Neural Basis of Vocal Communication in Songbirds

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Outline of Talk

Part I:

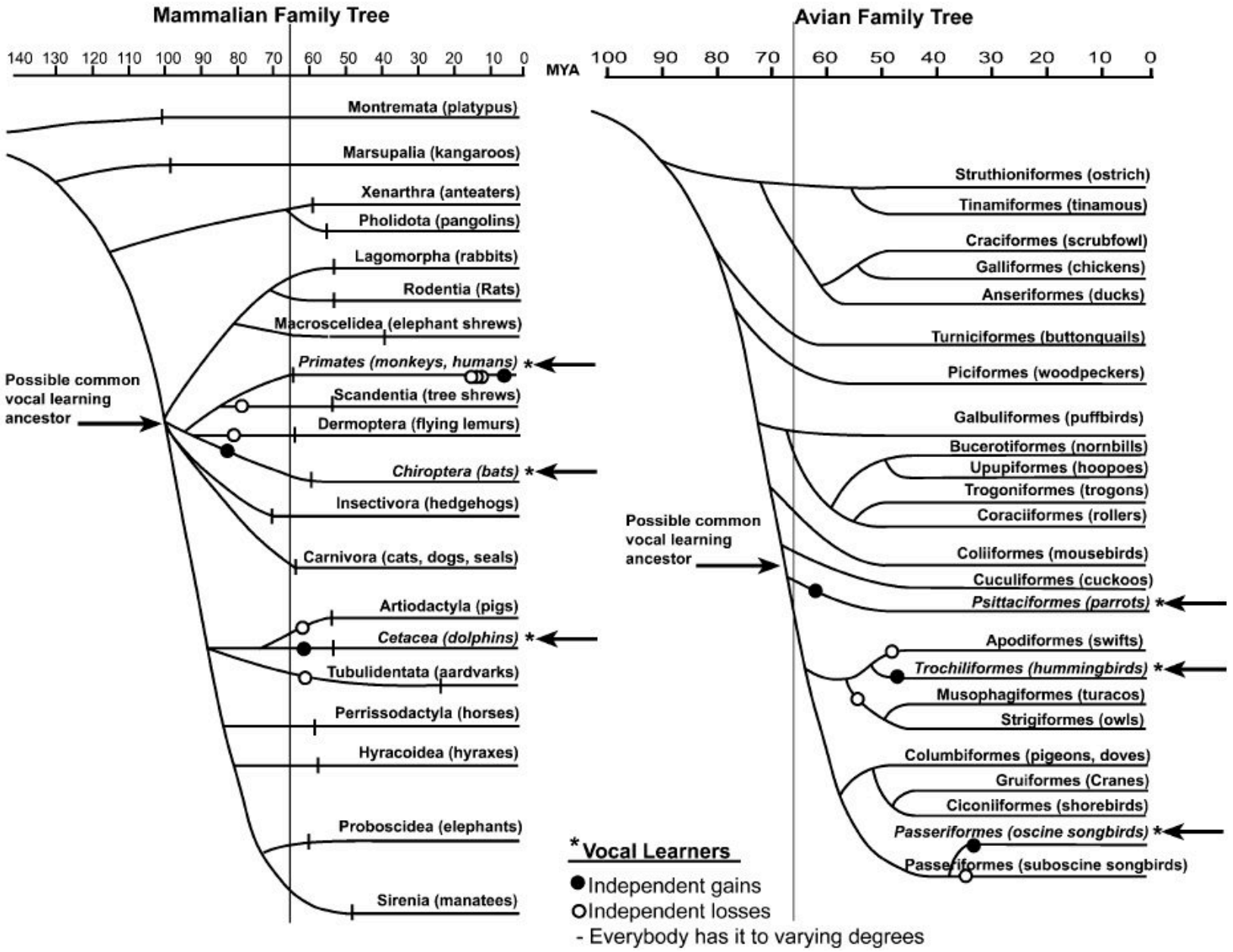
- Why songbirds?
- Vocal learning: auditory processing and auditory memories.
    - The songbird NCM:
    - Song selectivity, auditory discrimination and perception.

Part II:

- Proteomics screening: song-regulated proteins.

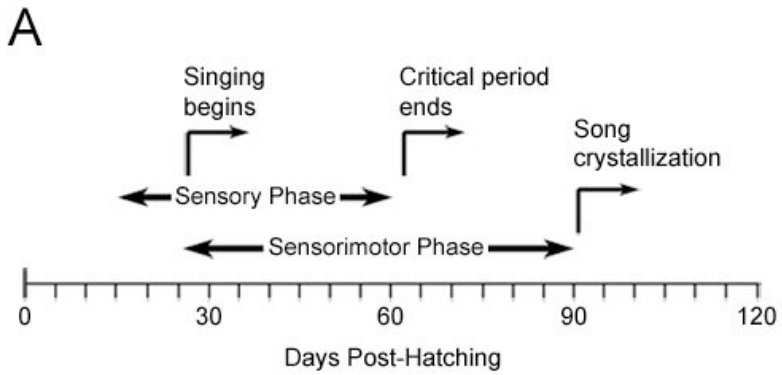
Conclusions and Acknowledgements

Songbirds As a Model for Vocal Learning and Auditory Processing

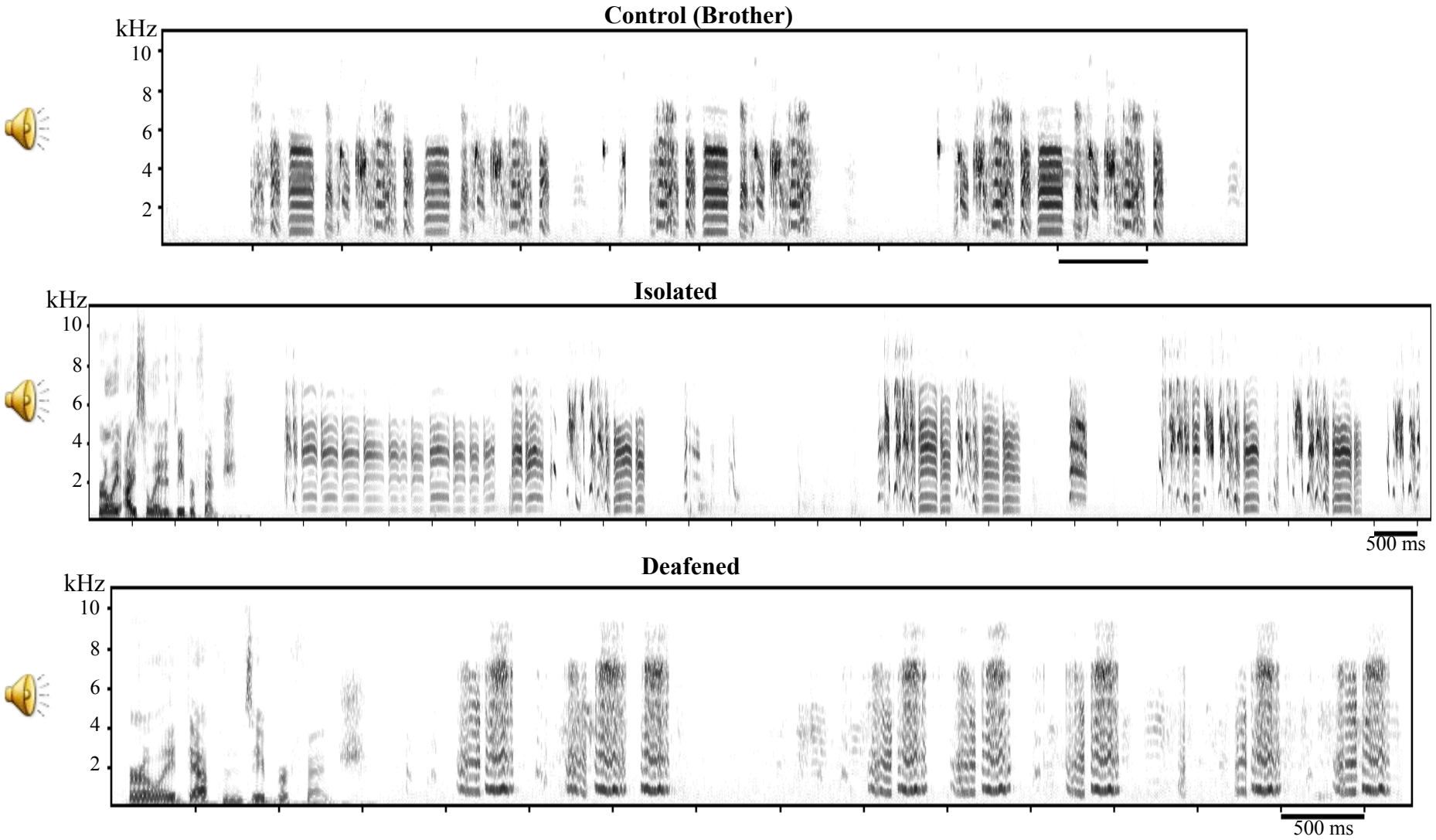


Songbirds As a Model for Vocal Learning and Auditory Processing

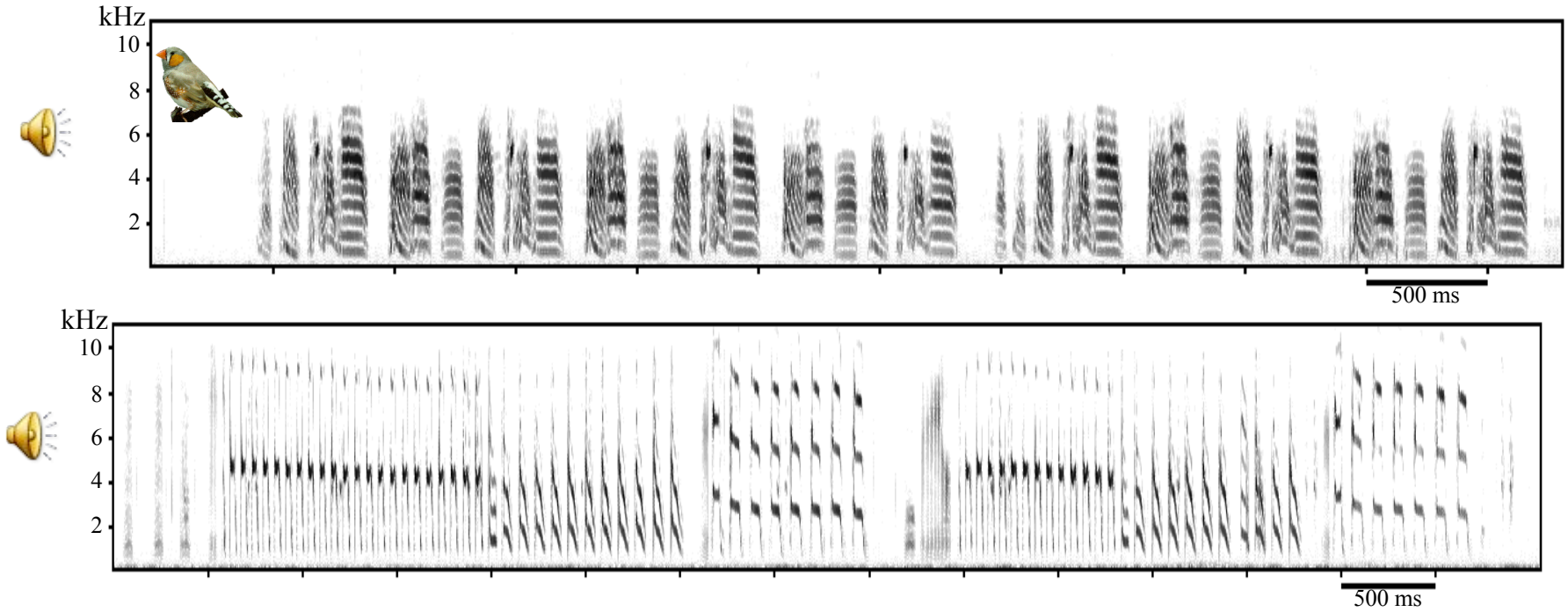
Zebra Finches (*Taeniopygia guttata*)



