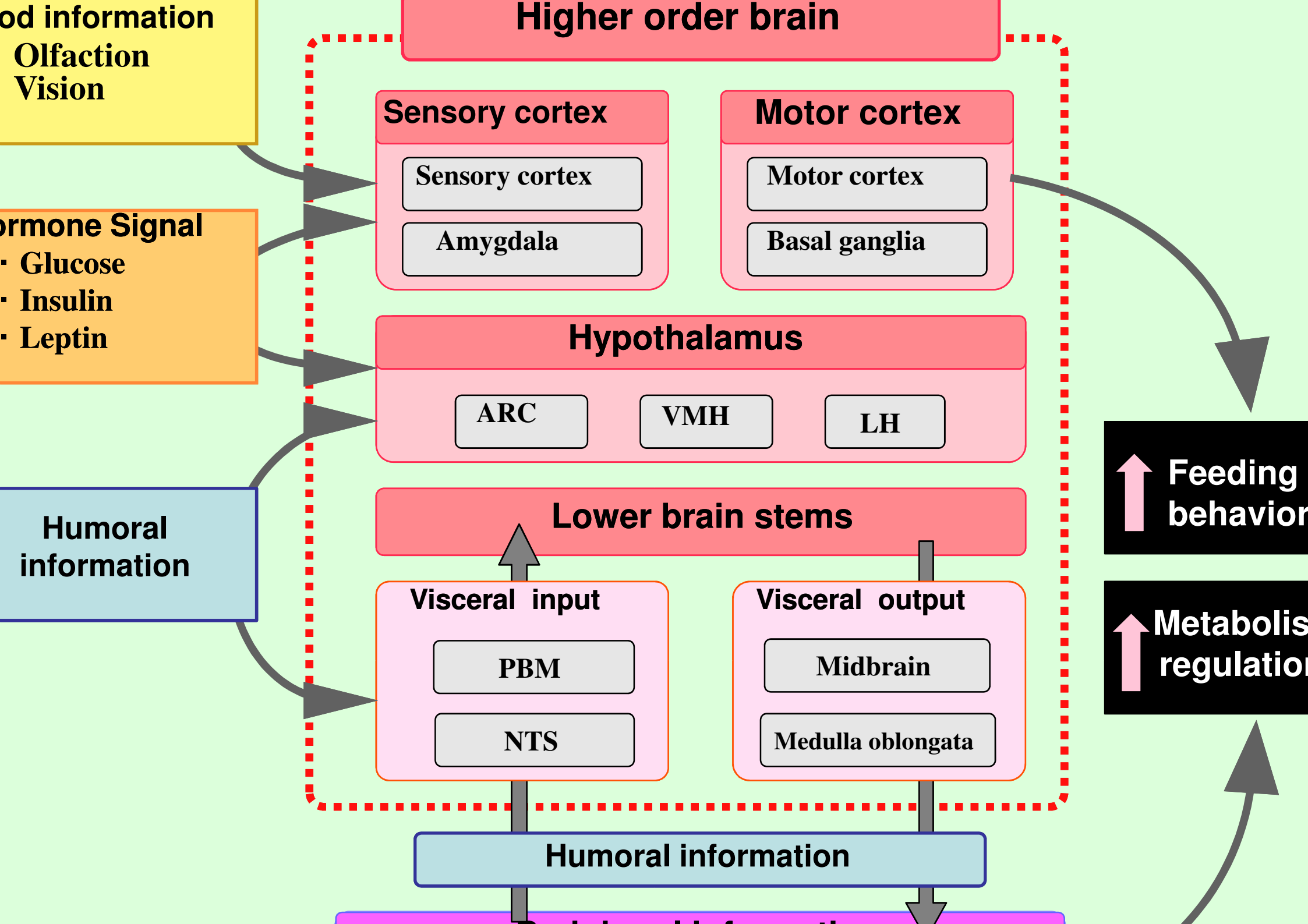
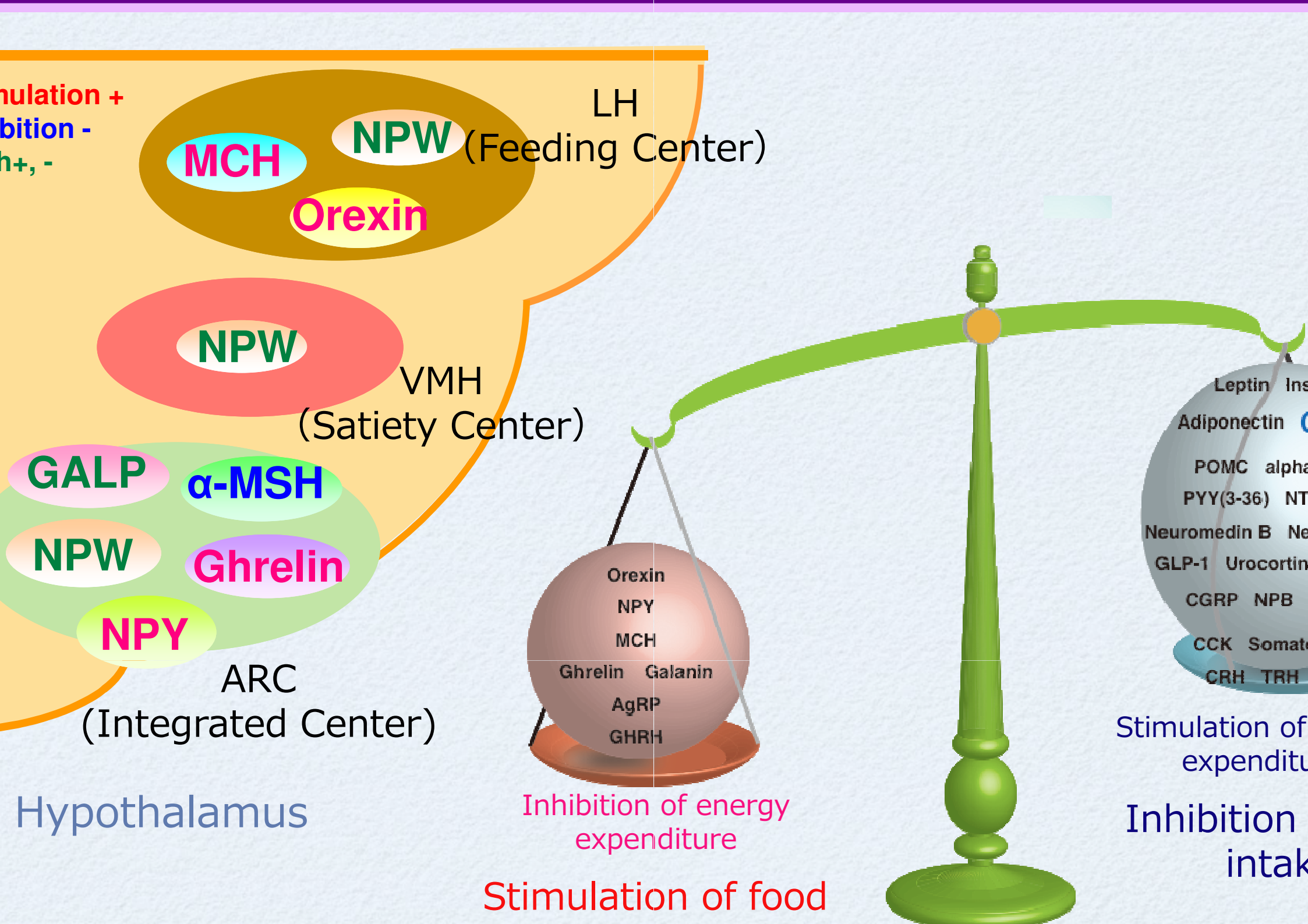
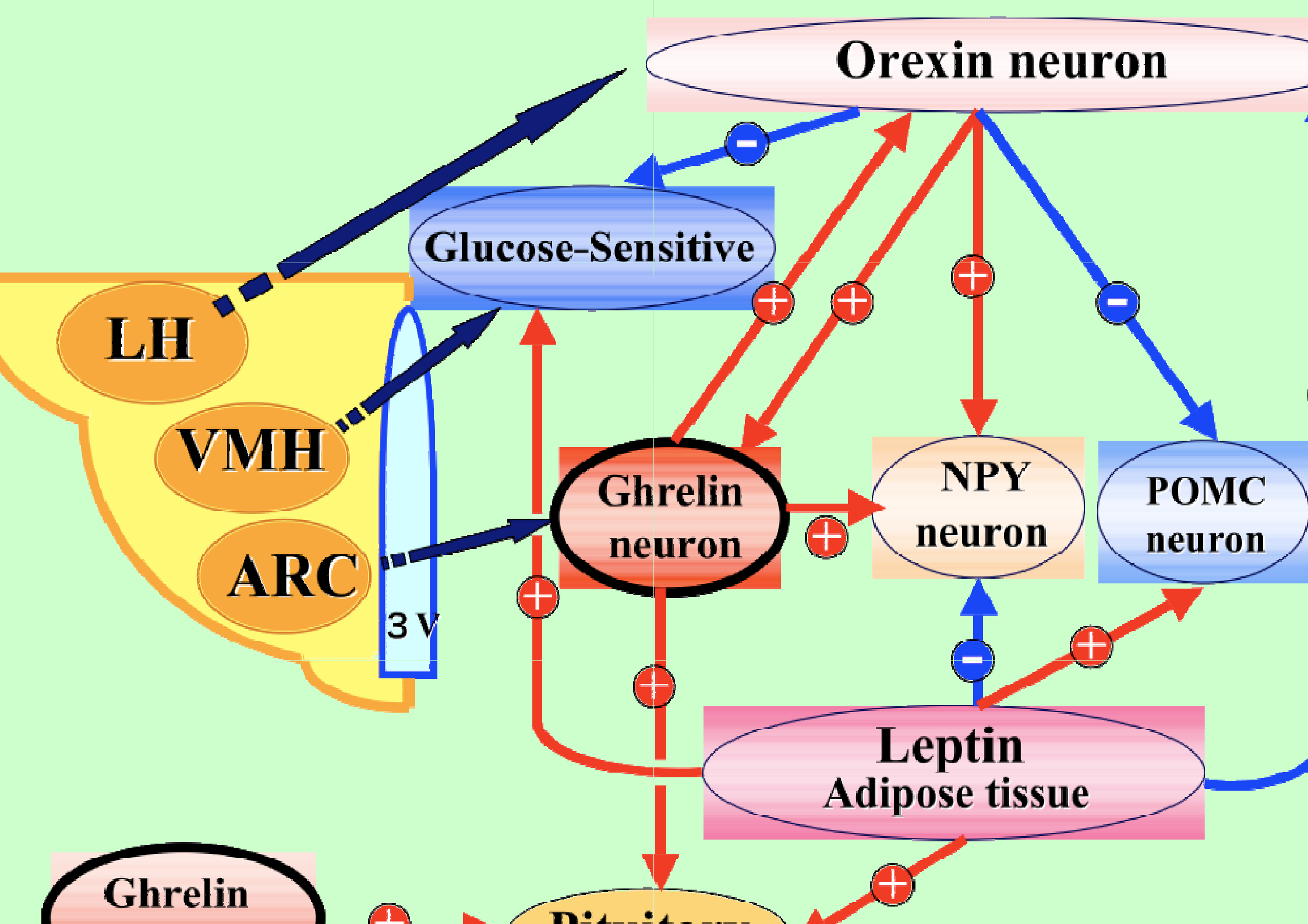
he Effect of Intranasal infusion of GALP on feeding and energy metabolism

Department of Anatomy,
Showa University School of Medicine Seiji Shioda


od information
Olfaction
Vision
ormone Signal
· Glucose
· Insulin
· Leptin
Humoral information
Higher order brain
Sensory cortex
Sensory cortex
Amygdala
Motor cortex
Motor cortex
Basal ganglia
Hypothalamus
ARC
VMH
LH
Lower brain stems
Visceral input
PBM
NTS
Visceral output
Midbrain
Medulla oblongata
Humoral information
Feeding behavior
Metabolis regulatio


nulation +
bition -
h+, -
LH
(Feeding Center)
MCH
NPW
Orexin
NPW
VMH
(Satiety Center)
GALP
α-MSH
NPW
Ghrelin
NPY
ARC
(Integrated Center)
Hypothalamus
Orexin
NPY
MCH
Ghrelin Galanin
AgRP
GHRH
Inhibition of energy
expenditure
Stimulation of food
Leptin Ins
Adiponectin
POMC alpha
PYY(3-36) NT
Neuromedin B Ne
GLP-1 Urocortin
CGRP NPB
CCK Somat
CRH TRH
Stimulation of
expenditu
Inhibition
intak


