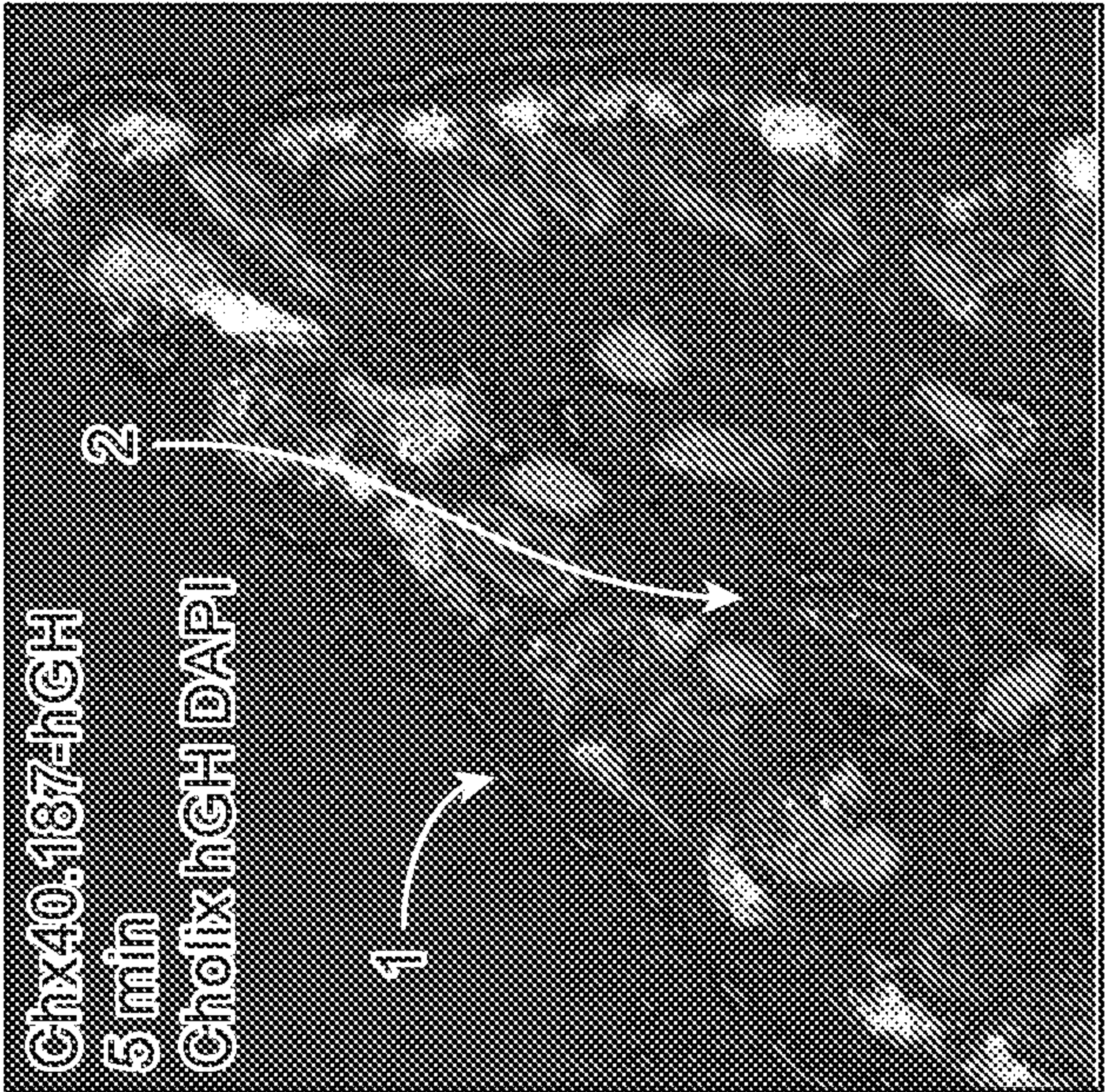


FIG. 8A

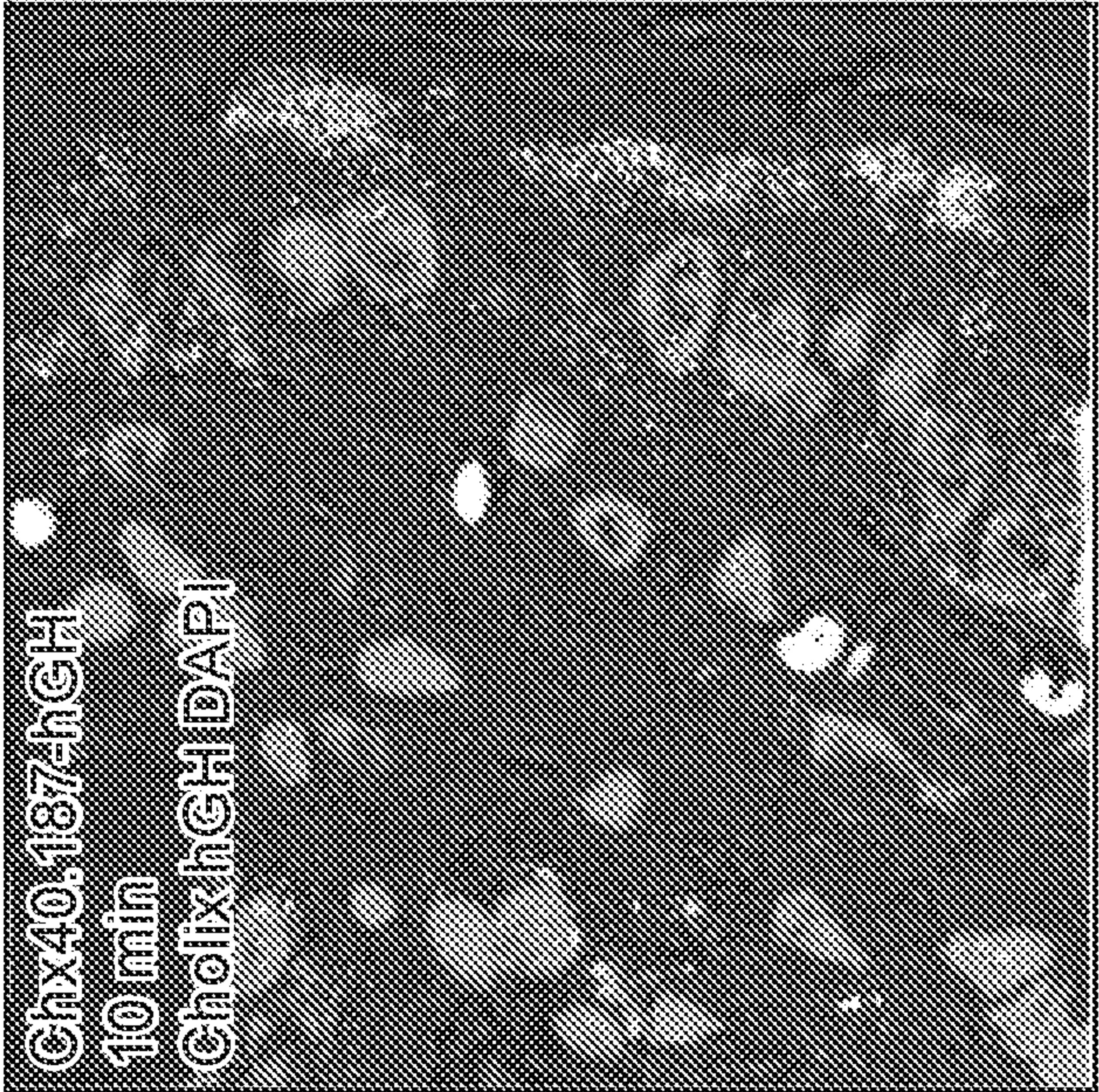


FIG. 8B

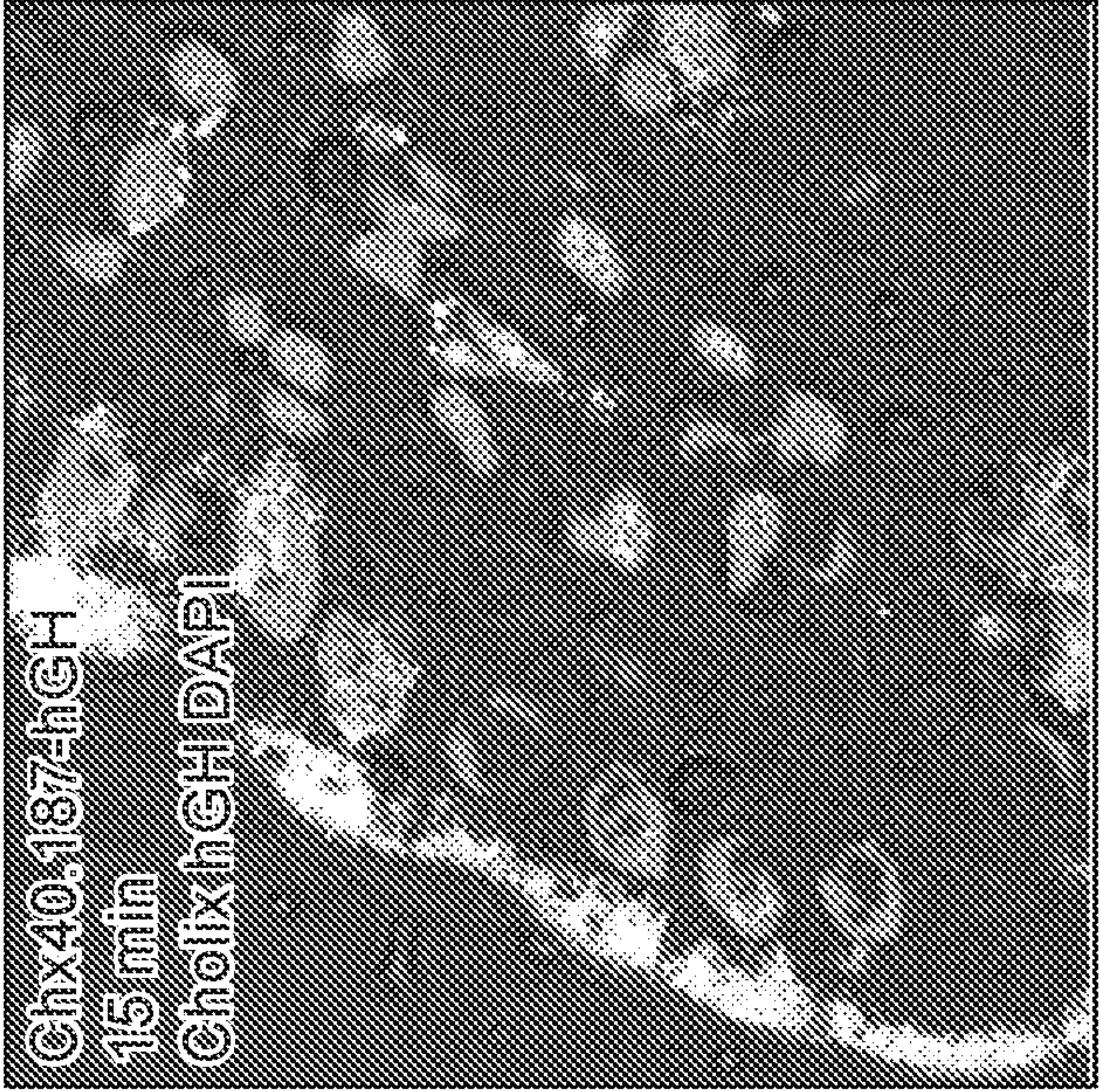


FIG. 8C

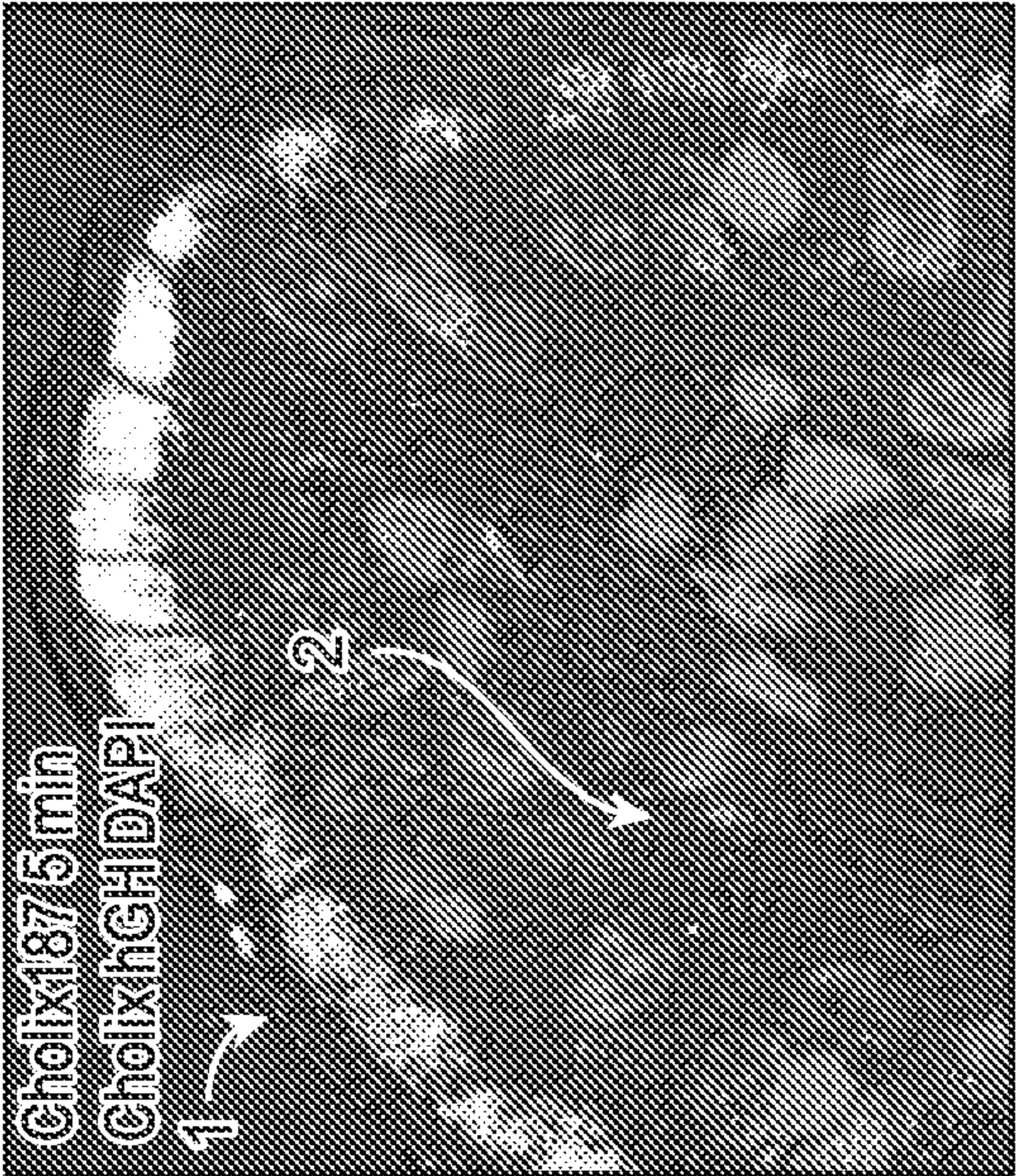


FIG. 9A

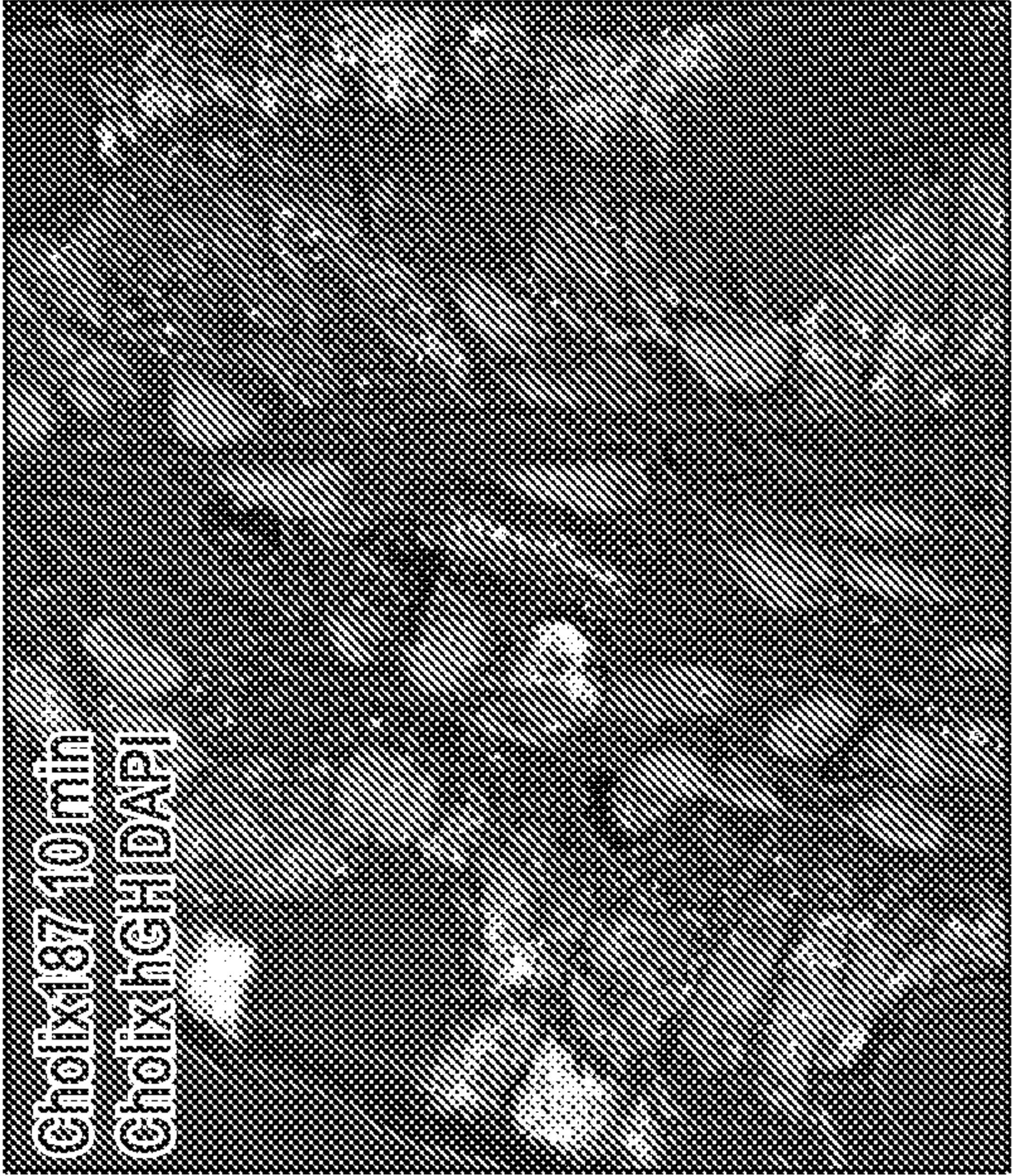


FIG. 9B

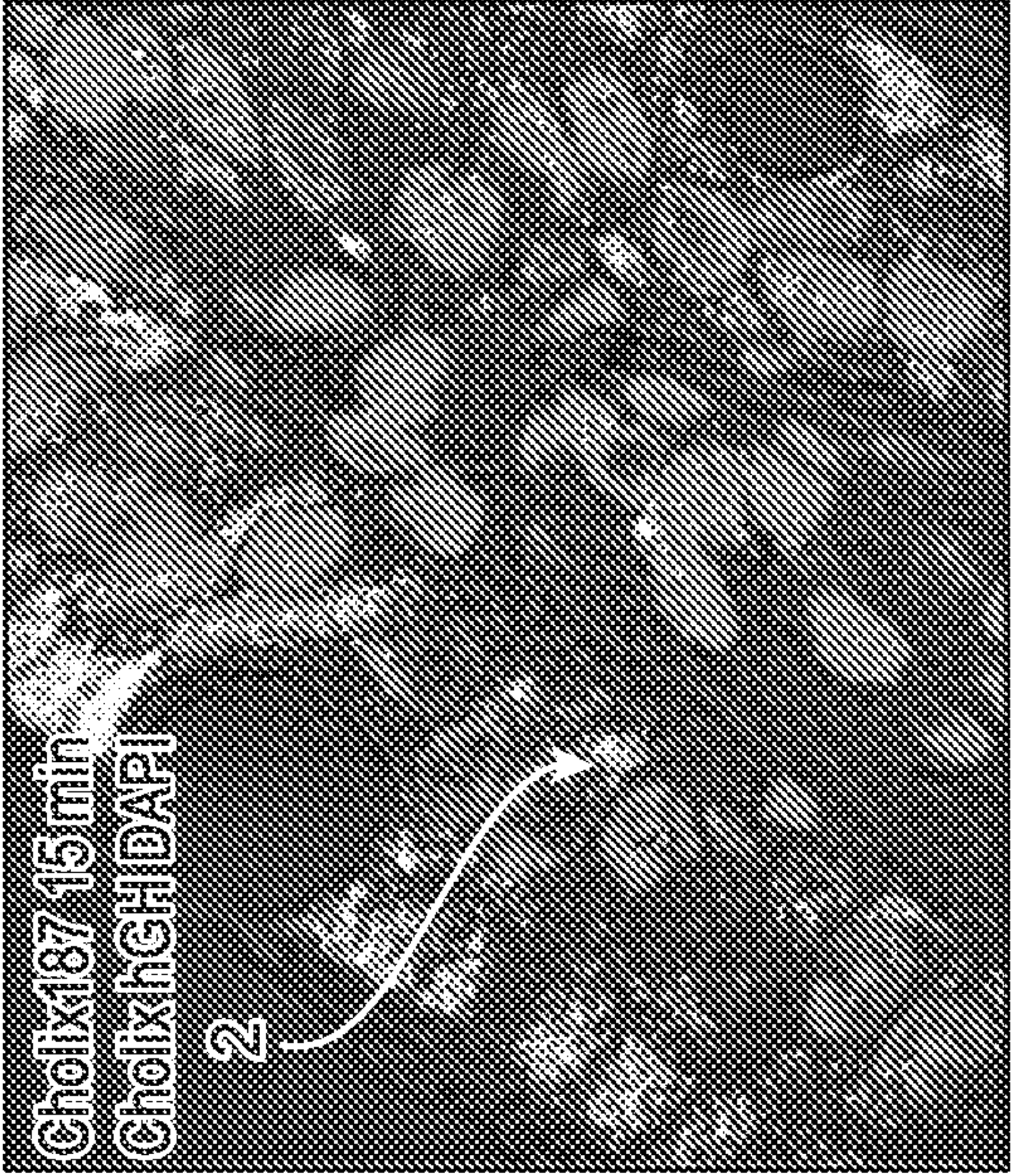


FIG. 9C

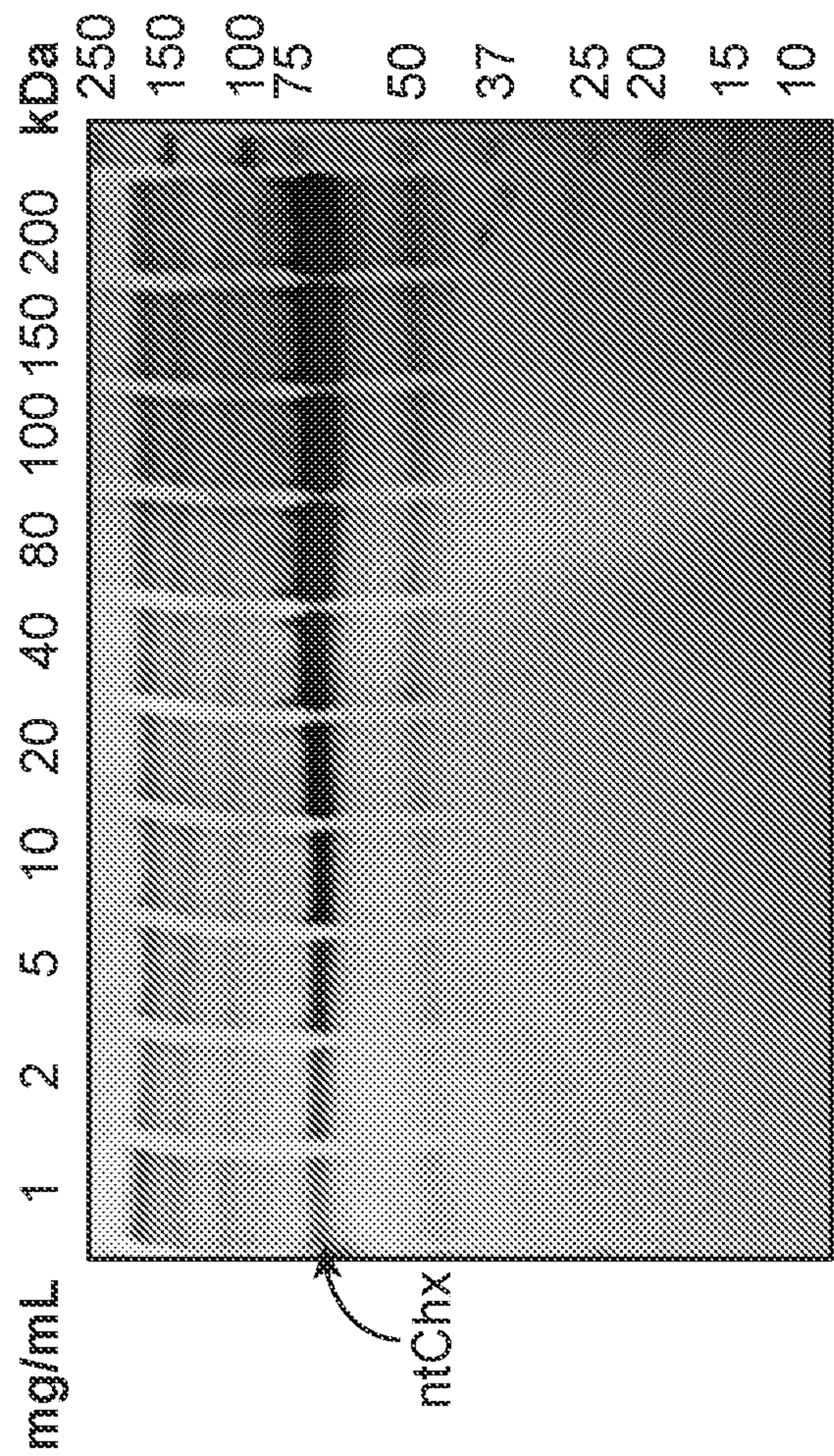


FIG. 10B

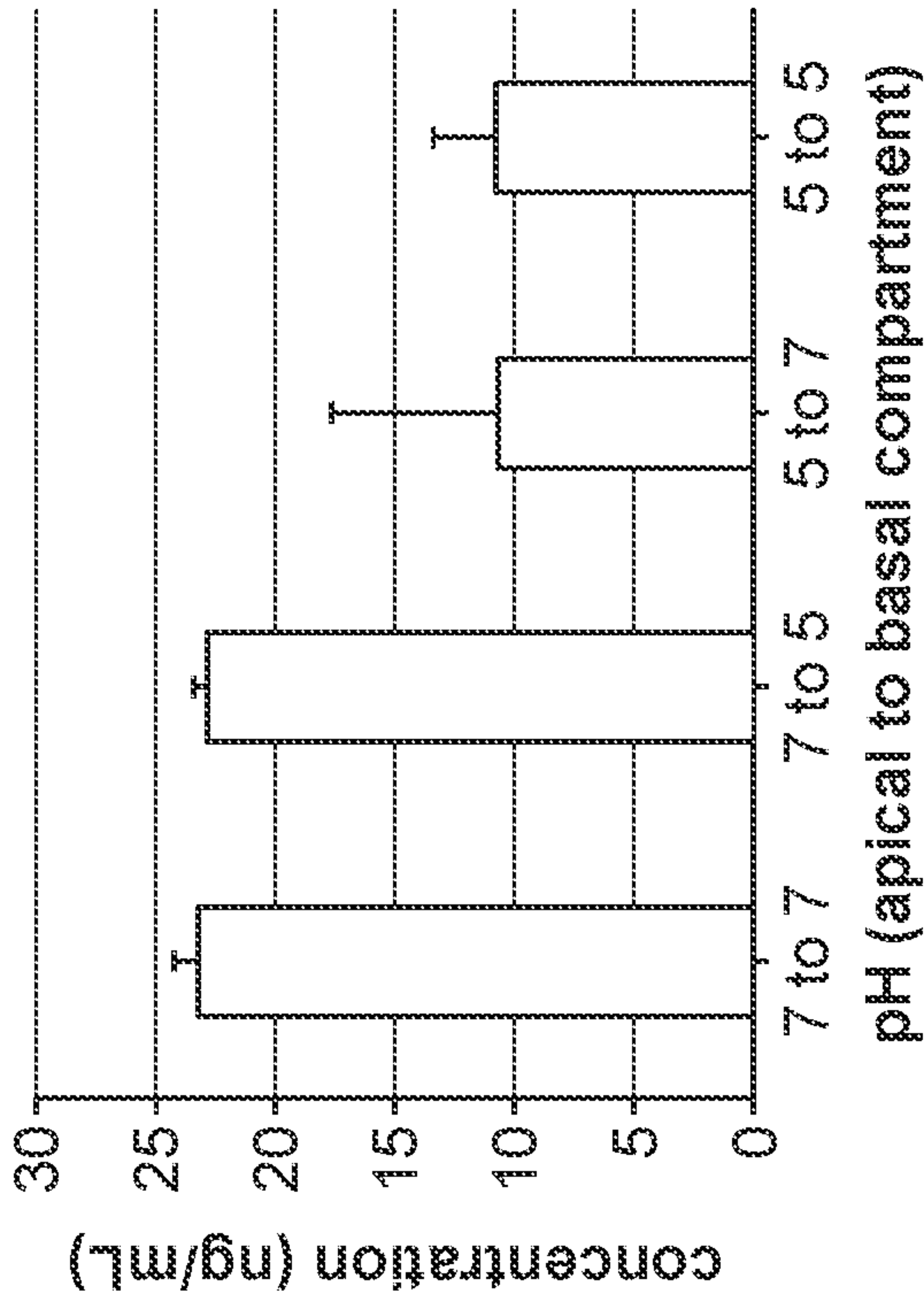