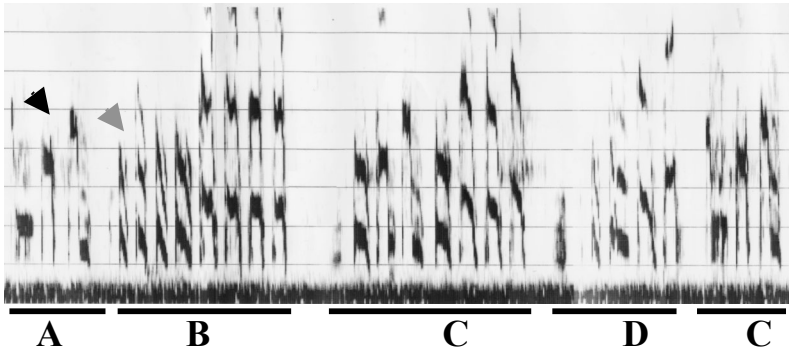
Vocal Learning and Maintenance of Learned Songs Requires Auditory Feedback



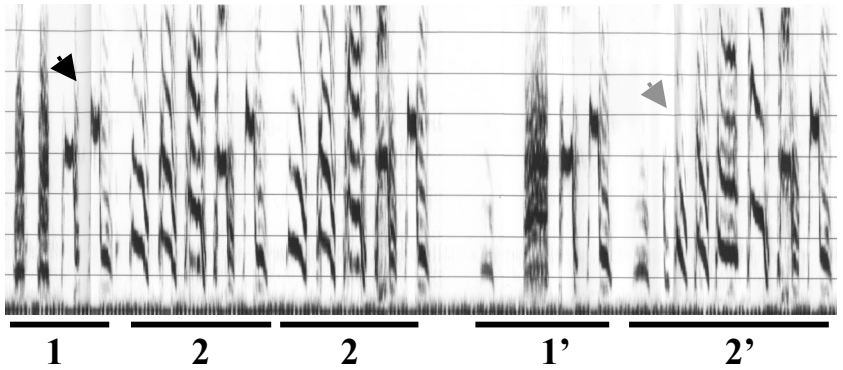
Cross Fostering Illustrates Vocal Learning



Bengalese Finch Tutor



Zebra Finch Pupil



Syntax and Phonology is Copied

Songbirds As a Model for Vocal Learning and Auditory Processing

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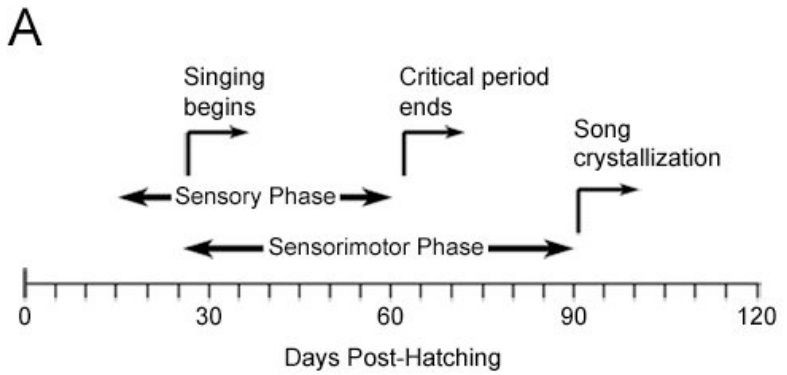
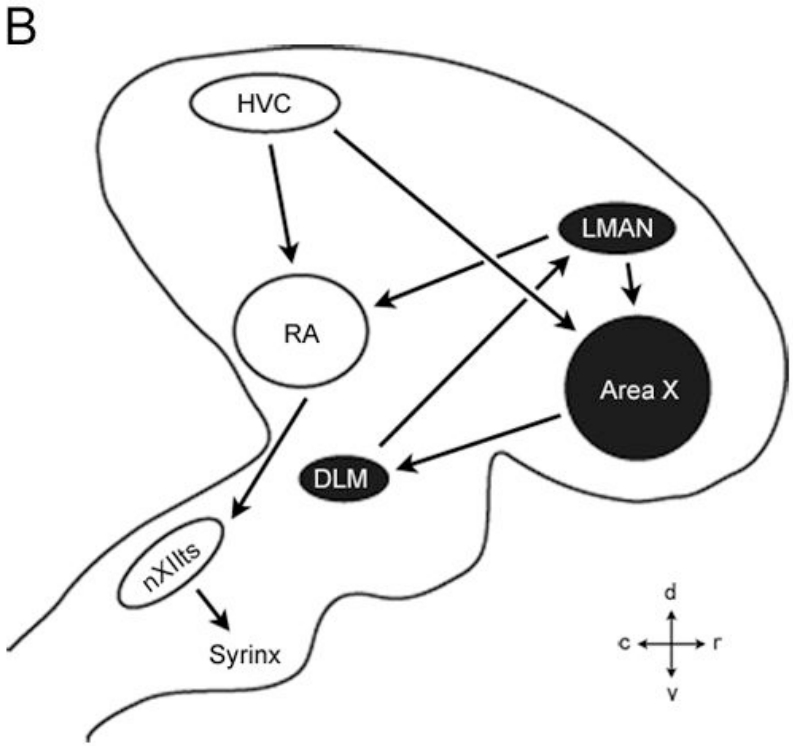