Orexin neuron
Glucose-Sensitive
LH
VMH
ARC
3 V
Ghrelin neuron
NPY neuron
POMC neuron
Leptin
Adipose tissue
Ghrelin

eding Regulation by Galanin-Like Peptide (GALP)

lanin

ALP

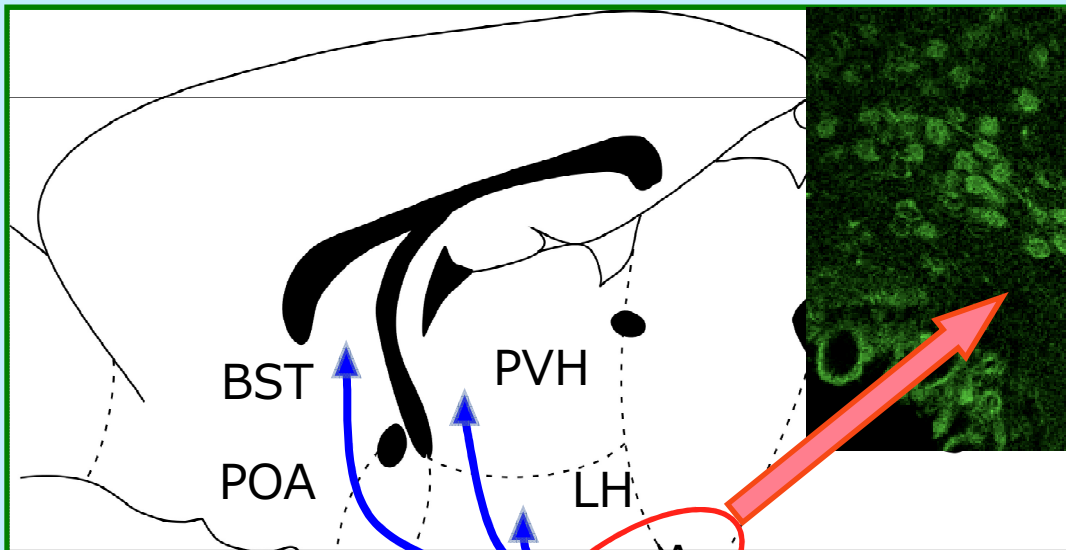
```
                1                 10                  20                  30
Human           GWTLNSAGYLLGPHAVGNHRSFSDKNGLTS
Monkey          GWTLNSAGYLLGPHAVGNHRSFSDKNGLTS
Pig             GWTLNSAGYLLGPHAIDNHRSFHDKYGLA
Rat             GWTLNSAGYLLGPHAIDNHRSFSDKHGLT
Mouse           GWTLNSAGYLLGPHAIDNHRSFSDKHGLT

                1                 10                  20                  30                  40                  50
Human           APAHRGRGGWTLNSAGYLLGPVLHLPQMGDQDGKRETALEILDLWKAIDGLPYSHP
Monkey          APAHQGRGGWTLNSAGYLLGPVLHLPQMGDQDRKRETALEILDLWKAIDGLPYSHP
Pig             APVHRGRGGWTLNSAGYLLGPVLHPPSRAEGGGKGKTALGILDLWKAIDGLPYPQS
Rat             APAHRGRGGWTLNSAGYLLGPVLHLSSKANQGRKTDSALEILDLWKAIDGLPYSRS
Mouse           APAHRGRGGWTLNSAGYLLGPVLPVSSKADQGRKRDSALEILDLWKIIDGLPYSHS
```

Primary structure of Galanin and GALP

hat is GALP ?

dogenous ligand of galanin, GPCR family
olated from porcine gut and brain
erlapped with amino acid sequence



Bed nucleus of
the stria terminalis

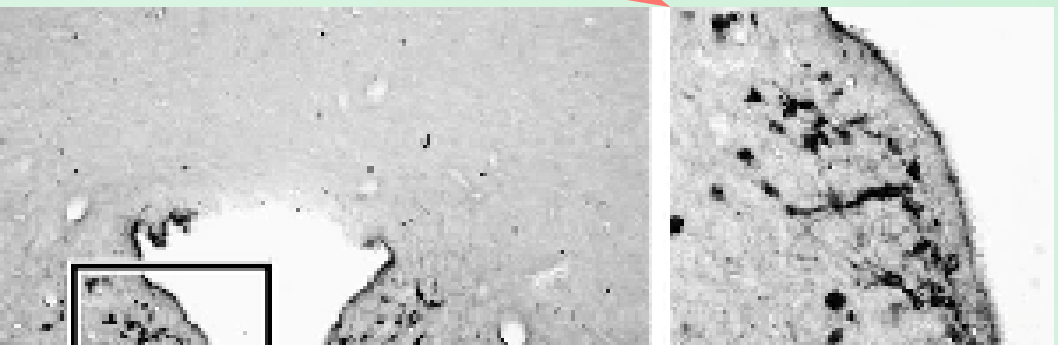
Medial preoptic area

Paraventricular nucleus

Lateral hypothalamus

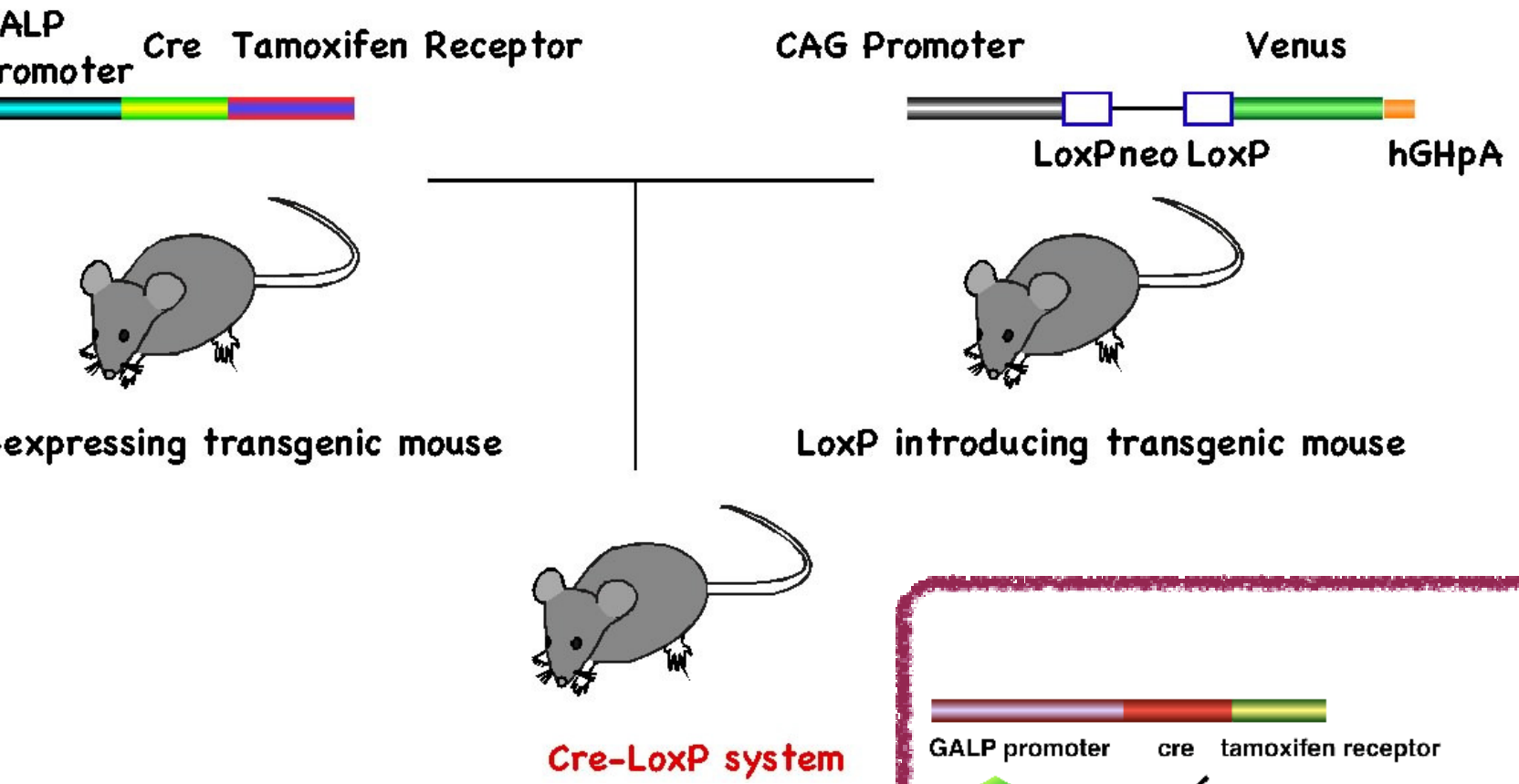
Arcuate nucleus

Pituitary

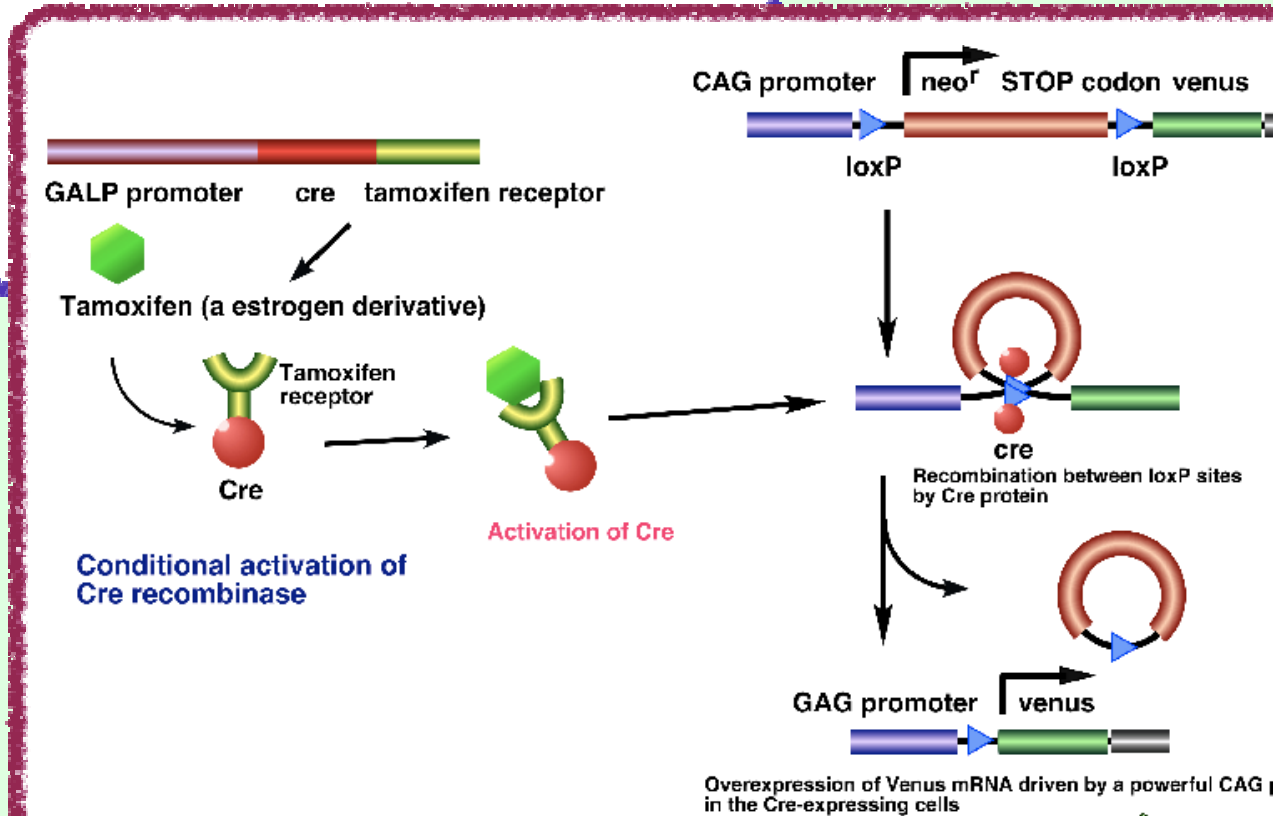


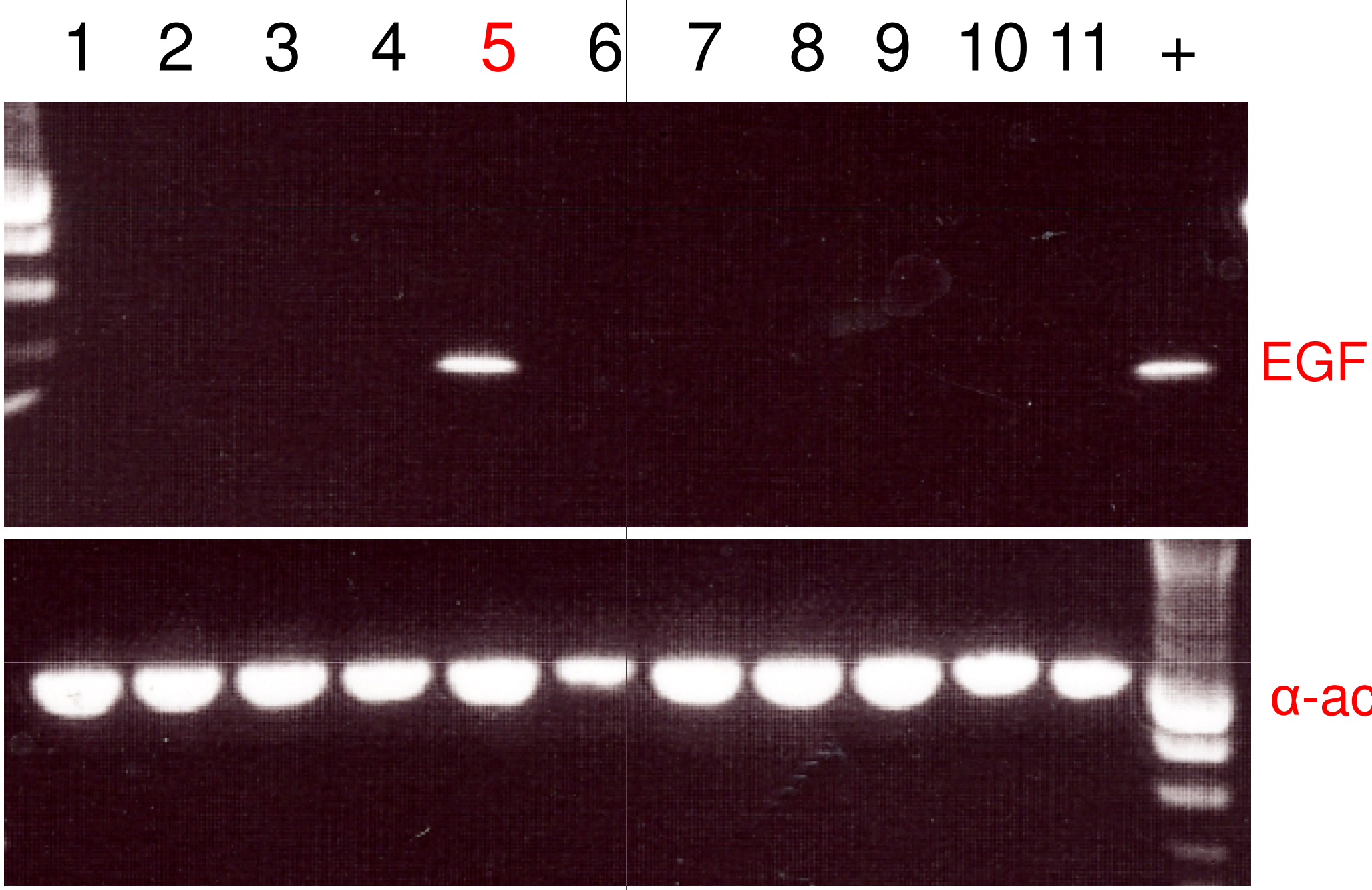
Guan, Shioda et al., Regul Pept 126, 200