FIG. 11

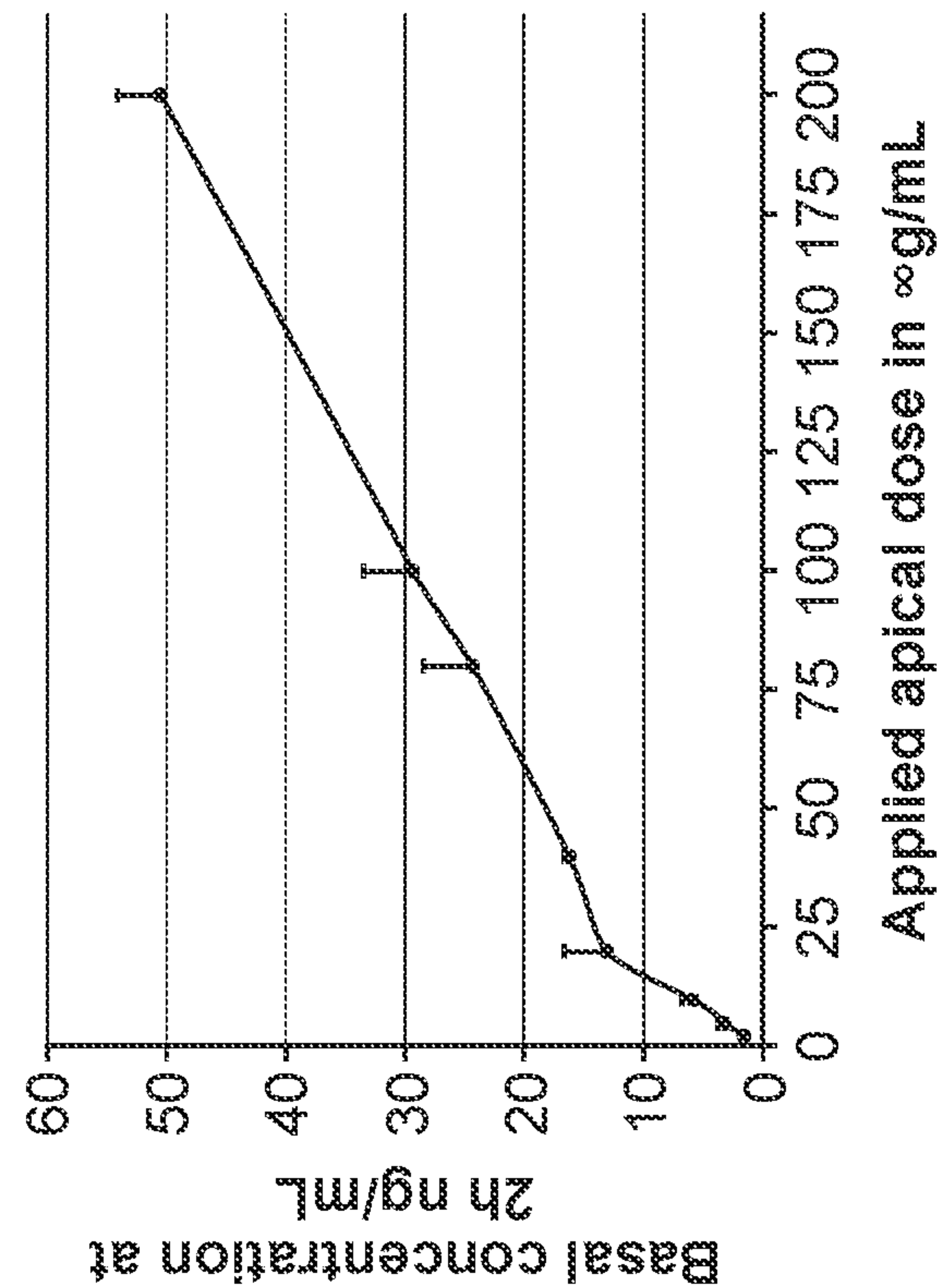


FIG. 10A

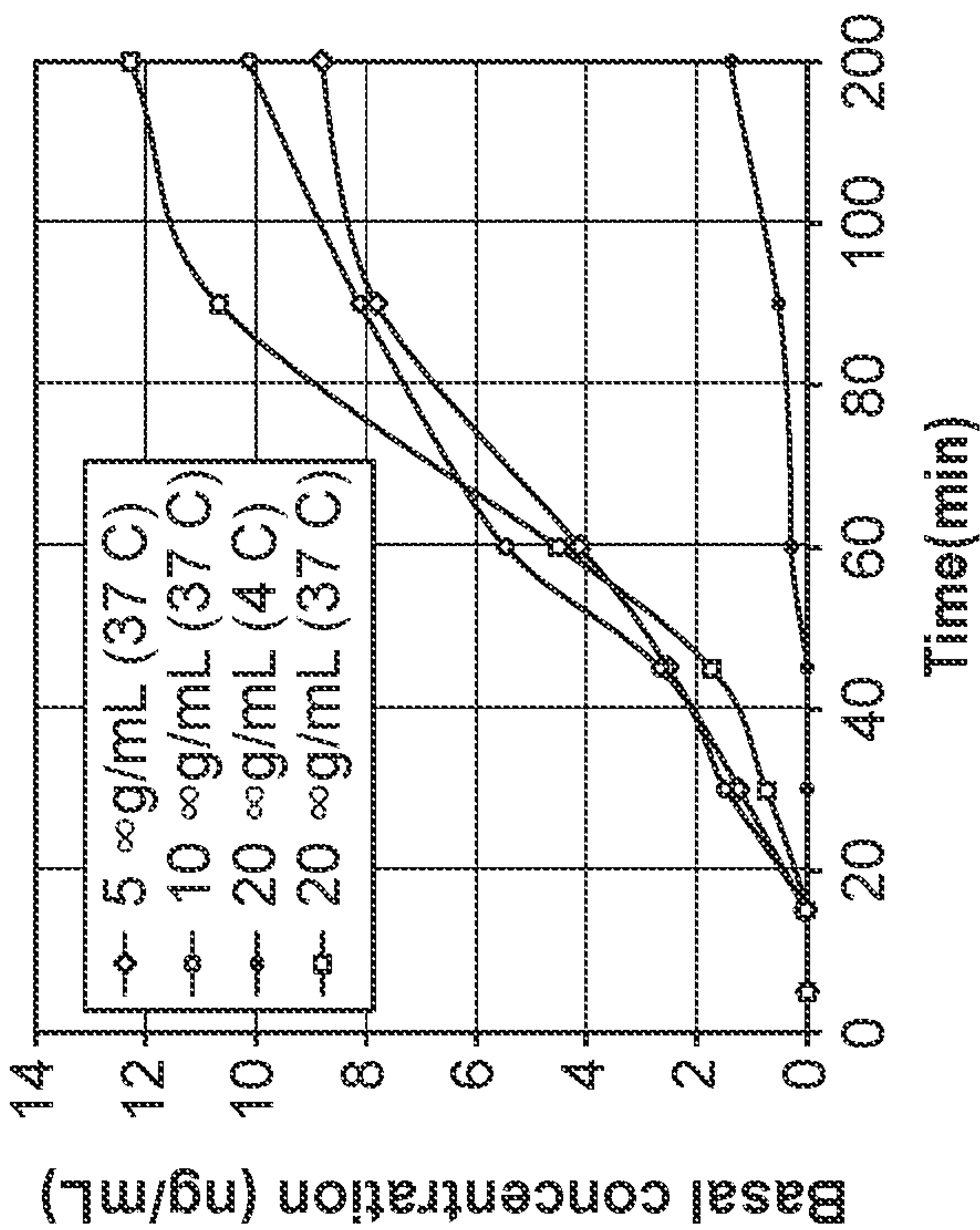


FIG. 10C

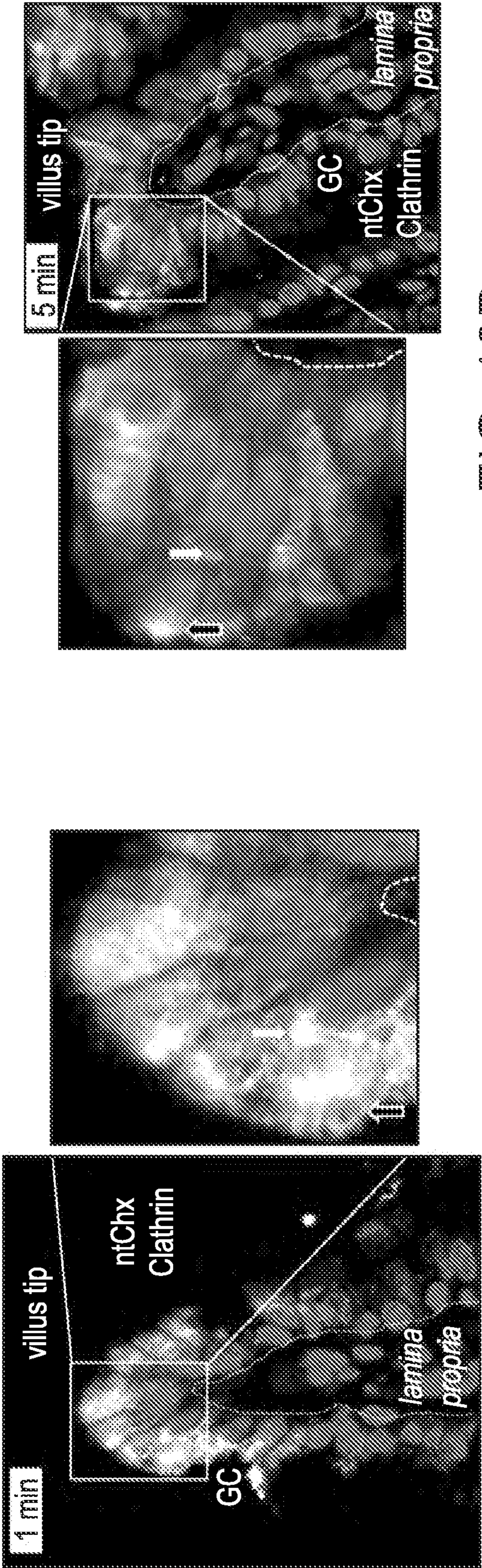


FIG. 12B

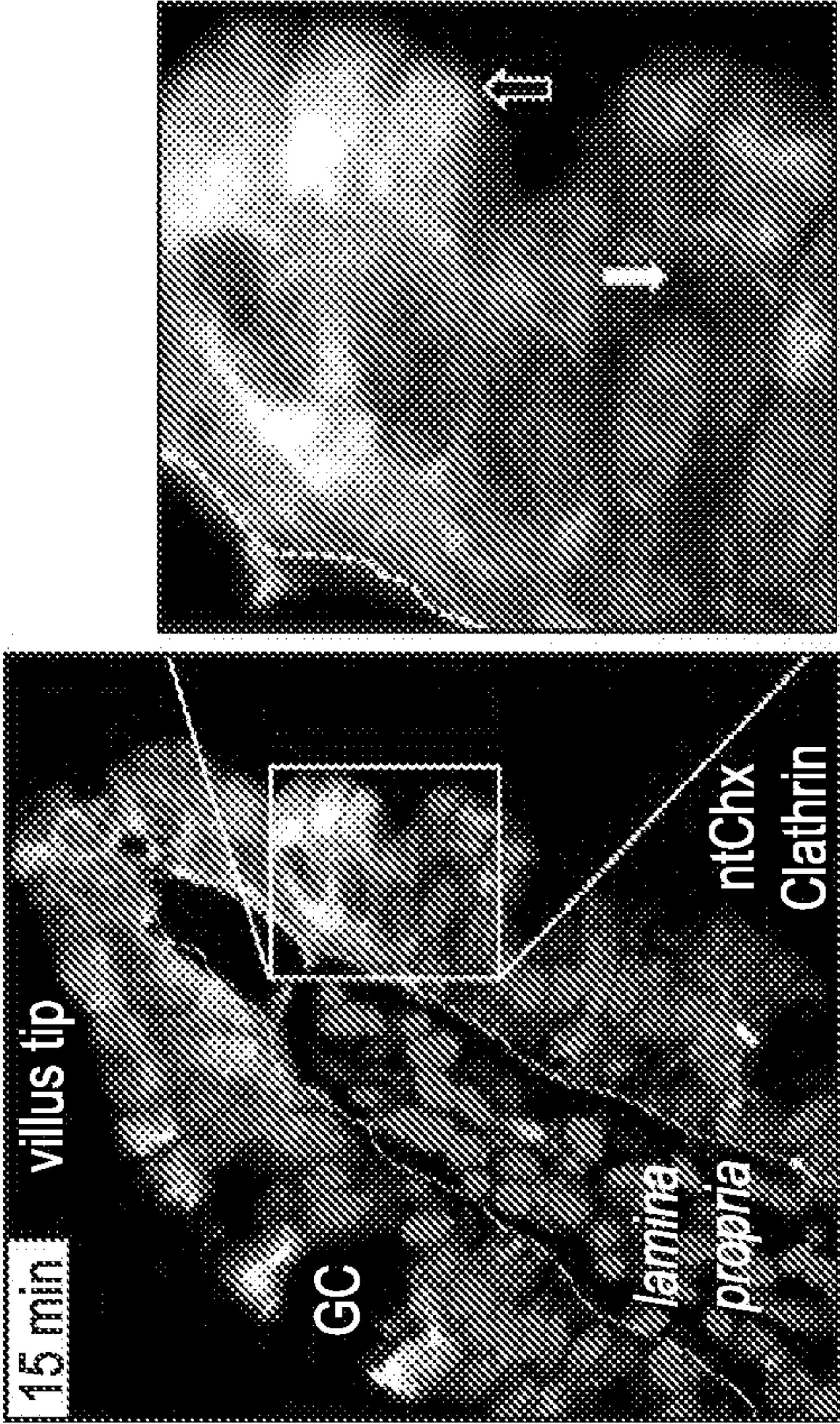


FIG. 12C

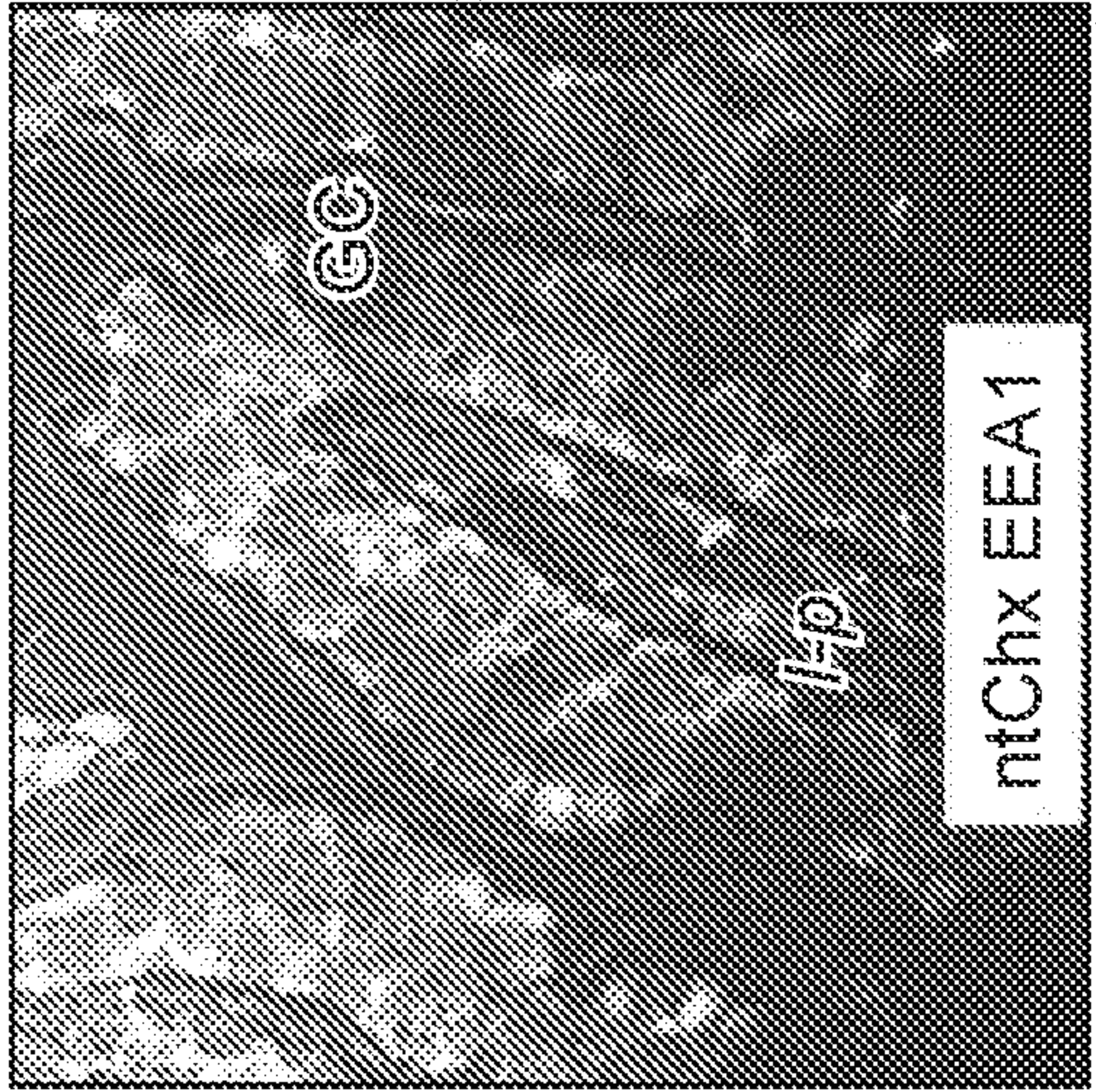


FIG. 13A

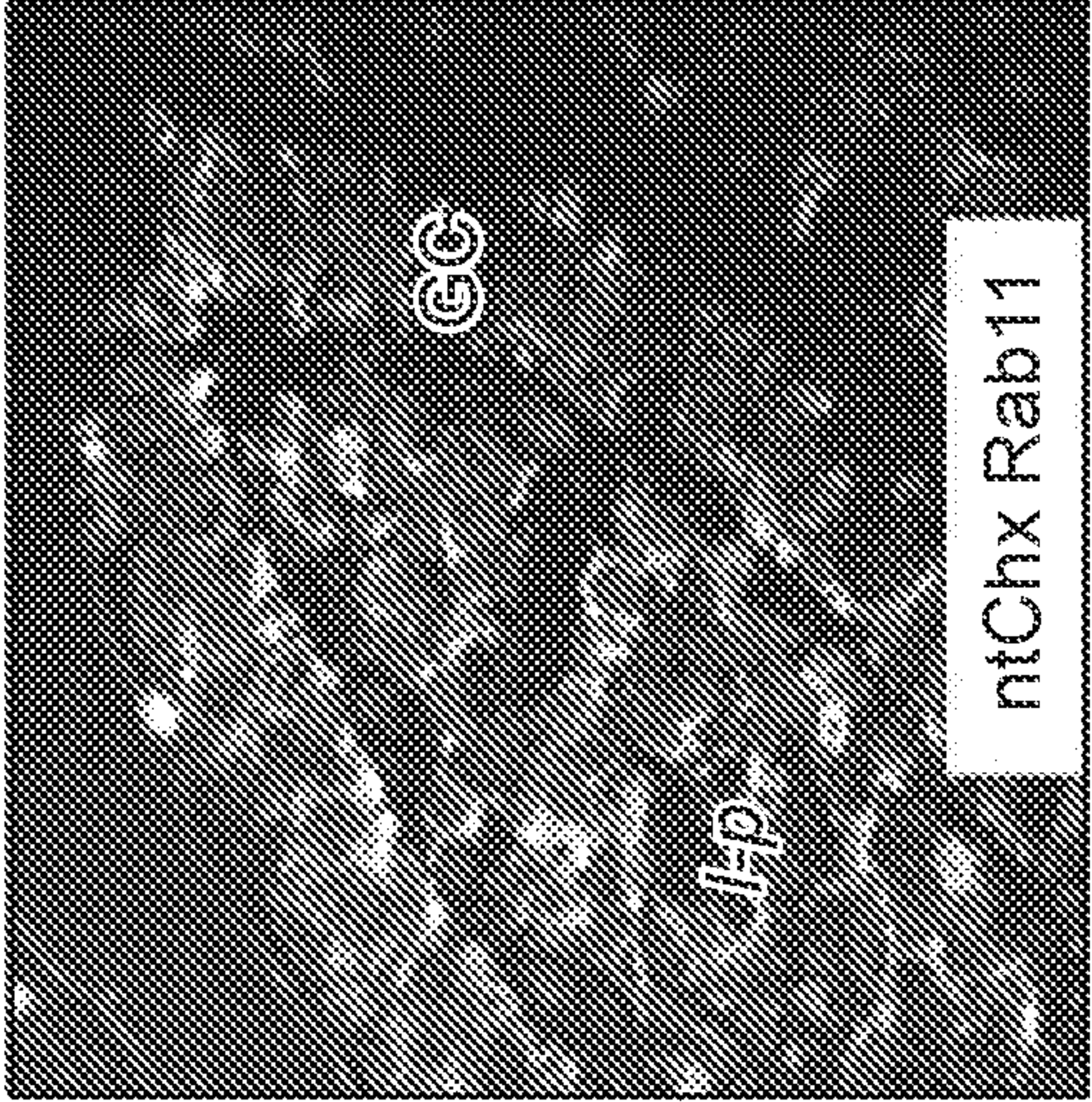


FIG. 13B

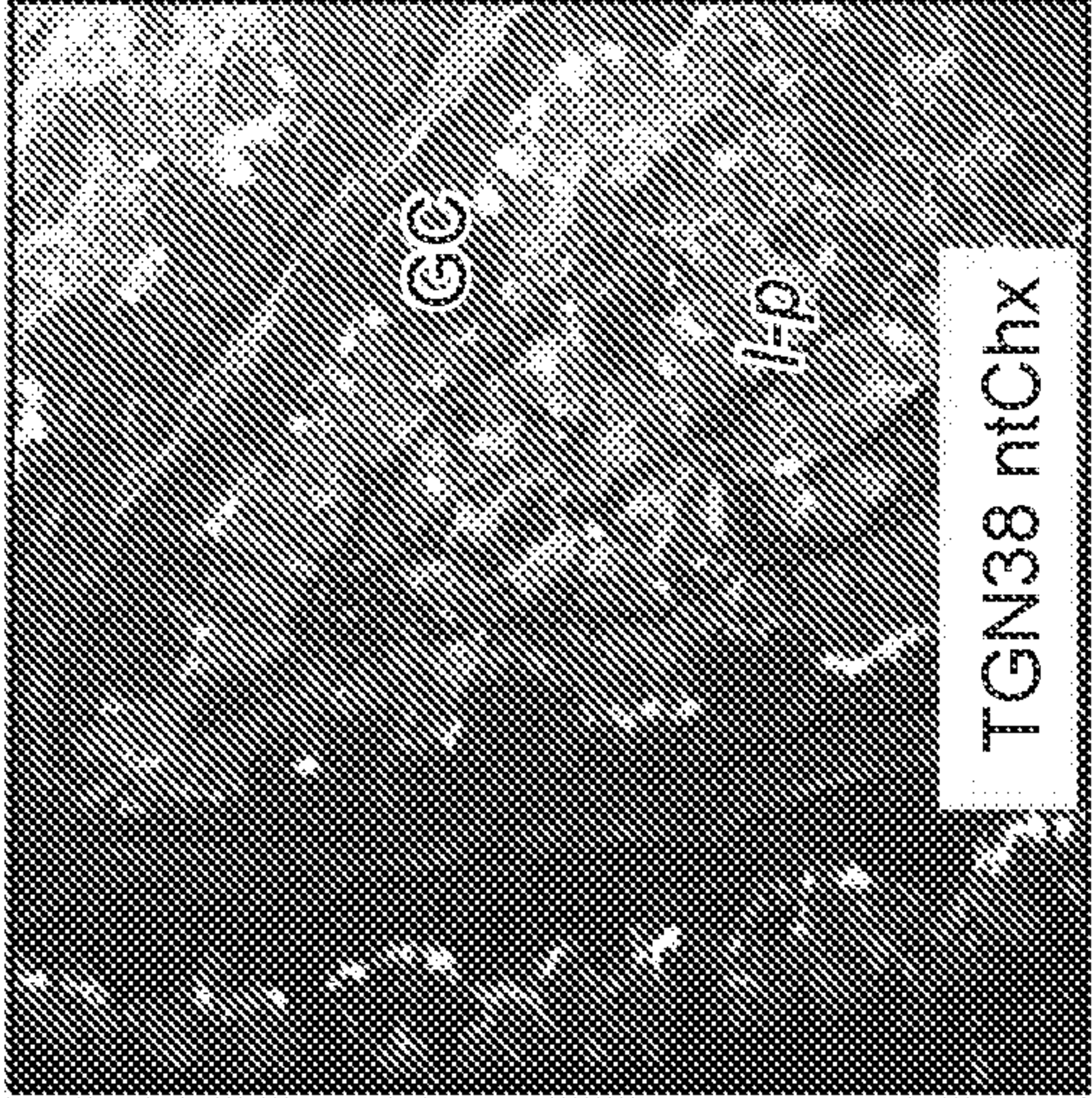


FIG. 13C

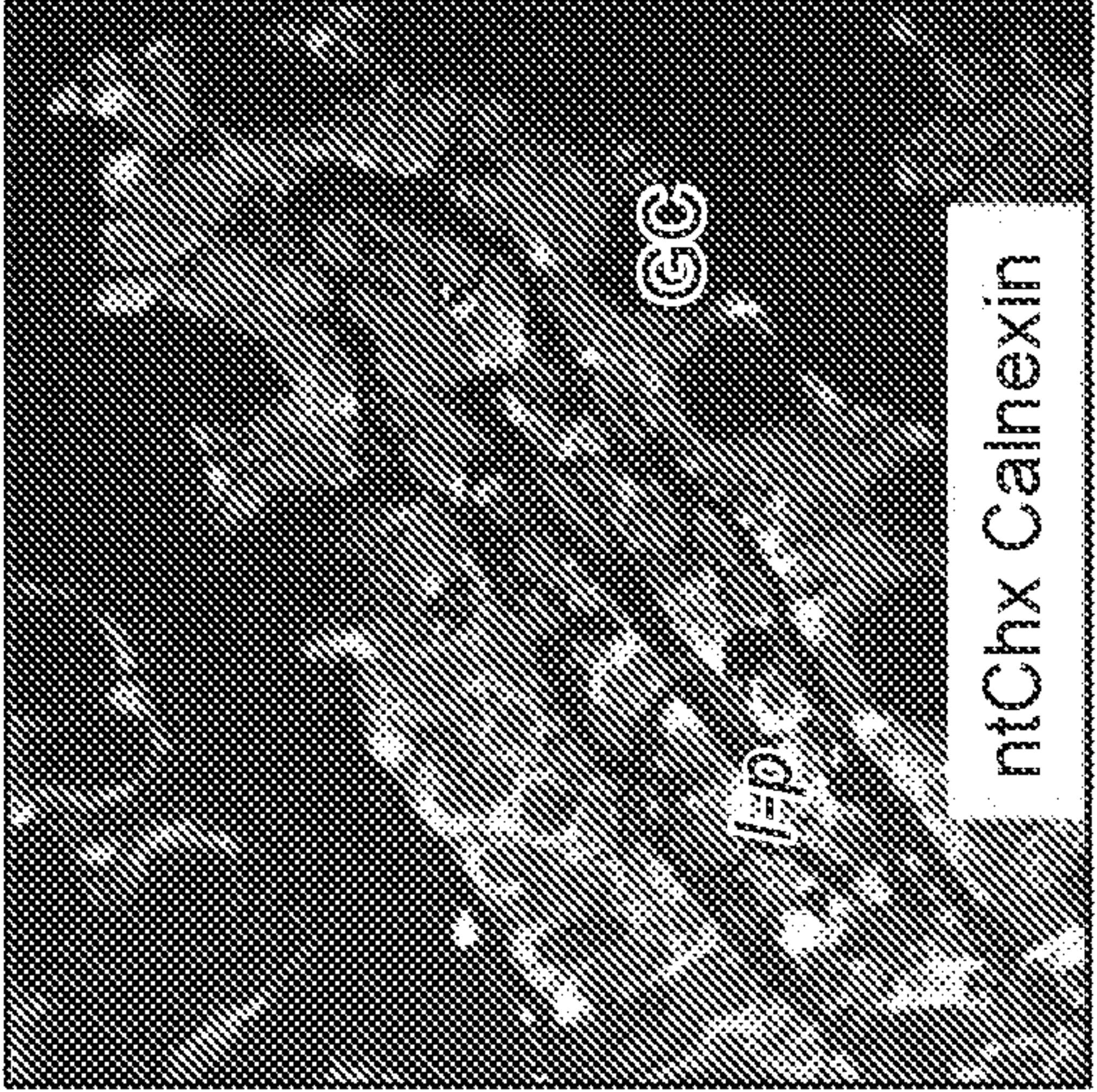