Sensory, sensorimotor & motor processes

<u>Auditory Processing is Required for:</u>

-Vocal Learning

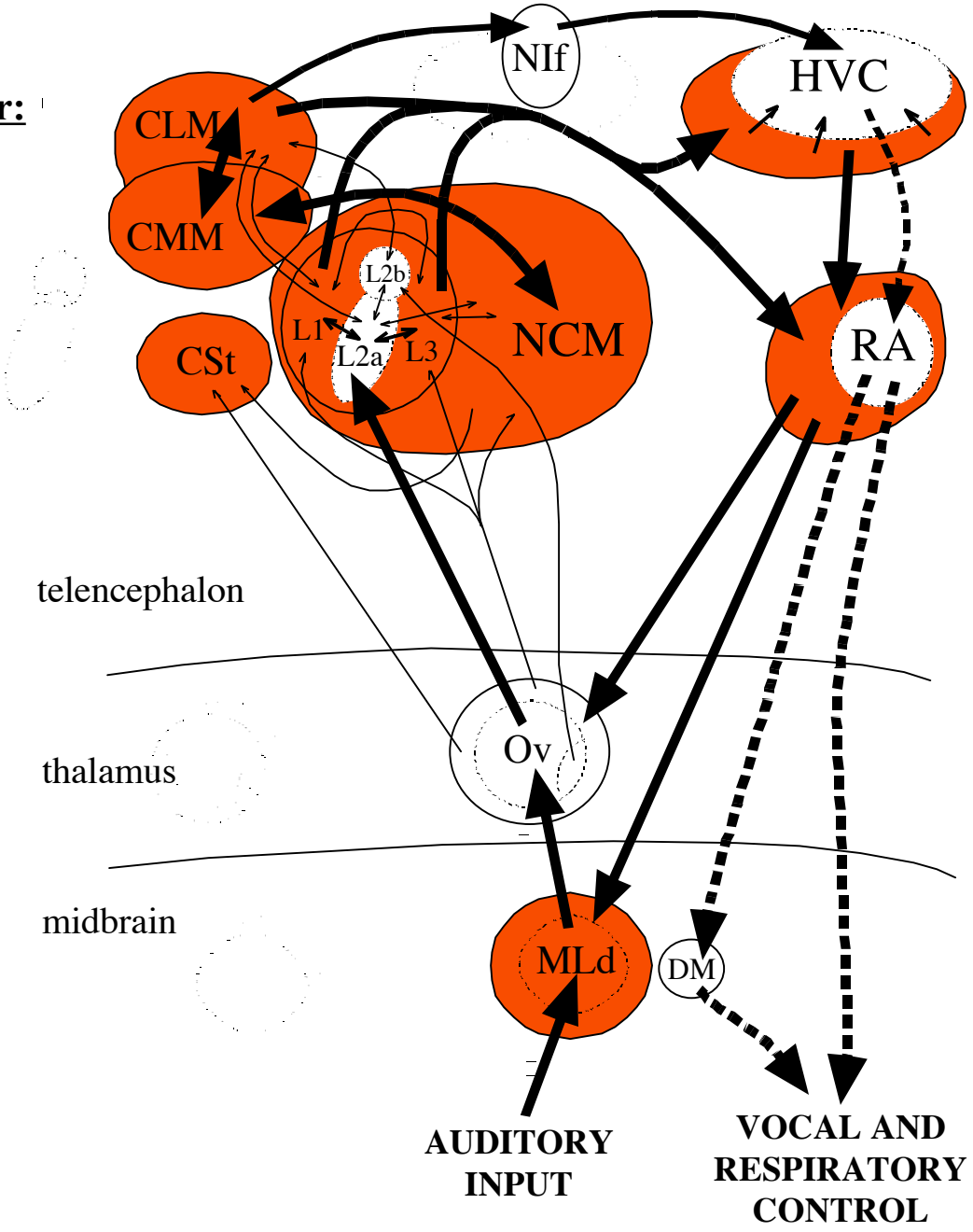
-Song Maintenance

-Perceptual Processing of Songs

-mate selection

-individual recognition

-territorial defense



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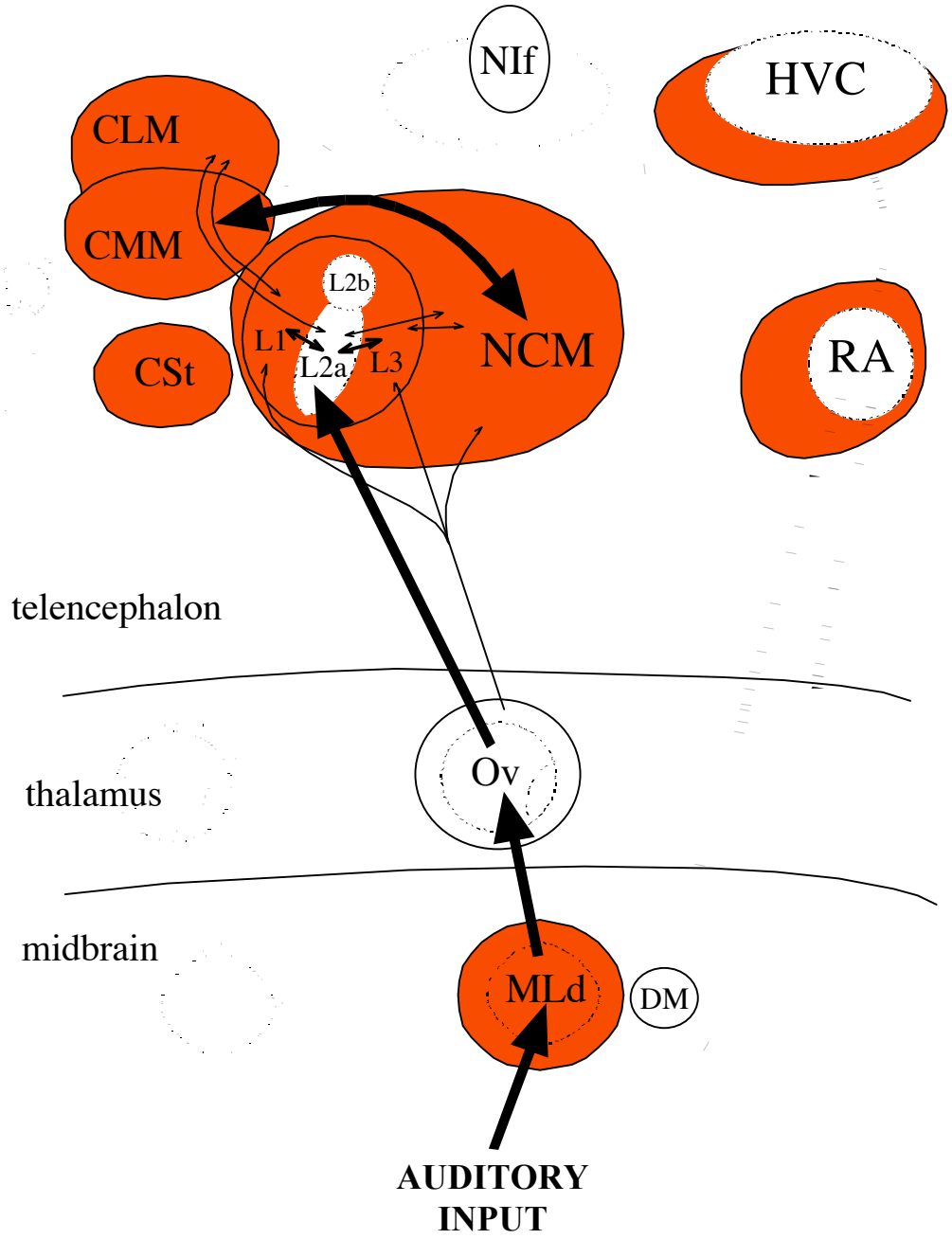


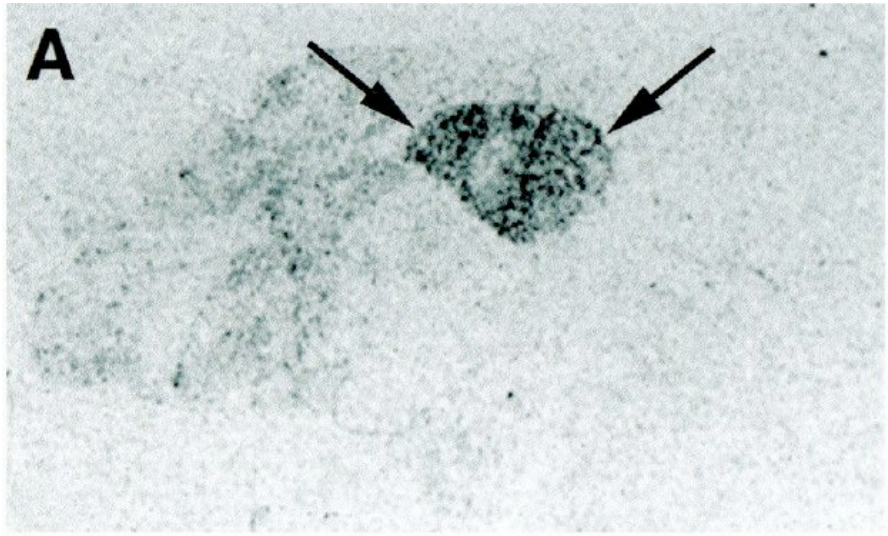
<u>NCM</u>:

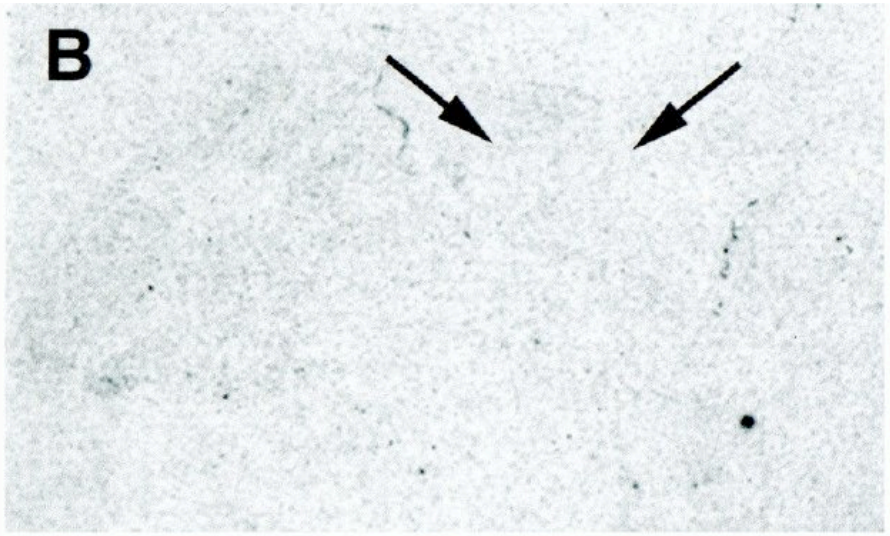
-Greater selectivity for complex stimuli

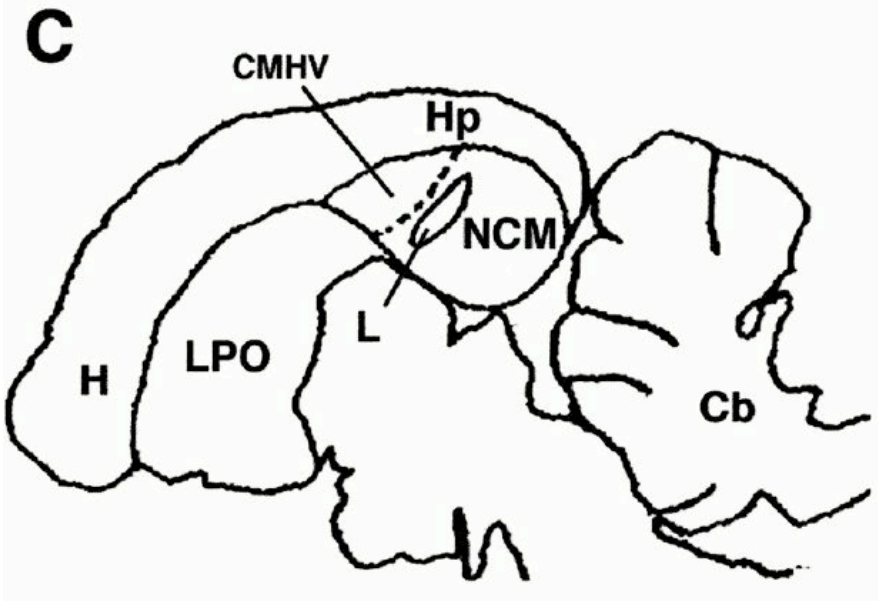
-Preference for conspecific songs

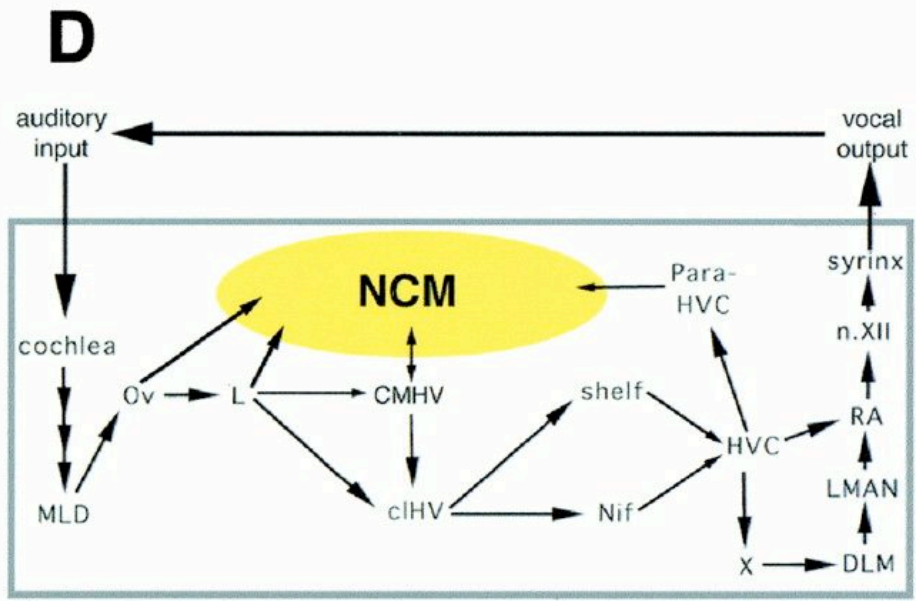
-Appears to be involved in discrimination of birdsong and maybe the formation of auditory memories.

