TARTU ÜLIKOOL
1632

High-molecular-mass hyaluronan mediates the cancer resistance of the naked mole rat

Tian X, Azpurua J, Hine C, Vaidya A, Myakishev-Rempel M, Ablaeva J, Mao Z, Nevo E, Gorbunova V, Seluanov A.

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Bioinformatics Journal Club

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Märt Roosaare

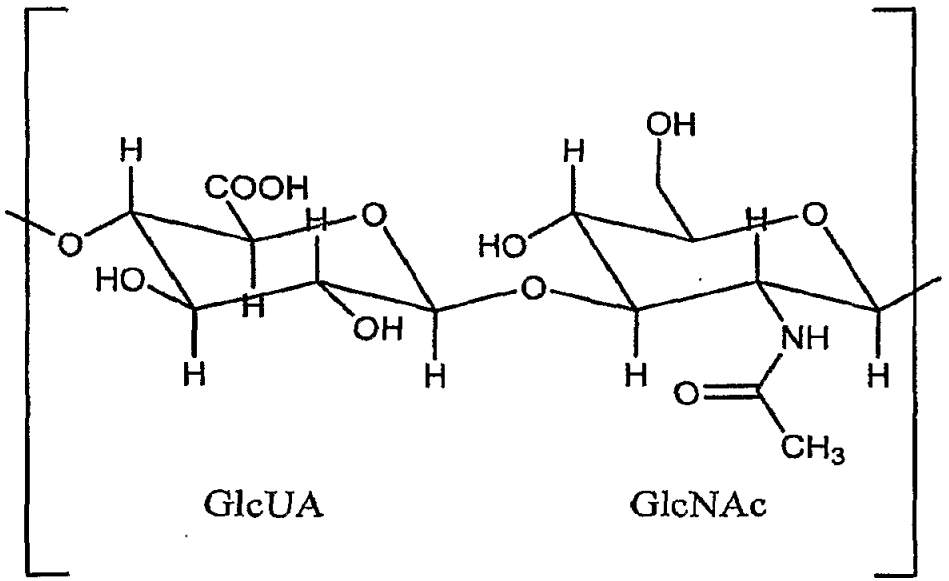
Naked mole rat (NMR)

- Subterranean, East Africa
- Colonies 60-80 individuals, large tunnel systems (3+ km)
- Skin does not feel pain (missing a neurotransmitter)
- Exceptionally long life span for its size (30+ years)
- Very resistant to cancer formation

Paljastuhnur
(*Heterocephalus glaber*)

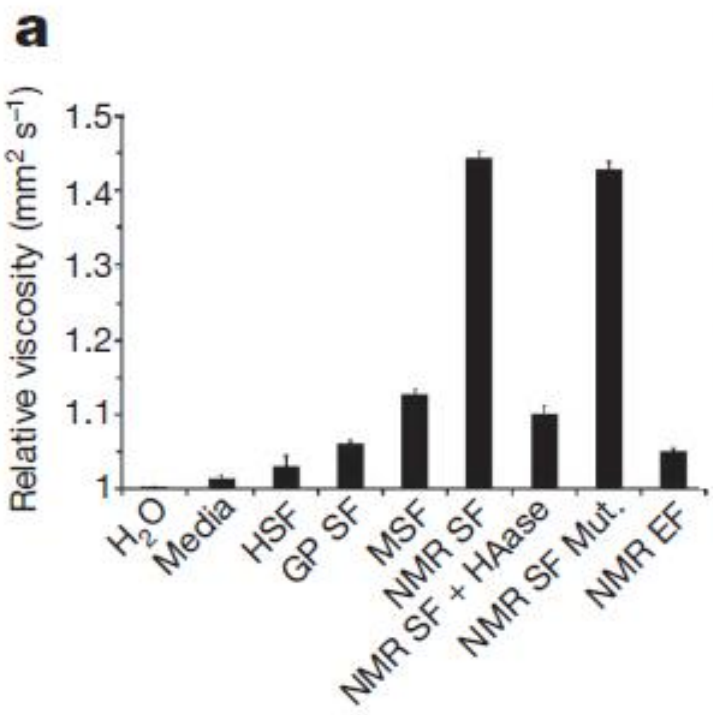
Hyaluronan (HA)