FIG. 13D

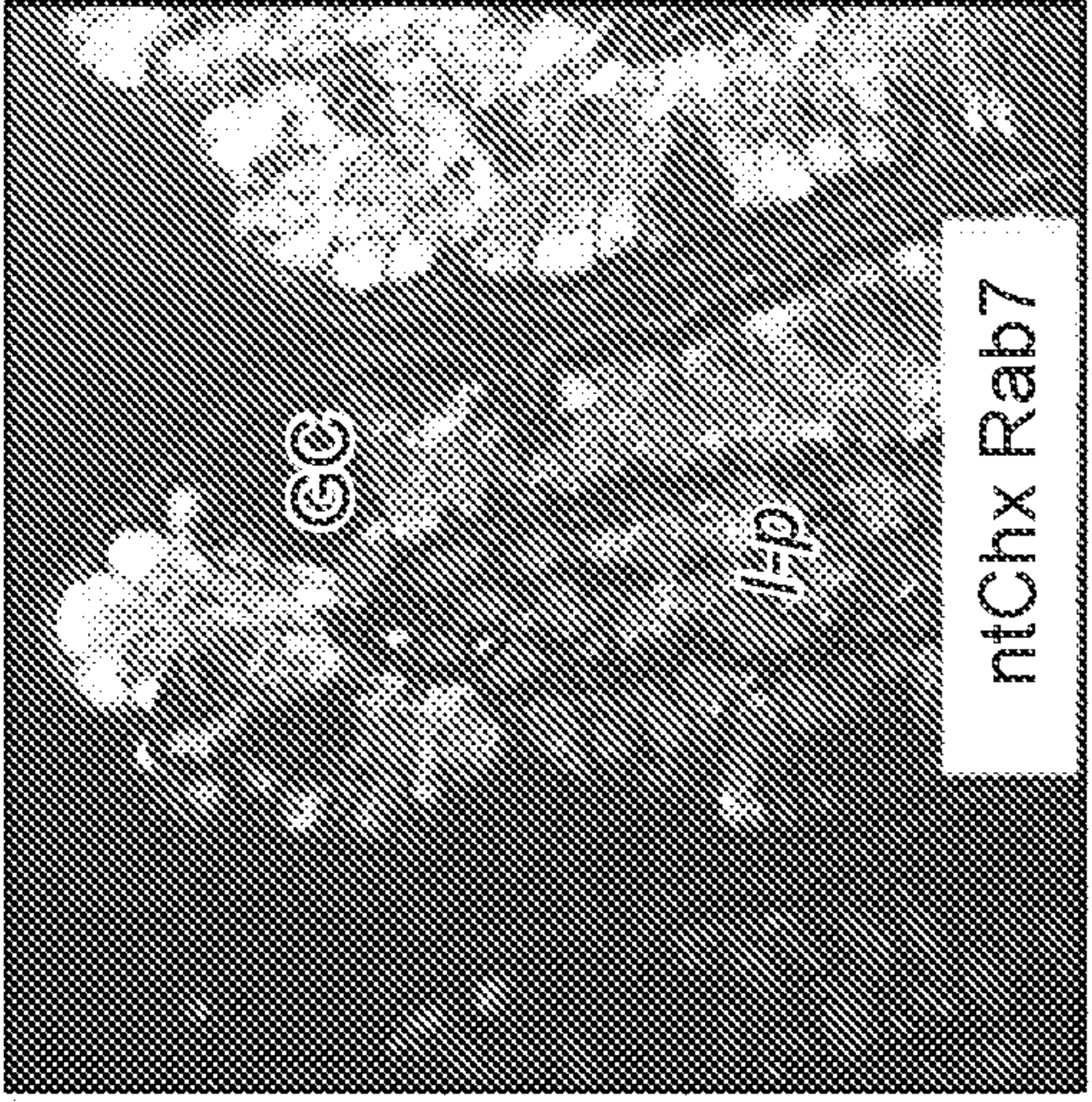


FIG. 13E

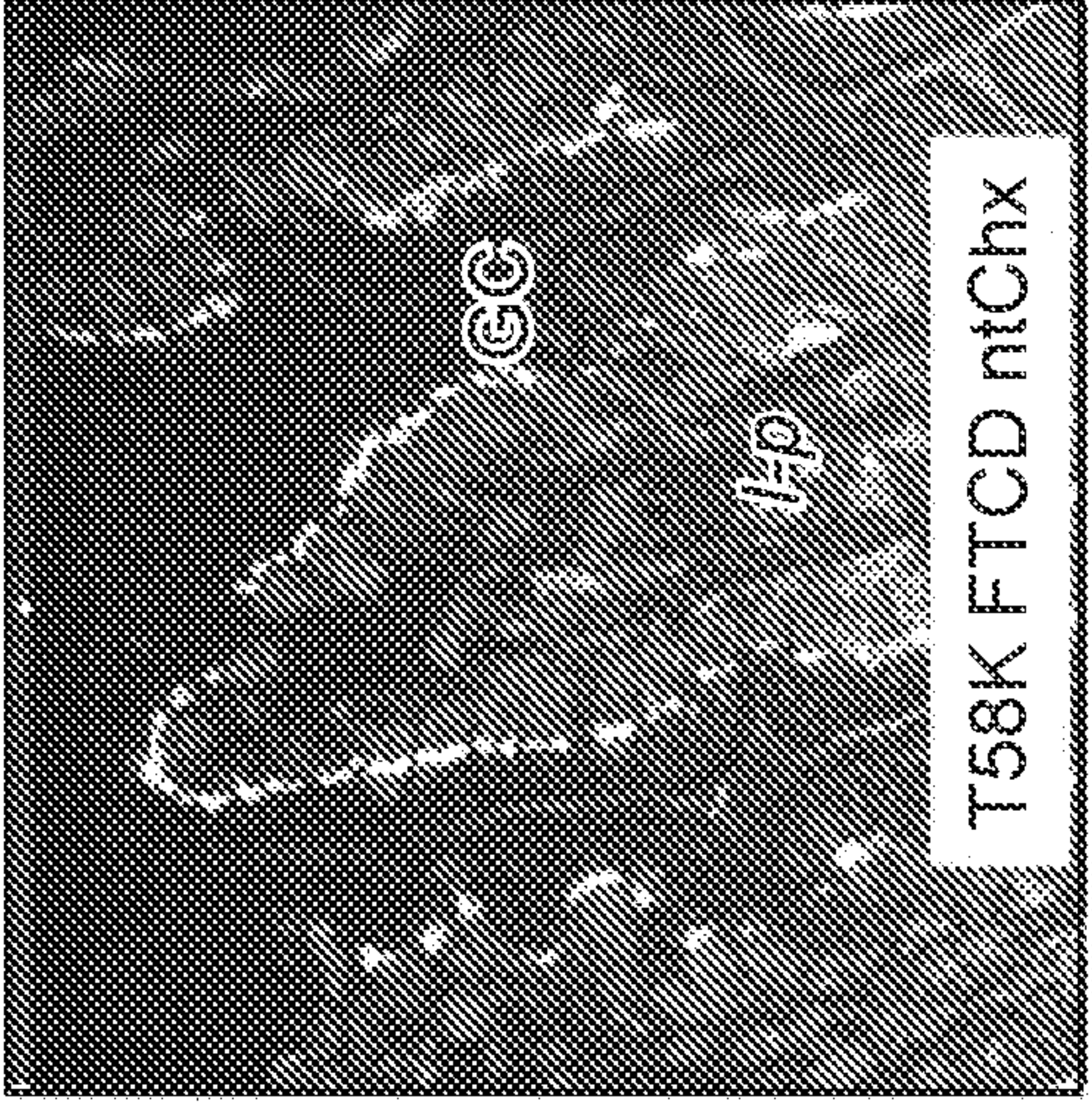


FIG. 13F

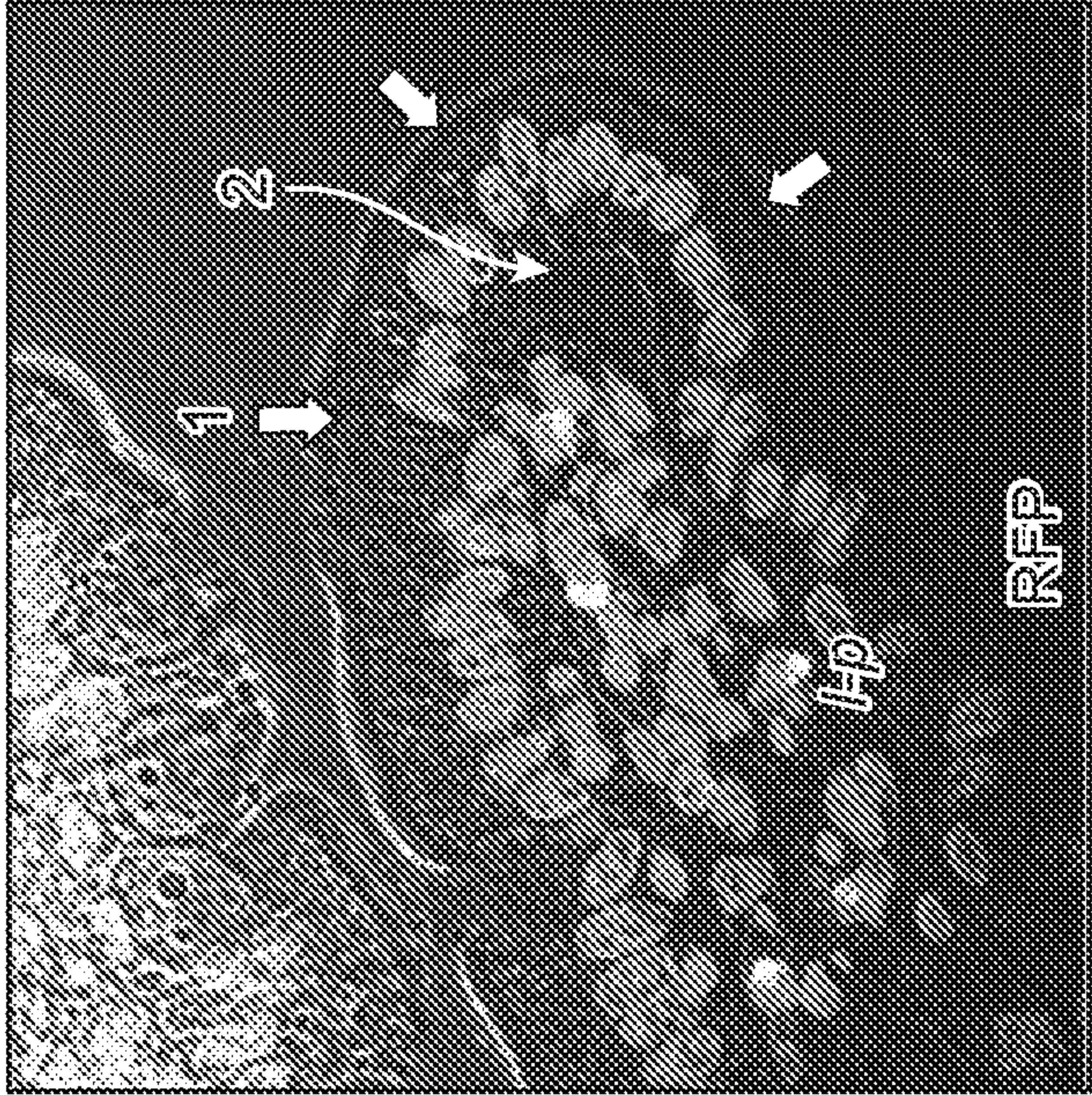


FIG. 14A

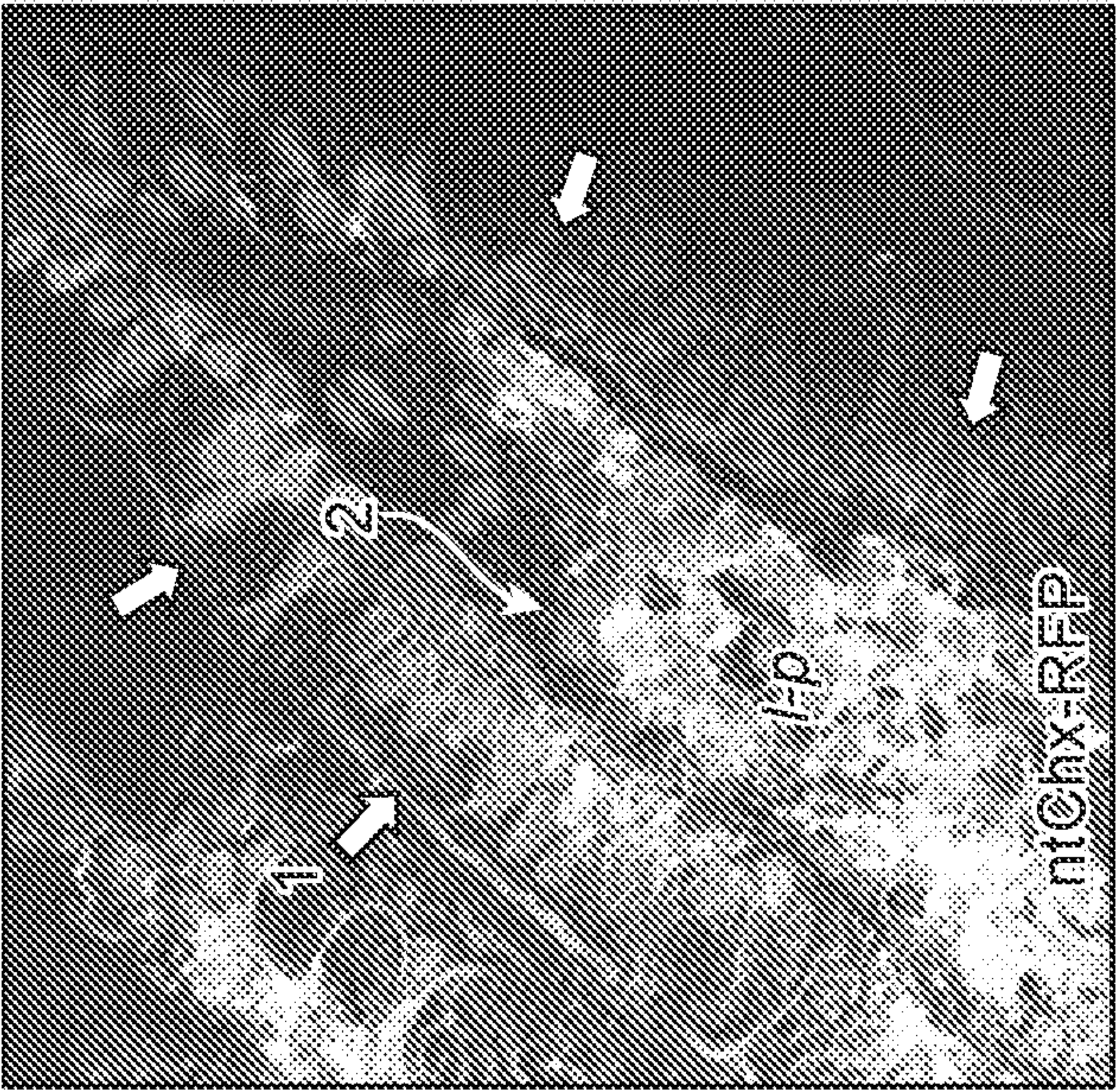


FIG. 14B

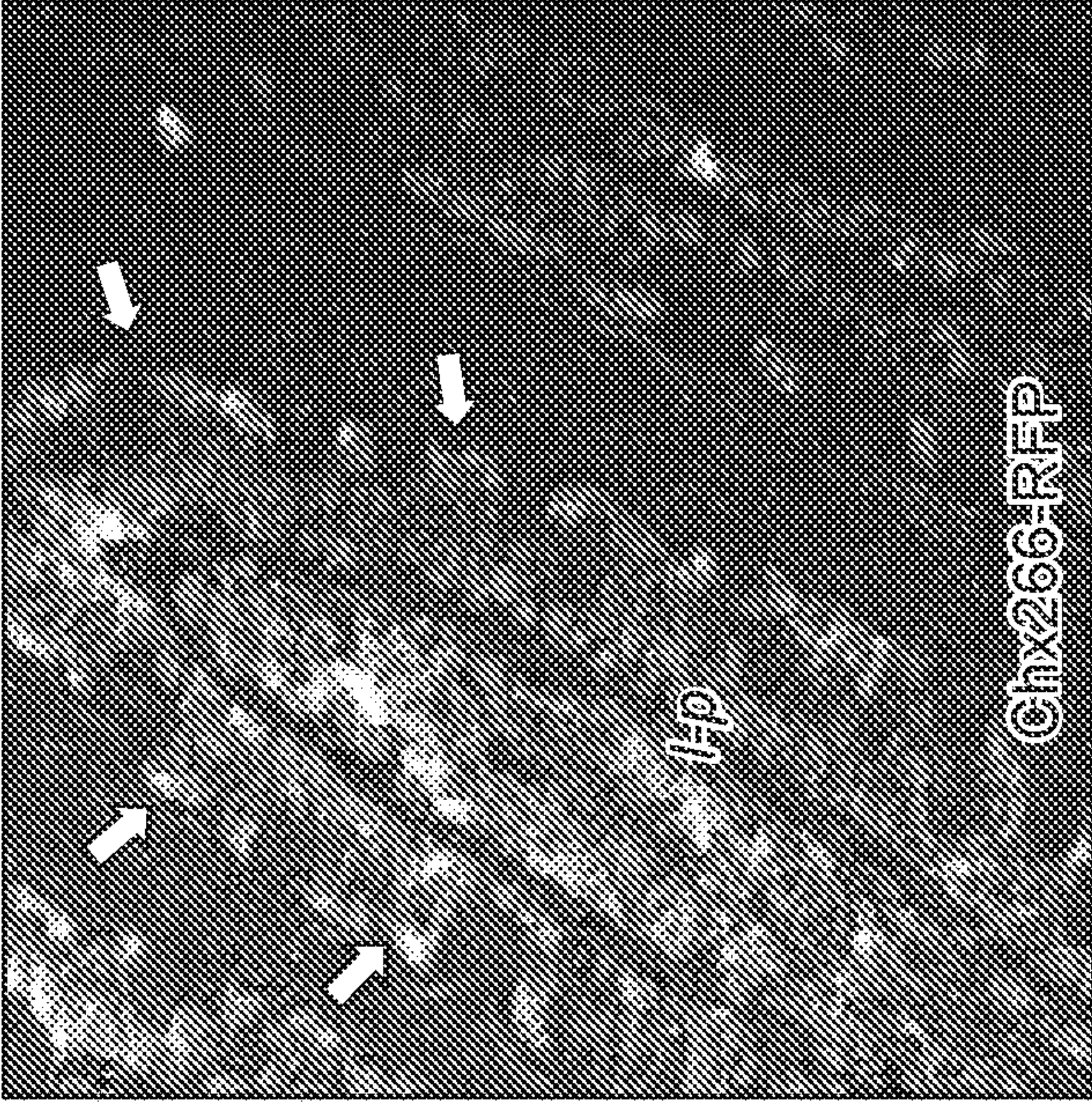


FIG. 14C

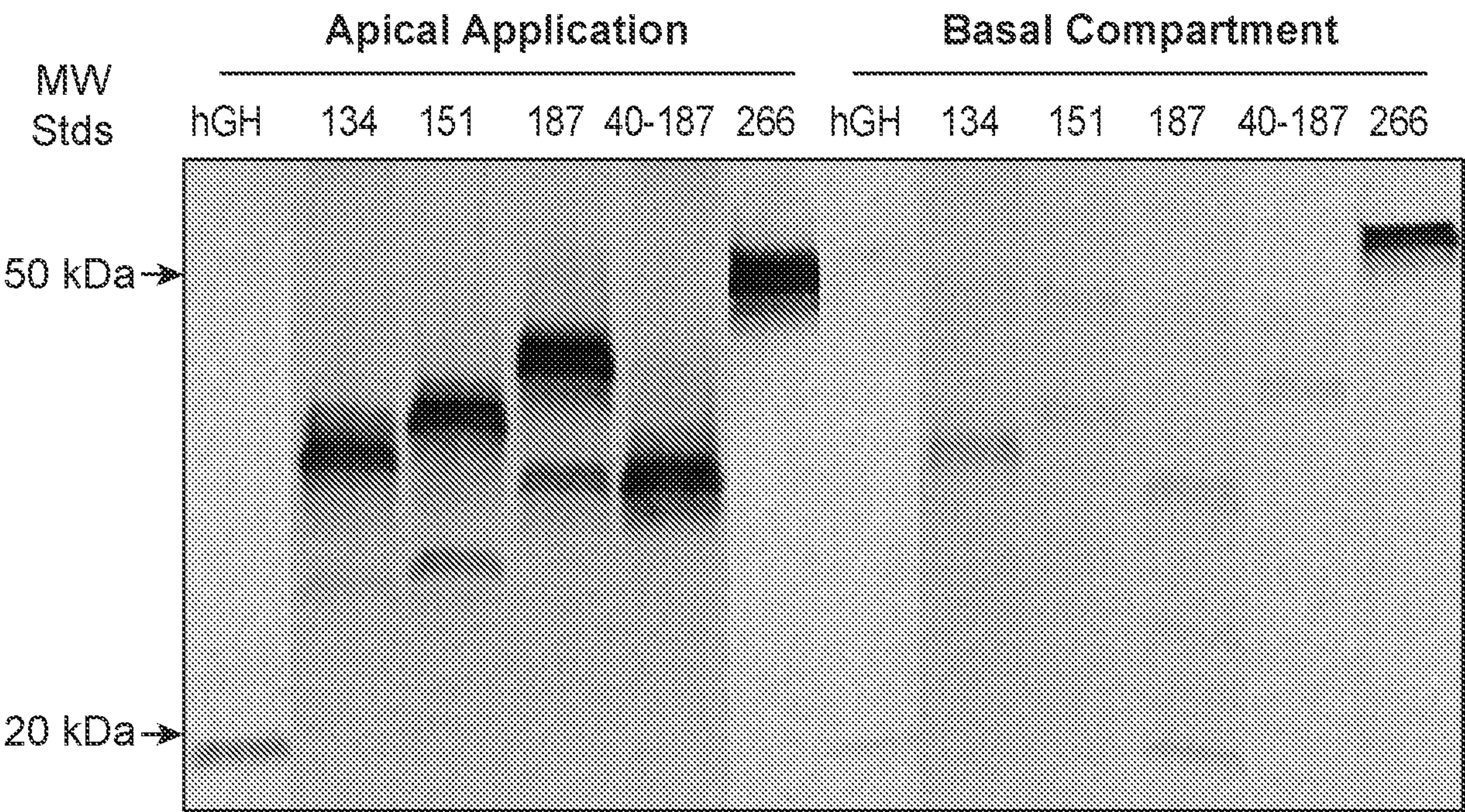


FIG. 15A

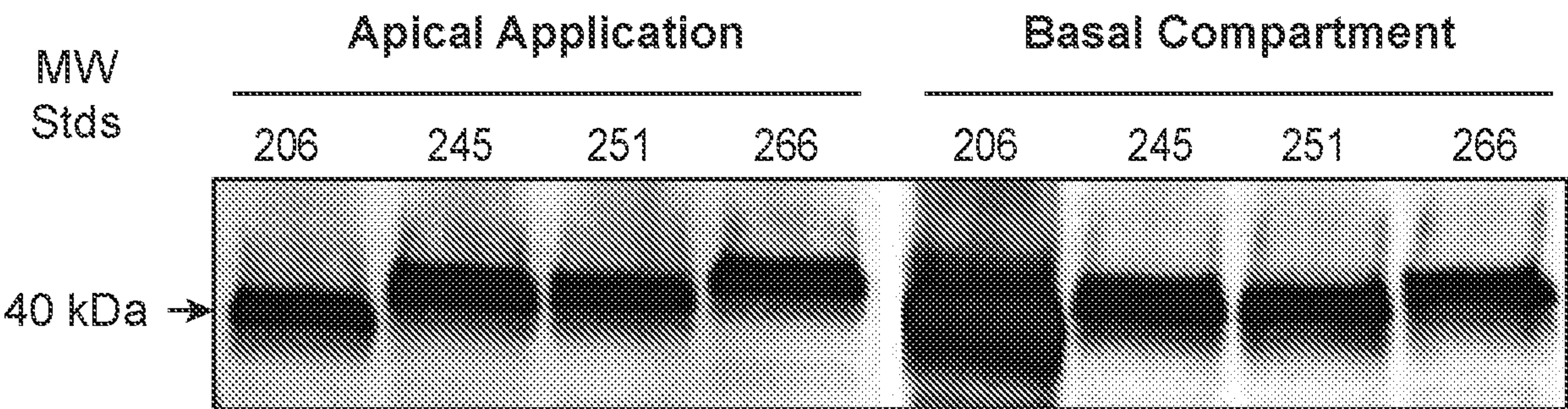


FIG. 15B

FIG. 15