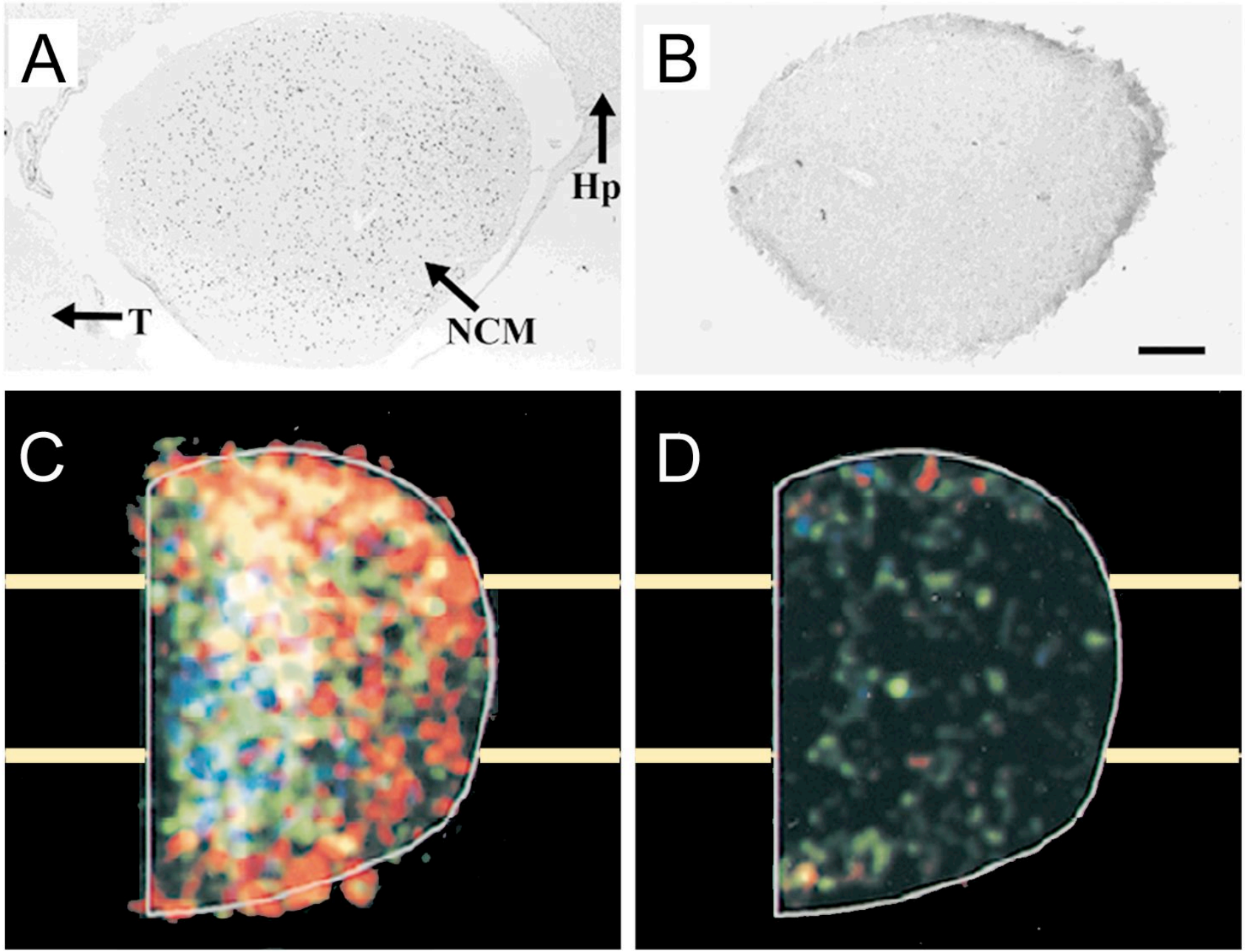
zenk


A

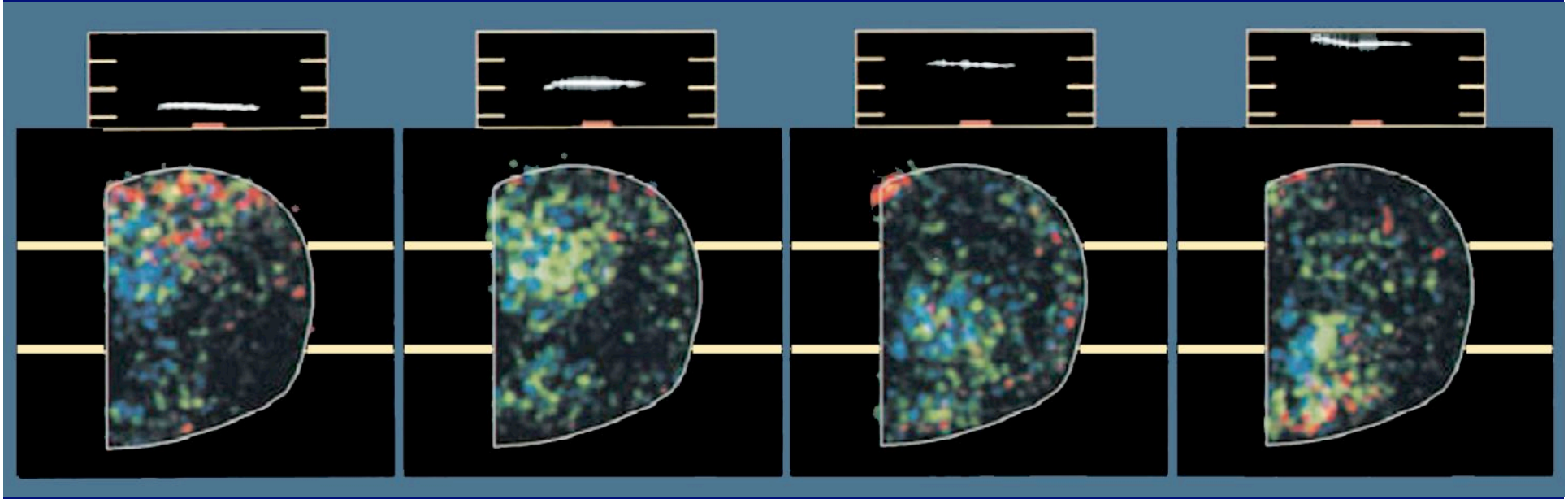

B


C
CMHV
Hp
NCM
L
H
LPO
Cb

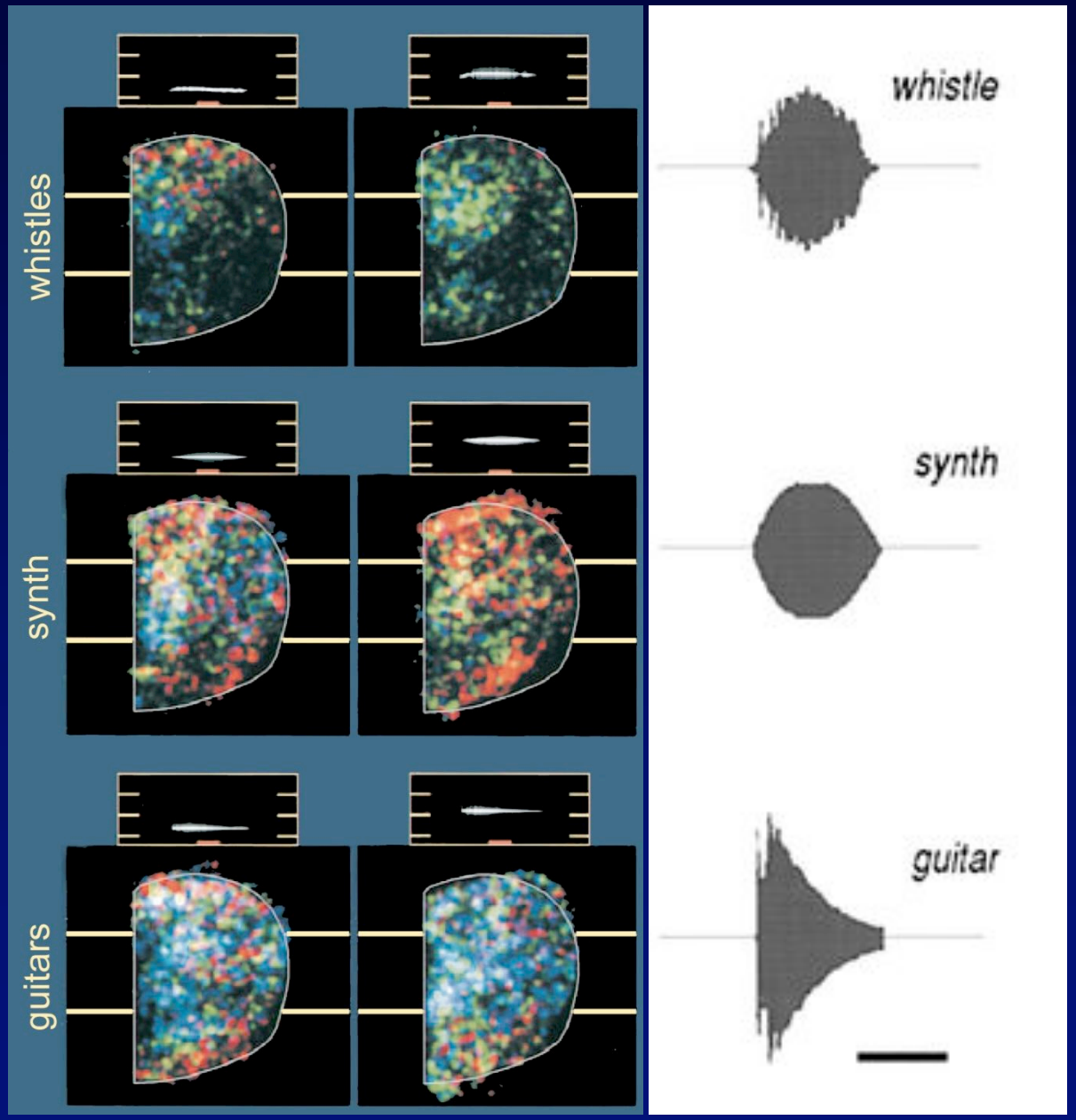

D
auditory input
vocal output
NCM
Para-HVC
syrinx
n.XII
cochlea
Ov
L
CMHV
shelf
HVC
RA
LMAN
MLD
clHV
Nif
X
DLM


A
Hp
T
NCM
B
C
D
Song-Stimulated
Silence

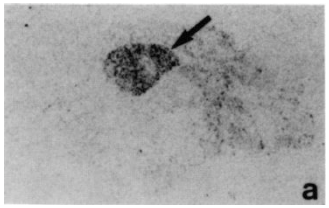
Frequency-Dependent Organization in NCM as Revealed by ZENK Expression



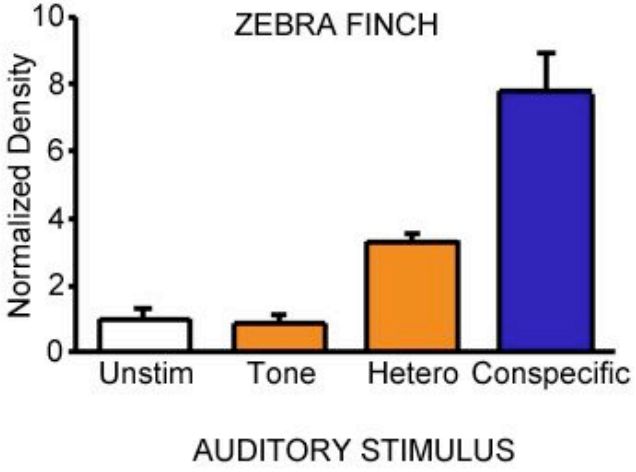
Discrimination of Natural Stimuli in NCM

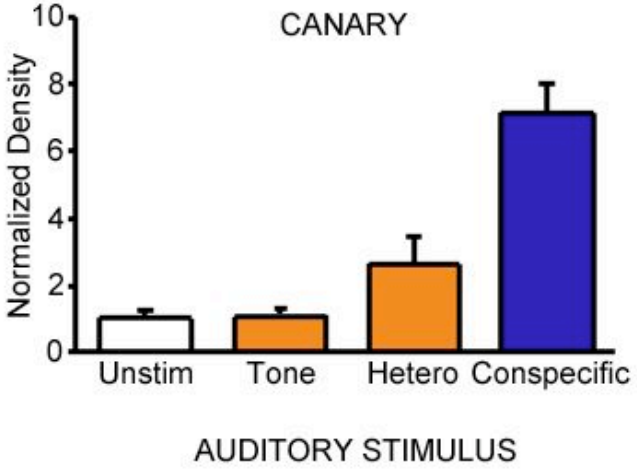


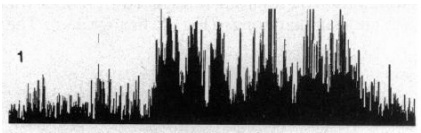
NCM Neurons are Selective to Conspecific Songs and Calls