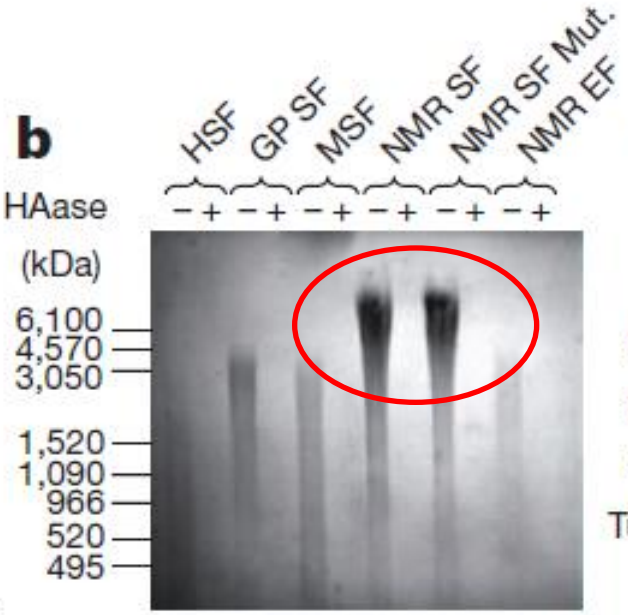
- Unbranched glucuronic acid/N-acetylglucosamine polymer
- One of the main components of extracellular matrix
- Very large molecular mass (human = 0,5 to 2, mouse 0,5 to 3 MDa)
- Biological effects depend on polymer length (high mass HA represses mitotic signalling and inflammation, low mass vice-versa)
- HAS1, HAS2, HAS3

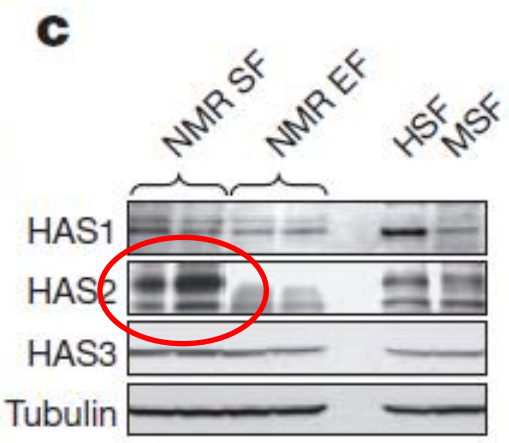


Accidental discovery?

- When culturing multiple NMR cell lines, some became very viscous after couple of days. Reason - high molecular mass HA synthesis