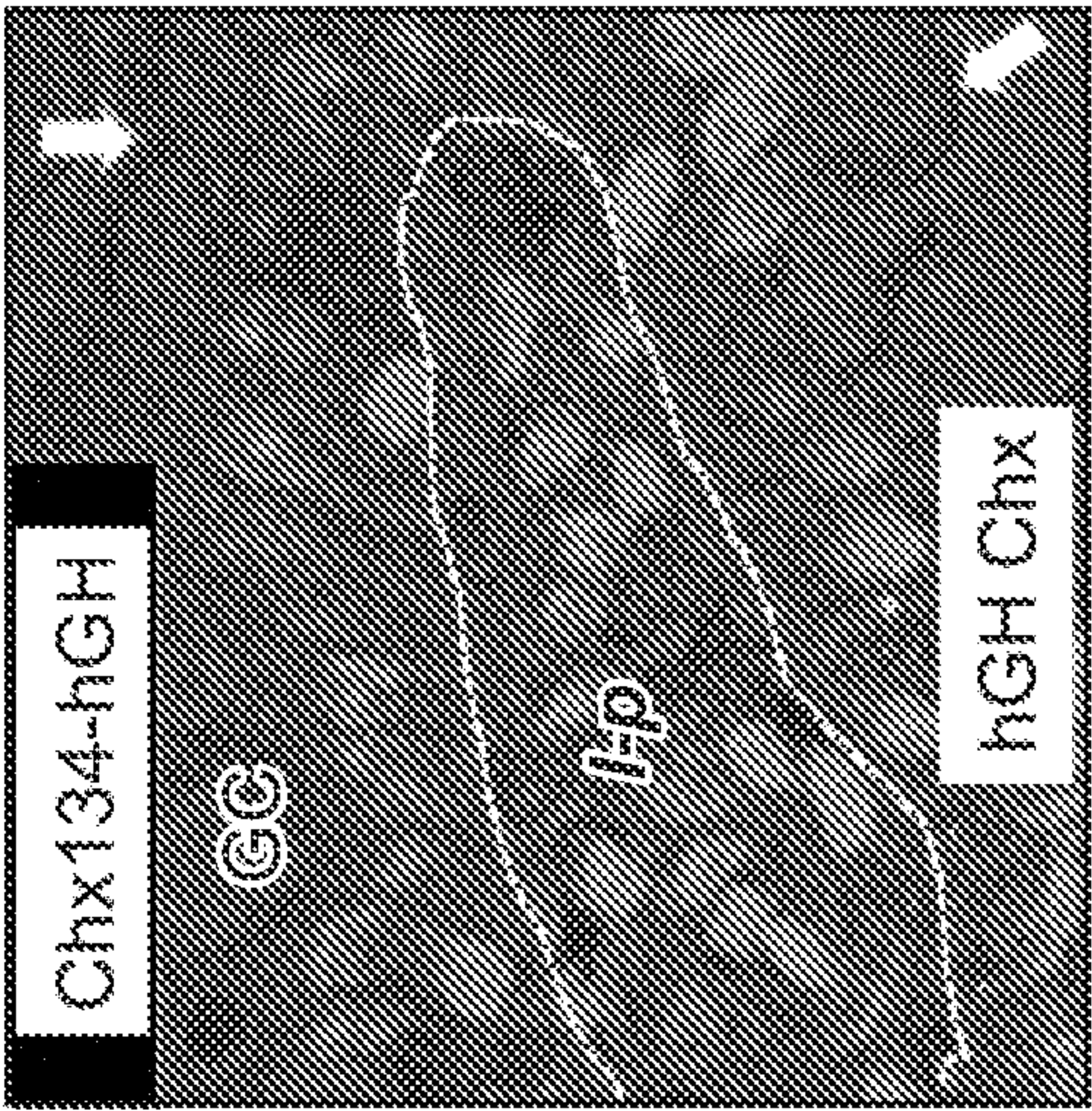


FIG. 16A

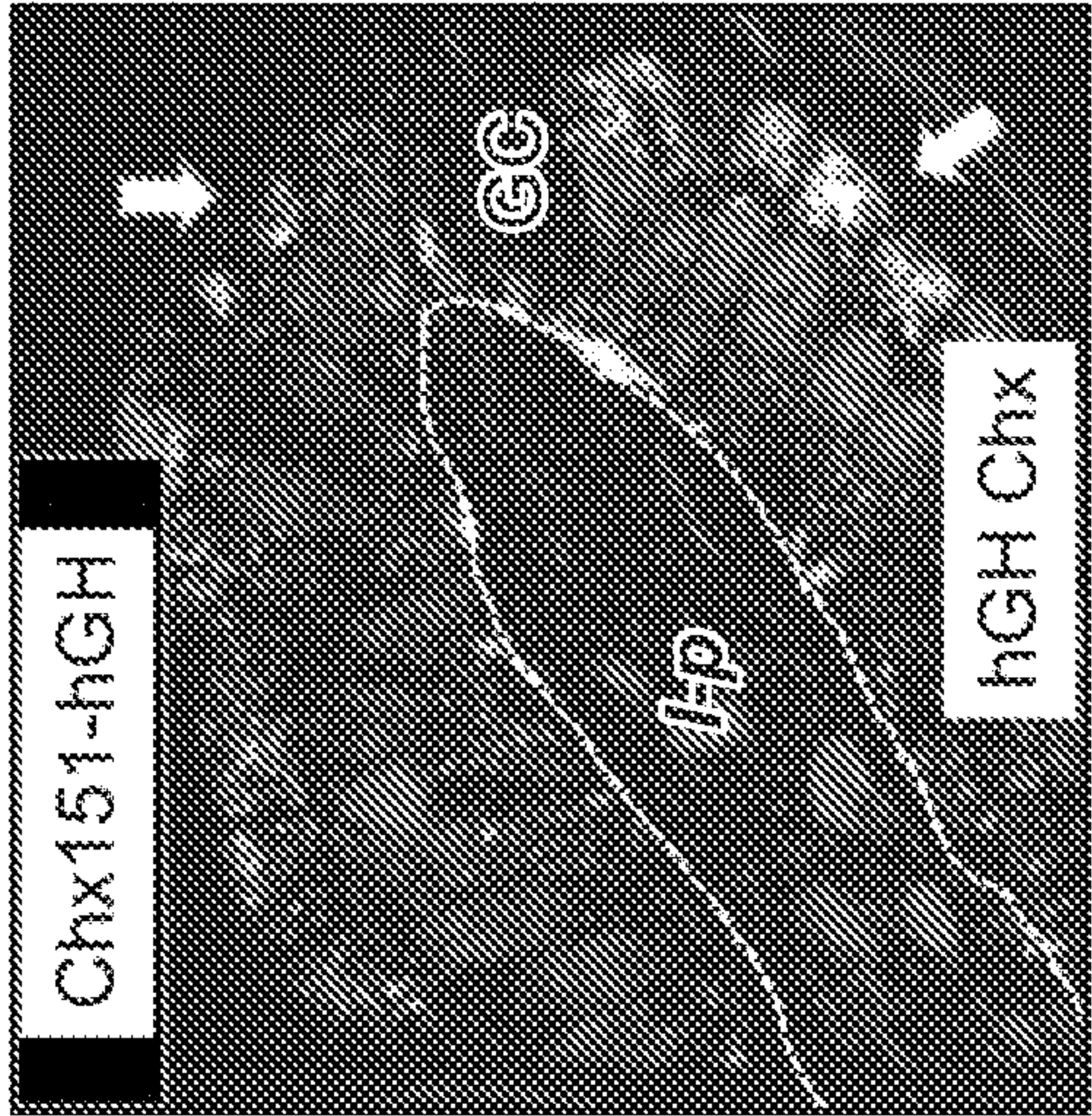


FIG. 16B

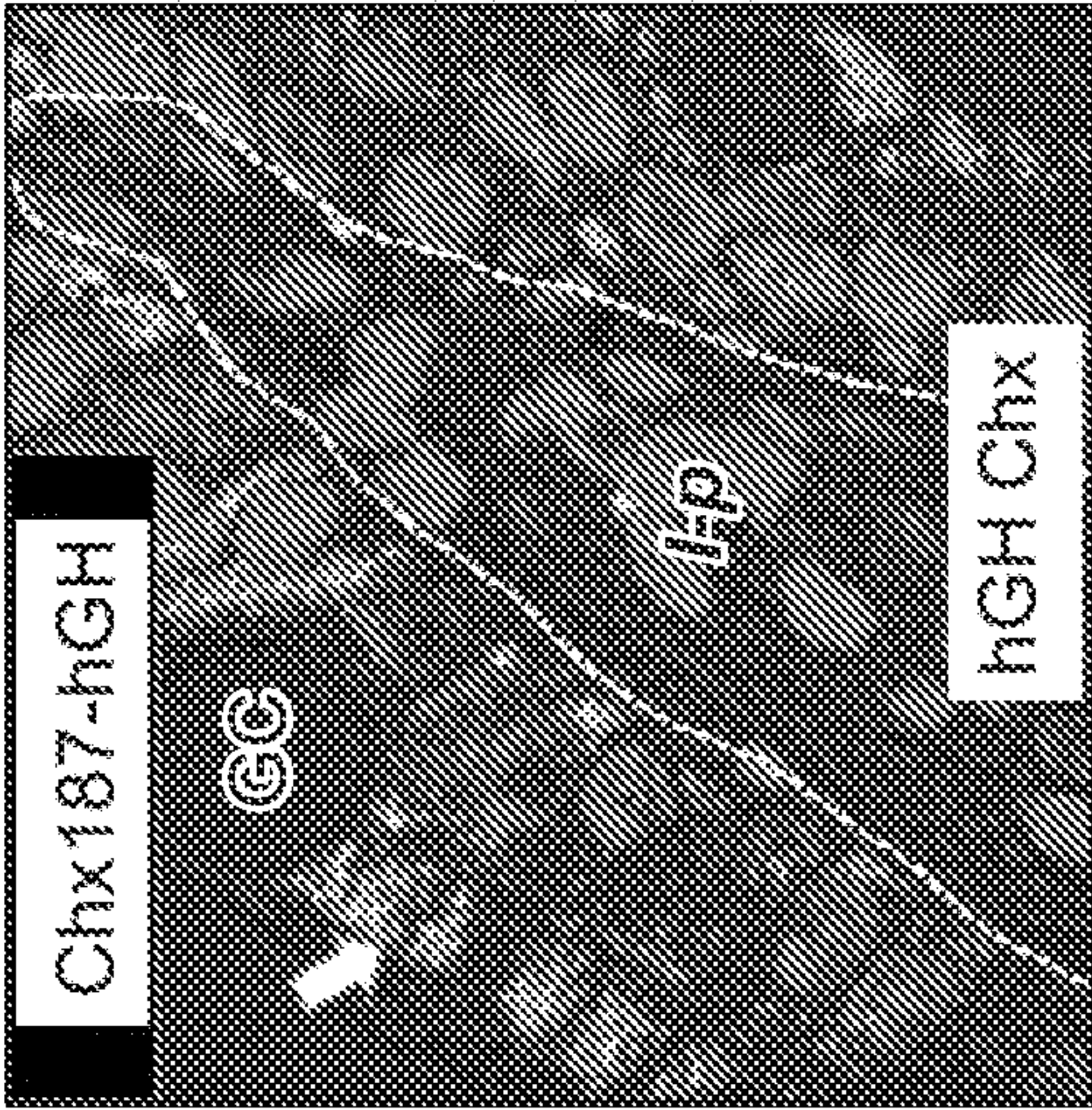


FIG. 16C

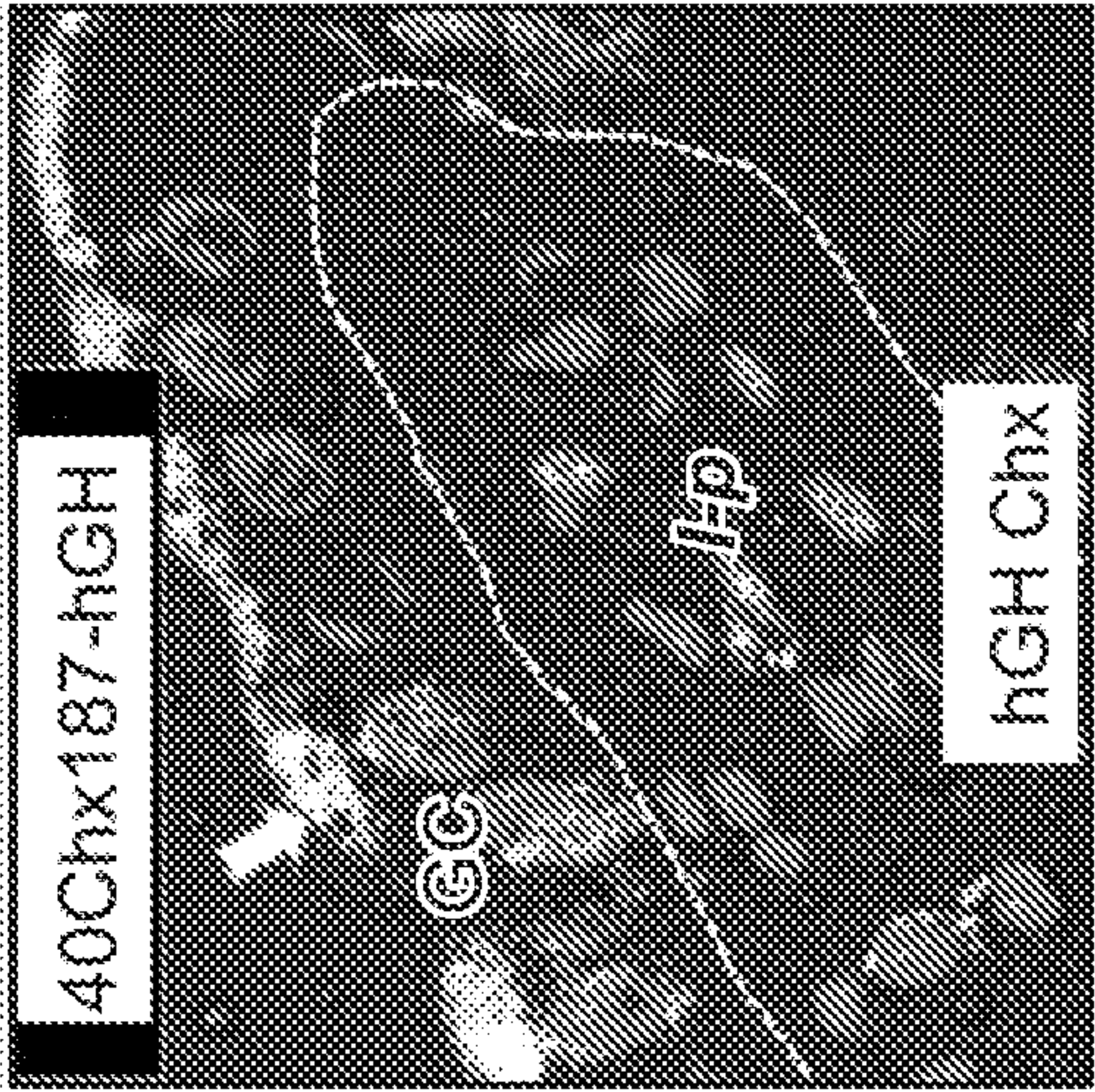


FIG. 16D

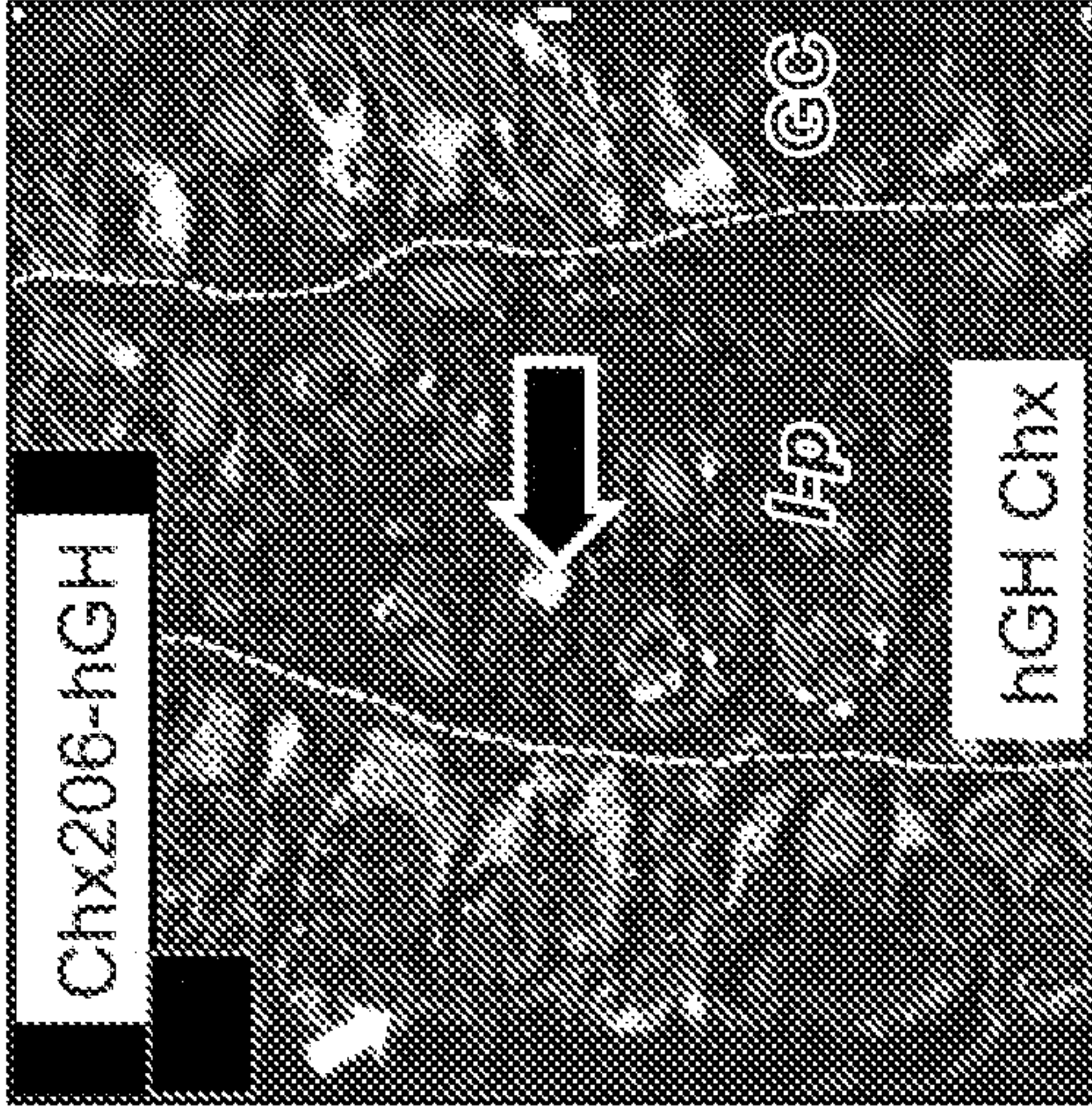


FIG. 16E

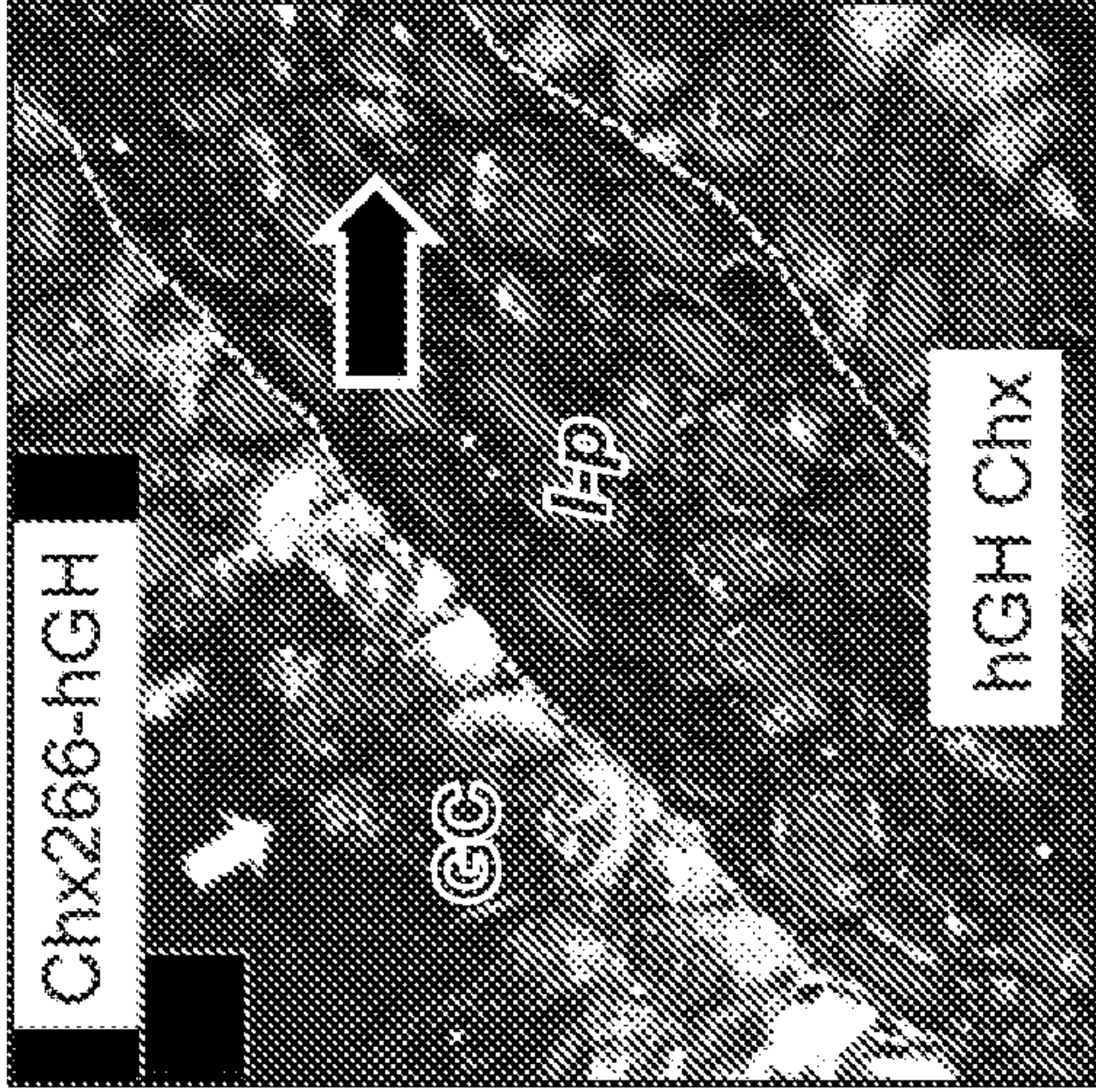


FIG. 16F

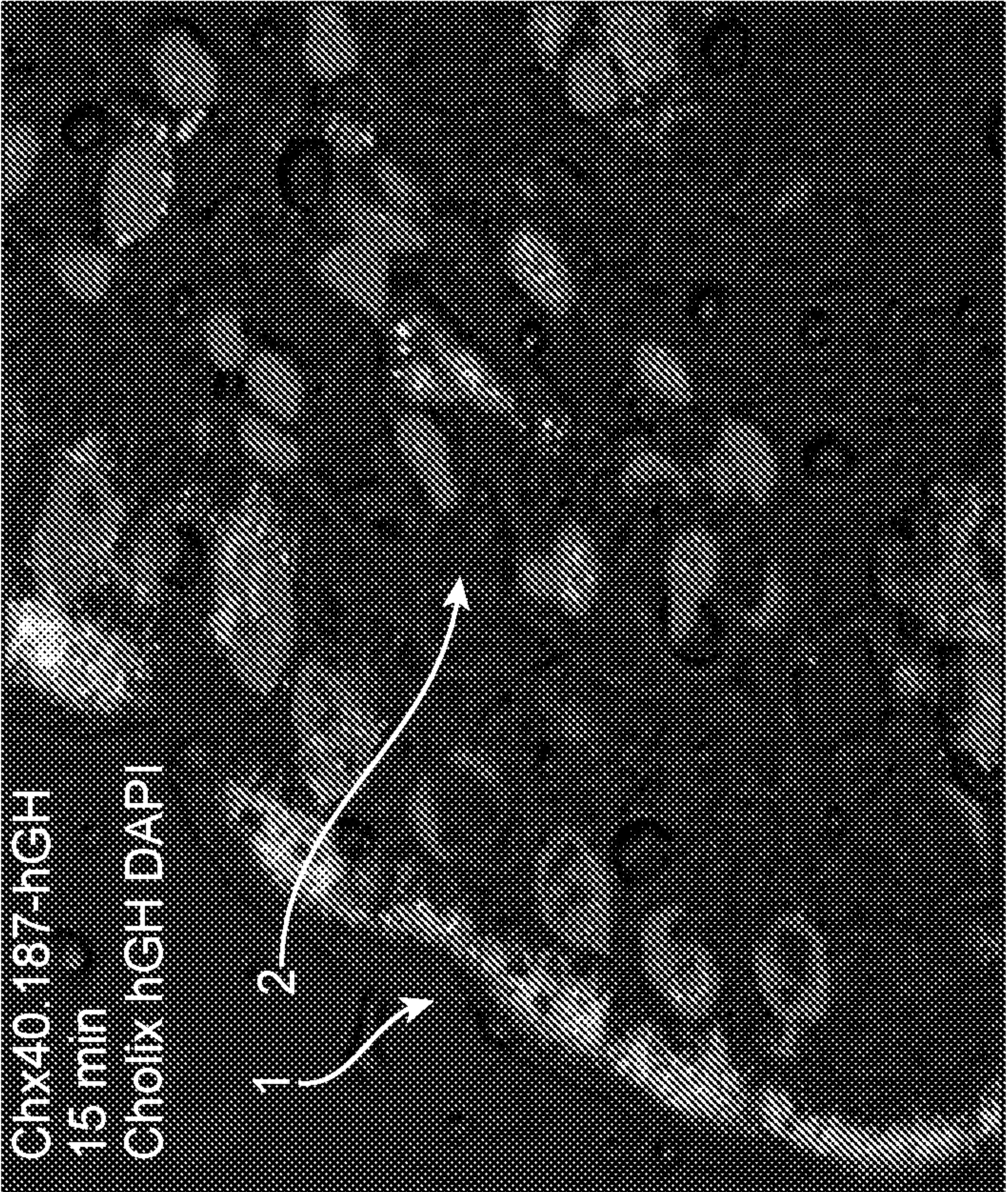
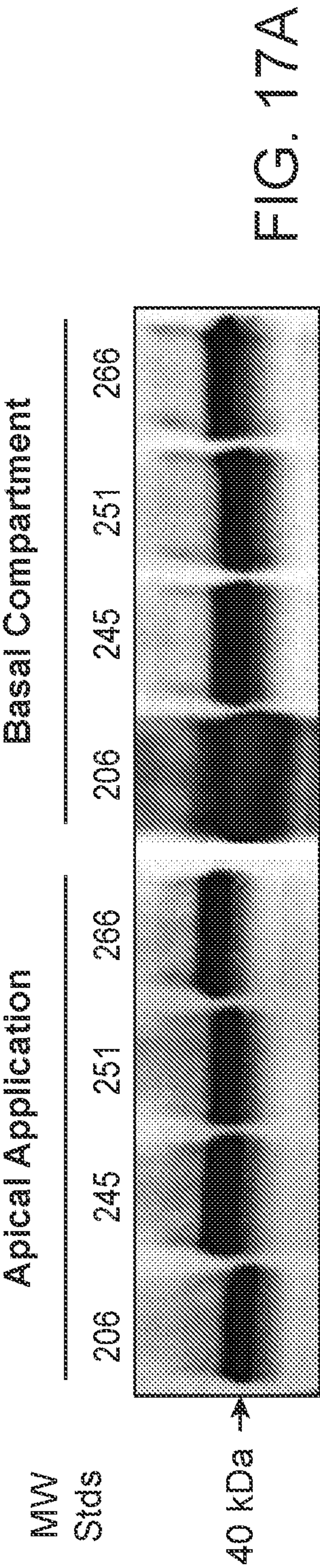