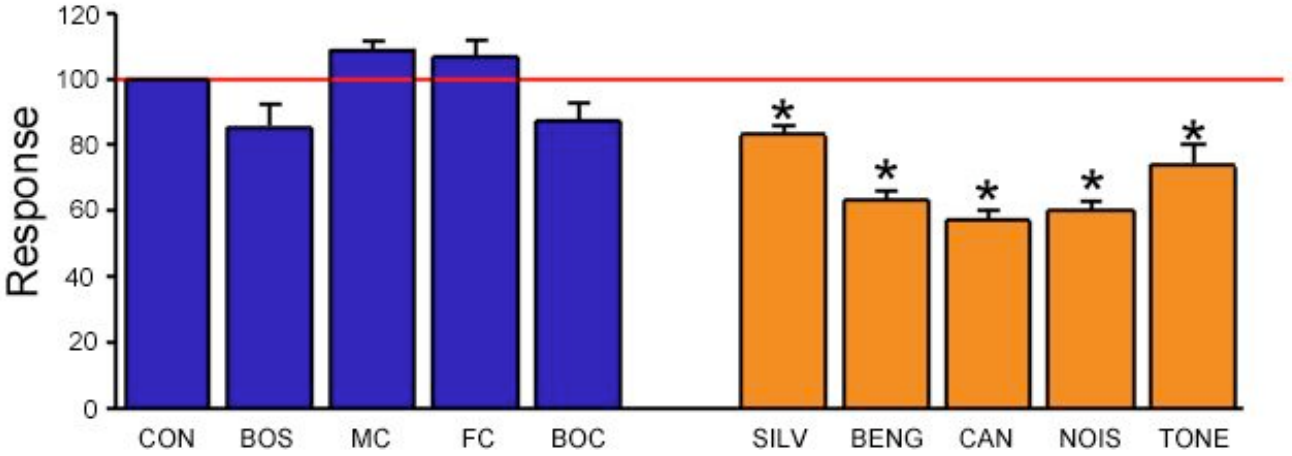
***zenk* expression**



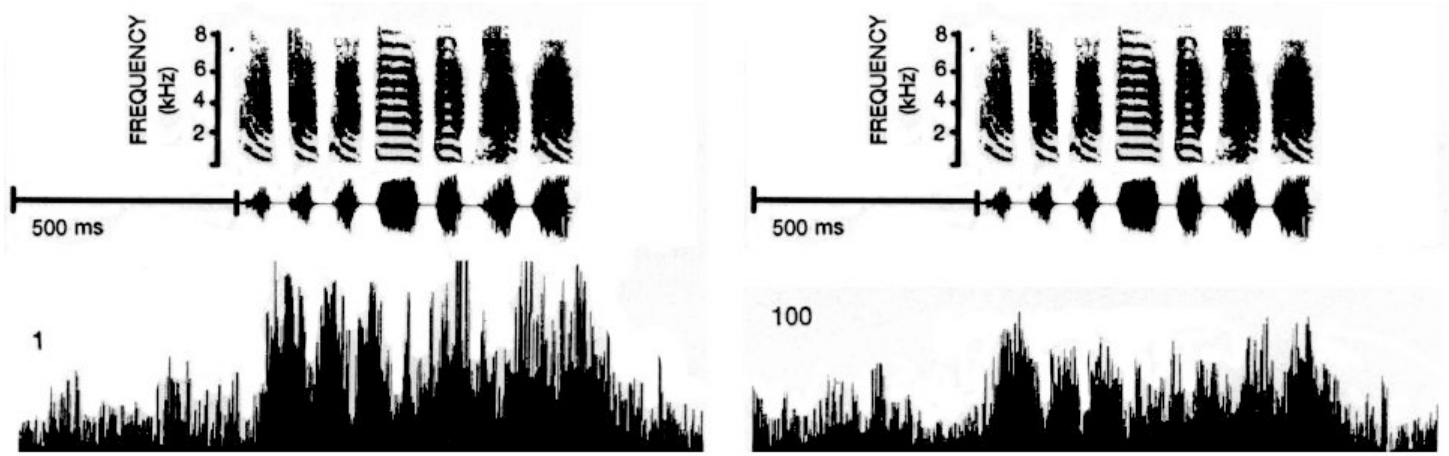


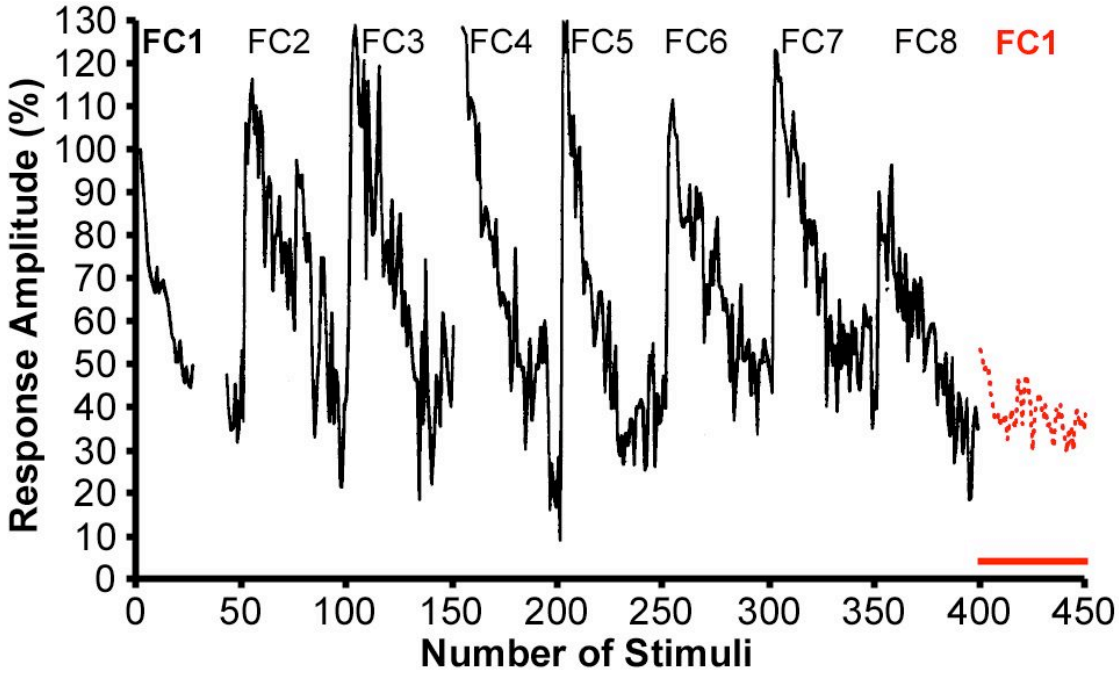


Electrophysiological responses



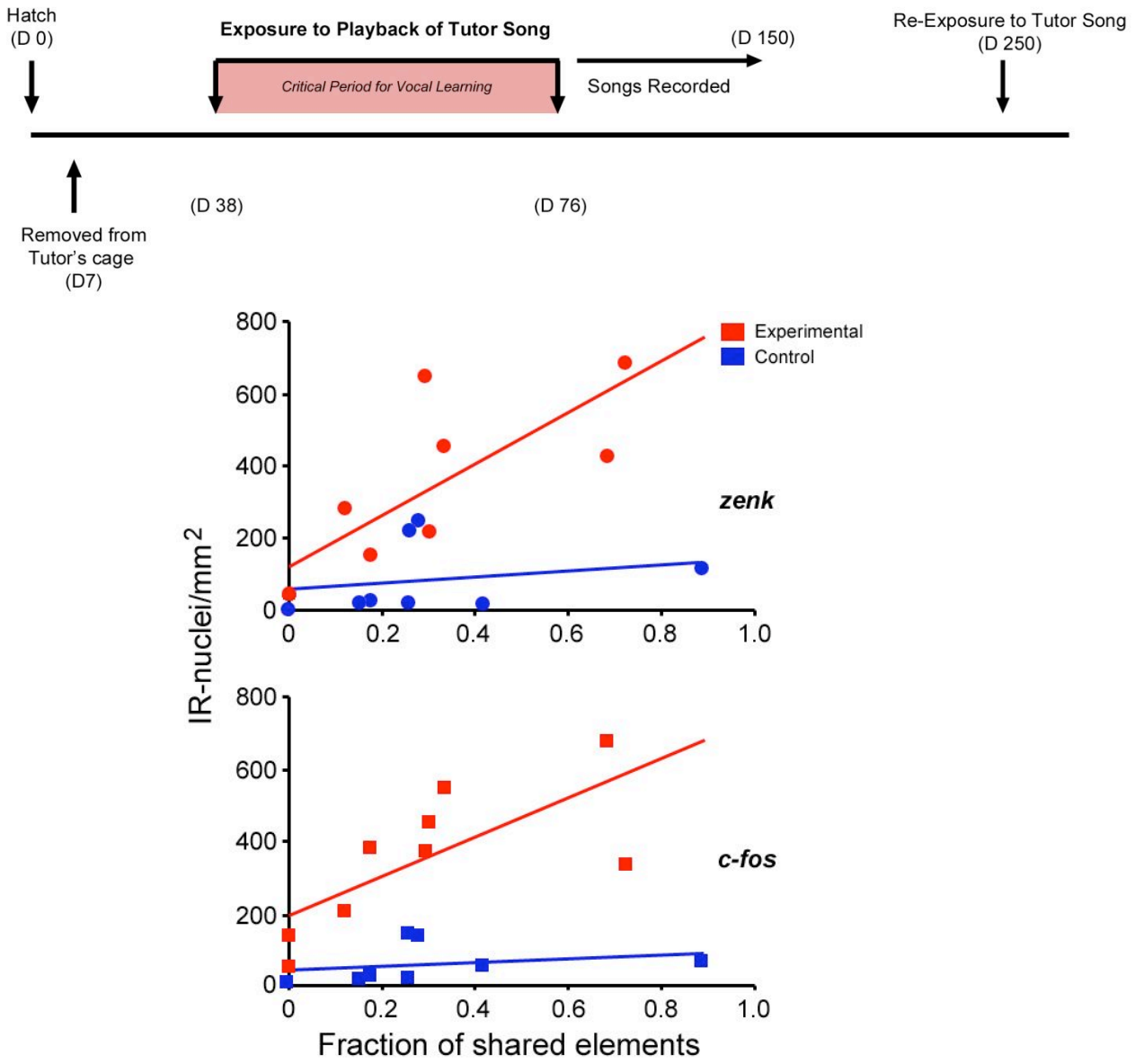
NCM Habituate and Respond Robustly to Novel Songs