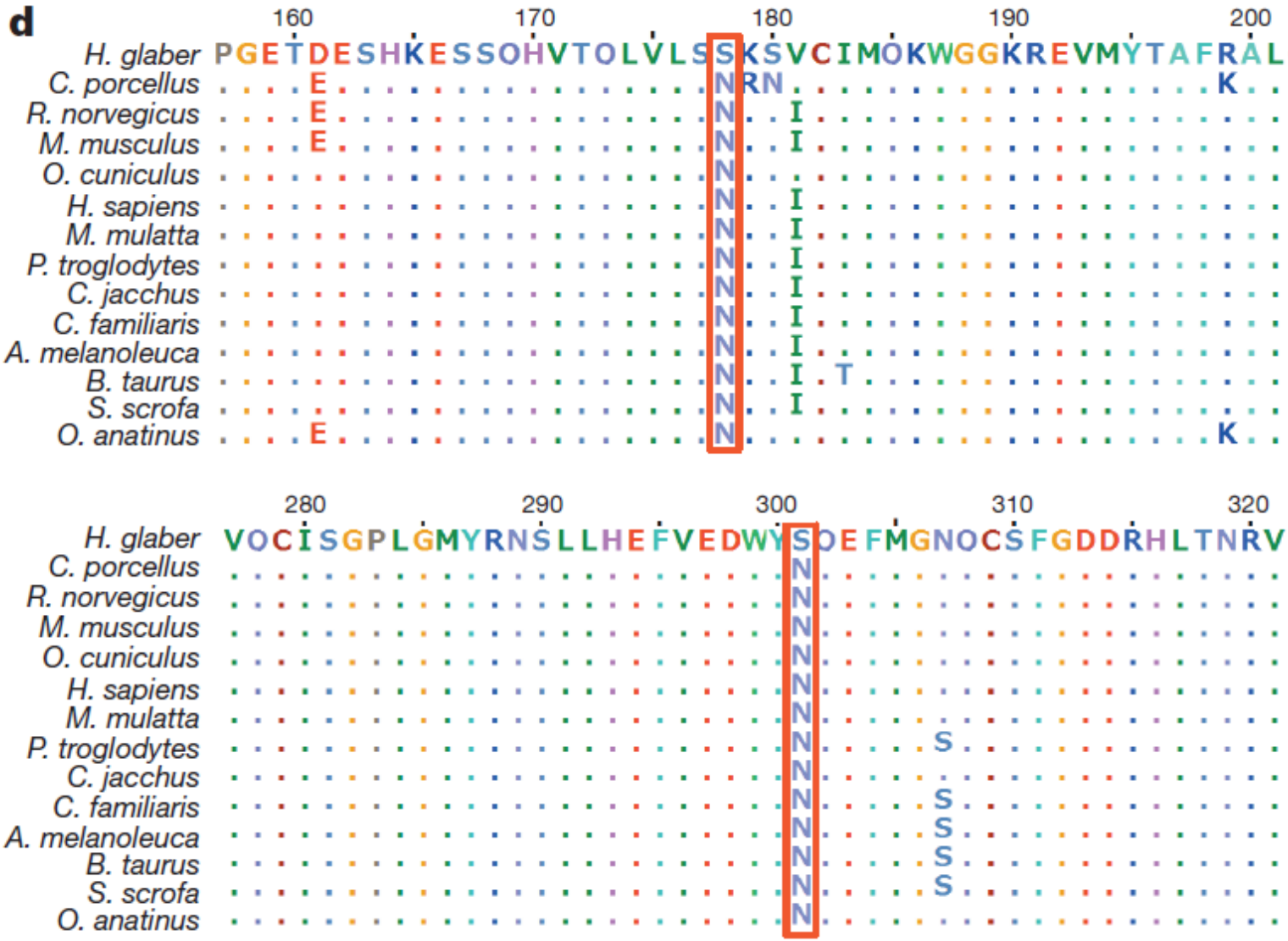




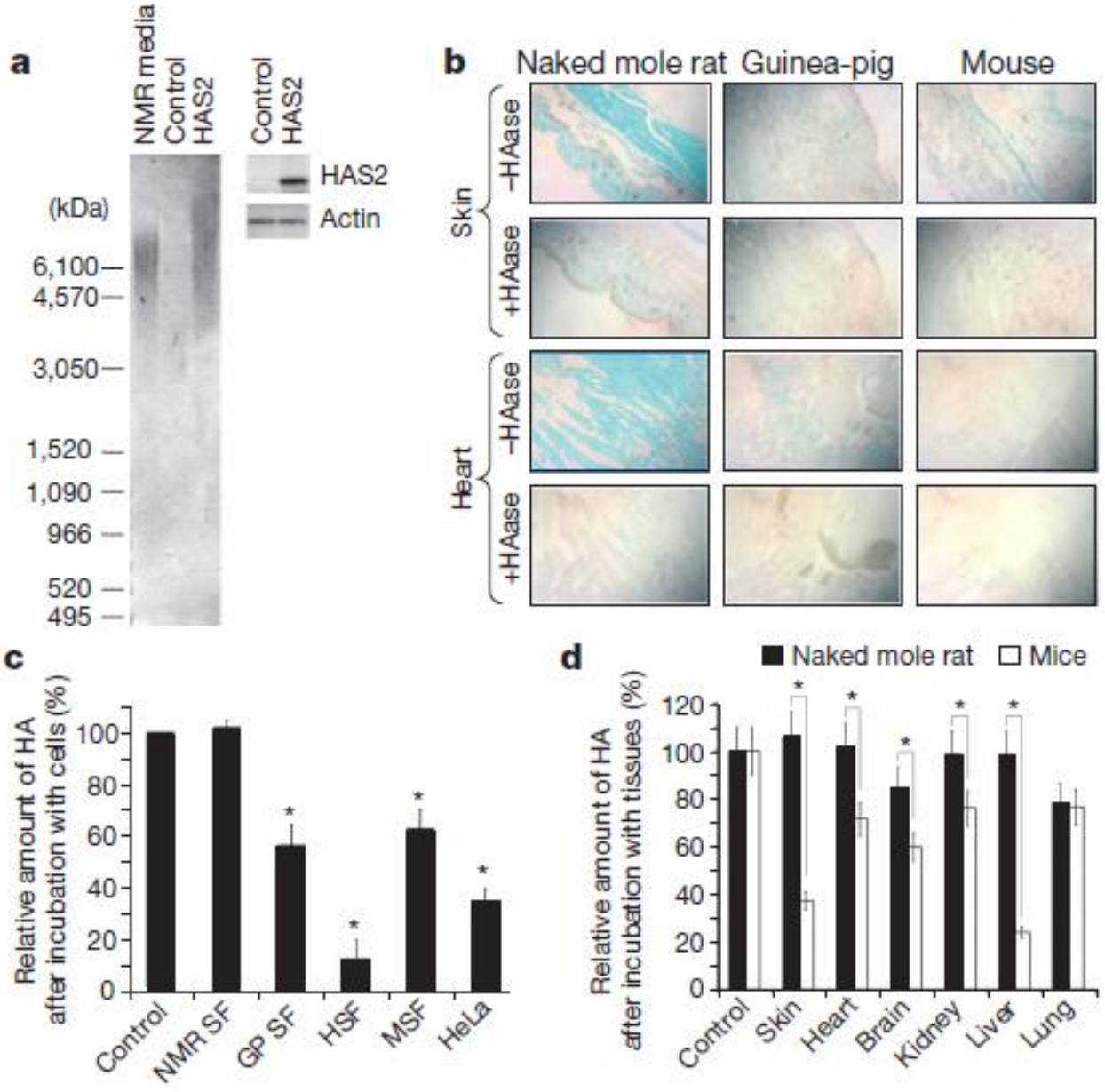


HAS 2

-
-
-


d
160
170
180
190
200
H. glaber PGETDESHKESSOHVTOLVLSSKSVCIMOKWGGKREVMYTAFRAL
C. porcellus
R. norvegicus
M. musculus
O. cuniculus
H. sapiens
M. mulatta
P. troglodytes
C. jacchus
C. familiaris
A. melanoleuca
B. taurus
S. scrofa
O. anatinus
280
290
300
310
320
H. glaber VOCISGPLGMYRNSLLHEFVEDWYSOEFMGNOCSFGDDRHLTNRV
C. porcellus
R. norvegicus
M. musculus
O. cuniculus
H. sapiens
M. mulatta
P. troglodytes
C. jacchus
C. familiaris
A. melanoleuca
B. taurus
S. scrofa
O. anatinus