GALP-expressing neurons



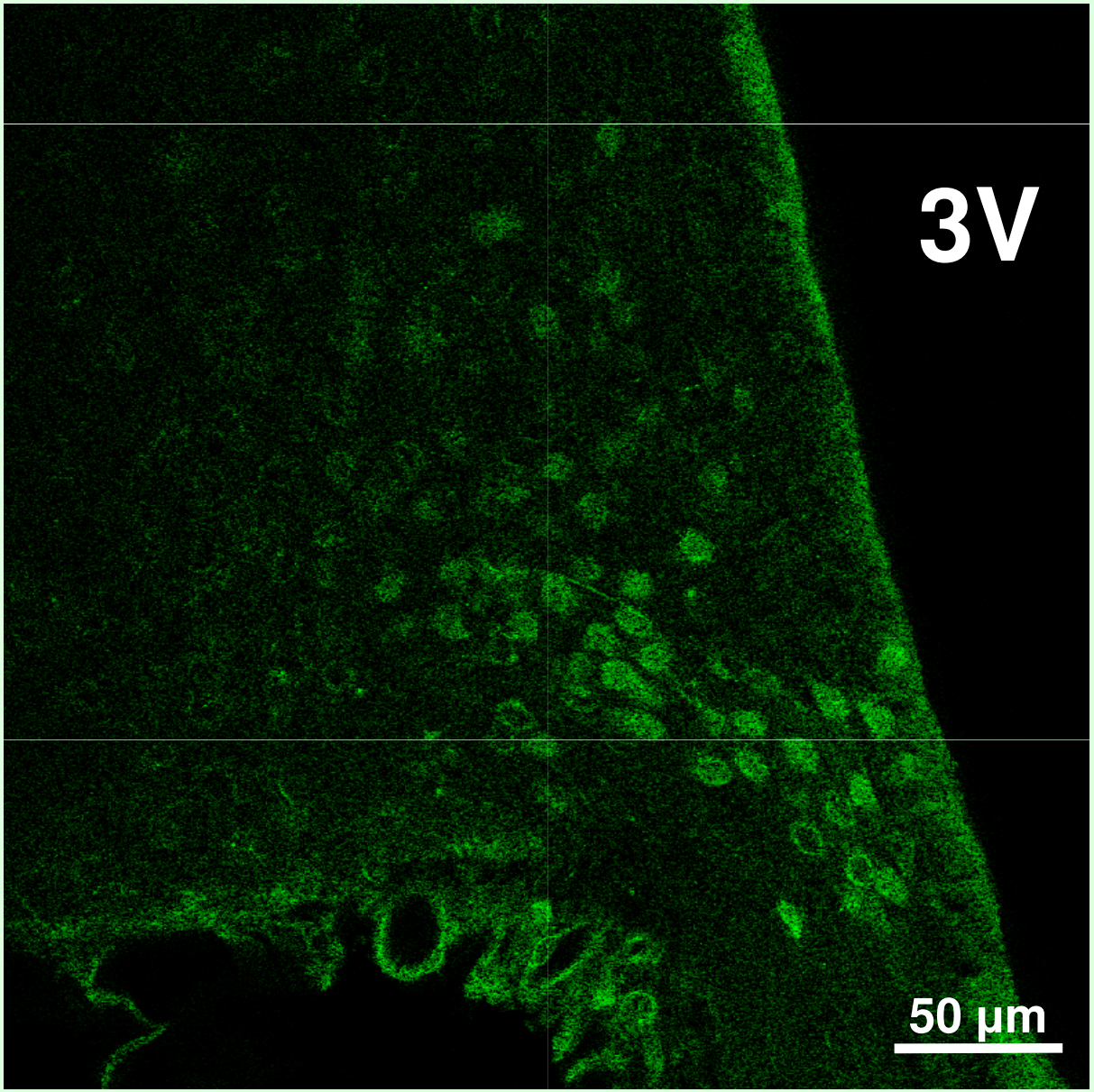
enus: mutant green fluorescent protein



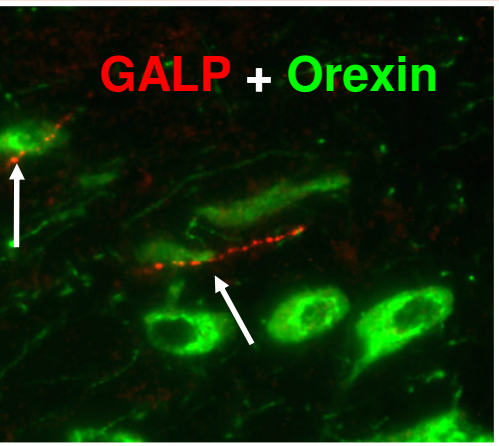

1 2 3 4 5 6 7 8 9 10 11 +
EGF
α-ac

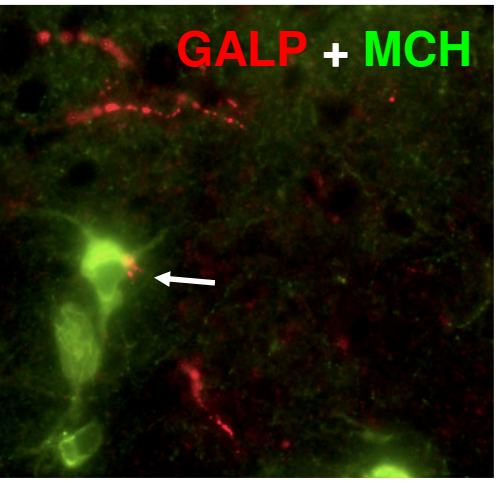
mouse carrying GALP-CreER/CAG-LNL-Venus gene

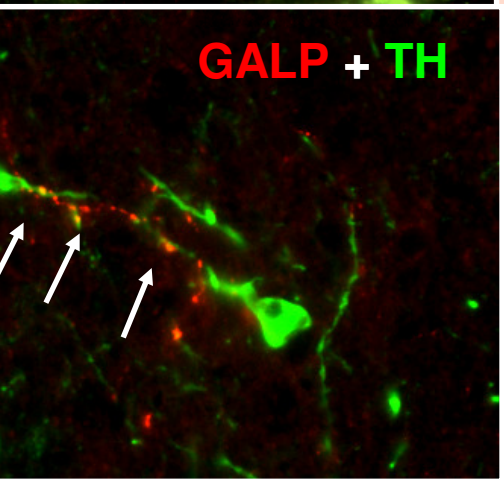
Venus fluorescence



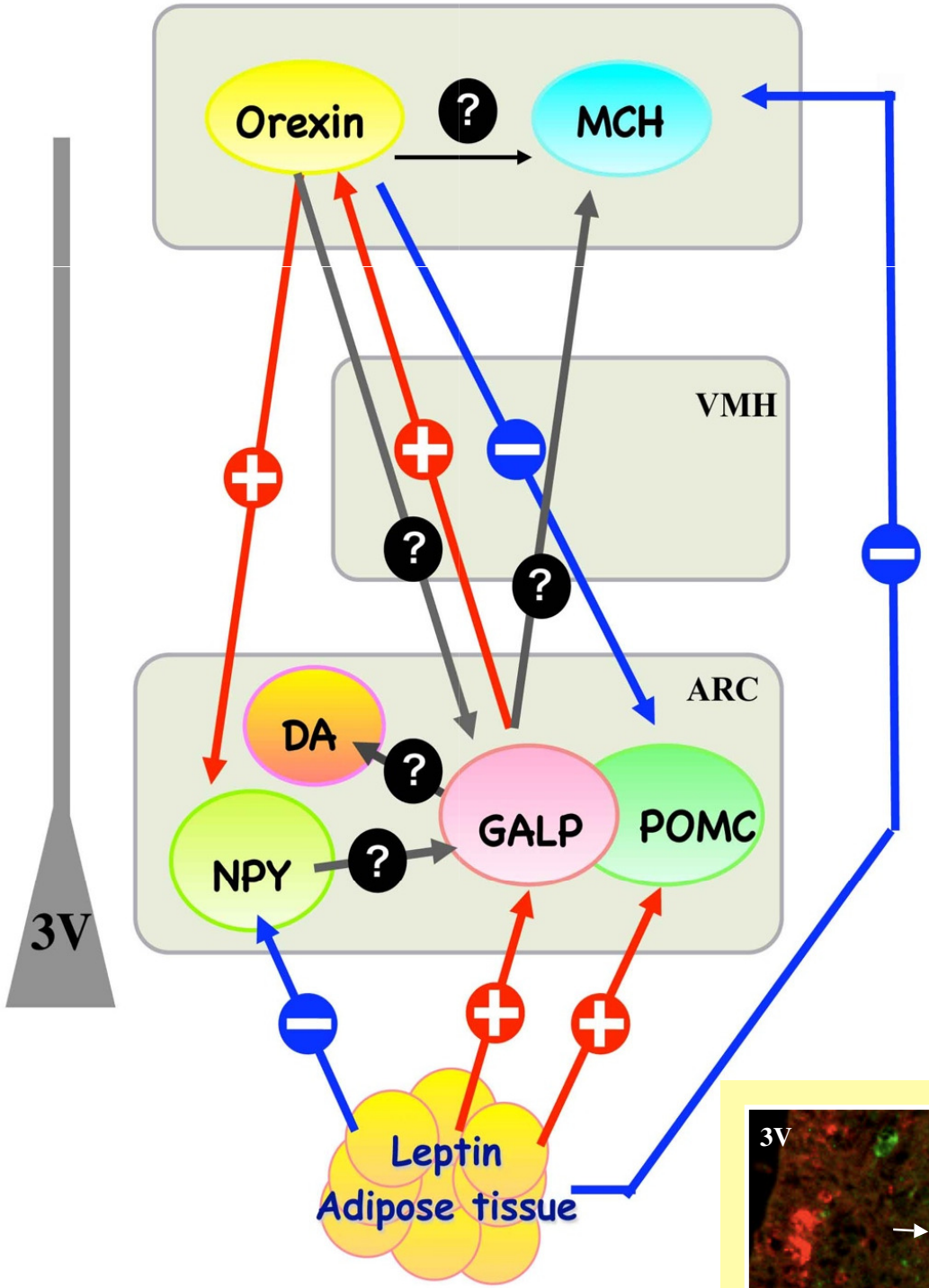
Output



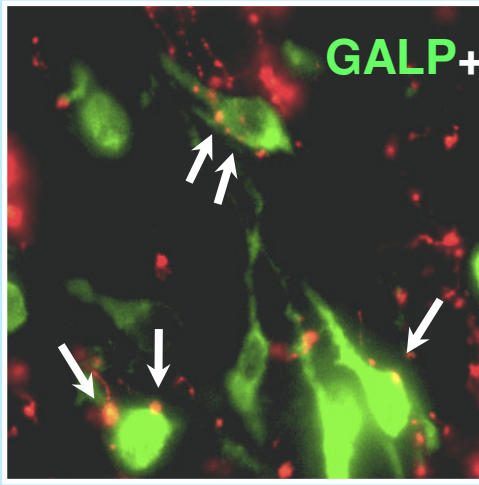


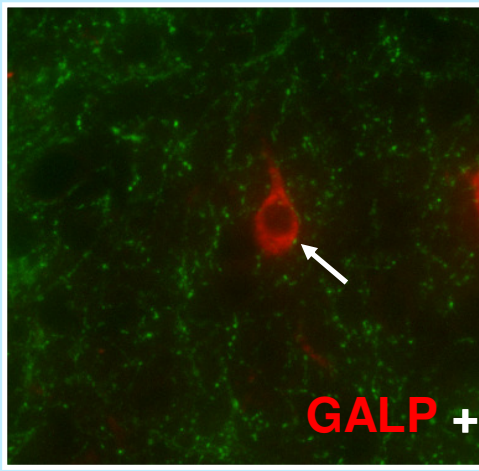


Hypothalamus

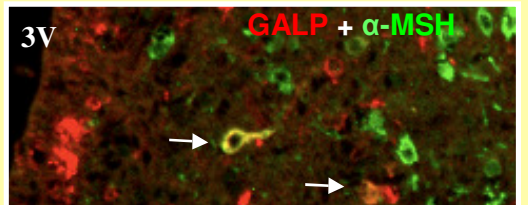


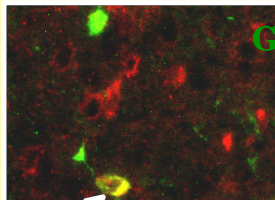
Input



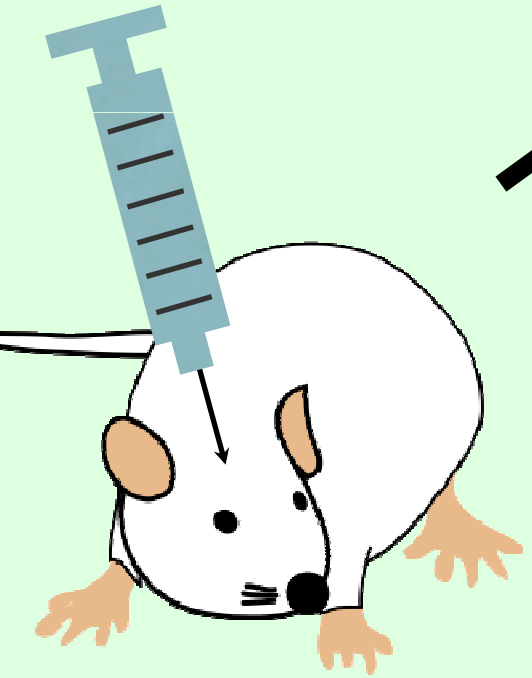


Co-localization





GALP



Food intake in short time

(Matsumoto et al., Neurosci Le

Body weight after 24 hrs ↓

(Lawrence et al., J Neuroendo

O_2 consumption ↑

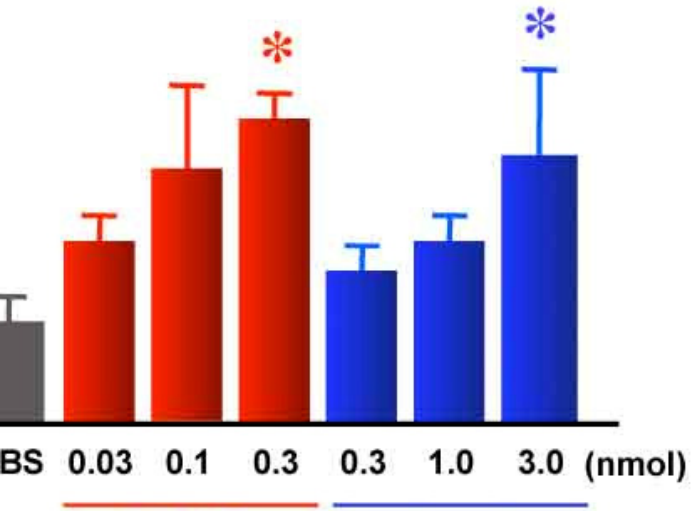
(Hansen et al., Endocrinology, 2003)