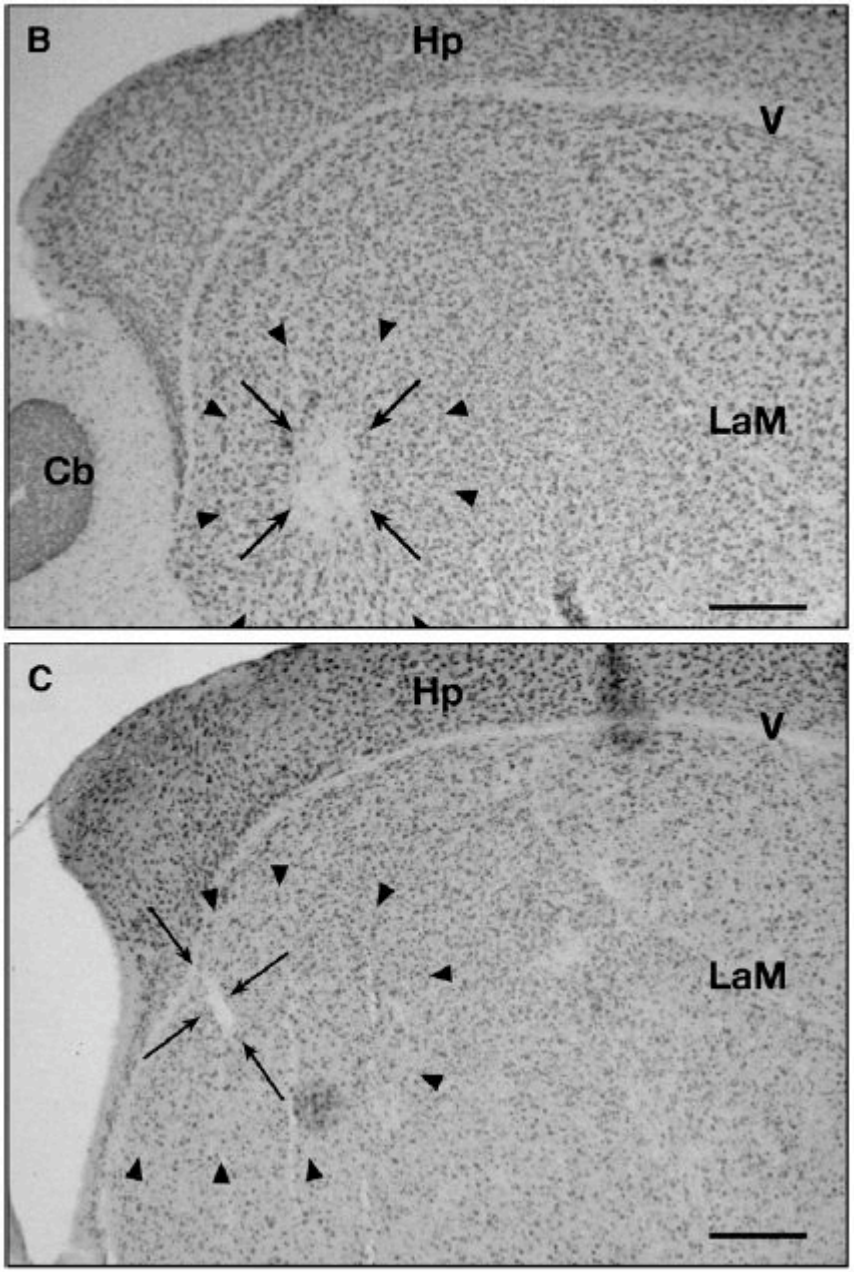


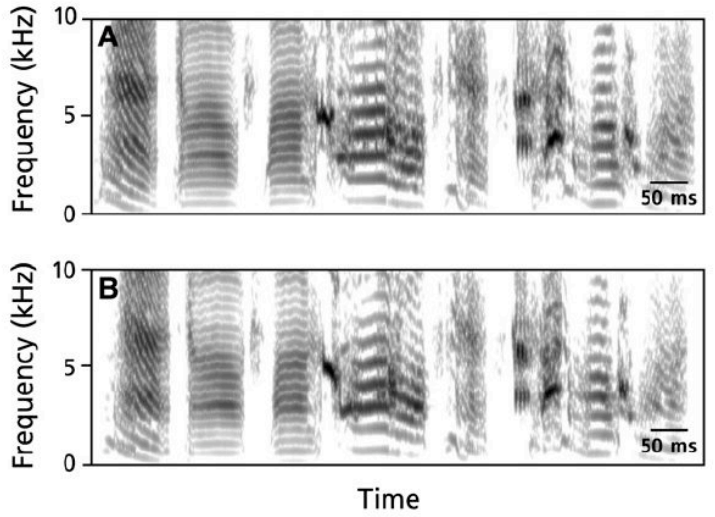
Role in Auditory Discrimination?
Auditory Memory Formation?

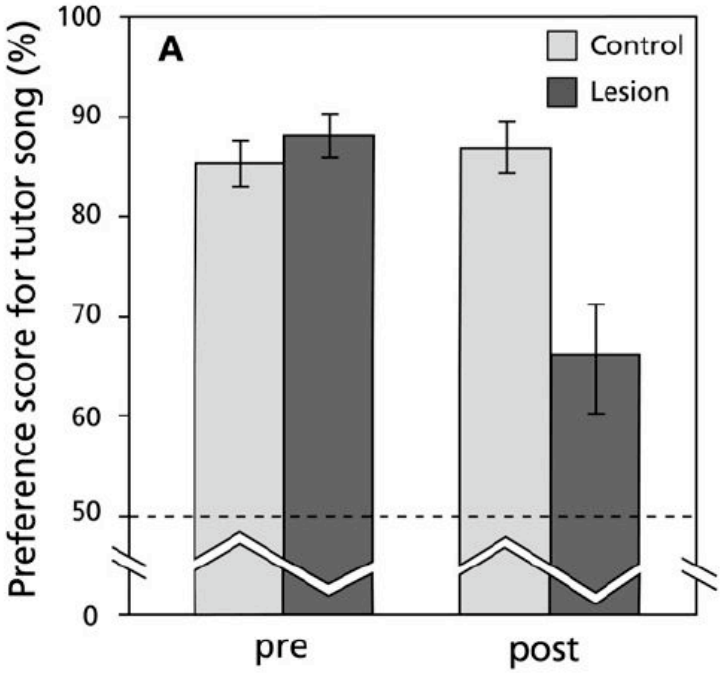
Neuronal Activation in NCM is Correlated with Strength of Learning



Lesions in NCM Impact Tutor Song Preference, but not Production







Conclusions – Part I

NCM as a key site involved in auditory discrimination and the formation of auditory memories

-Auditory stimuli drive activation of NCM neurons.

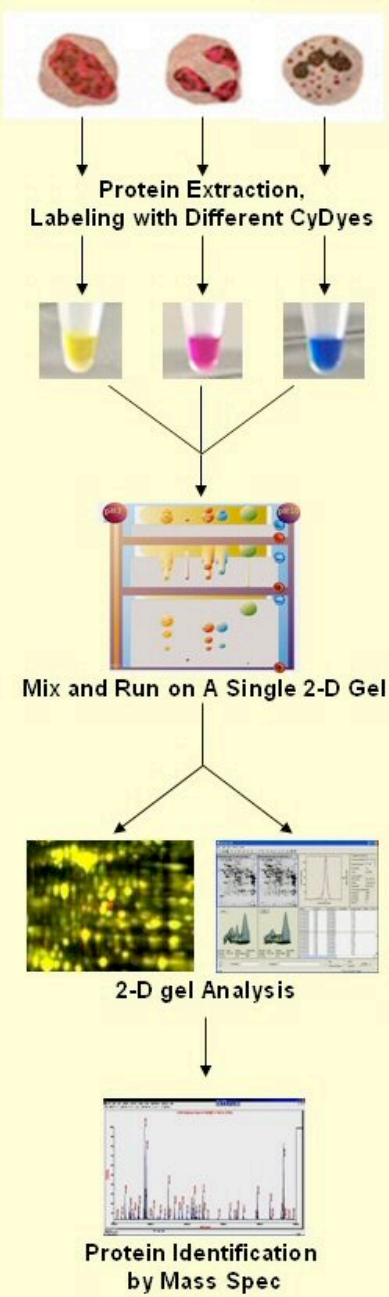
-These responses are selective to species-specific stimuli, habituate and are vigorously reinstated upon presentation of novel songs.

-Activation of neurons in NCM is positively correlated with strength of song learning.

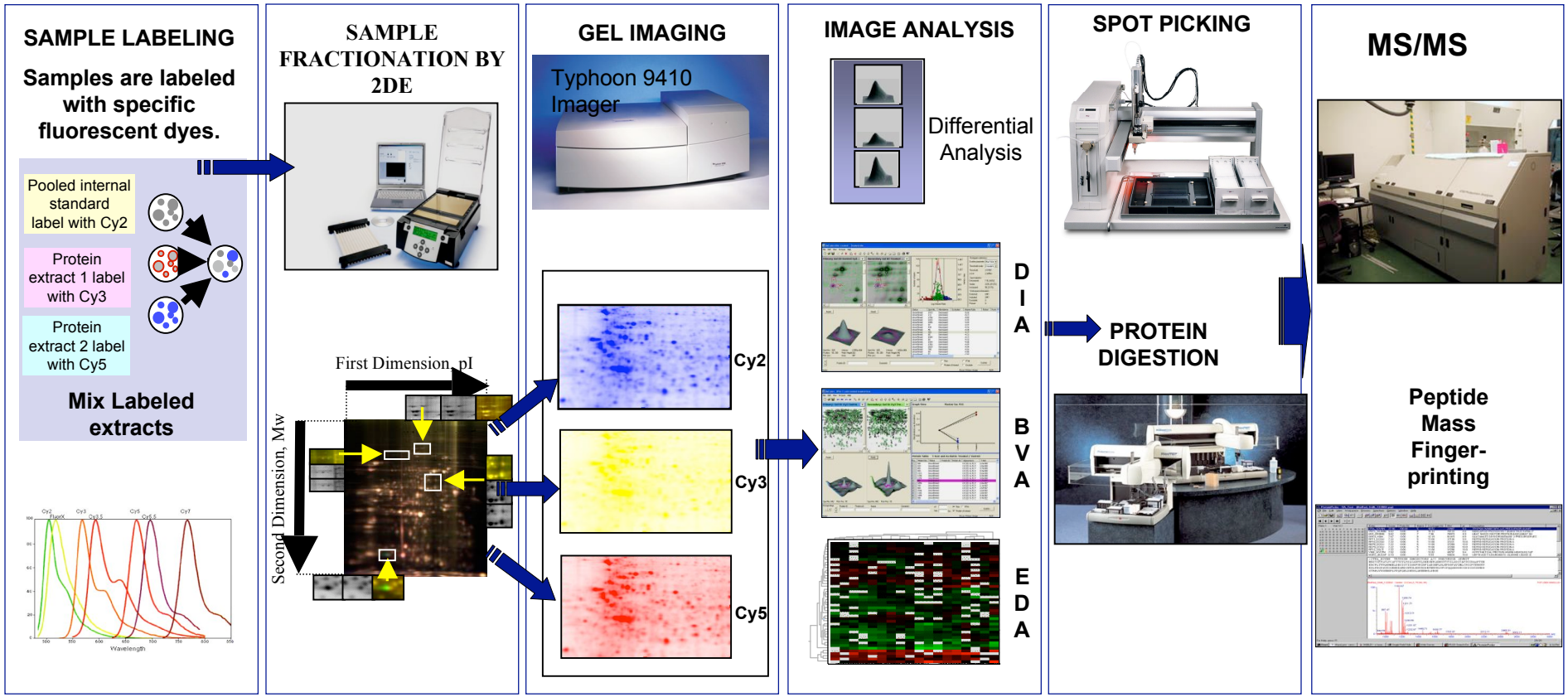
-Lesions of NCM impact discrimination of species-specific songs (possibly formation of auditory memories).