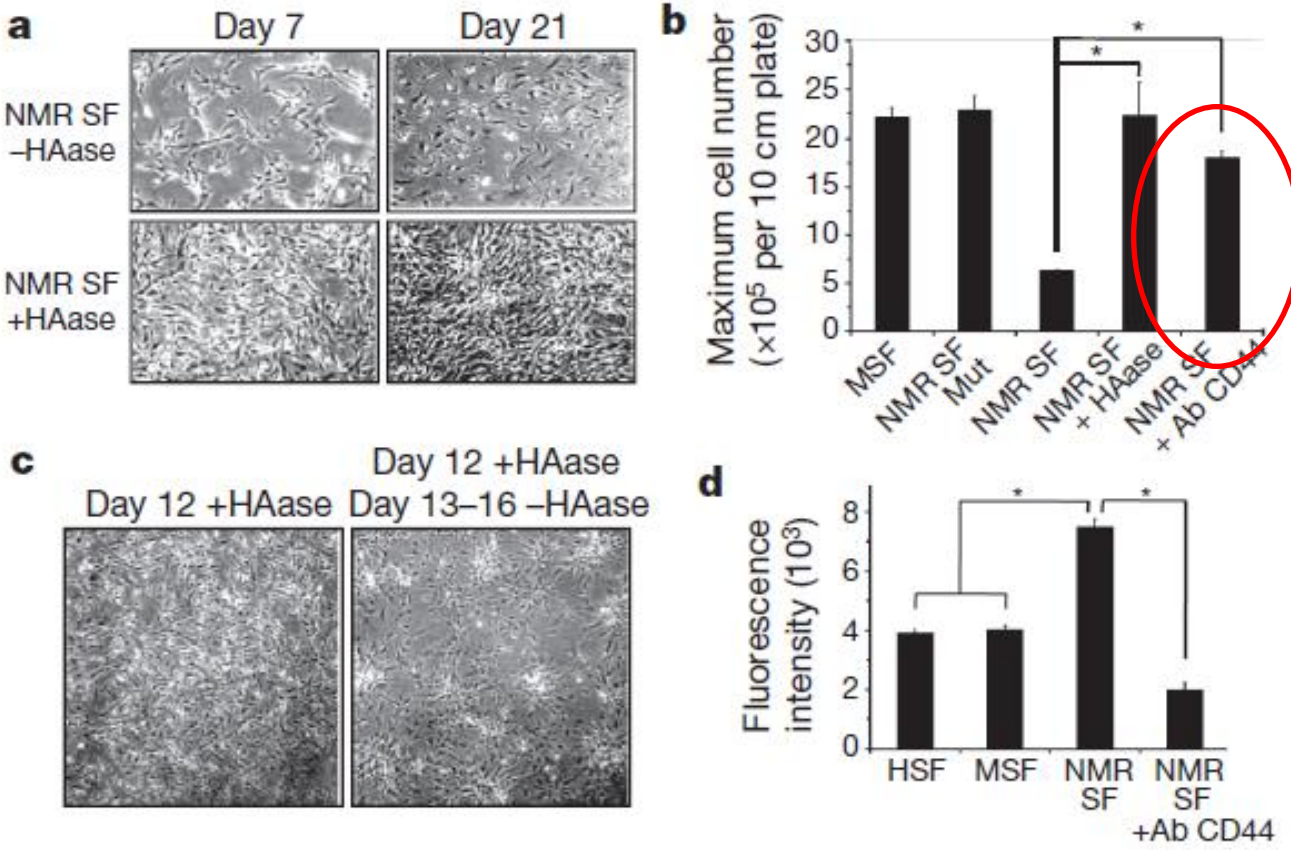
- A - NMR HAS2 overexpressed in HEK293 cells
- B - blue stained tissues (staining specific to HA)
- C - NMR skin fibroblasts have low HA-ase activity (incubated in media containing HMM-HA)
- D - NMR tissues have also low HA-ase activity



Contact inhibition (CI)