FIG. 17B

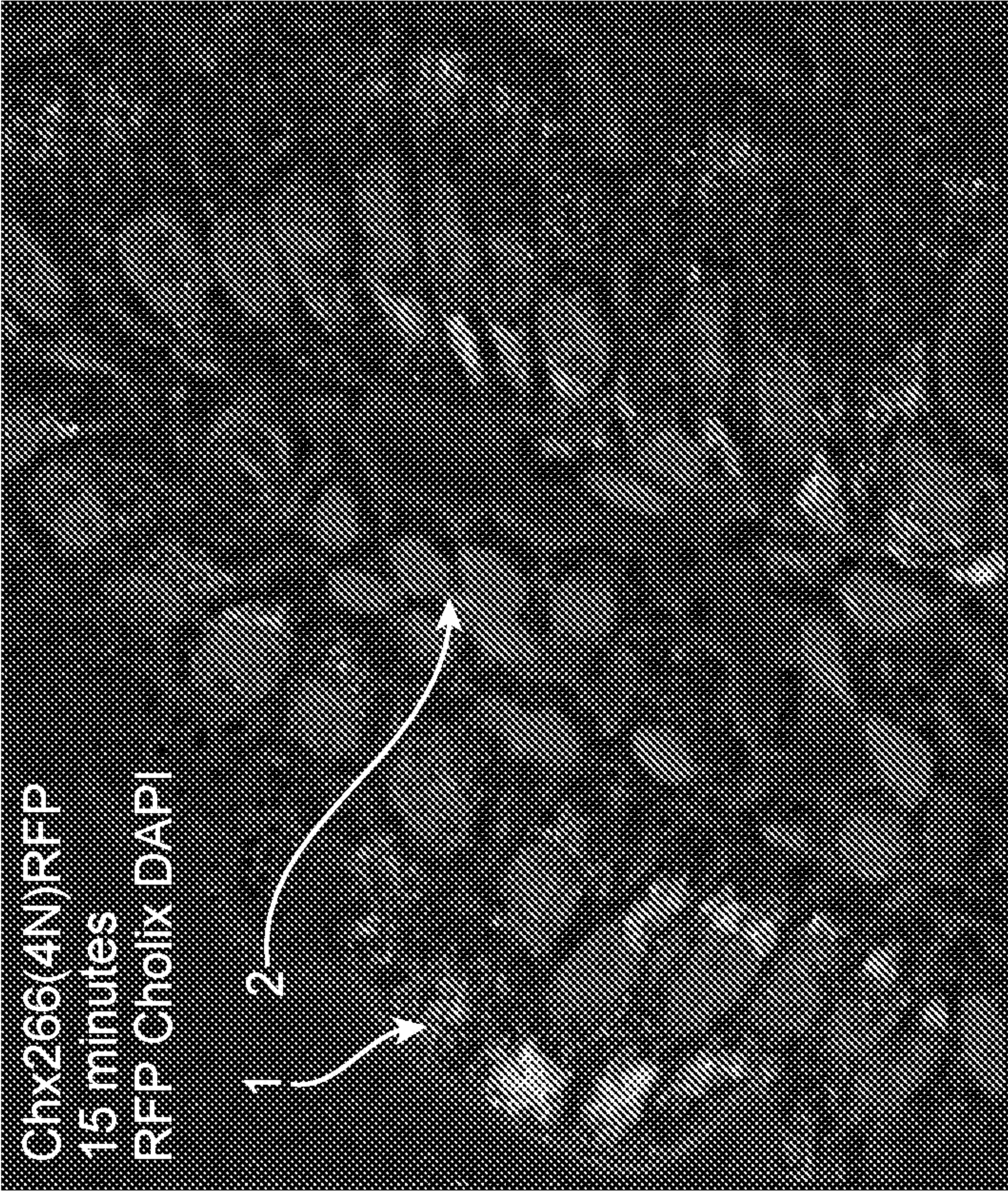


FIG. 17C

FIG. 17

Cholix domain | highlighting critical peptide sequences

1-40

134-151

151-186

187-206

MVEEALNIFDECRSPCSLTPEPGKPIQSKLSIPSDVVLD[EGVLYYSMTINDEQNDIKDEDKGESIITIGEFAT
VRATRHYVNQDAPEFGVIHLDITTENGTKTYSYNRKEGEFAINWLVPIGEDSPASIKISVD[ELDQQRNTEVEE
KLYSTIDIDNQTLFEQWKTOGNYSFSSVPRPEHNIAISWPSYKAAQKEGSRHKRWAHNHETGLAE[CWLVP
MDAIYNYITQQNCTLGDNWFGGSYETVAGTPKVIITVKQGIEQKPV[EQRIHFSSKGGGGSGGGSGGG
GS
(SEQ ID NO: 221)

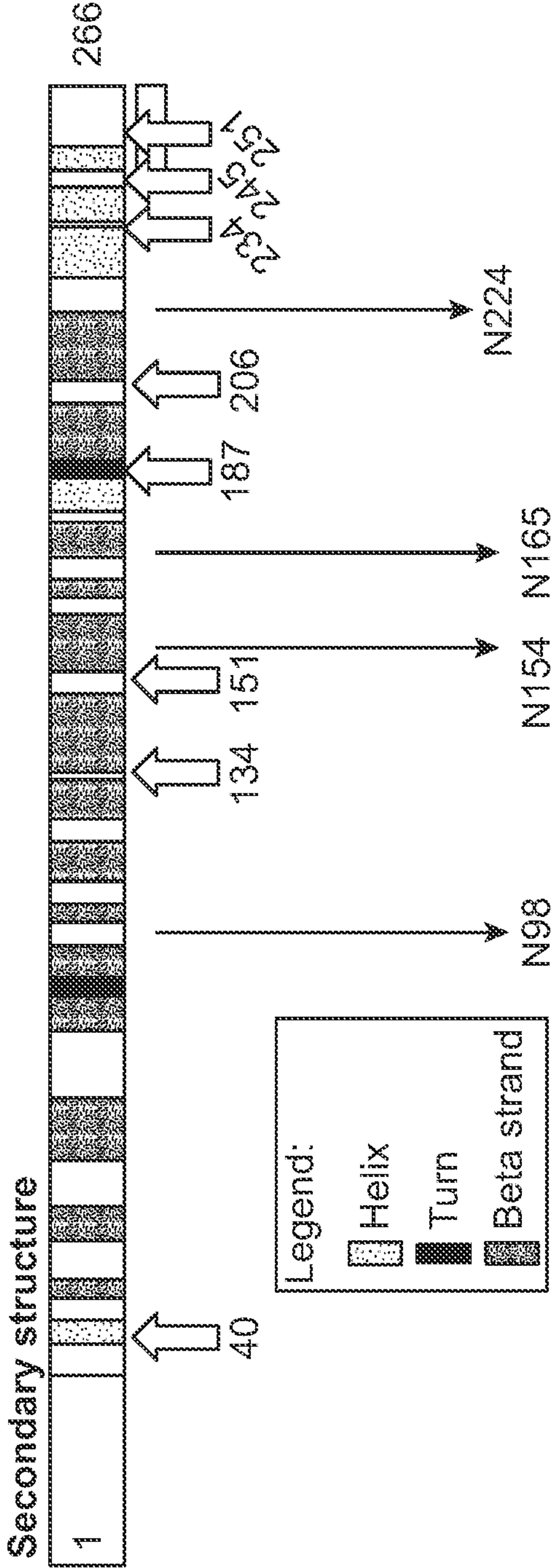


FIG. 18

Cholix domain I with linker (G₄S)₃ marking potential glycosylation sites

N = sites of potential glycosylation

MVEEALNIFDECRSPCSLTPEPGKPIQSKLSIPSDVVLDEGVLYYSMTINDEQNNDIKDEDKGESIITIGEEFAT
VRATRYVNQDAPFGVIHLDIITTENGTKTYSYNRKEGEFAINWLVPAGEDSPASIKISVDELDQQQRNIIEVP
KLYSIDLDNQTLEQWKTCQNVSFVSVTRPEHNIAISWPSVSYKAAQKEGSRHKKRWAHWHTGLALCWLVP
MDAIYNYITQQNCTLGDNWFGGSYETVAGTPKVIITVKQGIEQKPVEQRIHFSSKGGGGSGGGSGGG

GS

(SEQ ID NO: 221)