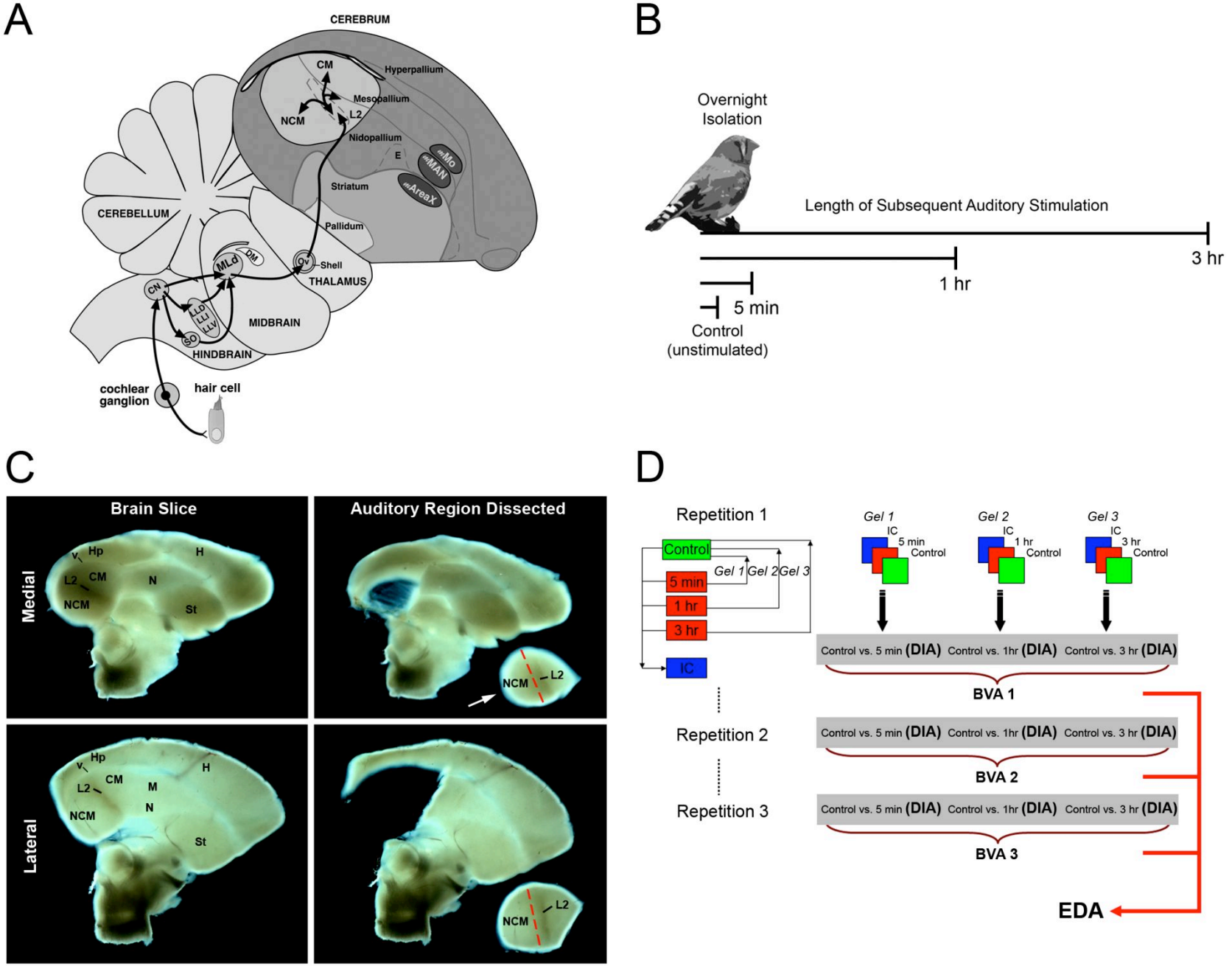
What are molecular mechanisms underlying these processes?

Proteomic Analysis – 2D-DIGE

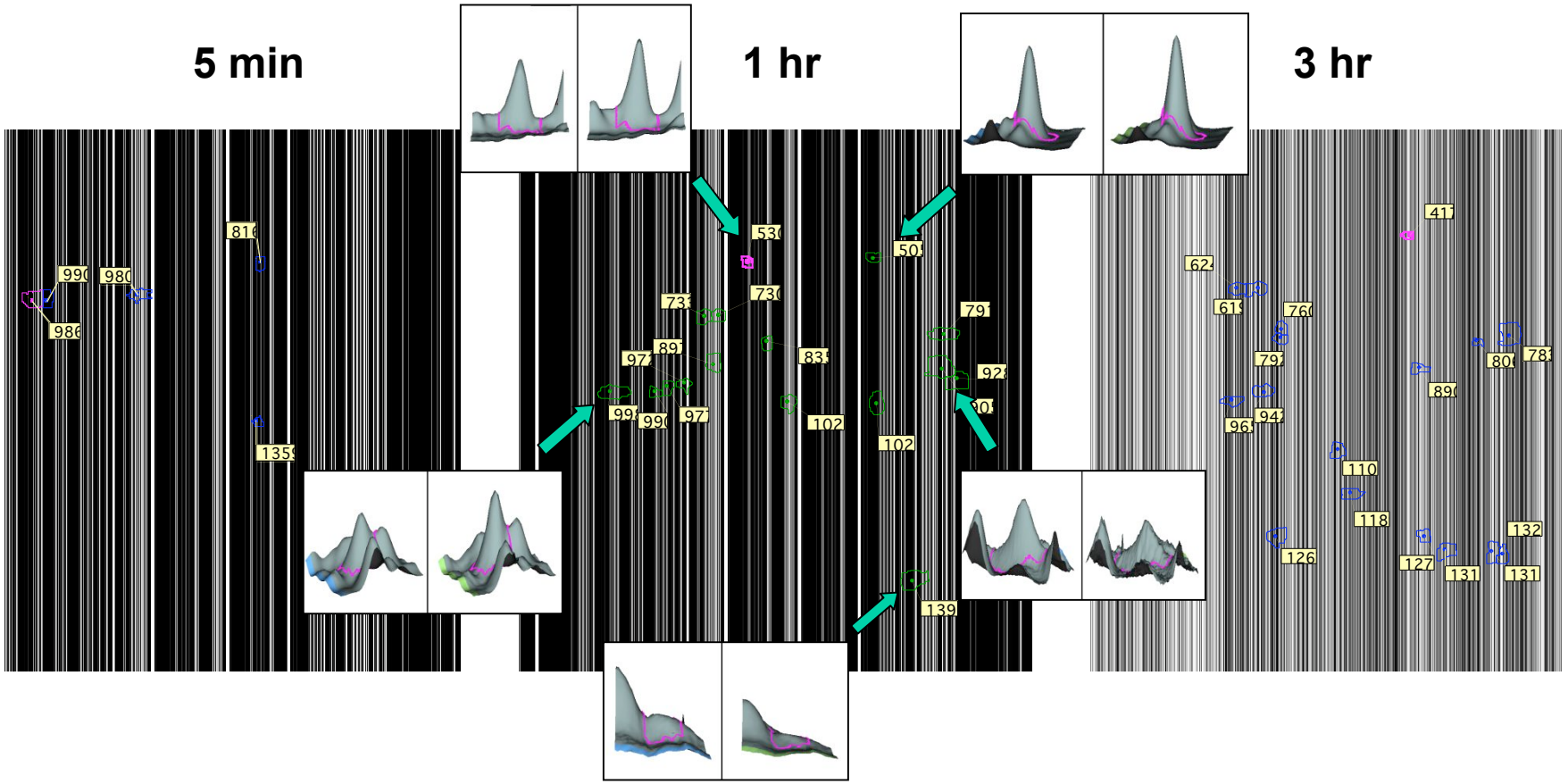


Proteomic Analysis – 2D-DIGE

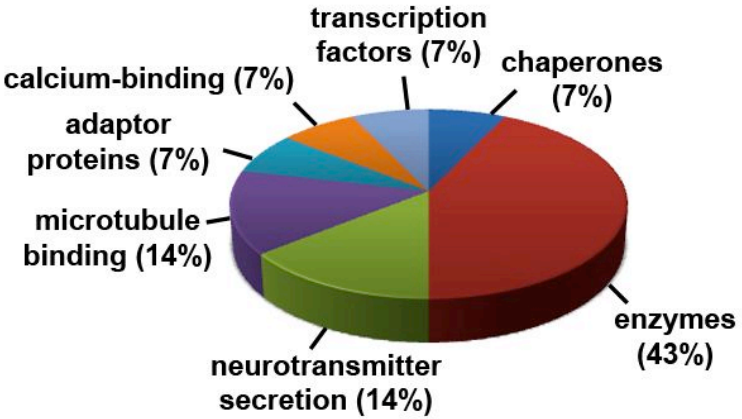



A
CEREBRUM
CM
Hyperpallium
Mesopallium
NCM
L2
Nidopallium
E
Striatum
Pallidum
CEREBELLUM
MLd
DM
Ov
Shell
THALAMUS
CN
LLD
LLI
LLV
SO
MIDBRAIN
HINDBRAIN
cochlear ganglion
hair cell
B
Overnight Isolation
Length of Subsequent Auditory Stimulation
3 hr
1 hr
5 min
Control (unstimulated)
C
Brain Slice
Auditory Region Dissected
Medial
Lateral
Hp
H
CM
L2
N
M
NCM
St
D
Repetition 1
Control
5 min
1 hr
3 hr
IC
Gel 1
Gel 2
Gel 3
Control vs. 5 min (DIA)
Control vs. 1hr (DIA)
Control vs. 3 hr (DIA)
BVA 1
Repetition 2
BVA 2
Repetition 3
BVA 3
EDA

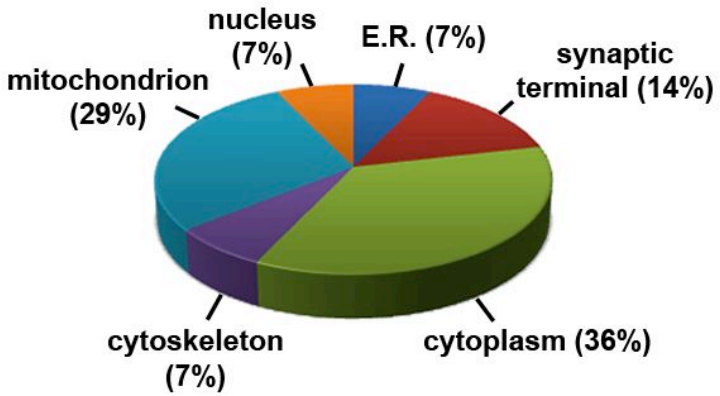
Representative Gels and Protein Identification