LH secretion ↑

(Matsumoto et al., Endocrinology, 2001)

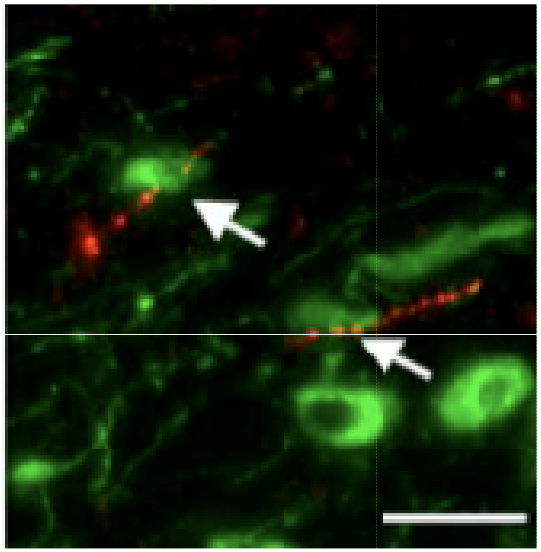
Testosterone secretio

ours food intake with GALP
and galanin (icv)



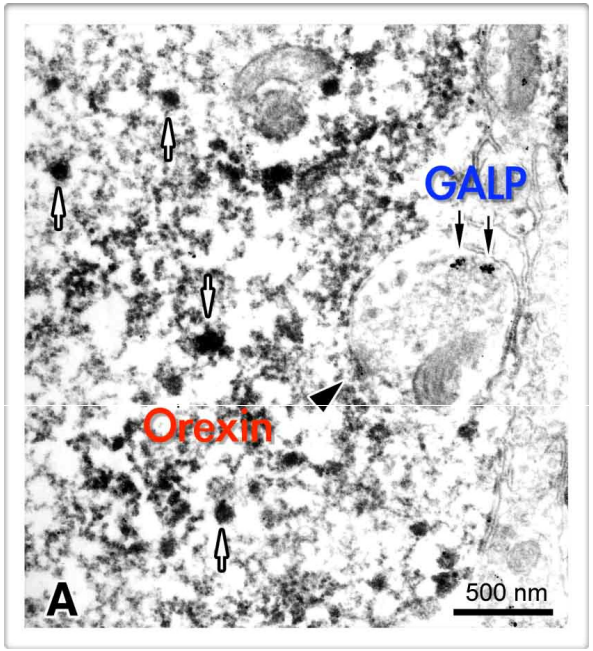
sumoto et al., Neurosci Lett, 2002)

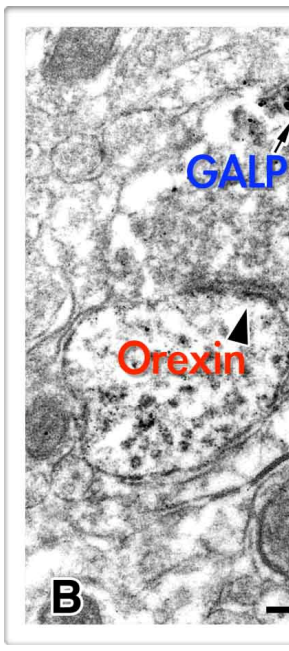
GALP → orexin



GALP and orexin

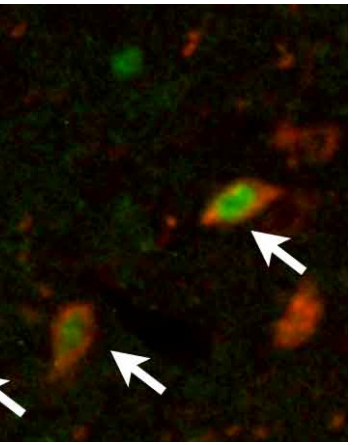
Takenoya et al., Reg Pept (2006)





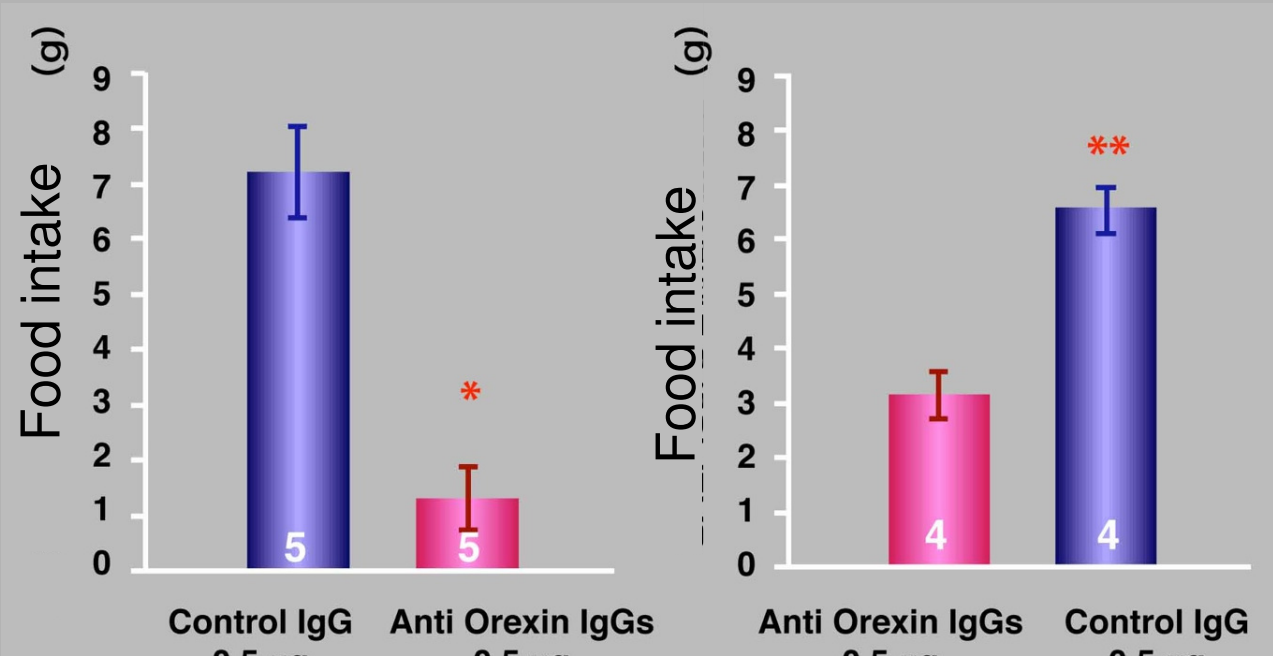
GALP-positive axon terminals making sy
with Orexin-positive neurons

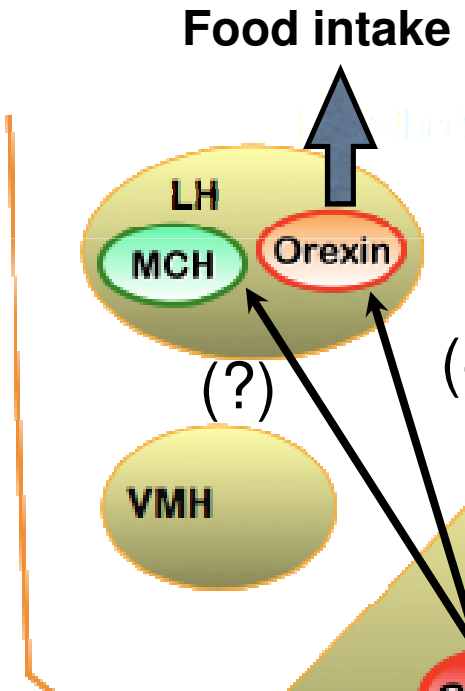
Fos-orexin



mulates c-Fos
on in orexin

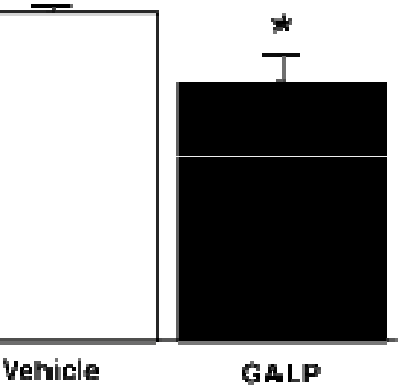
Anti orexin IgG icv induces food intake by GALP