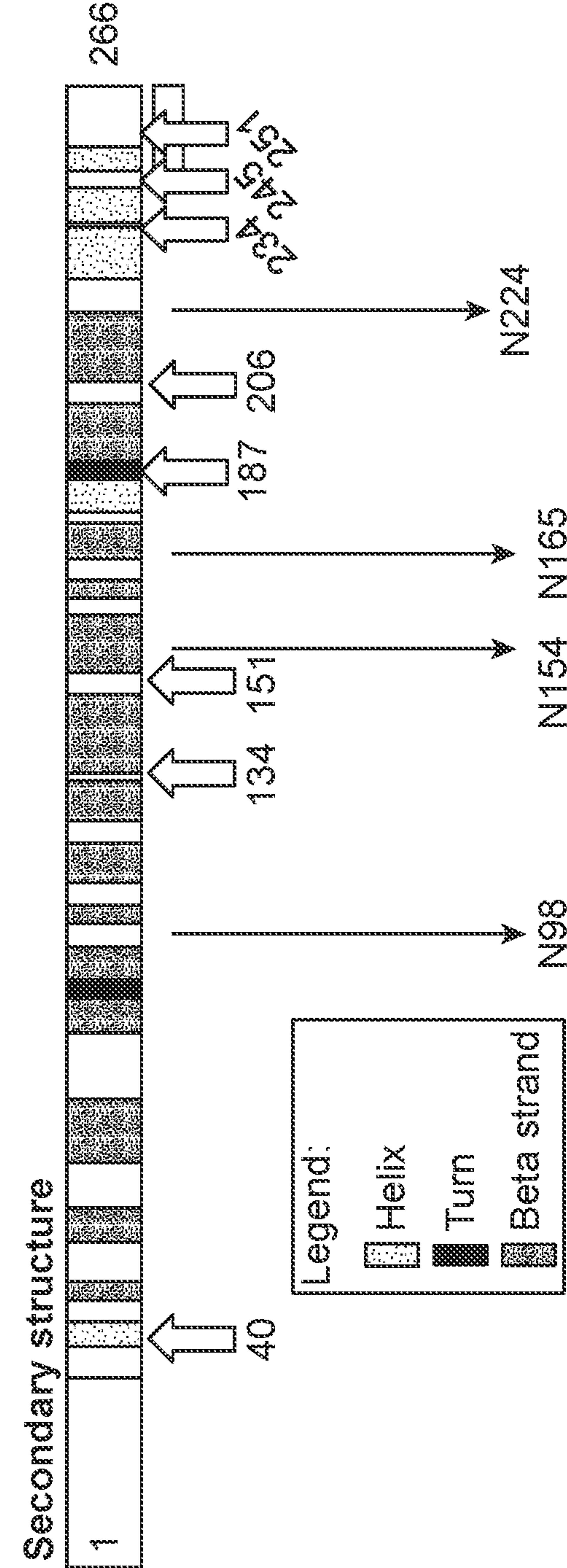


FIG. 19



FIG. 20A

FIG. 20B

FIG. 20C

FIG. 20

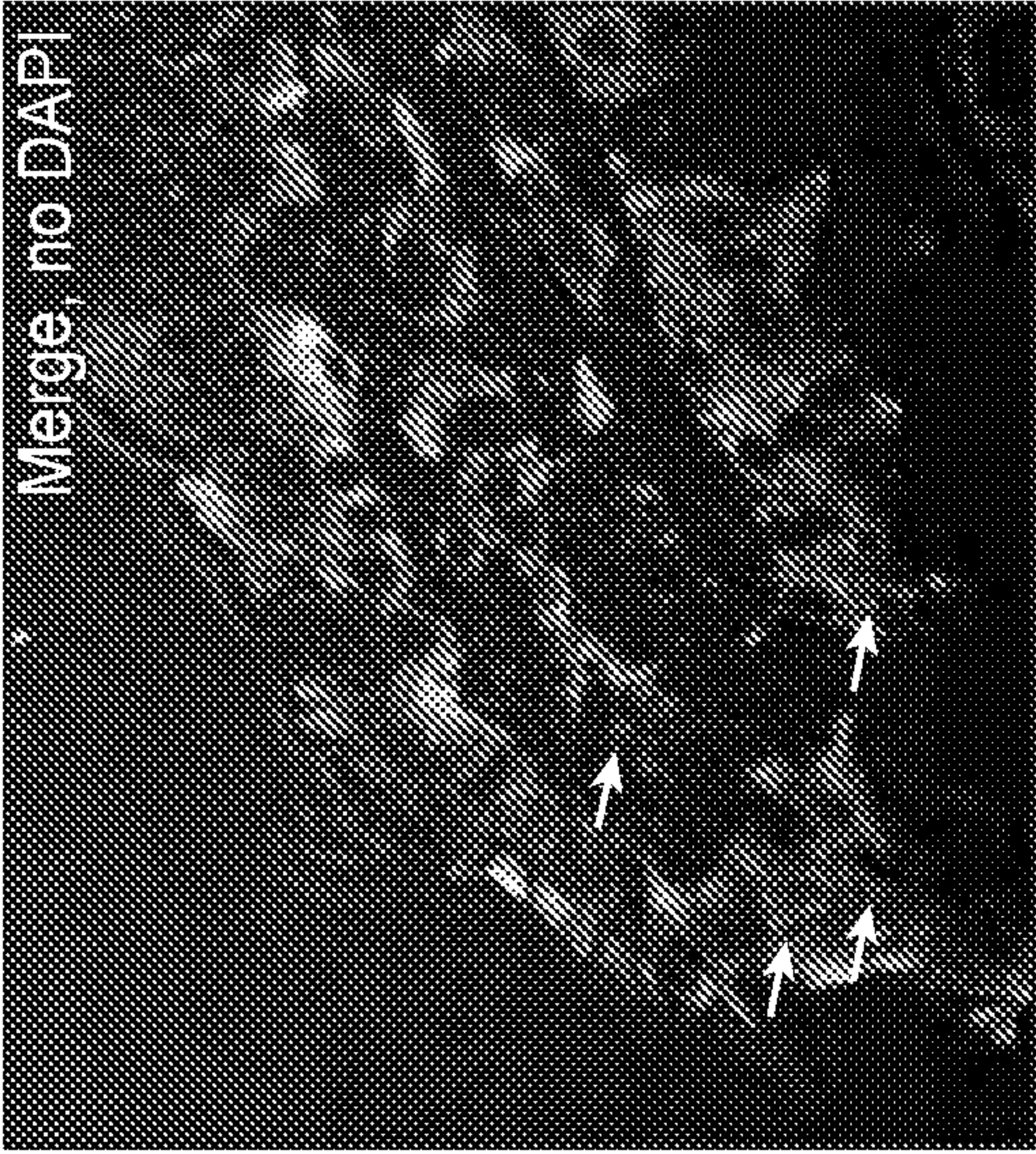


FIG. 21C

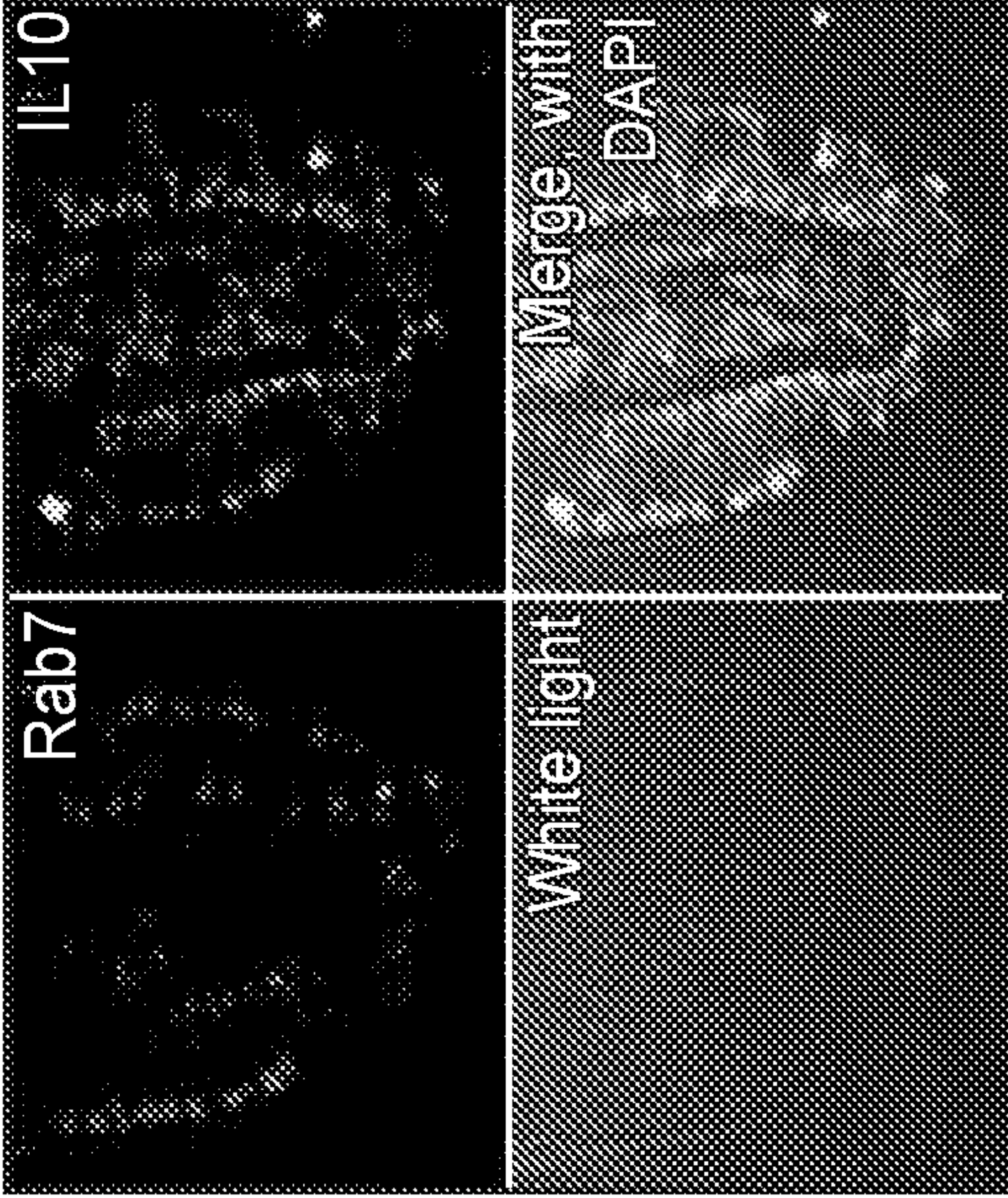


FIG. 21B



FIG. 21E

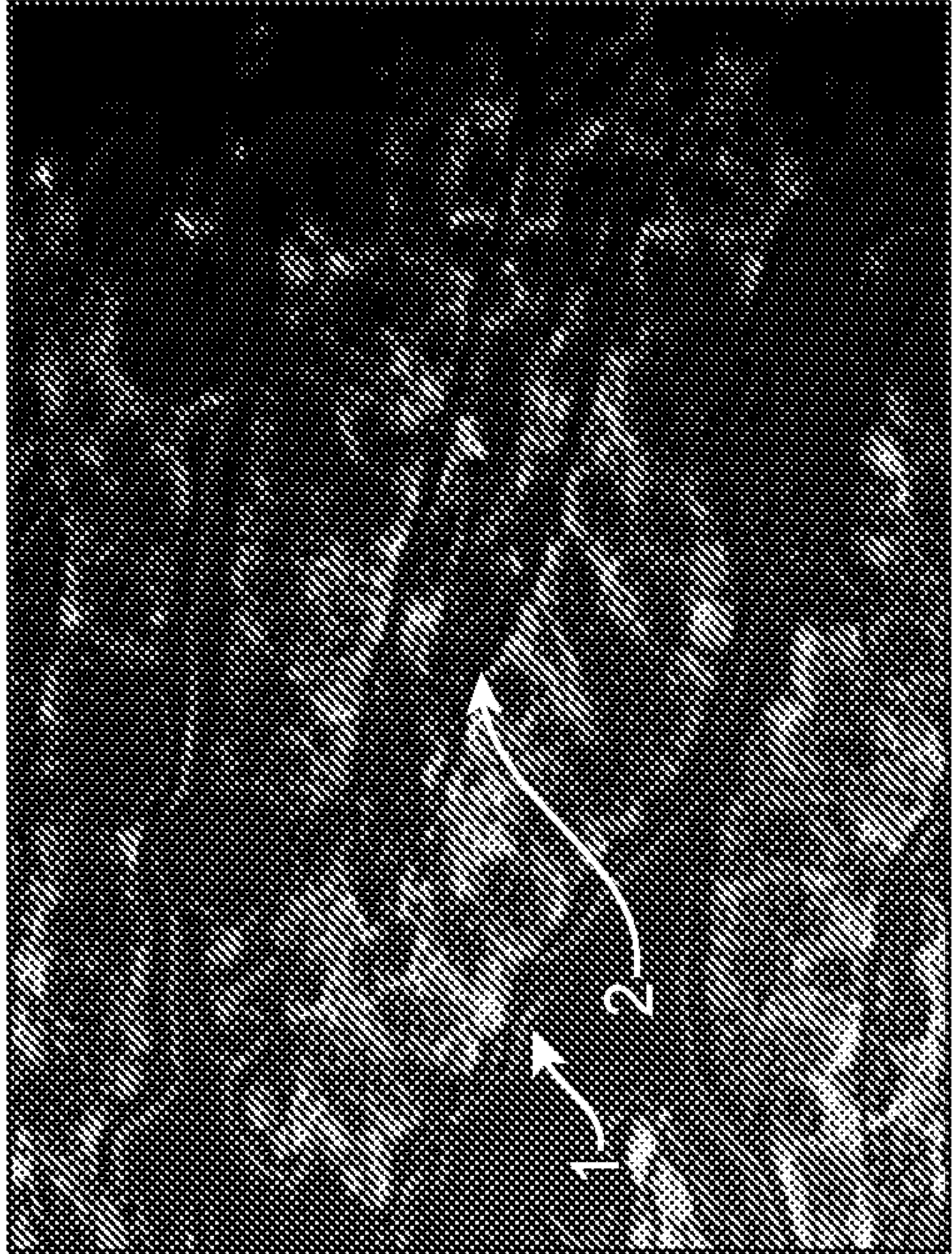


FIG. 21A

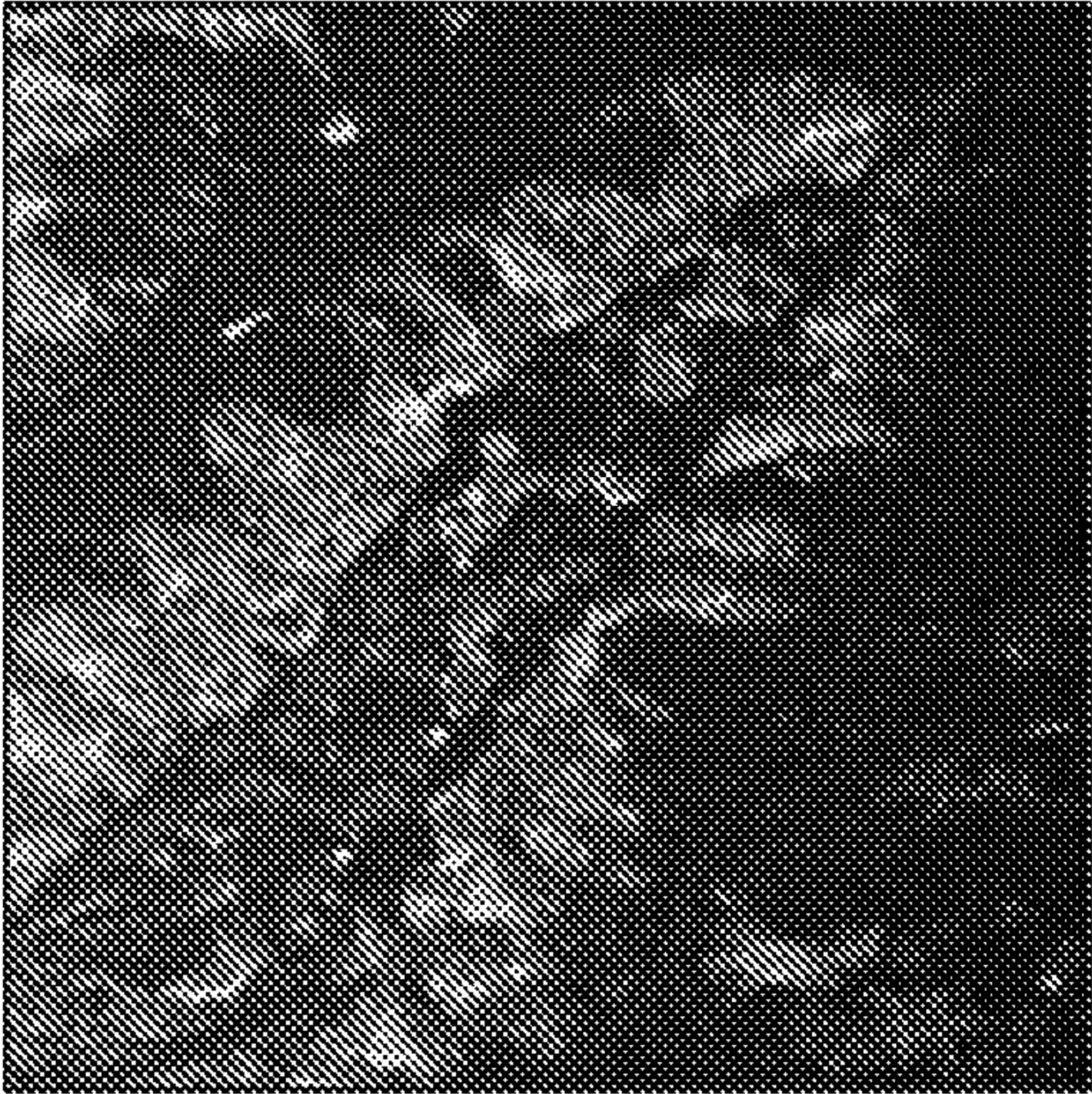


FIG. 21D

FIG. 21

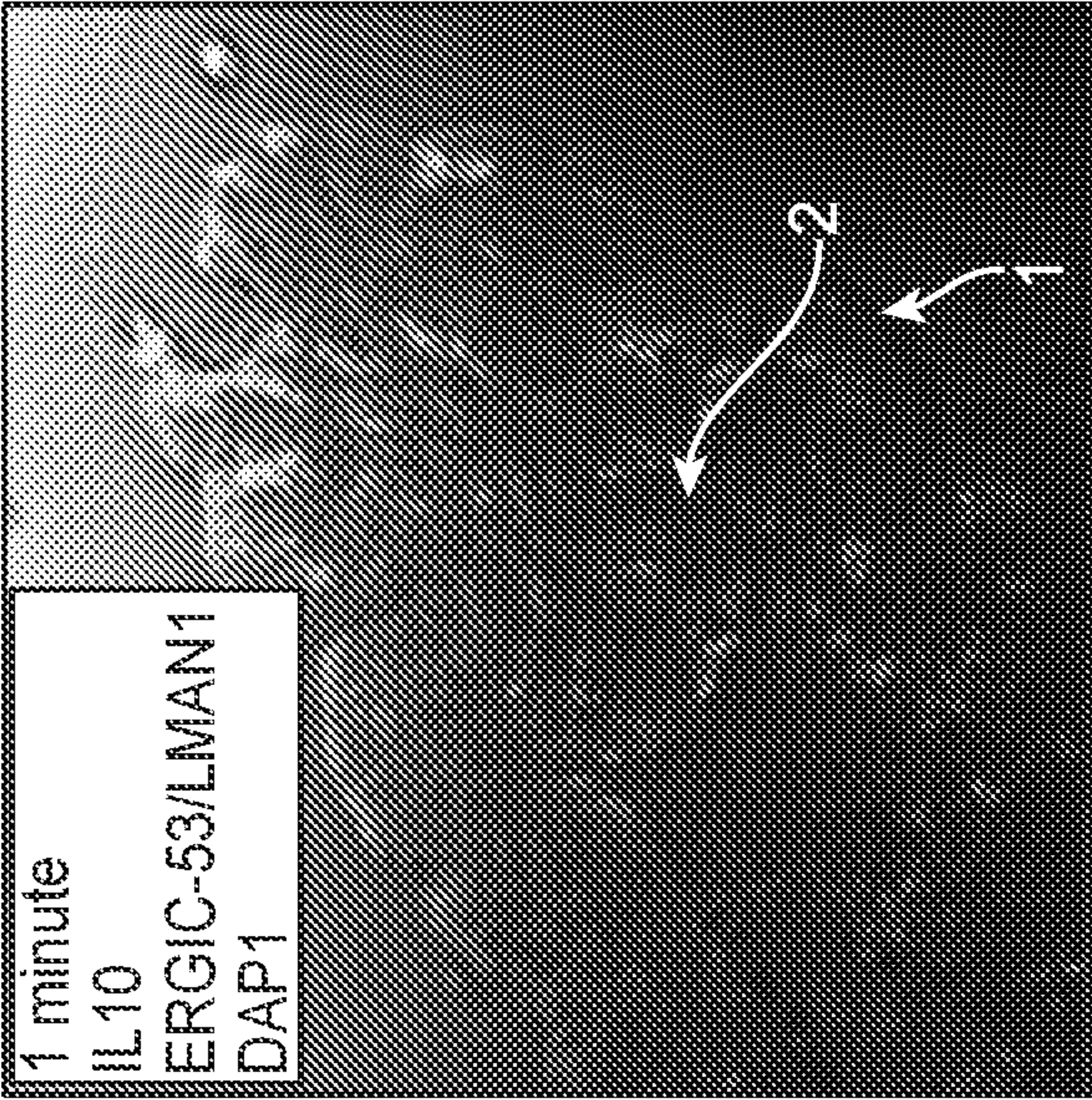


FIG. 21F

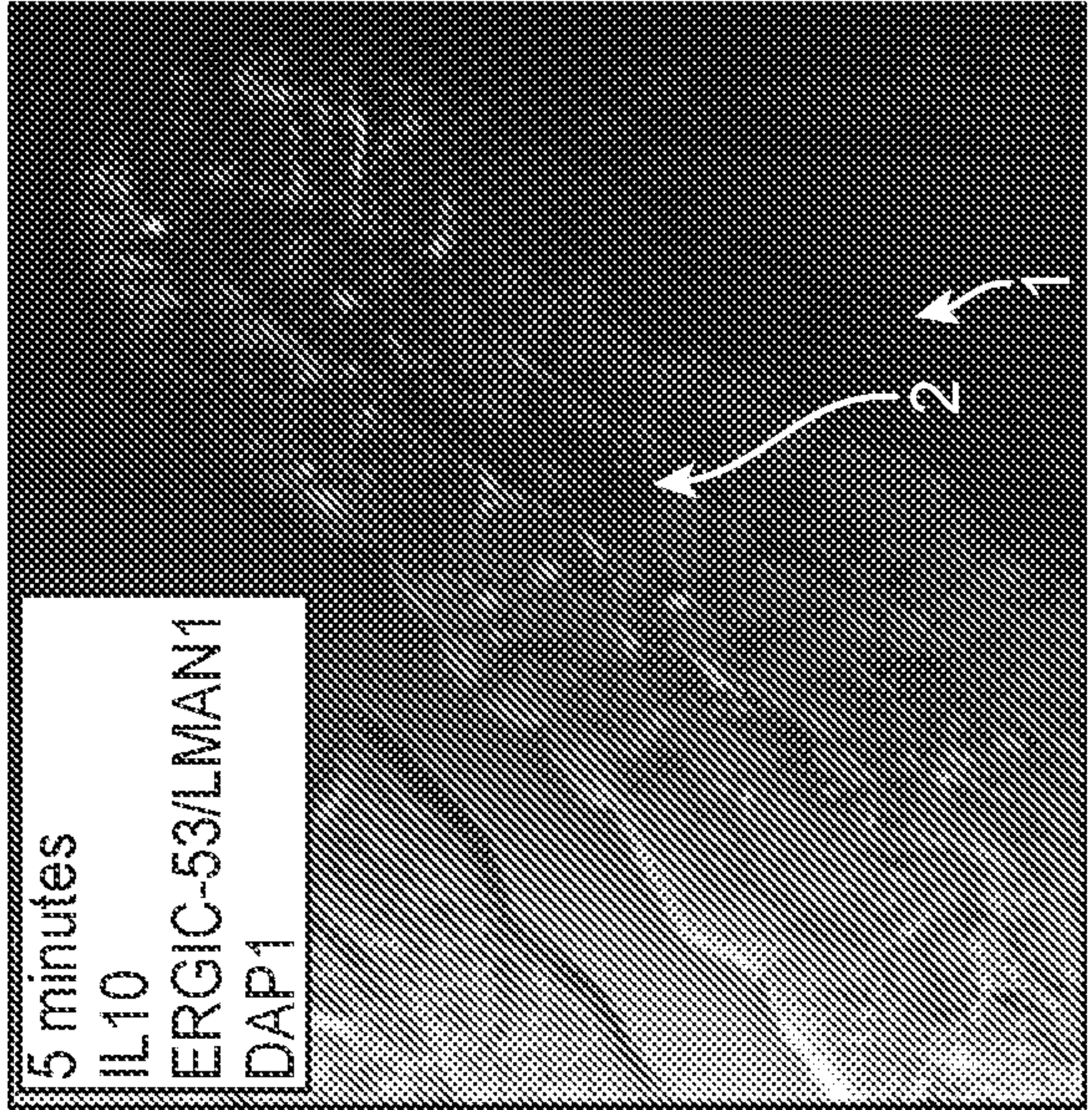


FIG. 21G

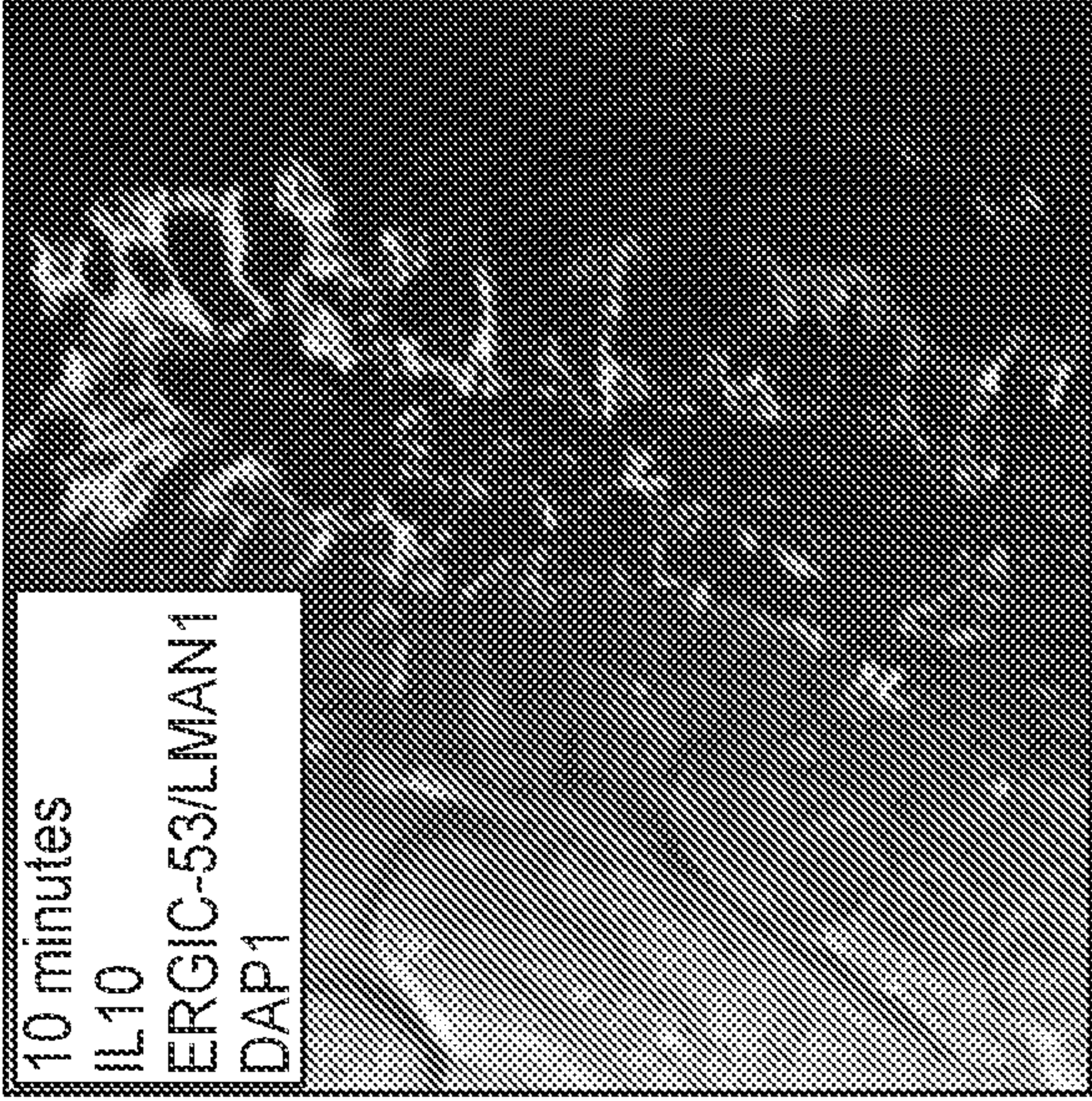


FIG. 21H

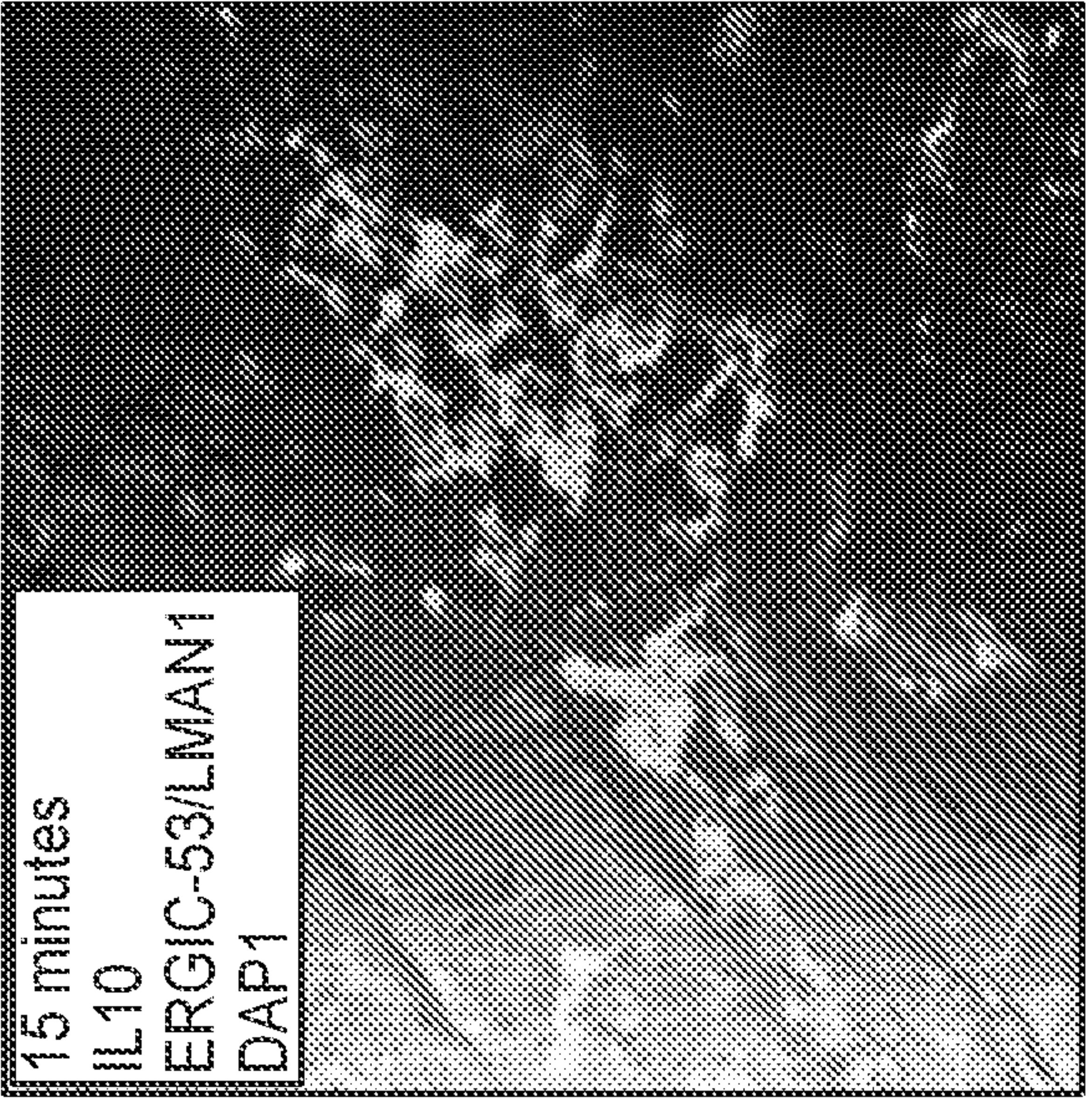


FIG. 21I

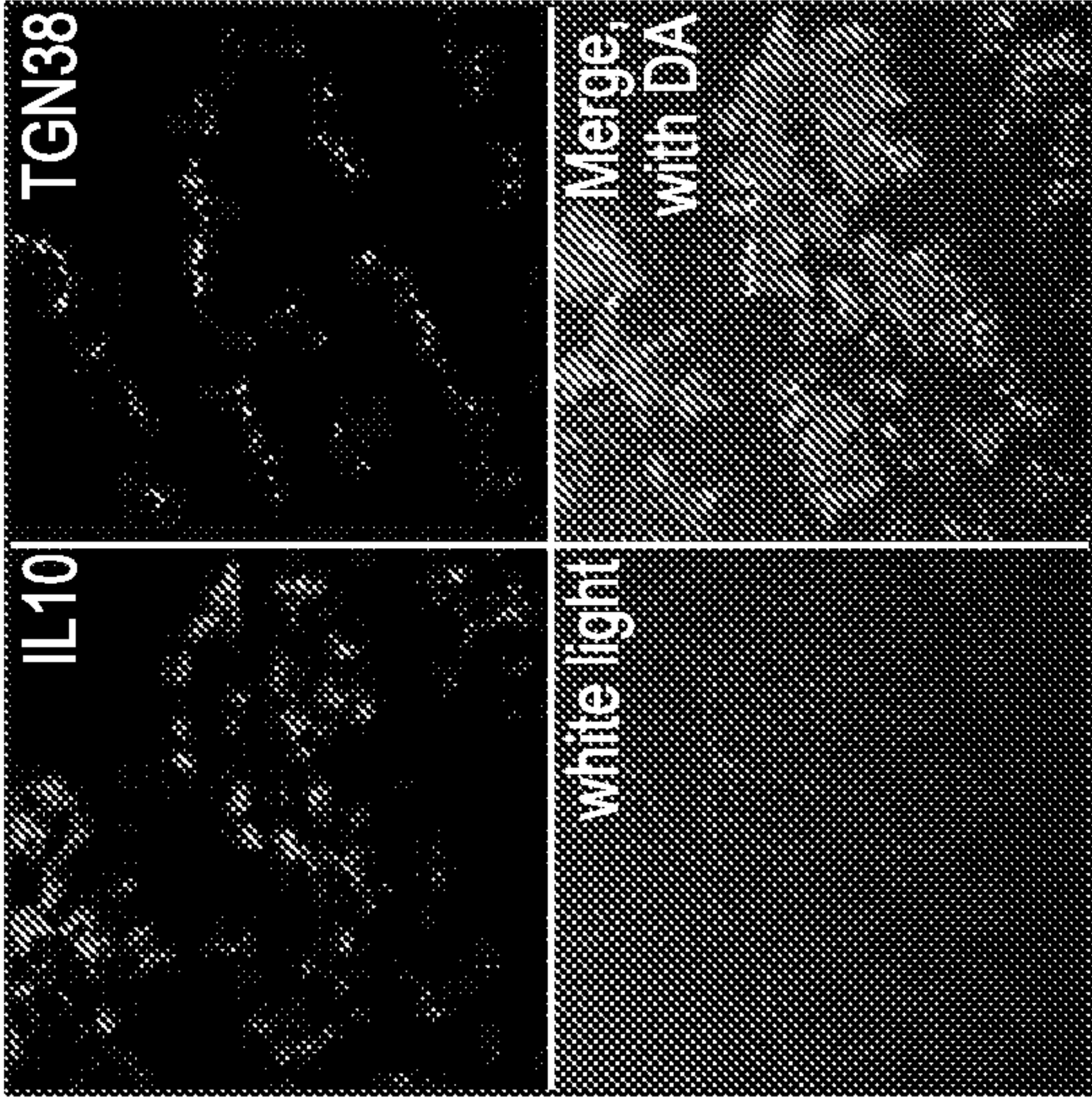


FIG. 21L

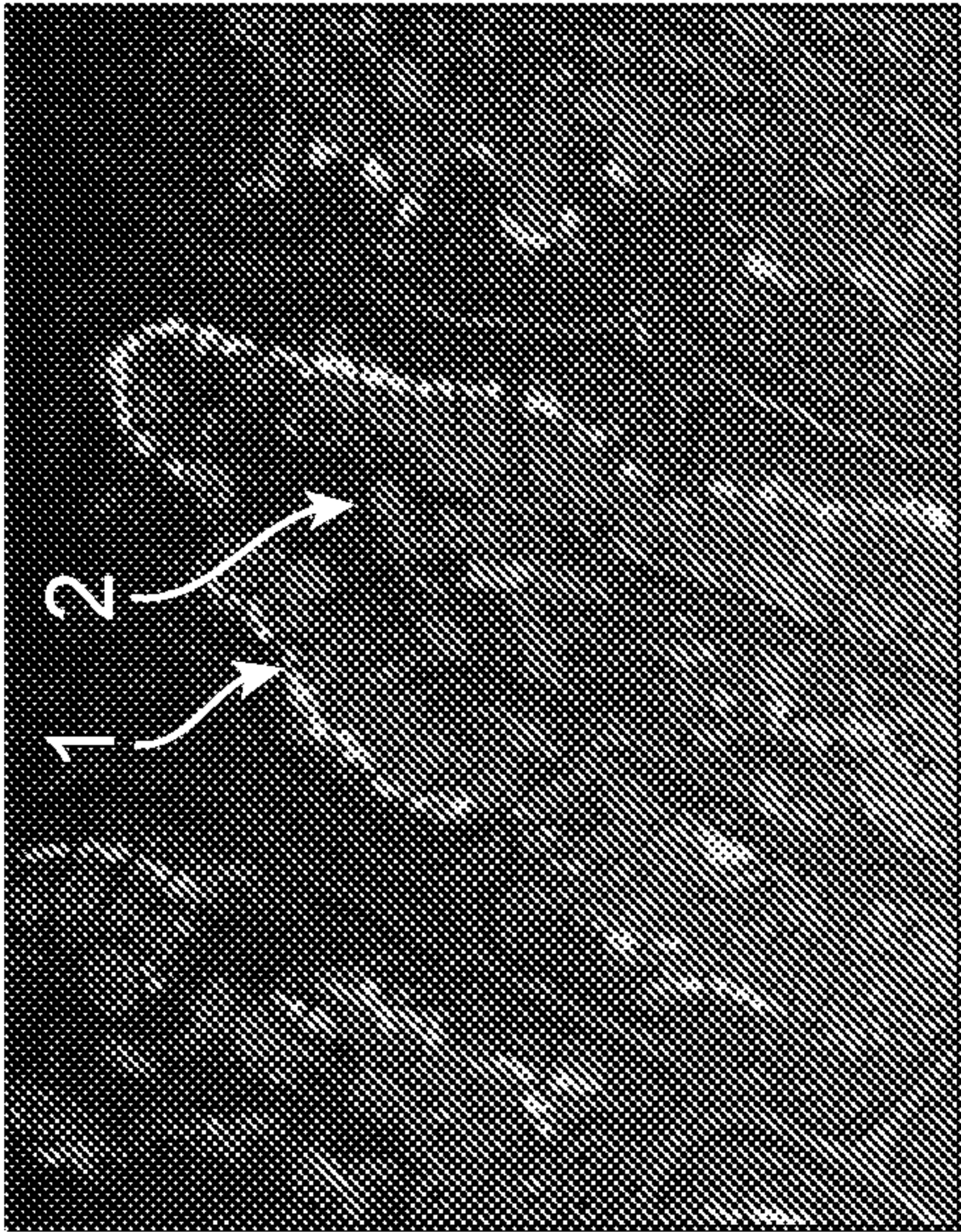


FIG. 21K

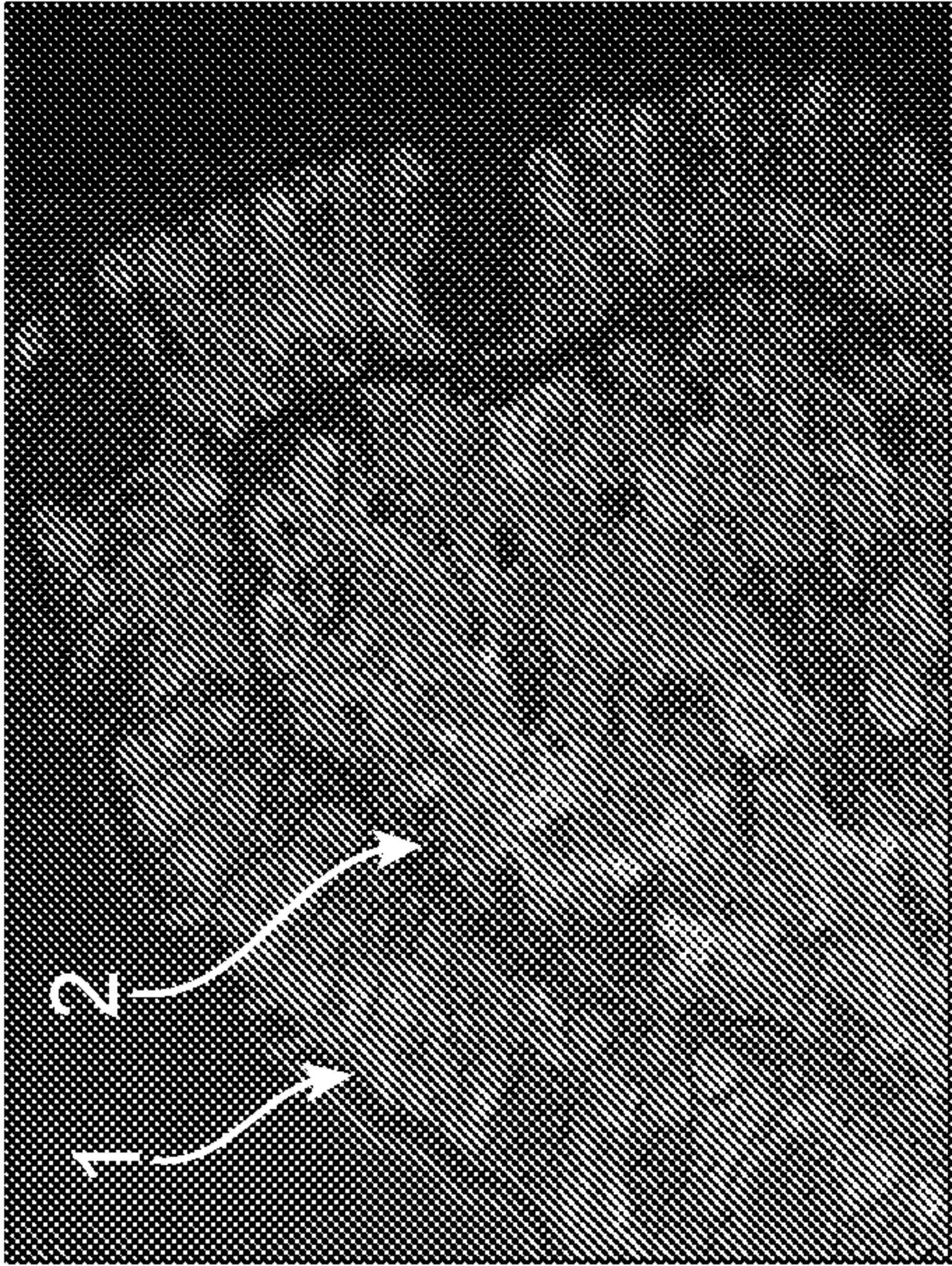


FIG. 21J

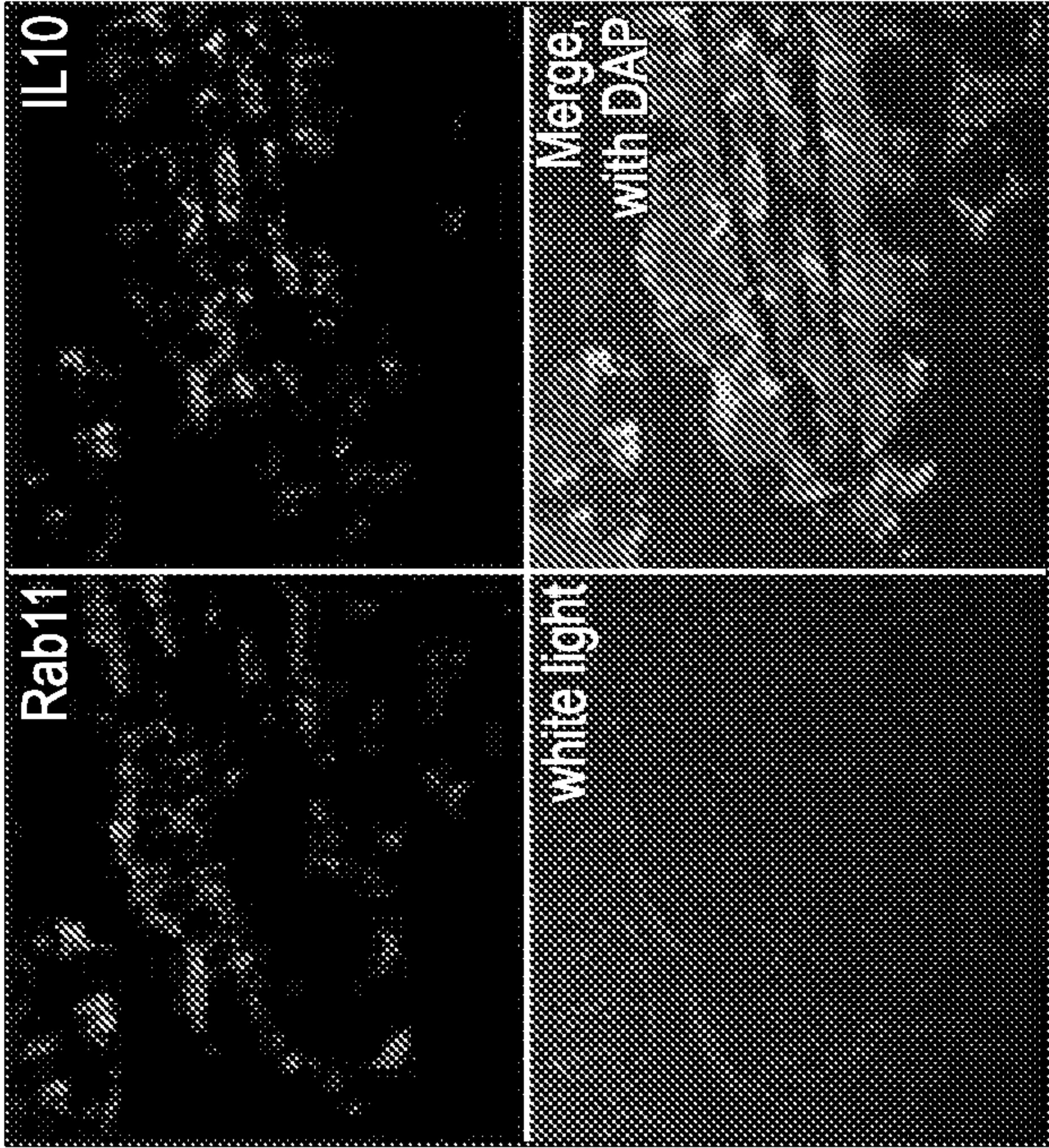


FIG. 21M

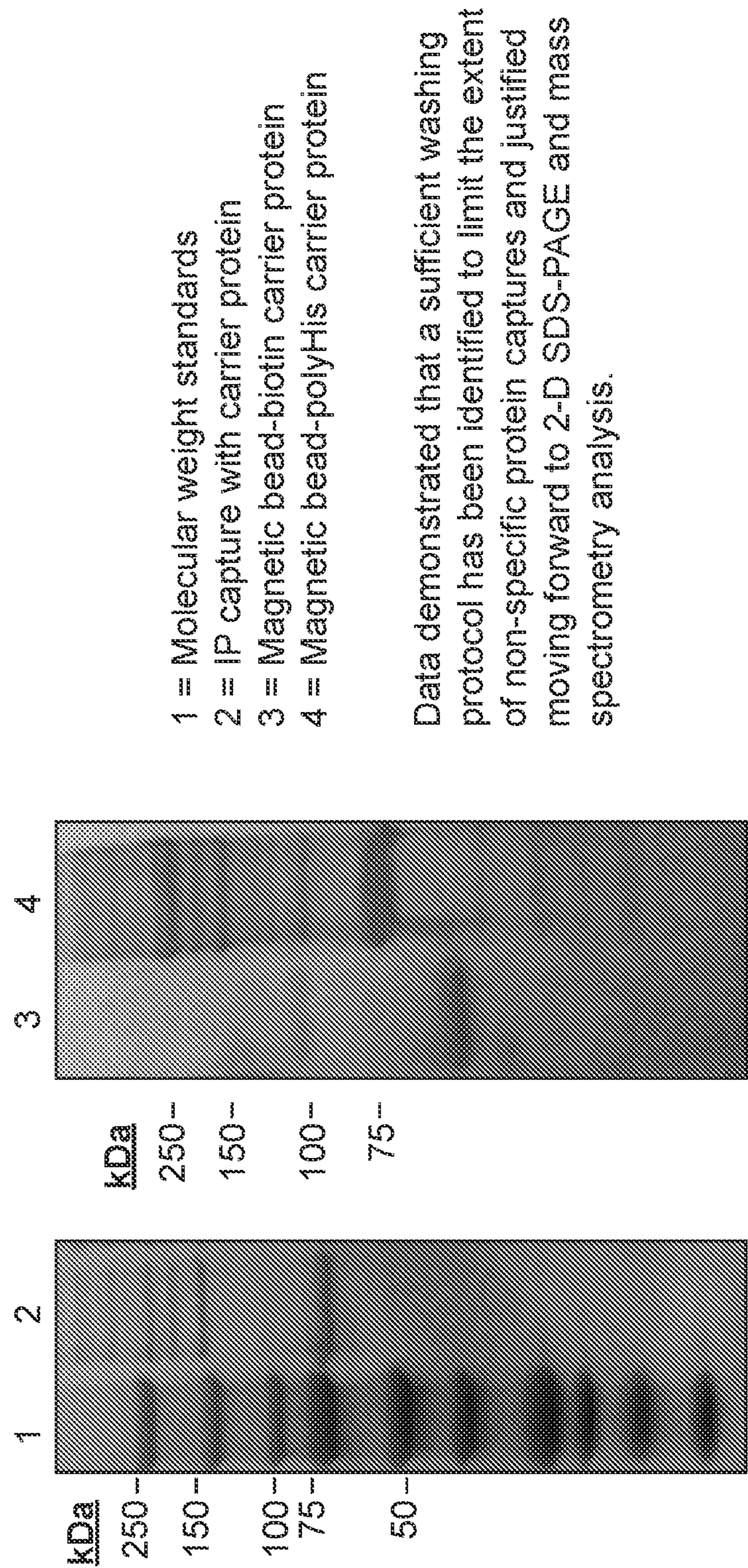