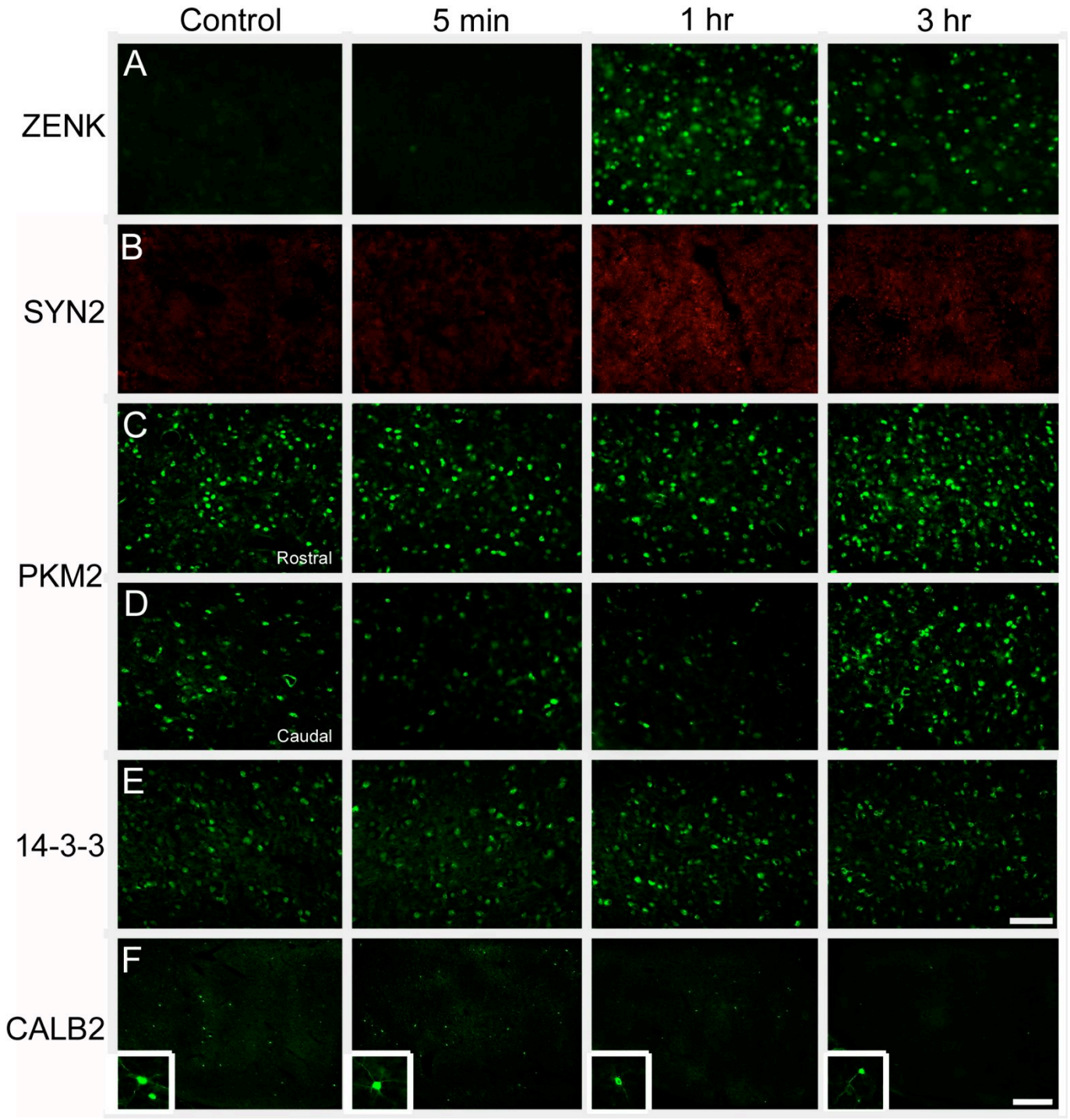


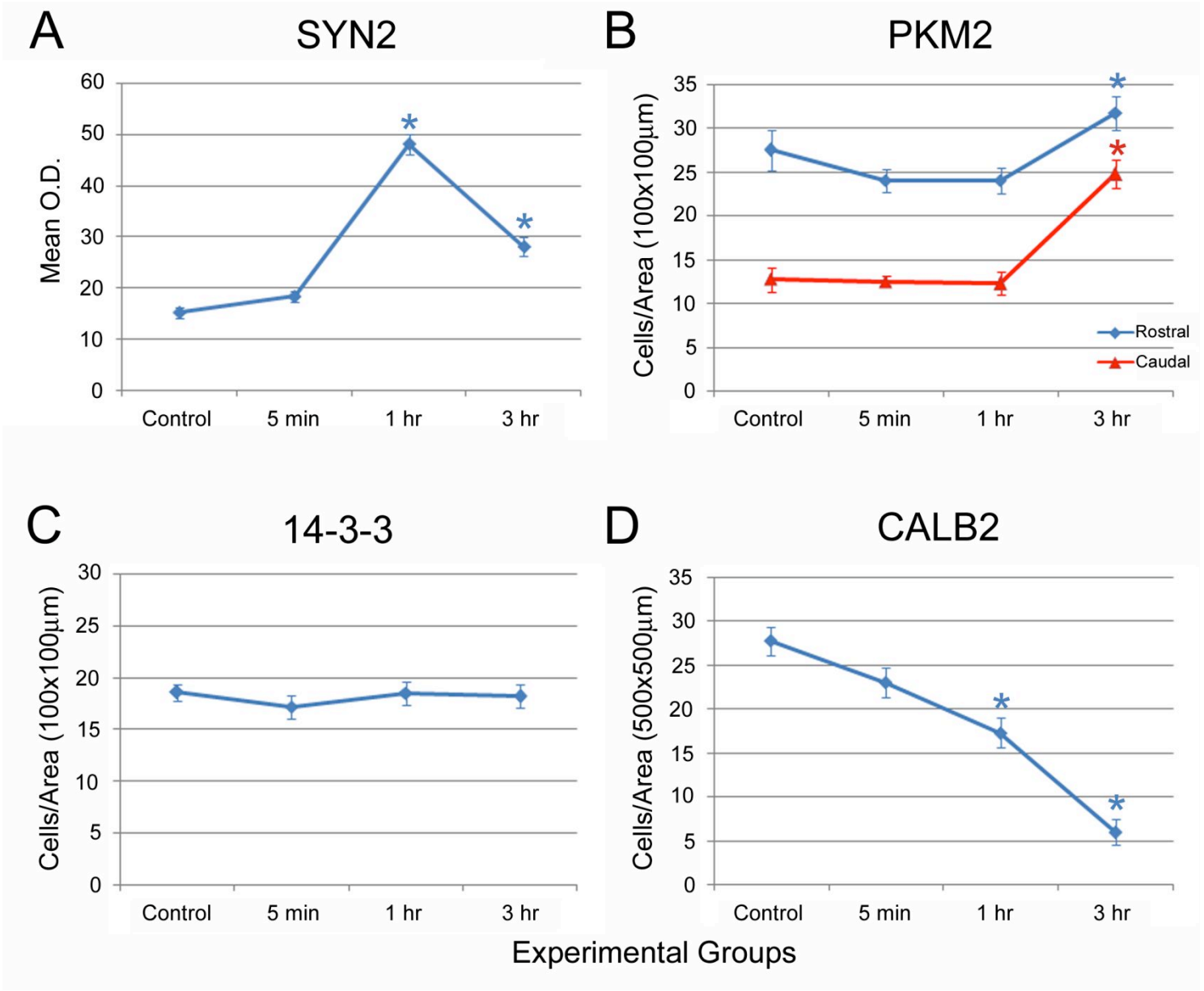
A

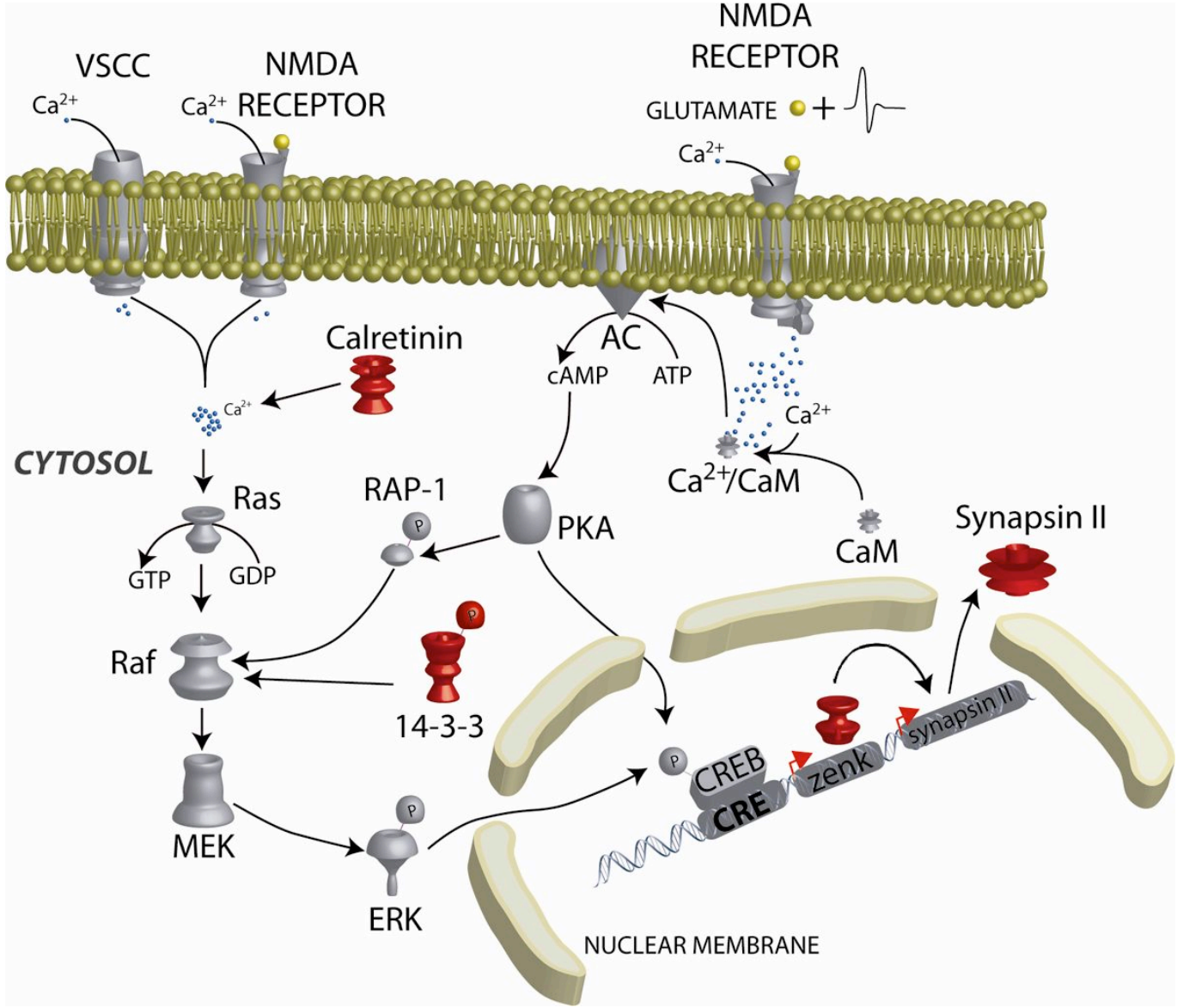

Function
transcription factors (7%)
chaperones (7%)
calcium-binding (7%)
adaptor proteins (7%)
microtubule binding (14%)
enzymes (43%)
neurotransmitter secretion (14%)

B


Location
nucleus (7%)
E.R. (7%)
synaptic terminal (14%)
mitochondrion (29%)
cytoskeleton (7%)
cytoplasm (36%)


Control
5 min
1 hr
3 hr
A
ZENK
B
SYN2
C
PKM2
Rostral
D
Caudal
E
14-3-3
F
CALB2


A
SYN2
Mean O.D.
60
50
40
30
20
10
0
Control
5 min
1 hr
3 hr
B
PKM2
Cells/Area (100x100µm)
35
30
25
20
15
10
5
0
Rostral
Caudal
C
14-3-3
Cells/Area (100x100µm)
D
CALB2
Cells/Area (500x500µm)
Experimental Groups


NMDA
RECEPTOR
VSCC
NMDA
RECEPTOR
GLUTAMATE
+
Ca2+
Ca2+
Ca2+
Calretinin
AC
cAMP
ATP
Ca2+
Ca2+
CYTOSOL
Ca2+/CaM
Ras
RAP-1
PKA
CaM
Synapsin II
GTP
GDP
P
Raf
14-3-3
P
P
CREB
CRE
zenk
synapsin II
MEK
P
ERK
NUCLEAR MEMBRANE

Conclusions – Part II

Calcium-regulated biochemical and gene expression programs in NCM may underlie long-term changes in neural circuitry required for song discrimination and the formation of auditory memories

-High throughput proteomics screening reveals a complex network of proteins regulated by auditory experience in NCM.

-The identified proteins belong to multiple functional classes and are present in several cell compartments.

-The MAPK pathway appears to be highly regulated by auditory stimuli in NCM.

Acknowledgments

Liisa Tremere
Ernest Nordeen
Kathy Nordeen
Thomas Terleph
Oscar Alzate
Erich Jarvis
Cristina Osorio
Robert Buechler