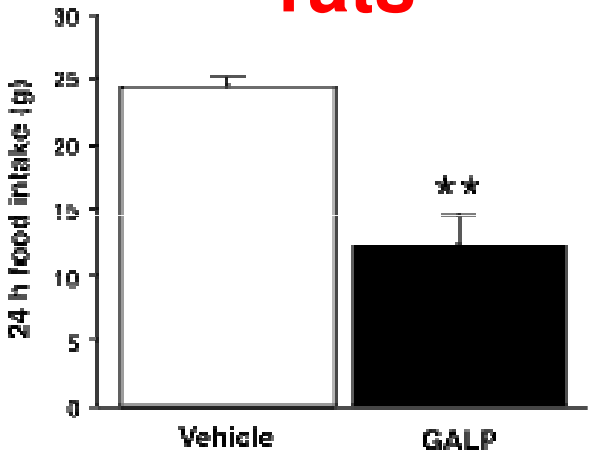


Food intake during 24 hrs

mice

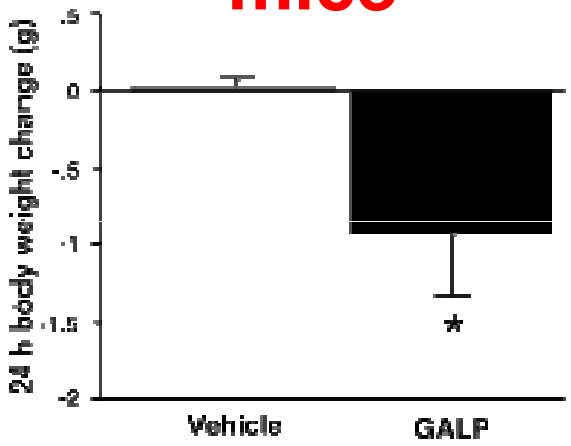


rats

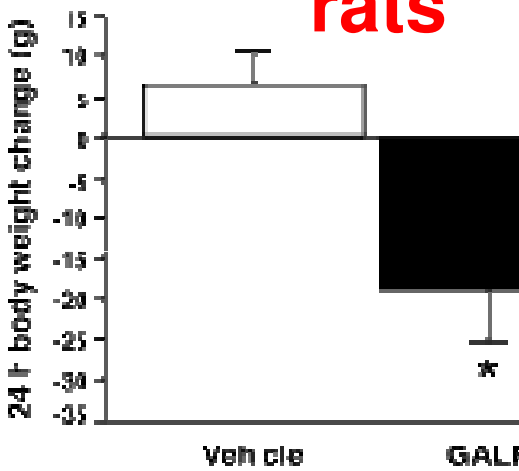


Body weight during 24 hrs (%

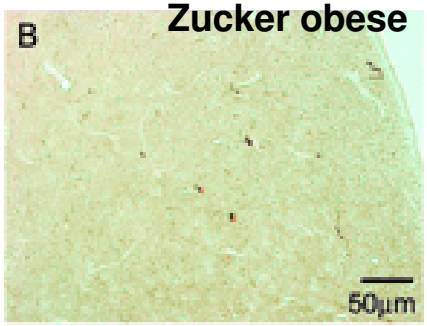
mice

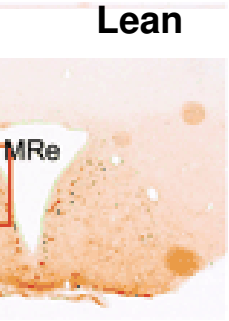


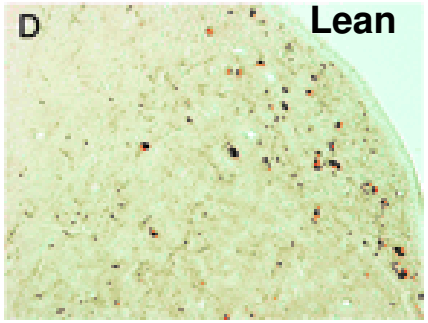
rats

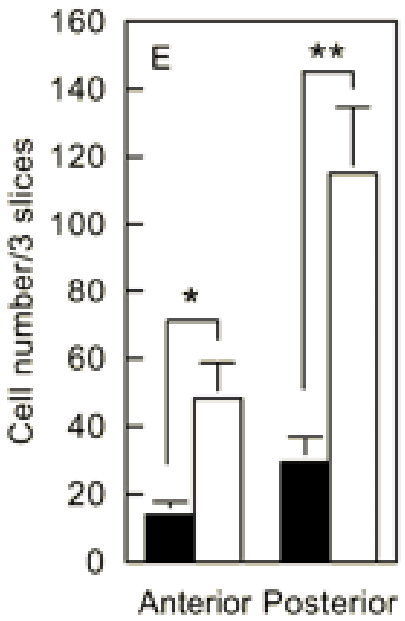


Krasnow et al. Endocrinol

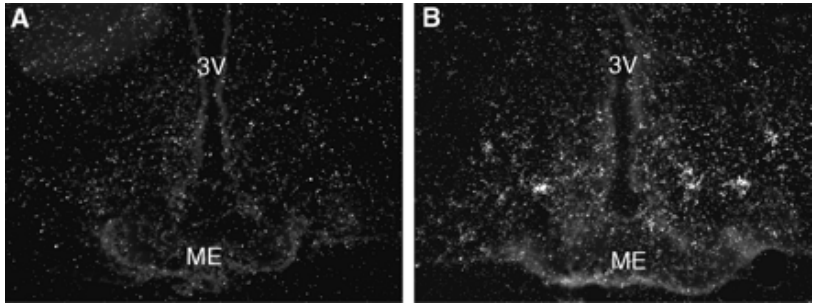






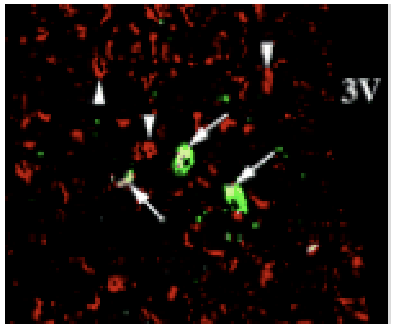


sion of GALP is decreased in Zucker rat

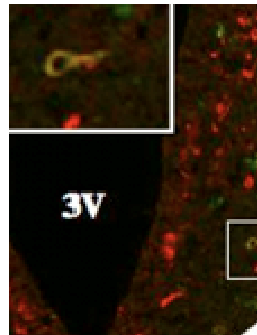


Leptin induc
mRNA expre
ob/ob mice

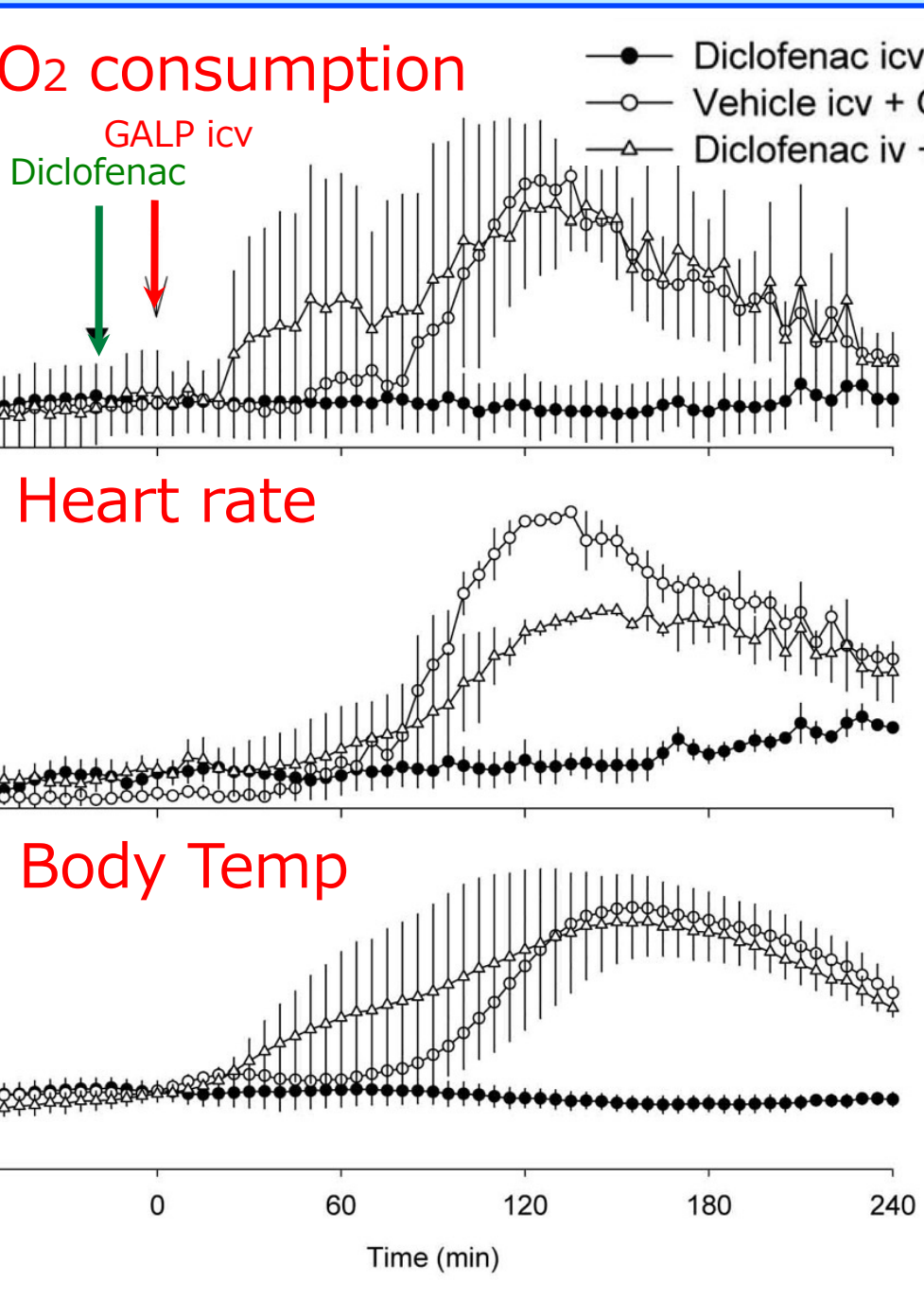
GALP + Leptin-R



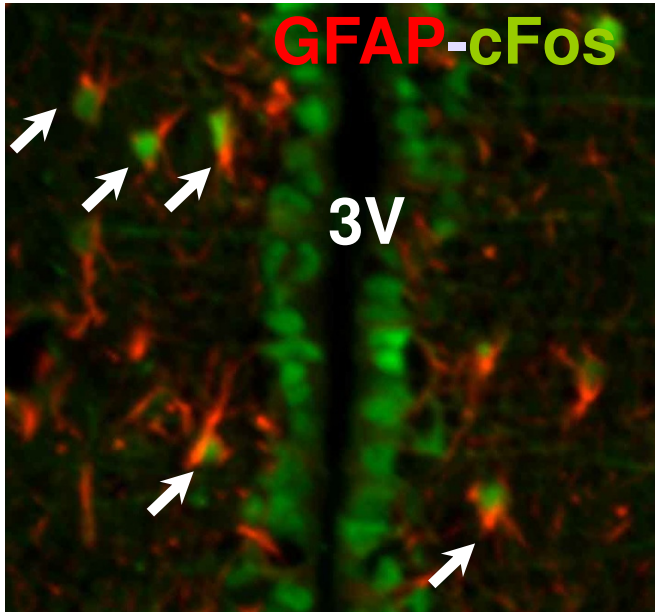
GALP + α–M



OX2 antagonist (icv) inhibits heat production by GALP

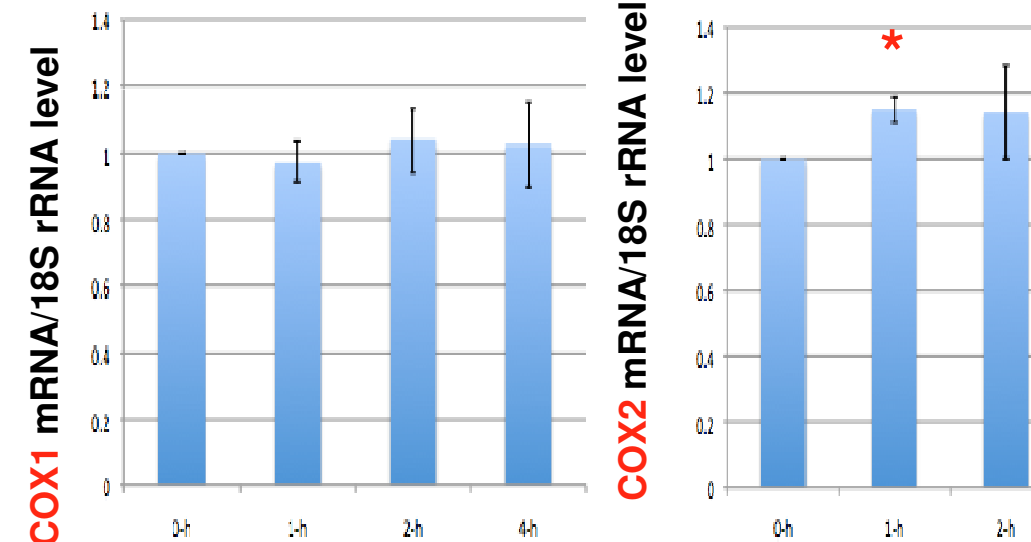


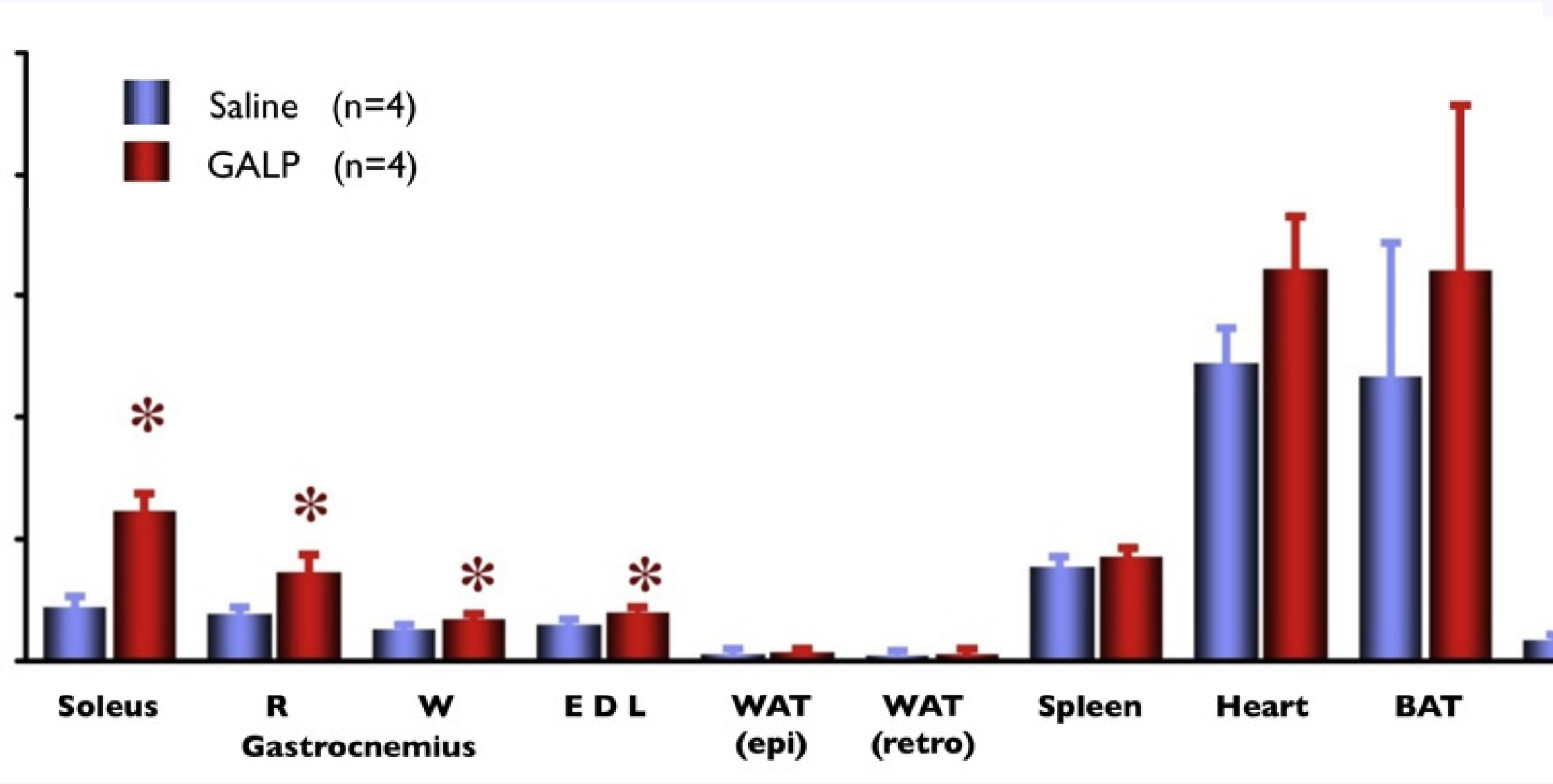
c-Fos expression in astrocyte by GAL



90 min after injection (icv)