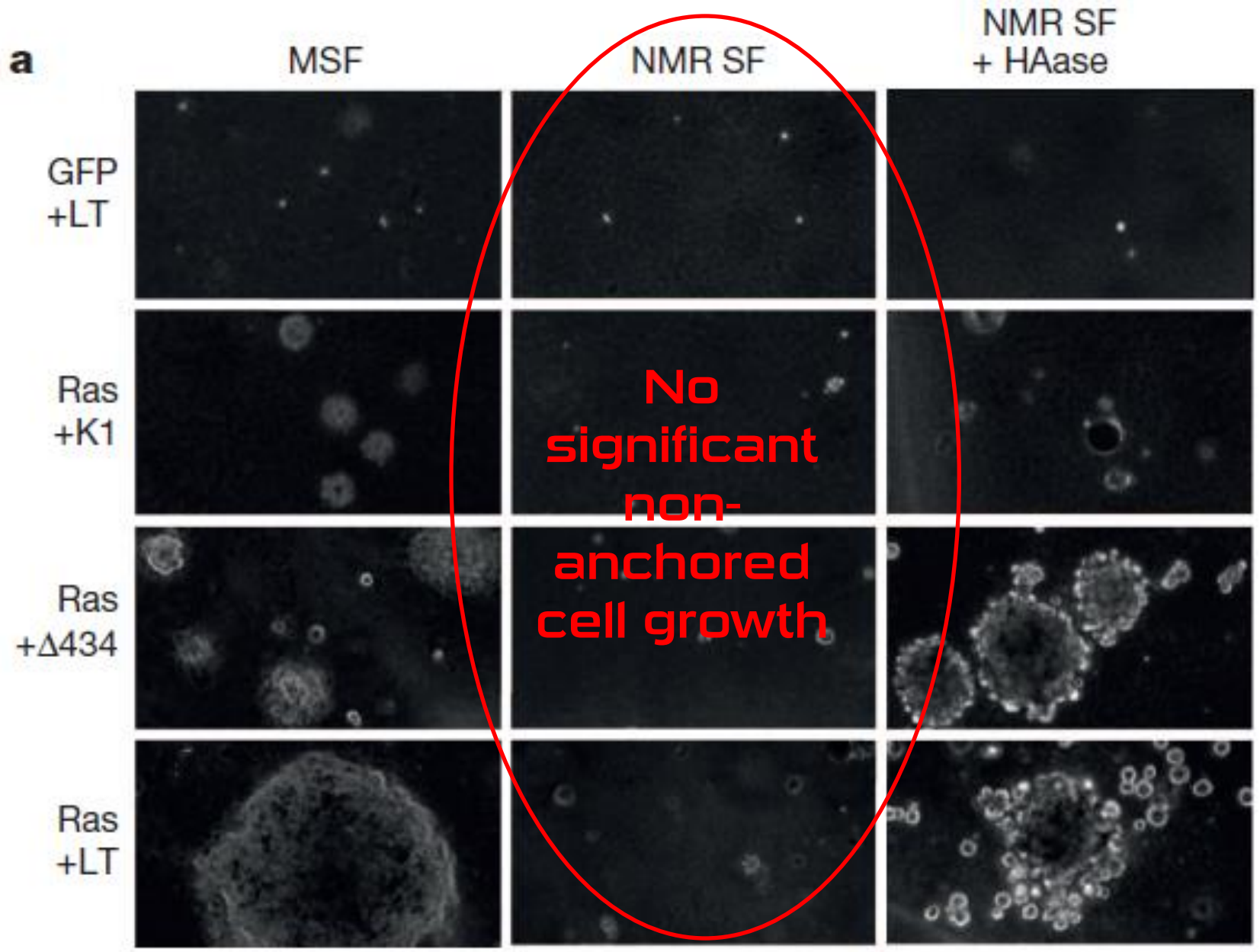
- Anti-cancer mechanism - cells stop growing when they encounter other cells/extracellular matrix
- NMR cells arrest at much lower densities

- B - HA signalling triggers CI, through CD44 receptor (B, CD44-blocking antibody)
- D - NMR skin fibroblasts show more affinity for HA



HA role in cancer resistance

- Anchorage-independent growth (cells growing without contact with each other/extracellular matrix) correlates with tumorigenicity
- Mouse and NMR skin fibroblasts
- Soft agar assay, transfection of different oncoproteins
  - Hras V12 (mutated GTPase, permanently active)
  - SV40 Large T Antigen (LT) - binds and disables p53 and pRb tumor suppressor proteins, whereas its mutants =>
  - LTK1 (K1) disables p53
  - Delta434 disables pRb


a
MSF
NMR SF
NMR SF
+ HAase
GFP
+LT
Ras
+K1
Ras
+Δ434
Ras
+LT
No significant non-anchored cell growth

Mouse xenografts

- Positive control – mouse skin fibroblasts transfected with SV40 LT and Ras V12
- Xenografts with transfected NMR cells (LT + V12)
  - Active HAS2
  - HYAL2 (HA-degrading) cDNA
  - RNA-induced silencing of HAS2

Conclusions

- Cancer resistance is derived from both lower HA-ase activity and very high molecular mass HA (HMM-HA)
- HMM-HA could have evolved as an adaption to subterranean lifestyle - gives flexible skin (a different subterranean rodent - blind mole rat - also secretes HMM-HA)
- Naked mole rat HMM-HA or suppressing HYAL2/targeting HA-signalling pathway could potentially be used for cancer prevention