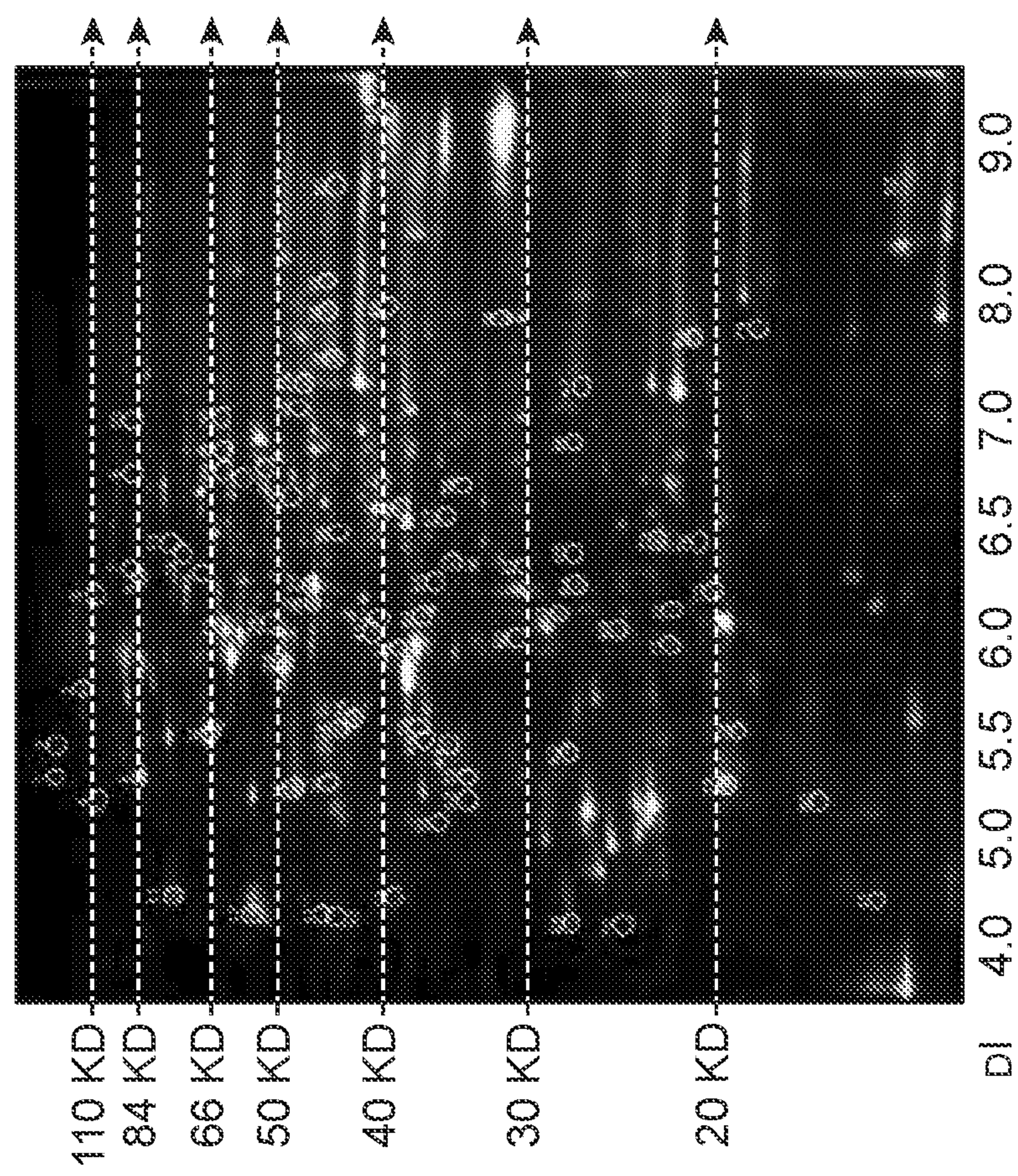


FIG. 22



Numbered circles refer to specific proteins that were targeted for mass spectrometry analysis for identification. These represent those that were augmented in the carrier-bead analyte pool and a number of control proteins used to validate the methodology.

FIG. 23

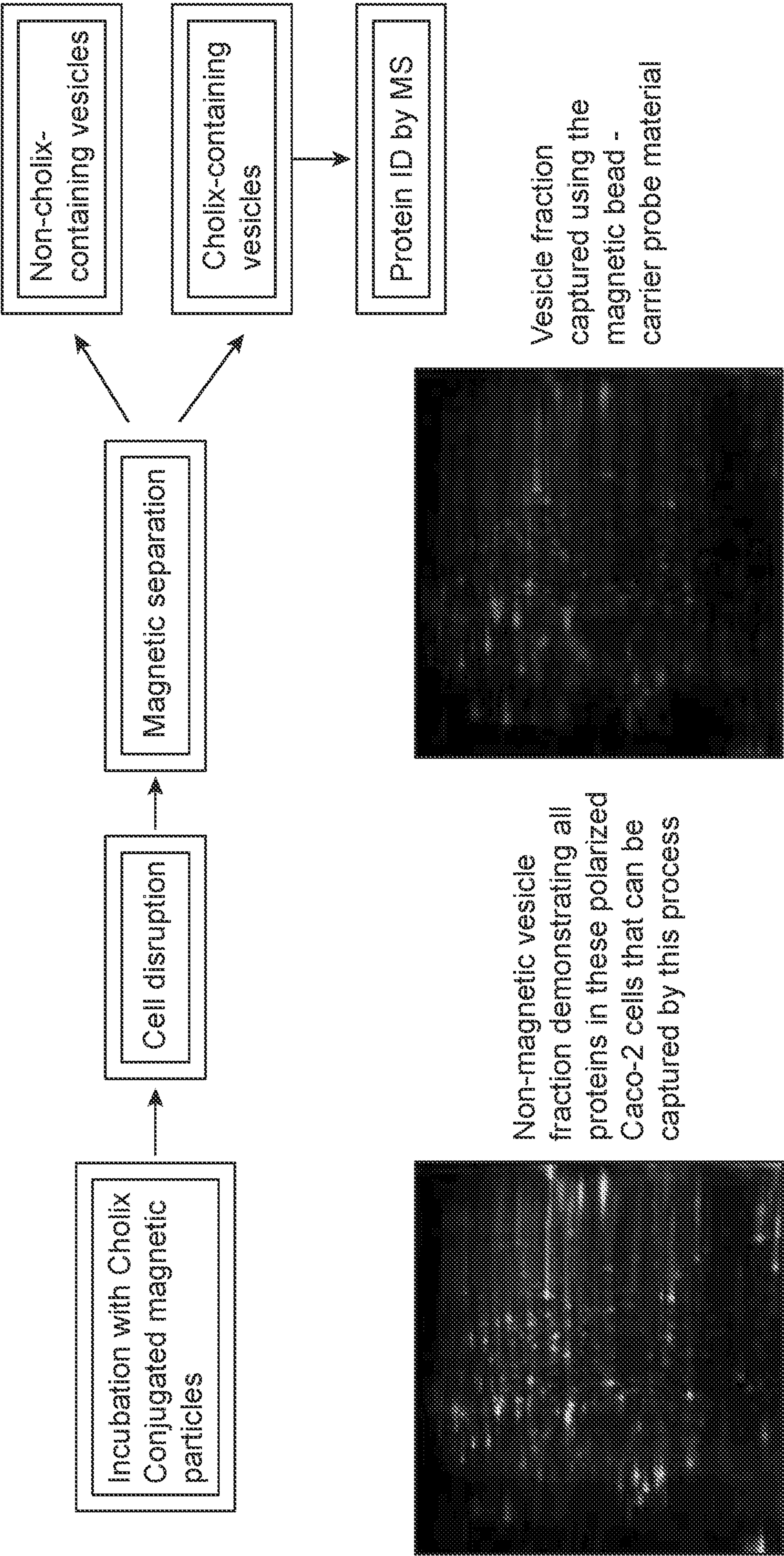


FIG. 24

Protein Candidate	Caco-2 vesicle	Caco-2 PD Cholix-His	Caco-2 PD Cholix-Biotin	Caco-2 IP anti-F4/80	Rat jejunum PD Cholix-His	Rat jejunum PD Cholix-Biotin	MS confidence
Cytokeratin	✓	✓	✓	✓	✓	✓	High
Heparan sulfate proteoglycan		✓					High
Heat shock protein	✓	✓			✓	✓	High
ATP synthase	✓	✓			✓	✓	High
Laminin	✓	✓		✓	✓	✓	Low
Tubulin	✓	✓		✓	✓	✓	Low
Actin	✓	✓	✓		✓	✓	Low
14-3-3	✓				✓	✓	High
Glial fibrillary acidic protein	✓				✓		
Dickkopf		✓					??