Up-regulation of COX2 mRNA by GALP





*p< 0.05 vs Vehicle co

2-DG uptake in several tissues at 2 hrs after GALP icv

Measurement of oxyge
consumption and cor
body temperature

Whole body

Thermogenesis and energy metabolism by GALP

rown adipose tissue

Heat

Non-Shivering
hermogenesis

xpression of UCP

Glucose Uptake

White adipose tissue

Skeletal Musc

Shivering thermoge

Lipid metabolism

Glucose metaboli

Activation of AMP-Ki

Expression of U

electromyogram

Glucose Uptak

tranasal infusion of Galanin-Like Peptide (GALP)

3 male ICR mice (4 month old)

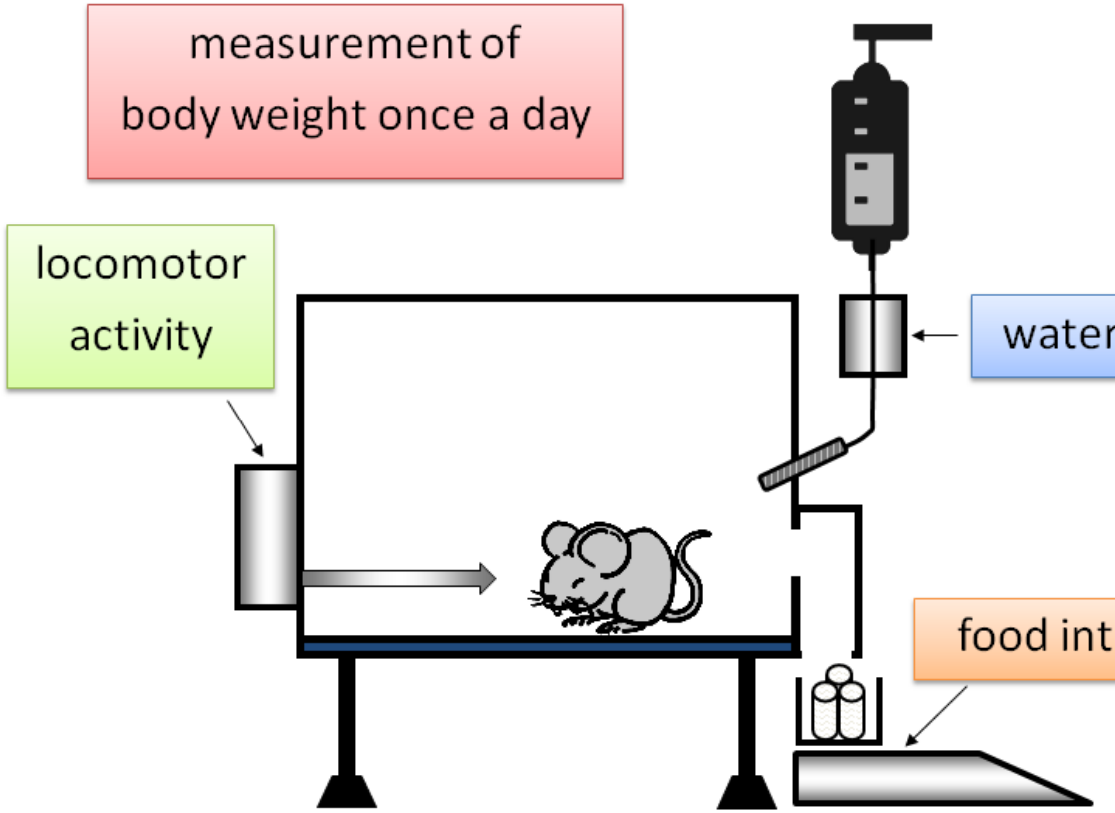
anesthetized with pentobarbital (i.p.)

intranasal administration (i.a.) of

ALP 2 nmol with alpha-cyclodextrin / 2µl

or

saline with alpha-cyclodextrin / 2µl



15:00 (0hr)
GALP or
saline i.a.

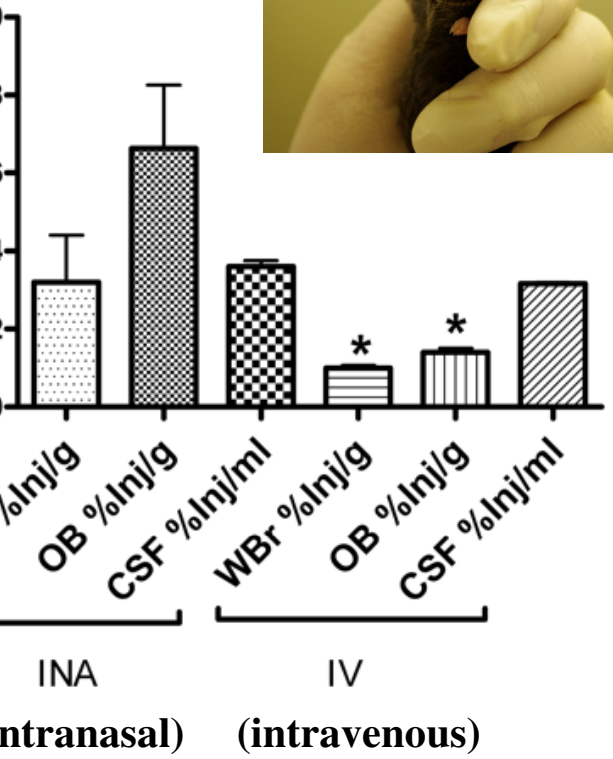
15:00 (24hr)
measurement
of body weight

15:00 (48hr)
measurement
of body weight

15:00 (72hr)
measuremen
of body weigh

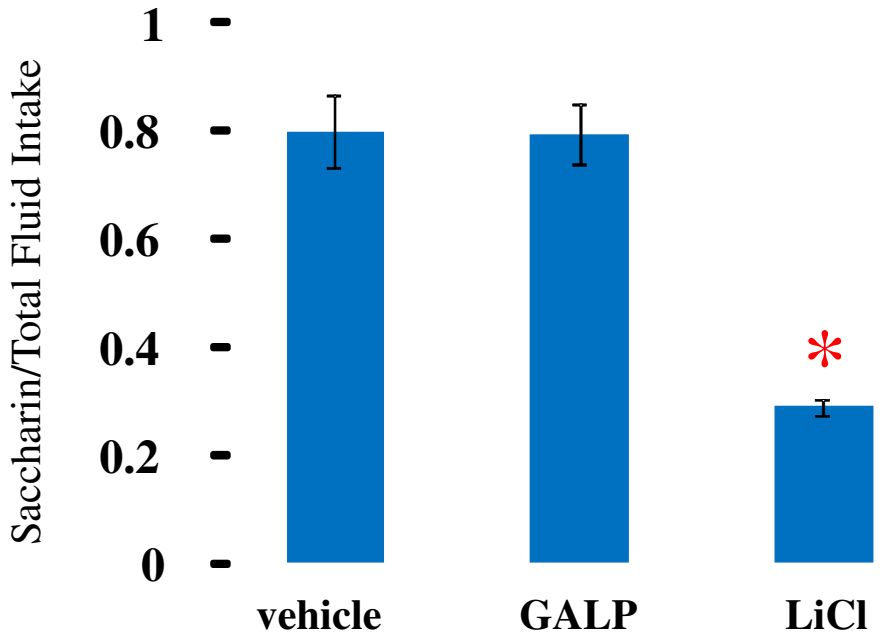
15:00 -20:00	20:00 -8:00	8:00 -20:00	20:00 -8:00	8:00 -20:00	20:00 -8:00	8:00 -15:00

P was higher in the brain
n. administration than i.v.
stration



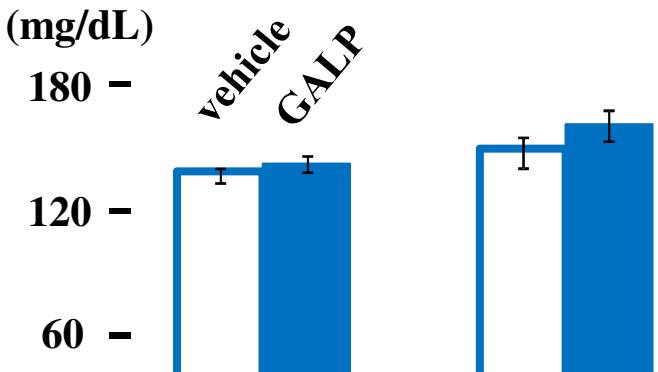
ke into brain of ^{131}I-GALP
ranasal > intravenous

Conditioned taste aversion study

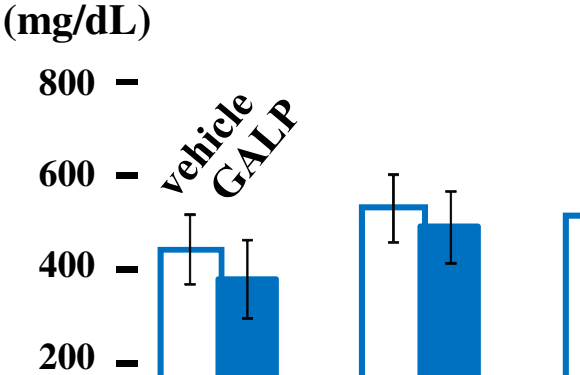


No change in blood glucose levels by GALP i.n. administration

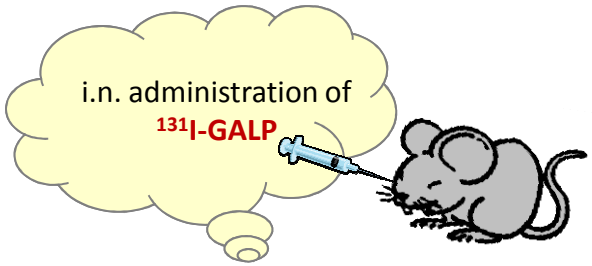
Blood glucose

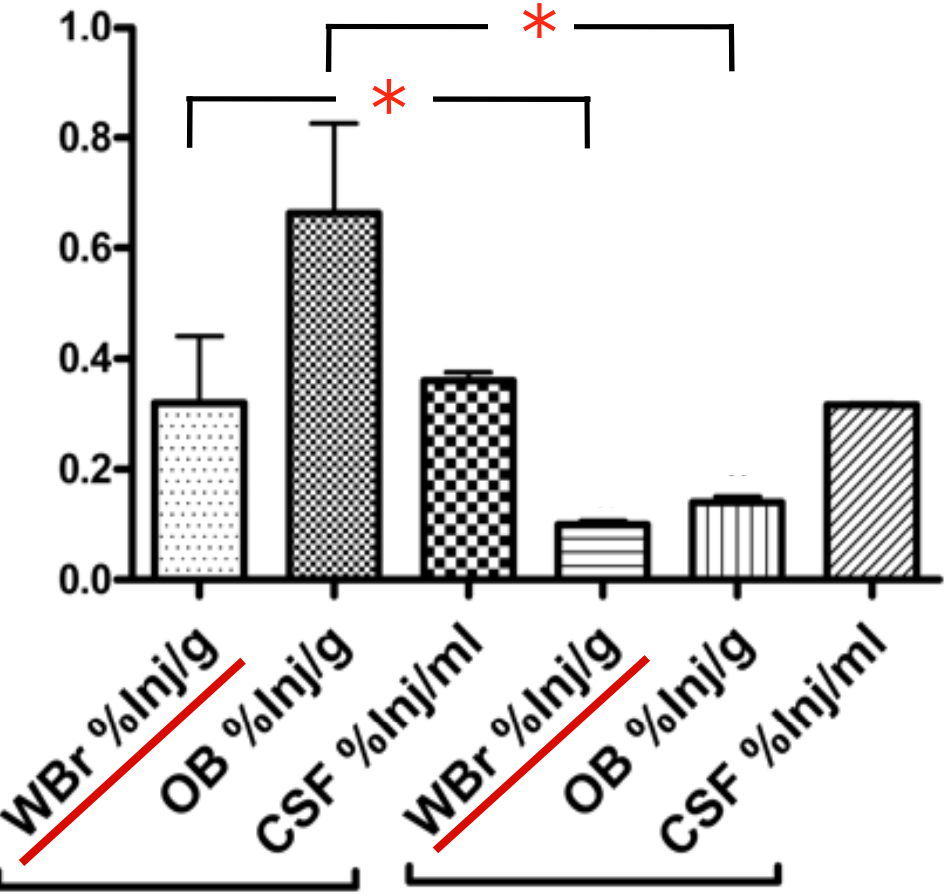


Blood glucos



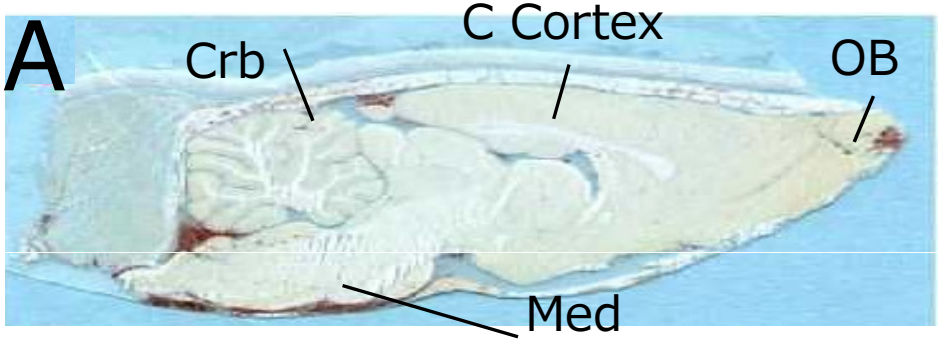
ransport activity of GALP in brain

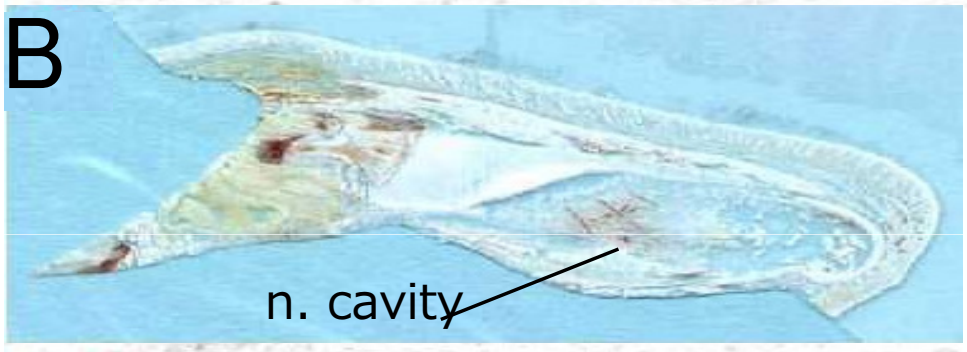


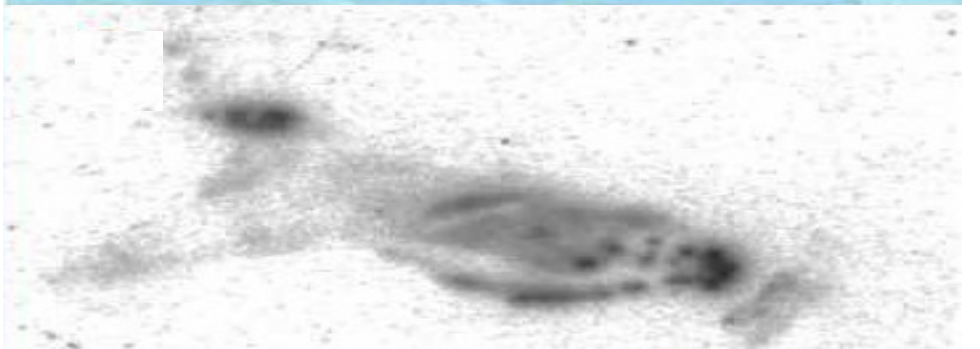


^{131}I-GALP transport activity

GALP is transported from nasal ca to brain capillary

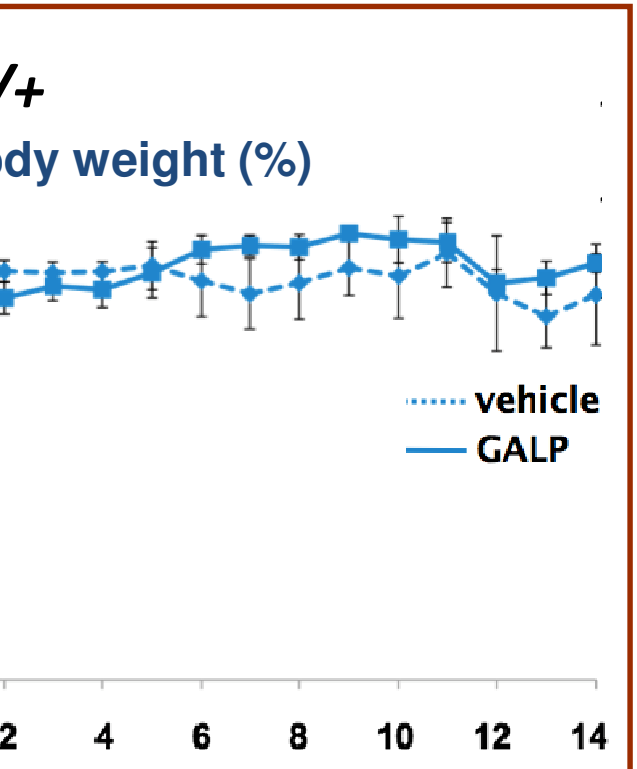


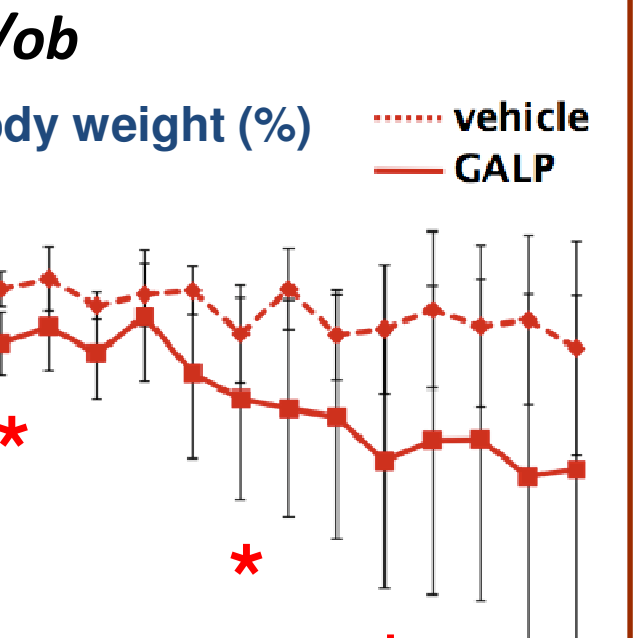


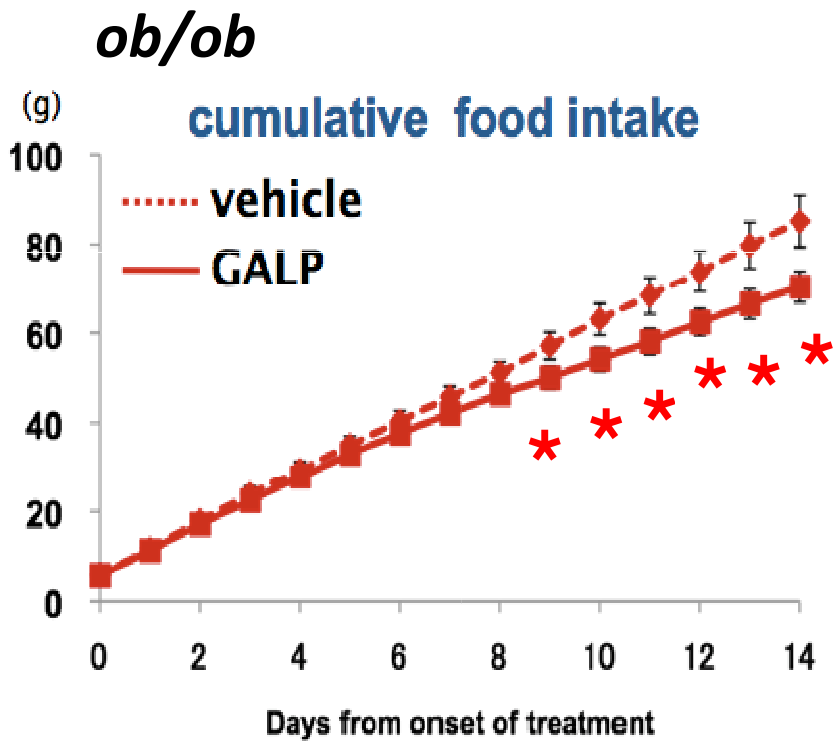


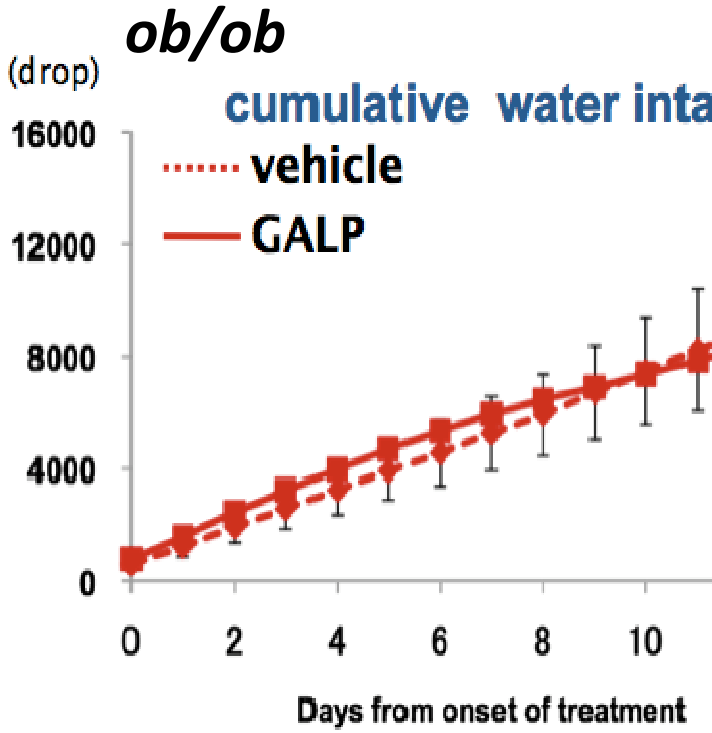
nasal treatment

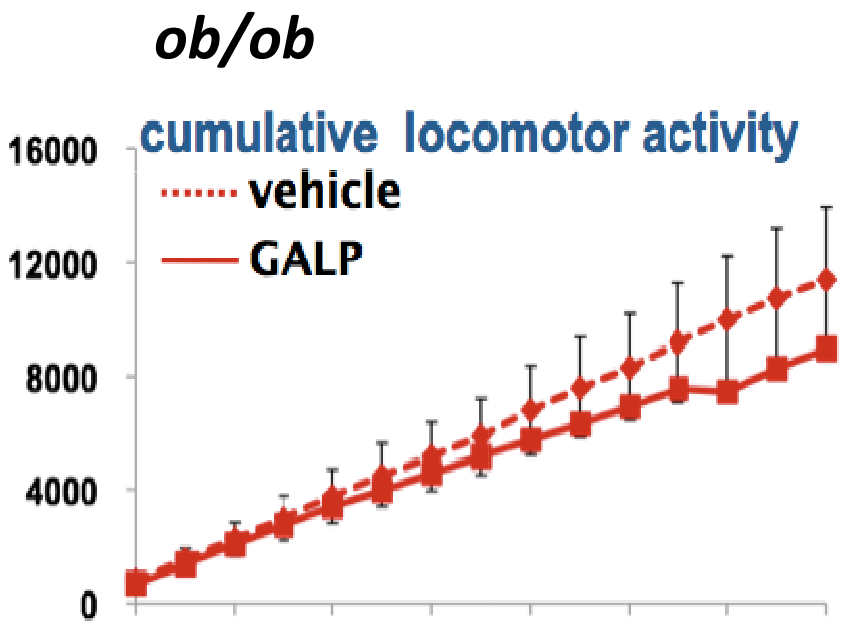
No anesthesia daily single inj. 19:00 2 weeks
vehicle or GALP (2 nmol) 2 µl with alpha-cyclodextrin

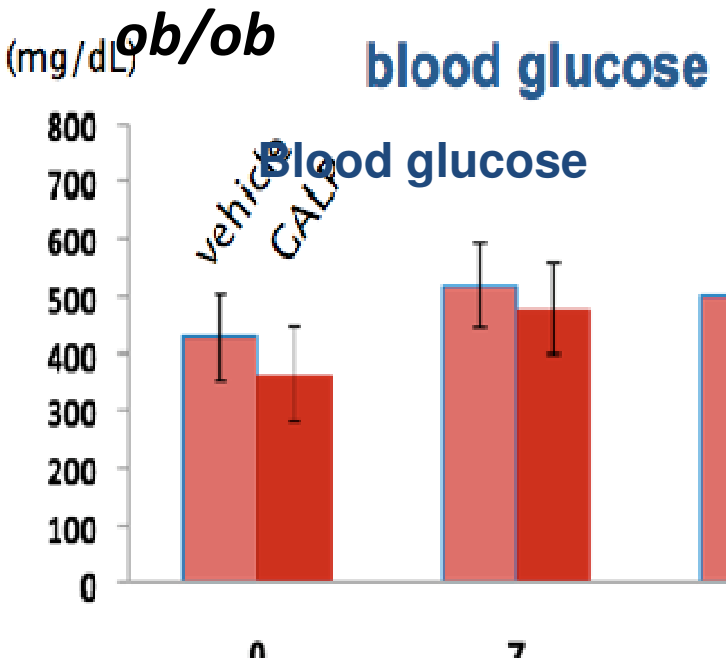












mals : DIO mice （18 weeks ♂ 22 wks n=7)

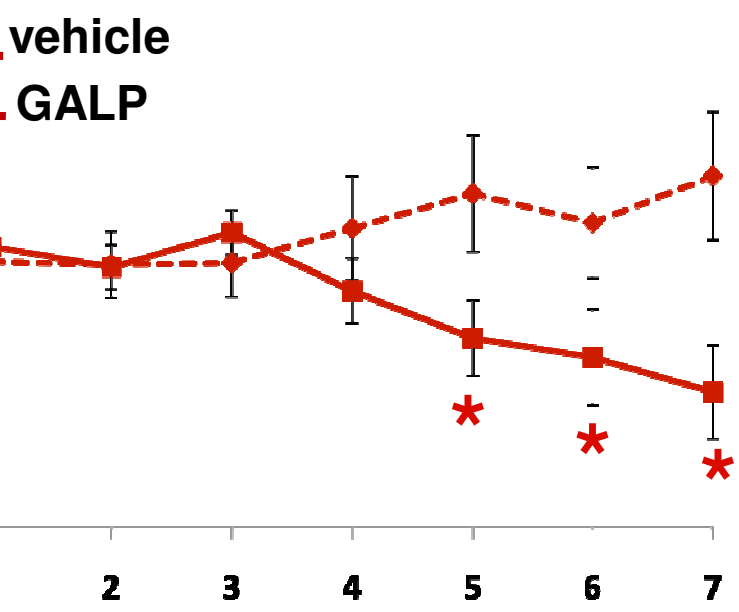
hice fed high fat chow (45% kcal)

4 ± 1.4 g, Blood glucose level : 135 ± 3.6 mg/dL)