FIG. 25

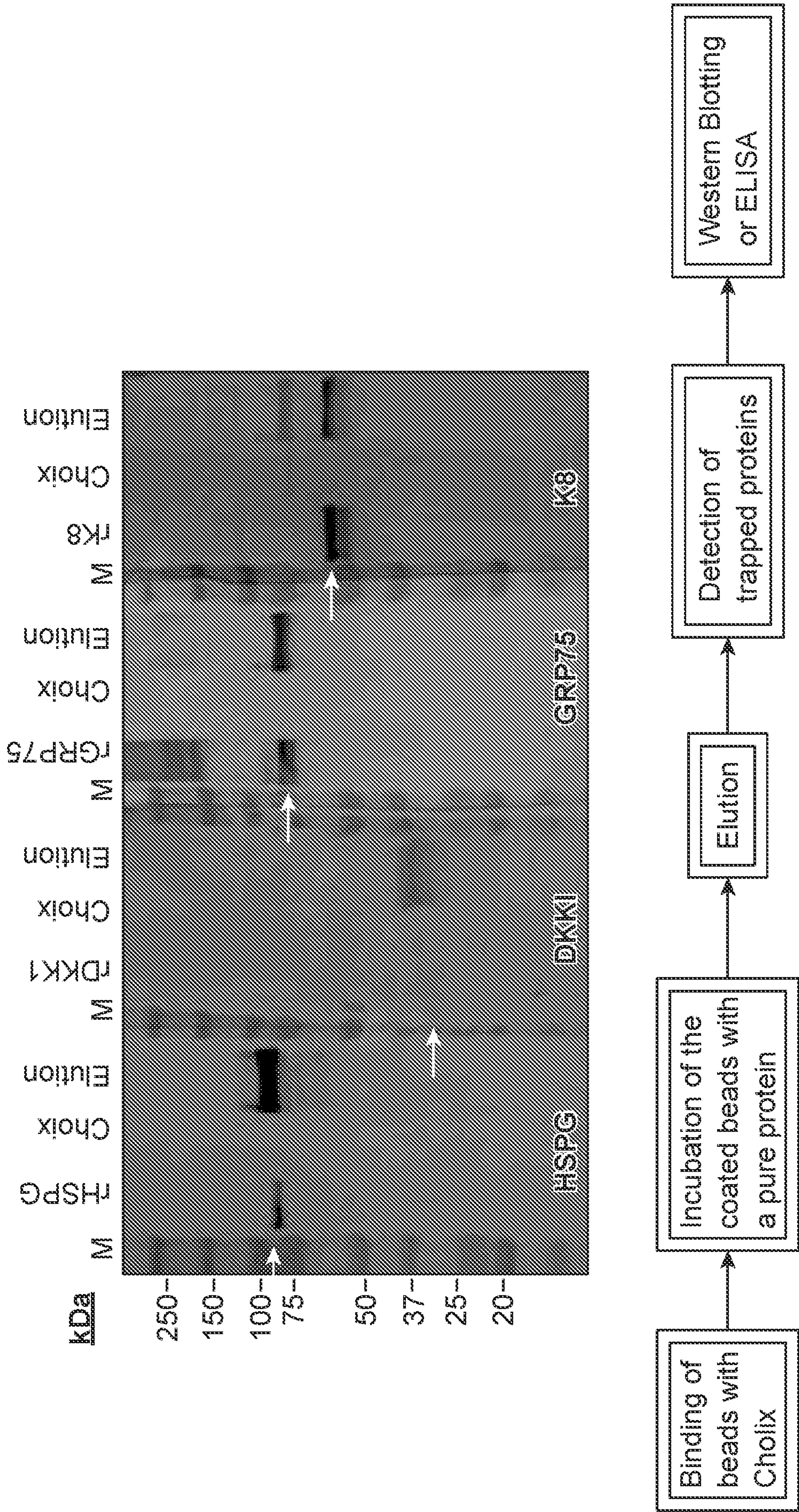


FIG. 26

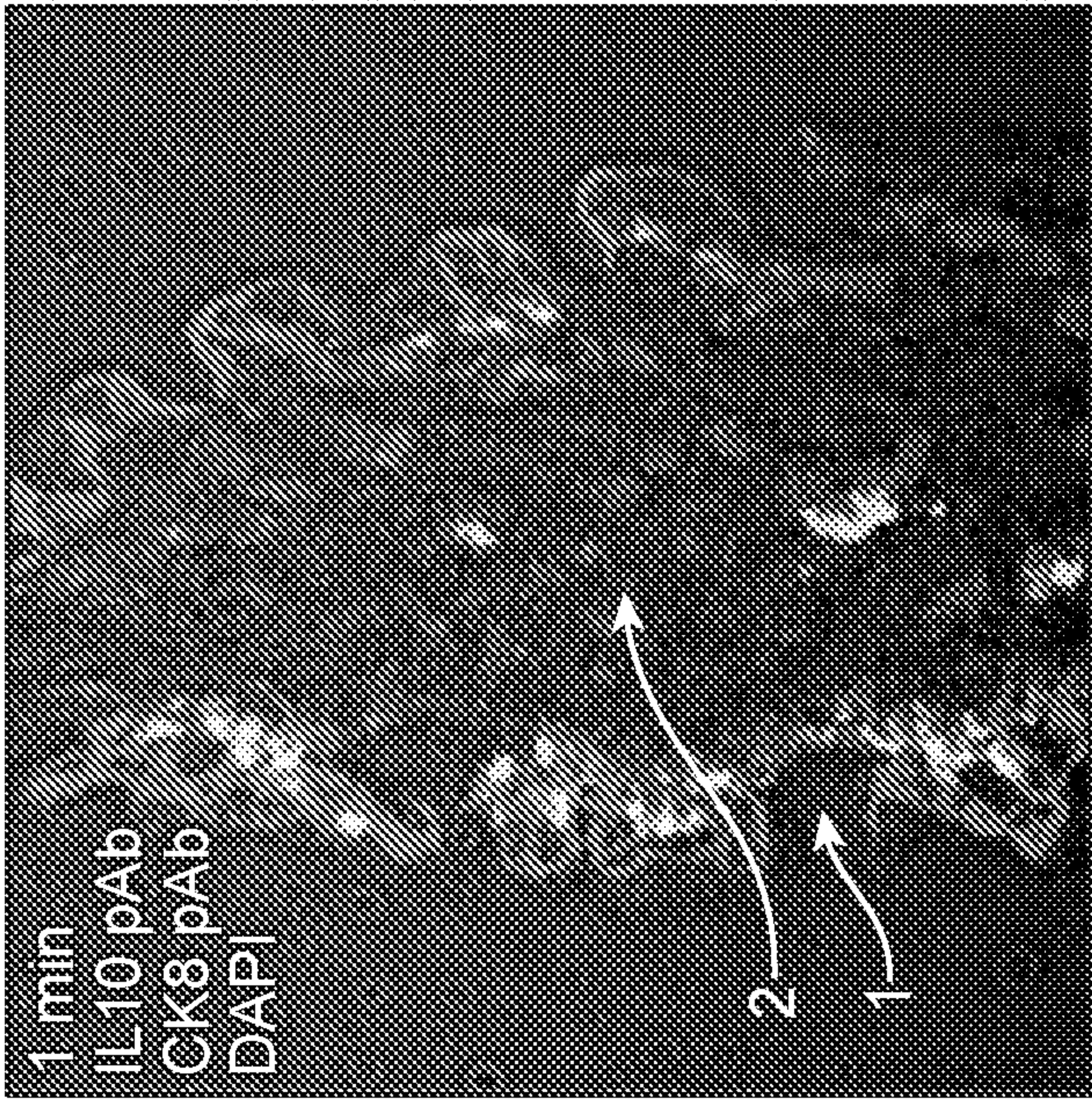


FIG. 27A

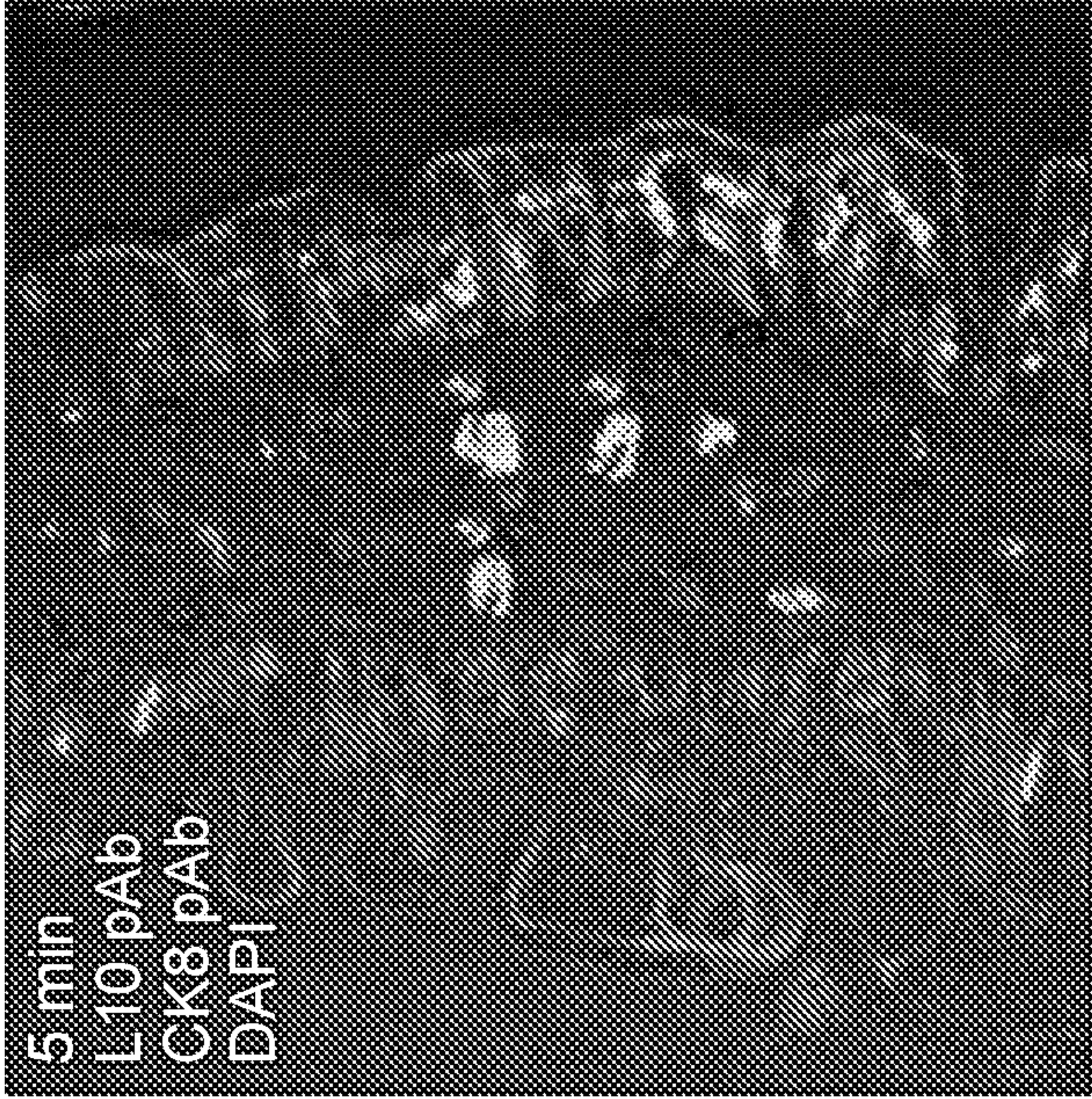


FIG. 27B

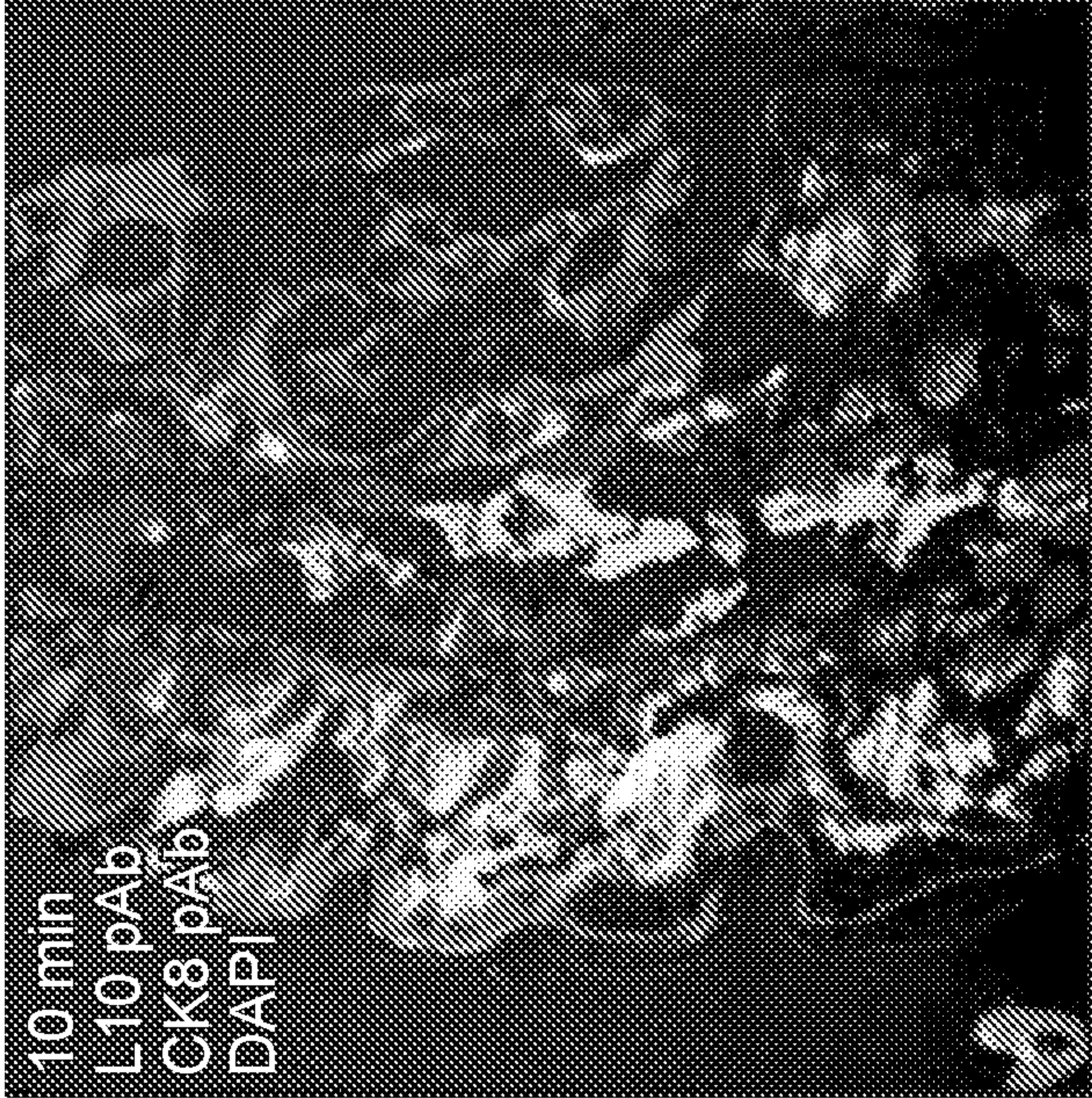


FIG. 27C

FIG. 27

29 / 33

GRP 75

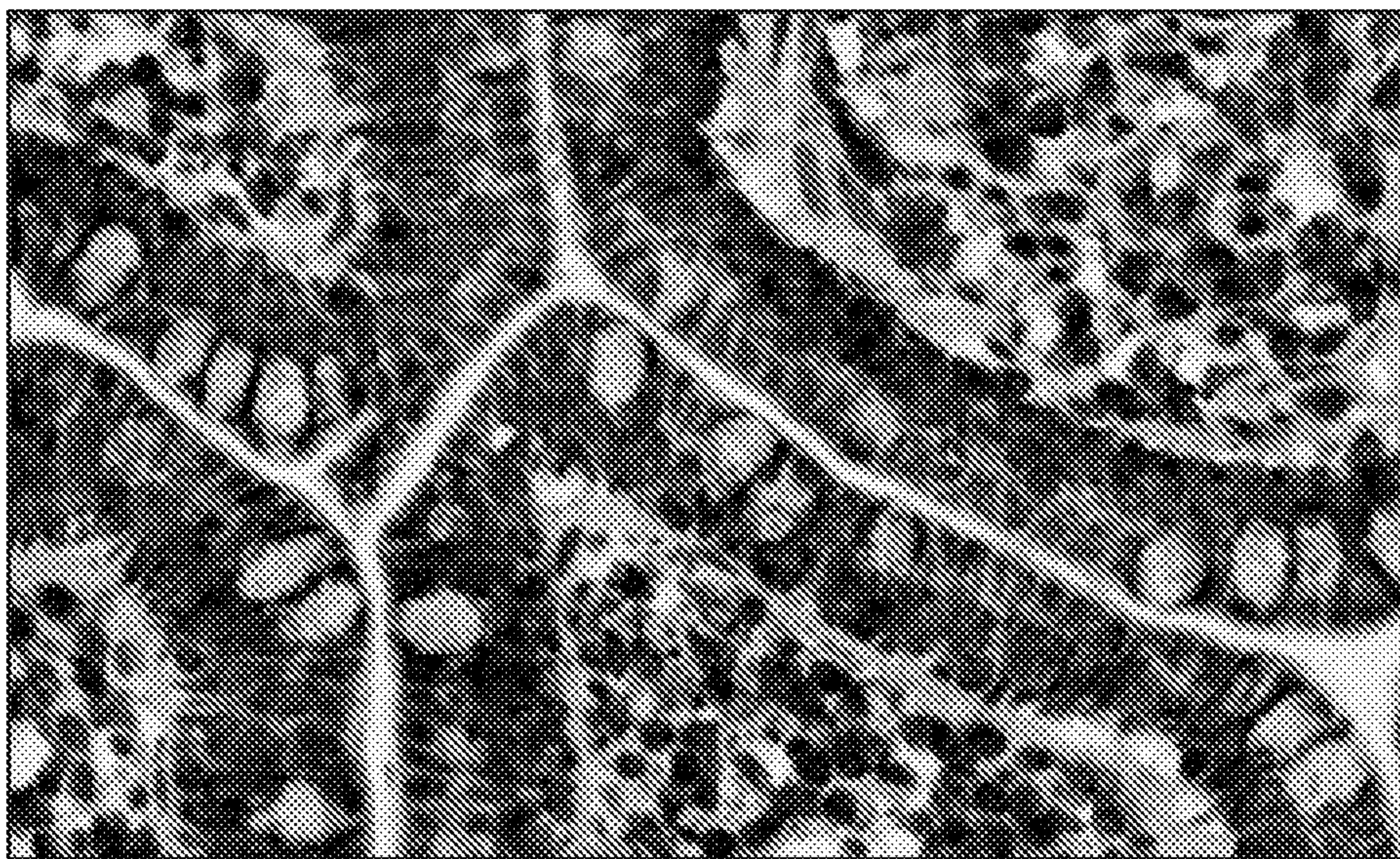


FIG. 28A

HSPG

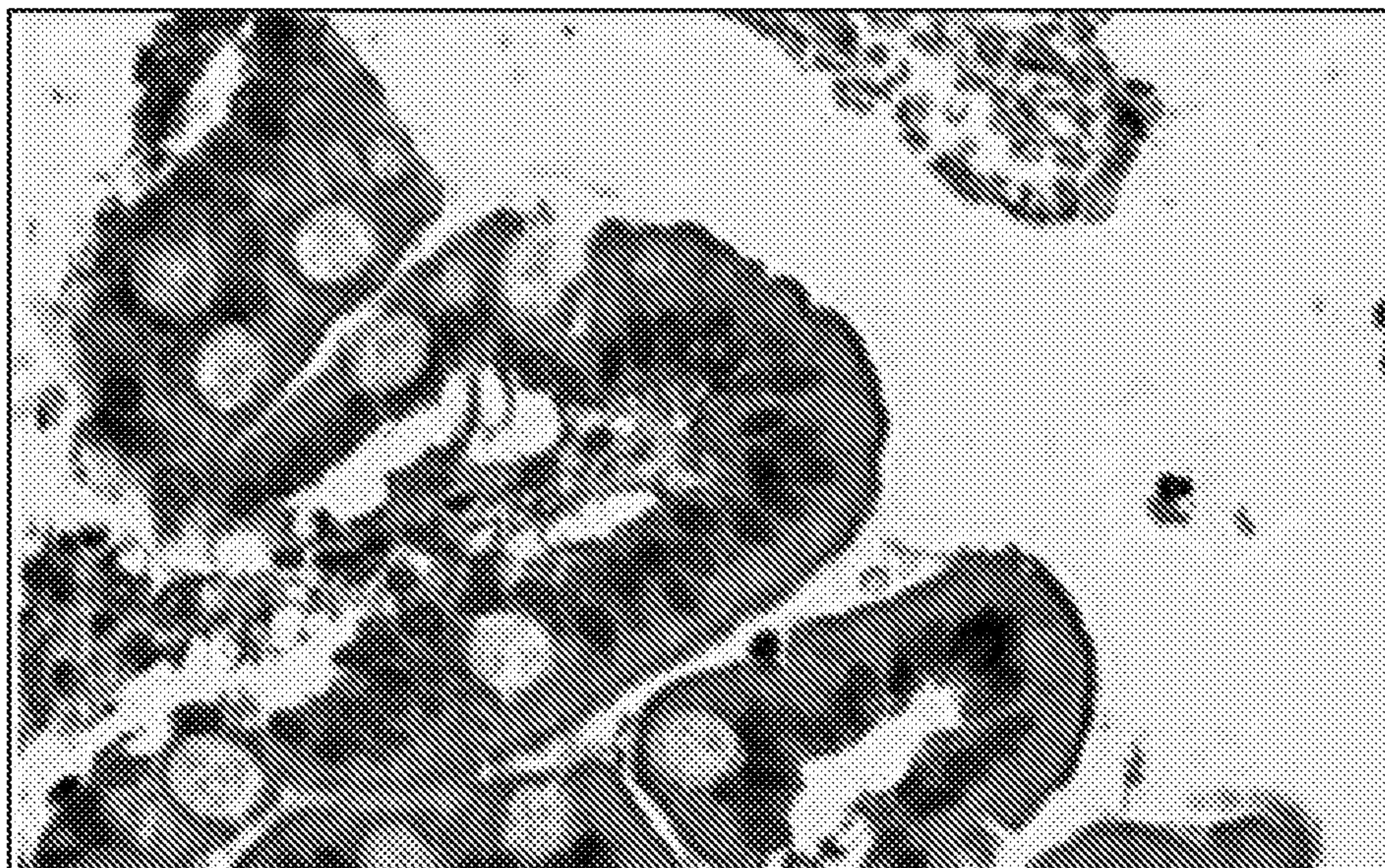


FIG. 28B

FIG. 28

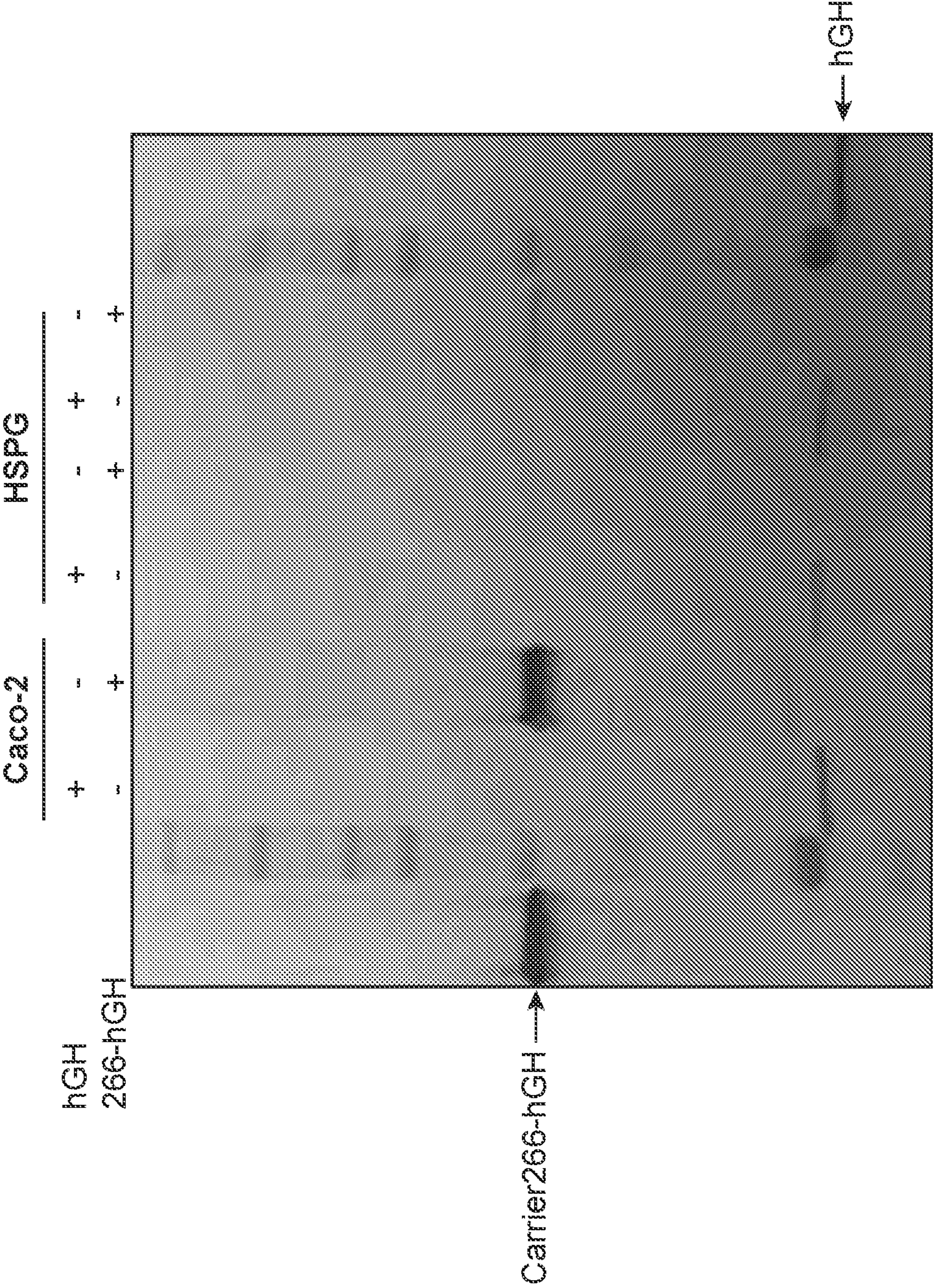


FIG. 29

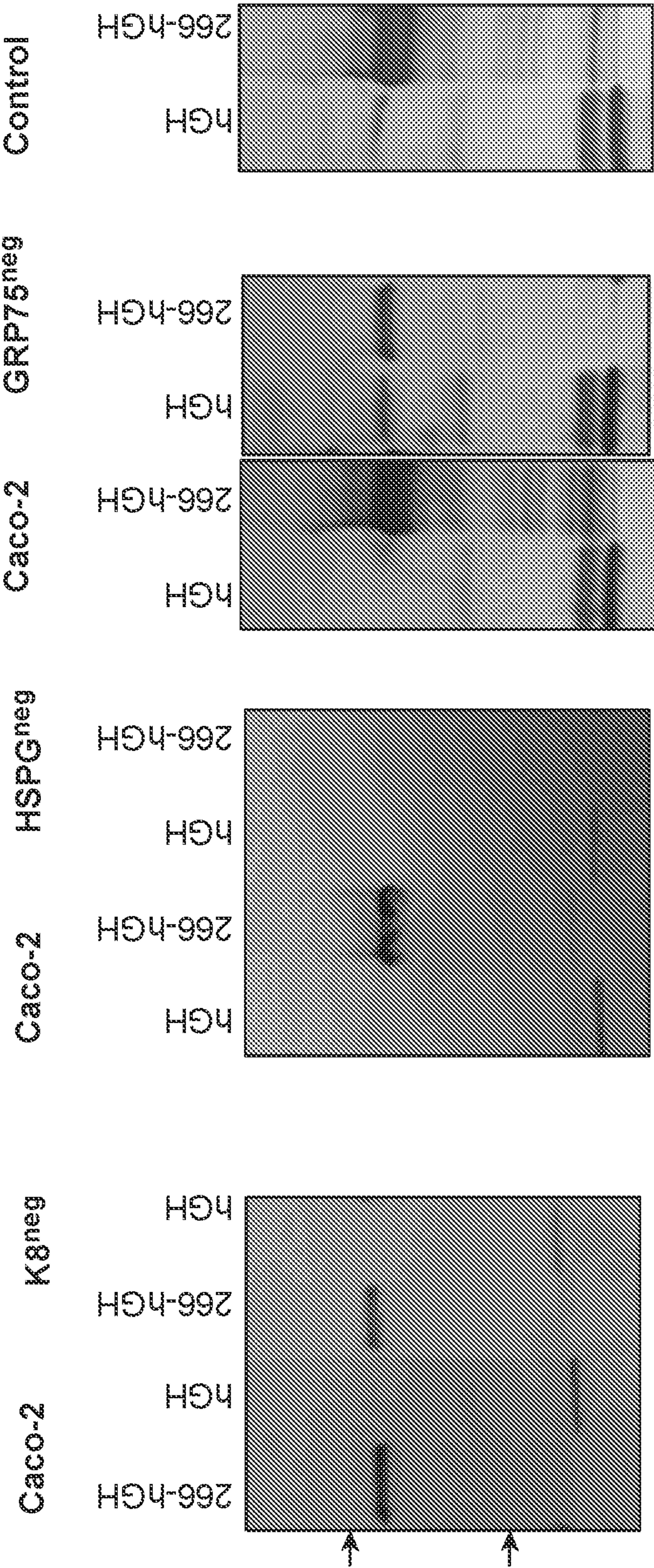


FIG. 30A

FIG. 30B

FIG. 30C

FIG. 30D

FIG. 30

Test Protein	Coated Protein	Test buffer	ka (1/Ms)	kd (1/s)	KD(M)	Affinity
Grp75	Chx-Biotin	1xTBS,pH5.5	1.64E+04	2.68E-04	1.63E-08	16.3 nM
Grp75	Chx-Biotin	1xTBS,pH6.5	3.77E+04	2.33E-05	6.17E-10	0.617 nM
Grp75	Chx-Biotin	1xTBS,pH7.5	2.87E+04	3.53E-04	1.23E-08	12.3 nM

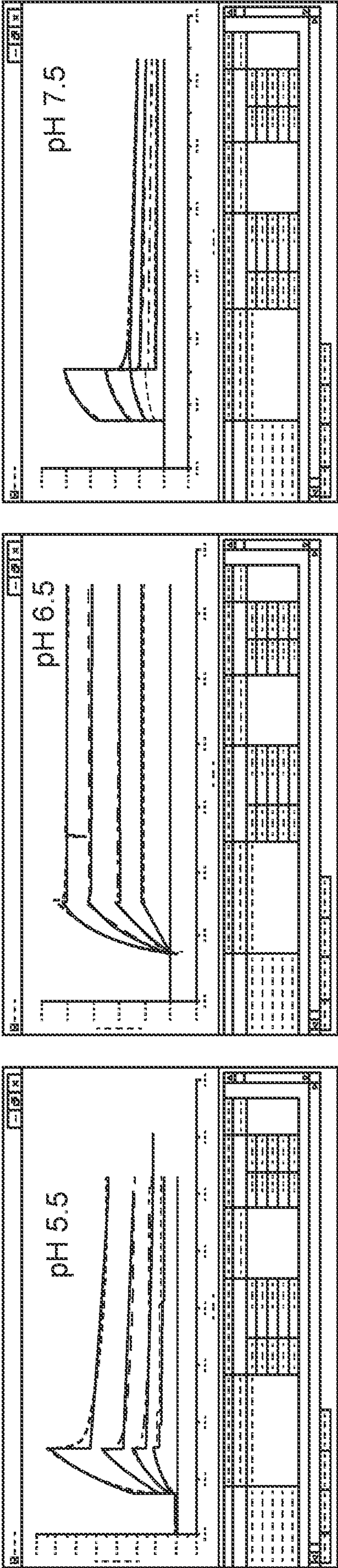


FIG. 31

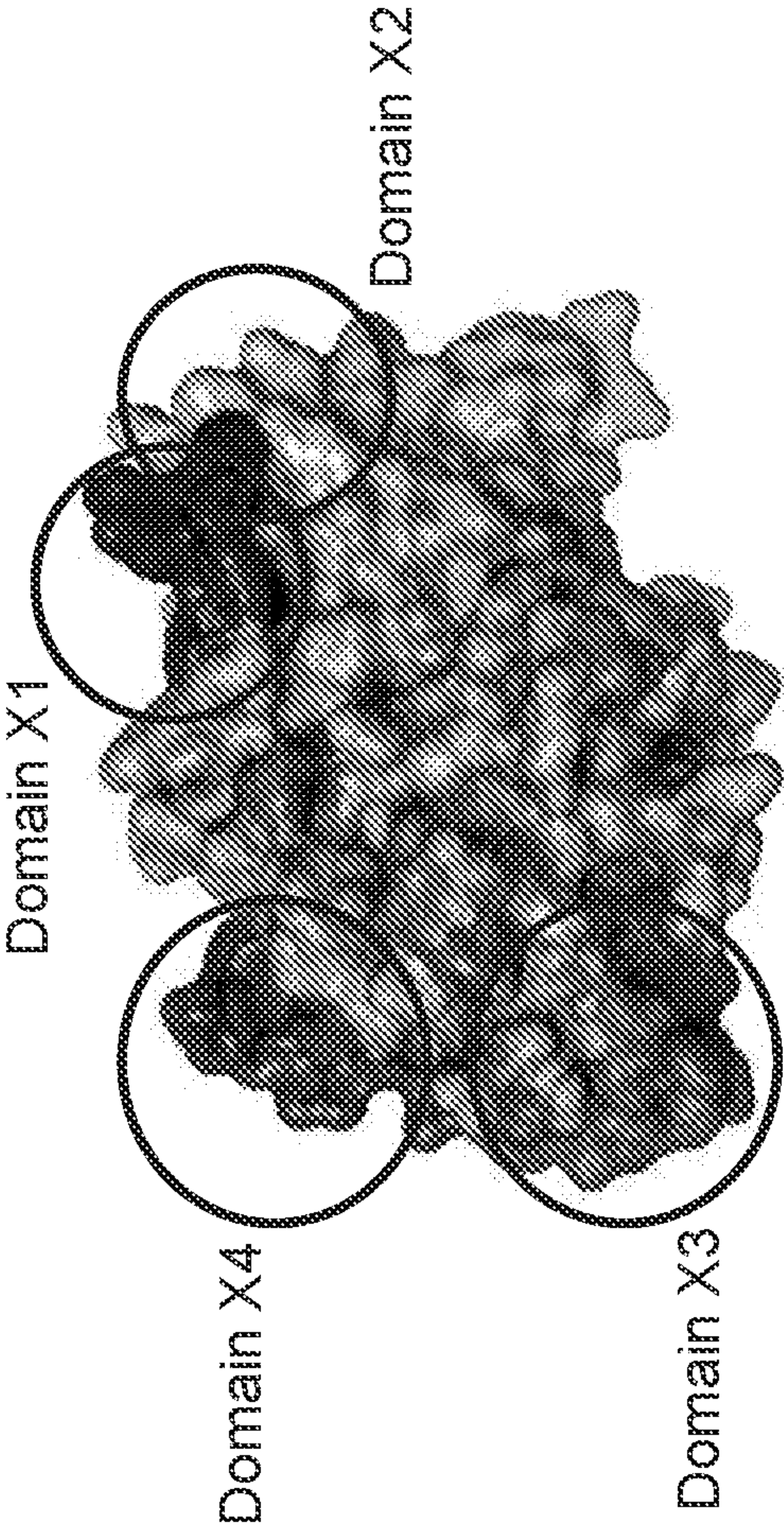


FIG. 32B

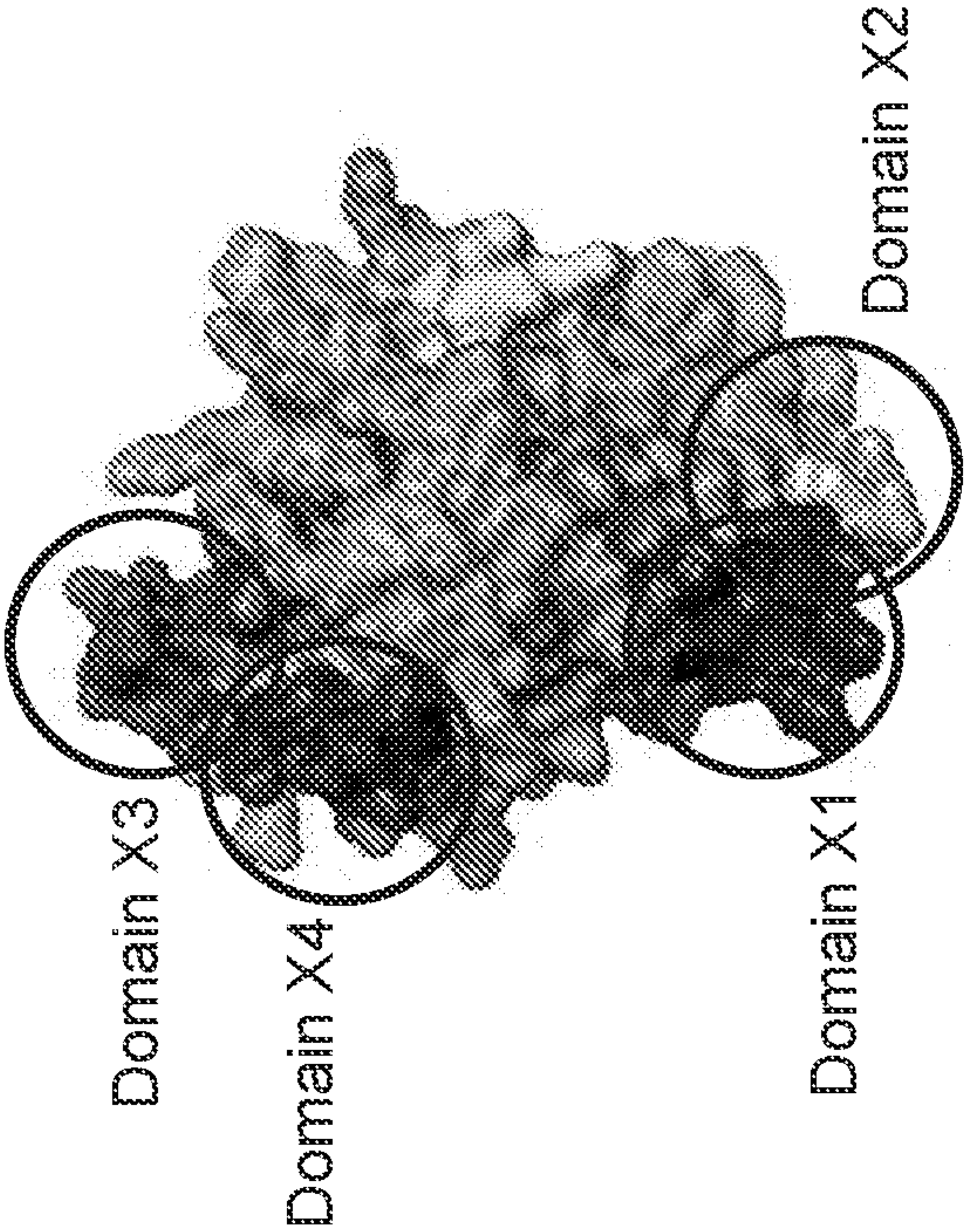


FIG. 32D

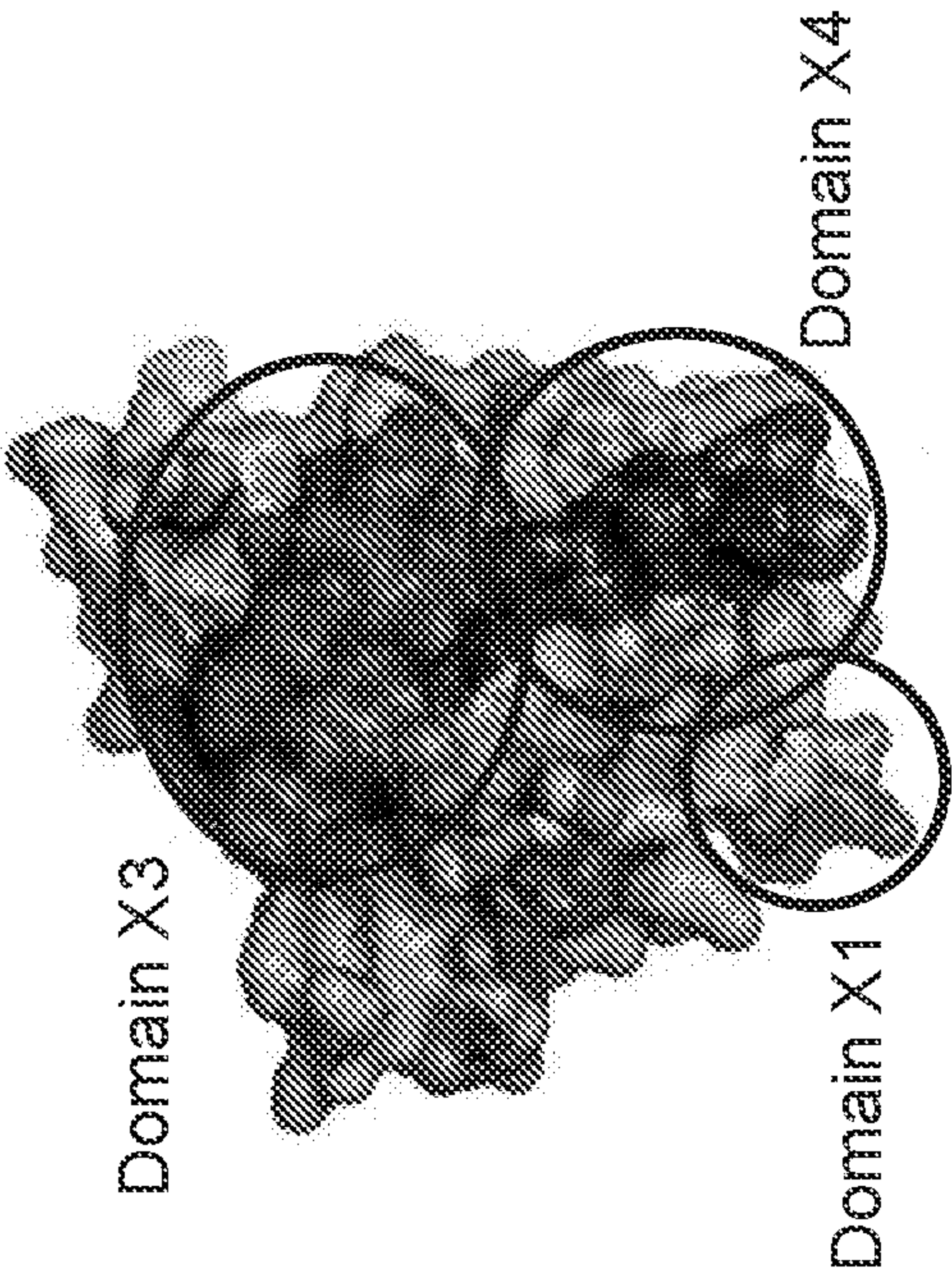


FIG. 32A

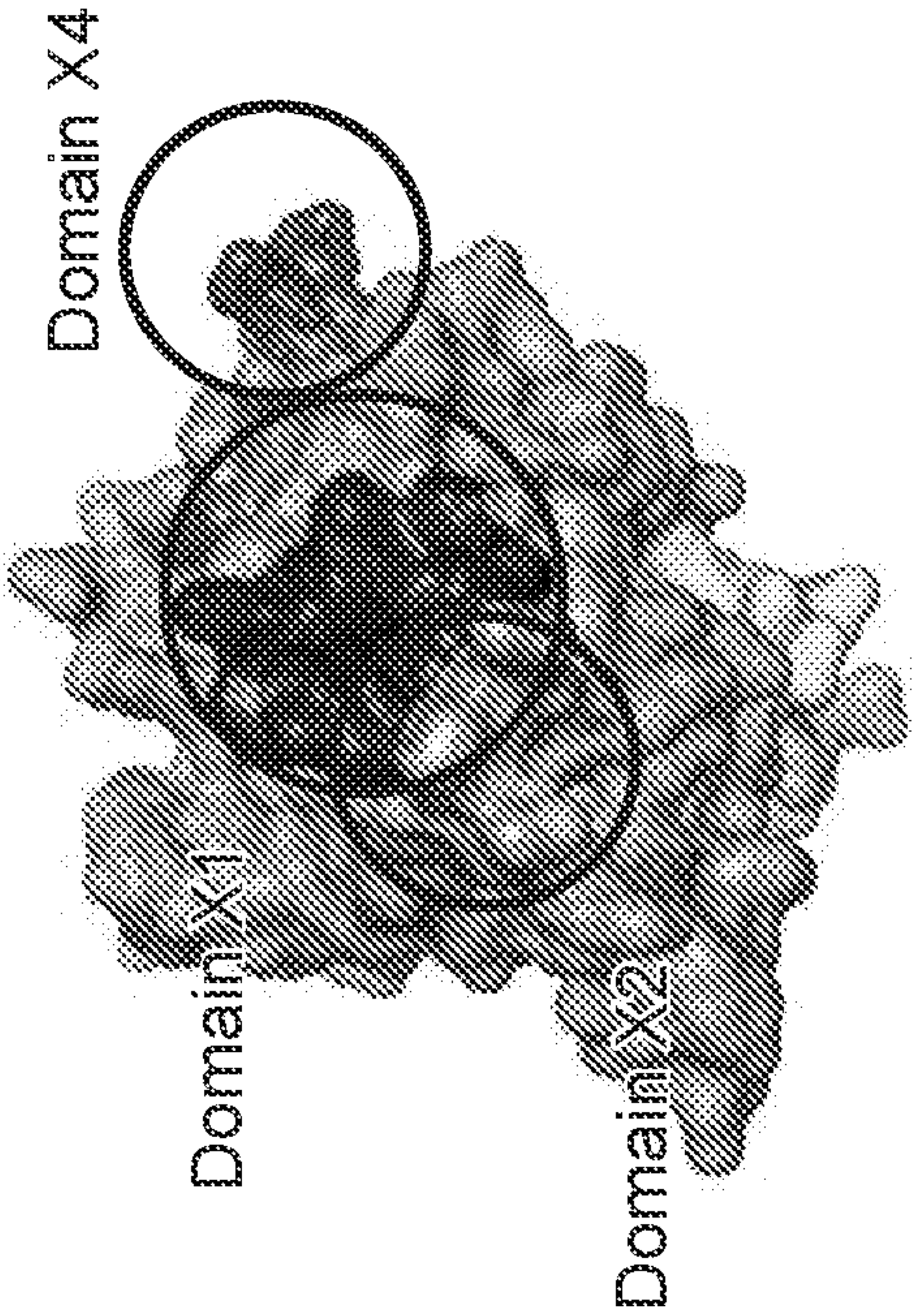


FIG. 32C

FIG. 32