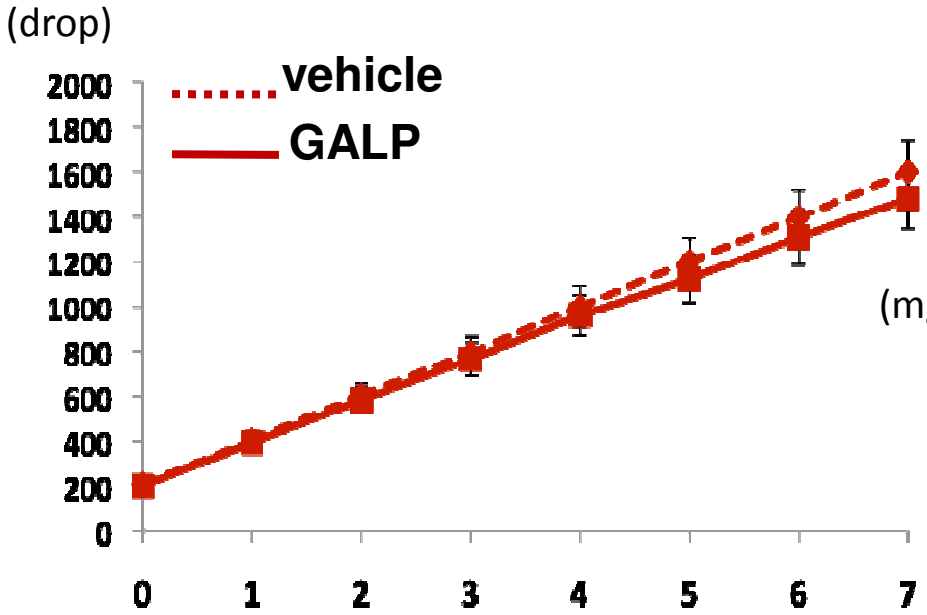
No anesthesia Daily inj. 19:00 7 days i.n.

vehicle or GALP (2 nmol) 2 μl with alpha-cyclodextrin

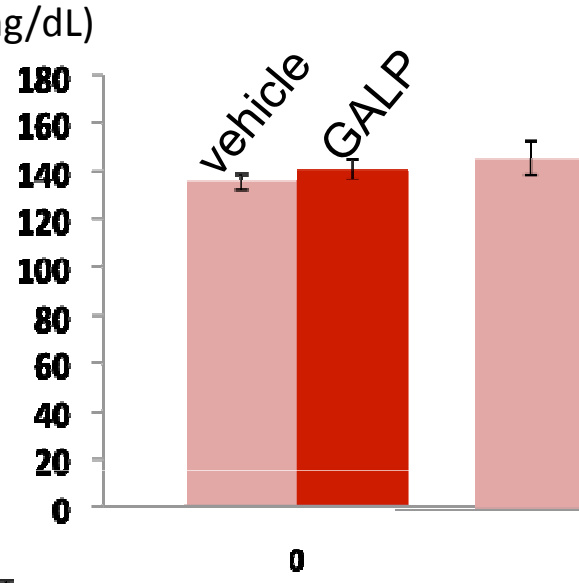
weight (%)



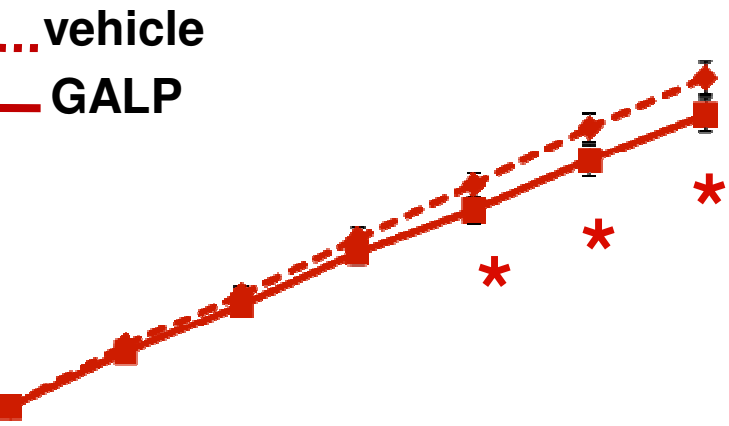
cumulative water intake



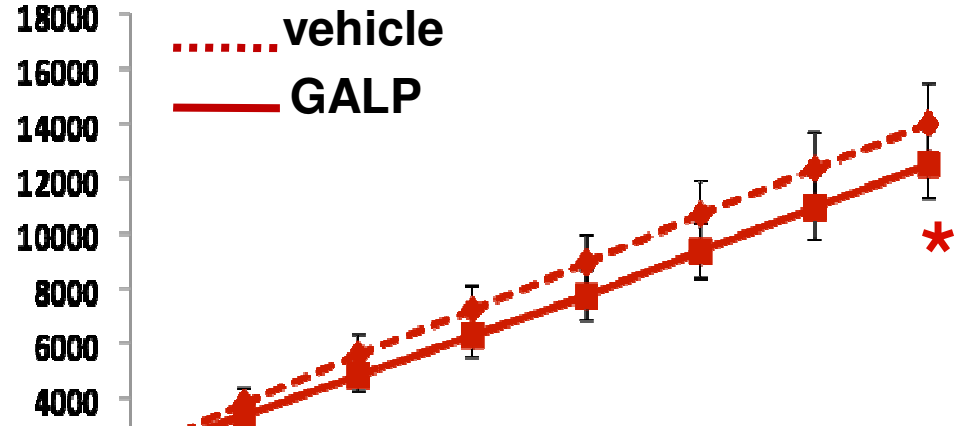
Blood glucos



mulative food intake



locomotor activity



GALP

Fat
metabolism ↑
？？

Summary

ntranasal infusion (i.n.) is more effective (3–5 times) than intravenous infusion

GALP (i.n.) is shown to transported from nasal cavity to blood capillaries in bra

GALP (i.n.) significantly decreases body weigh and food intake especially in ob/
rather than wild type animals

GALP (i.n.) has an anti-obesity effect on DIO mice

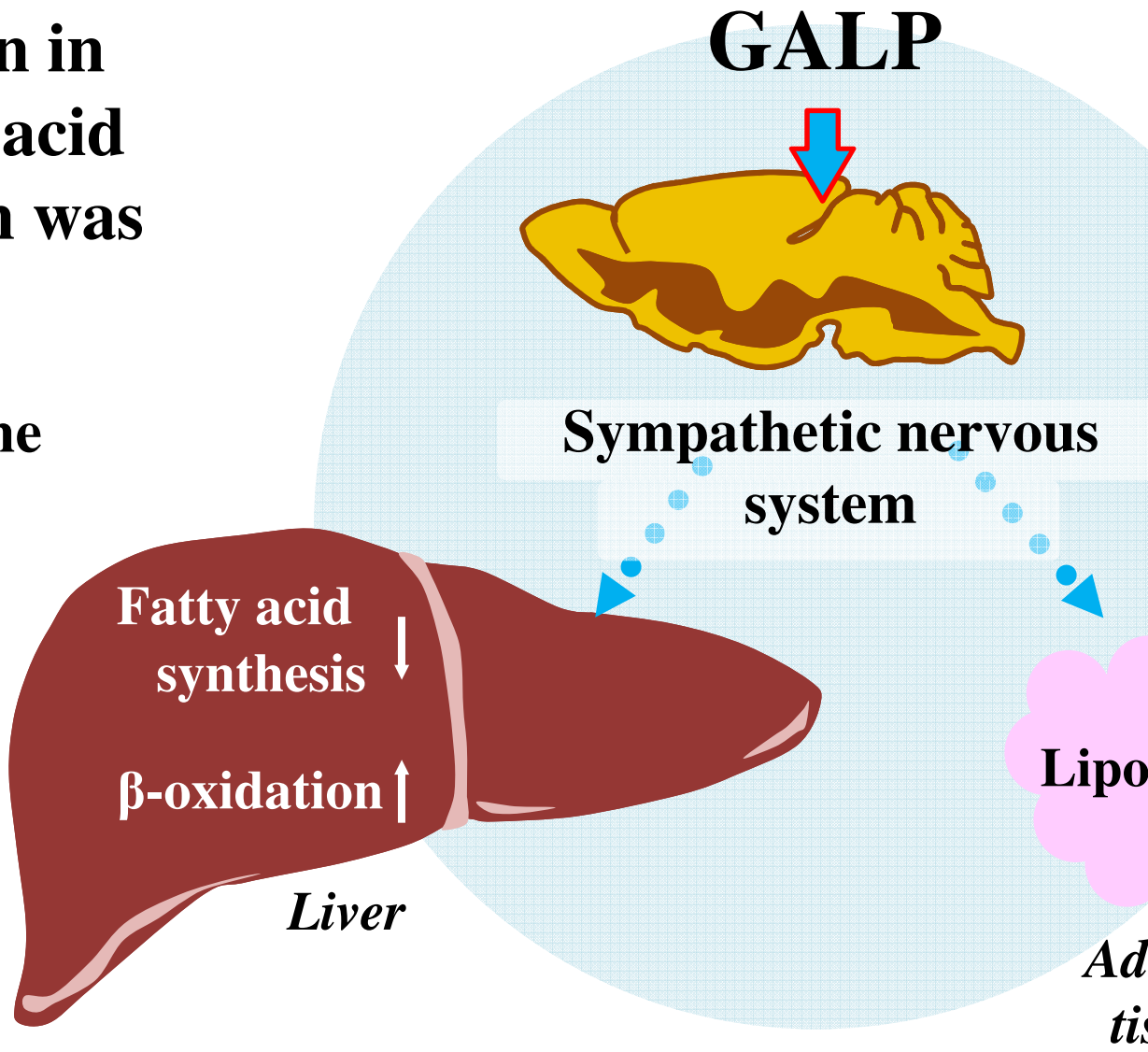
GALP (i.n.) does not induce visceral discomfort or unpleasantness

GALP may play very important role in lipid metabolism

Lipogenic-related gene expression in
he liver is reduced and the fatty acid
oxidation-related gene expression was
ncreased.

Lipolysis-related gene expression in the
WAT is increased.

The effect of GALP was
anceled by the blocking of the
ympathetic nervous system.



P stimulates the hepatic lipid metabolism
ncreasing the lipolysis in WAT and anti-
effect of GALP may be caused by

Anti obesity effec

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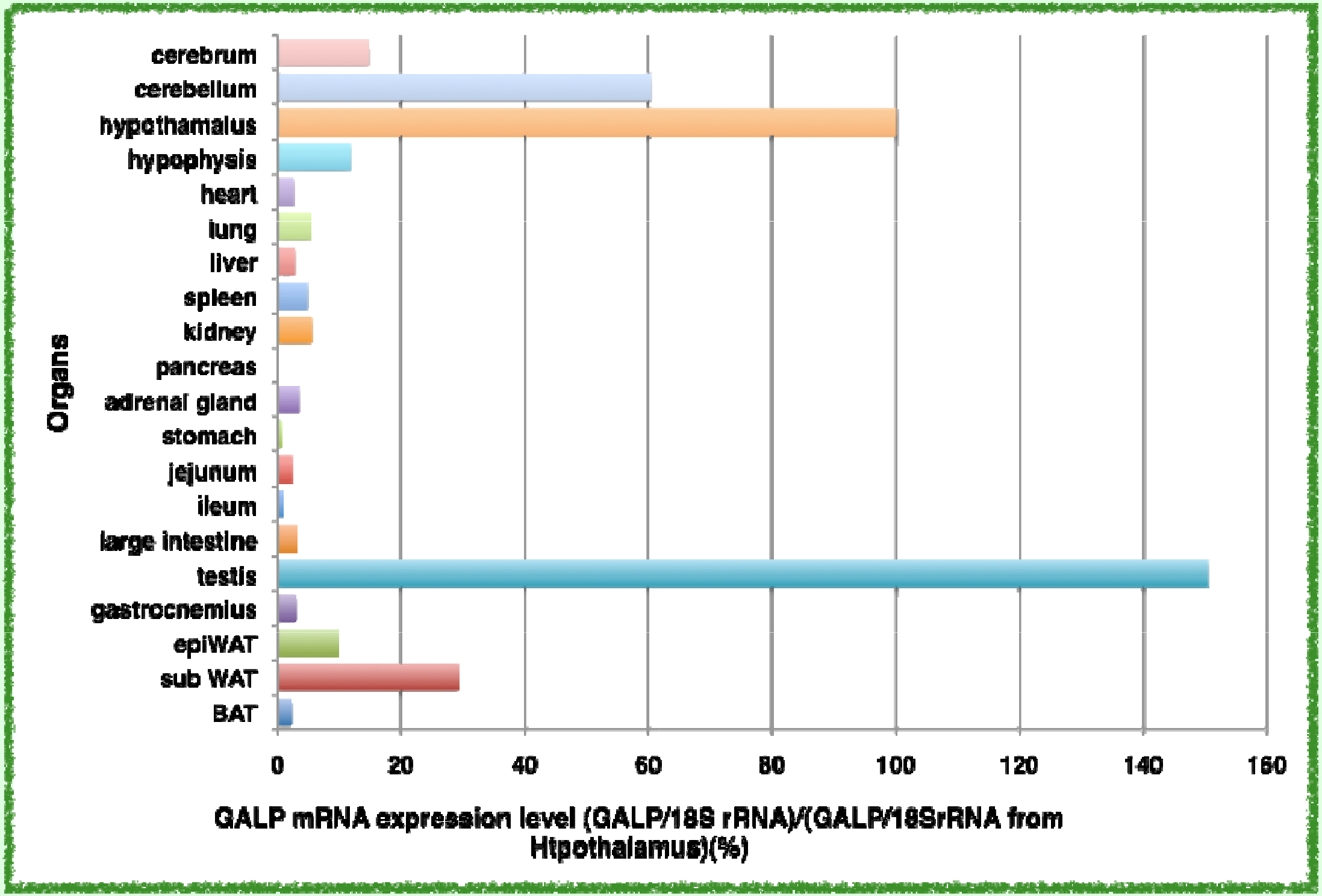
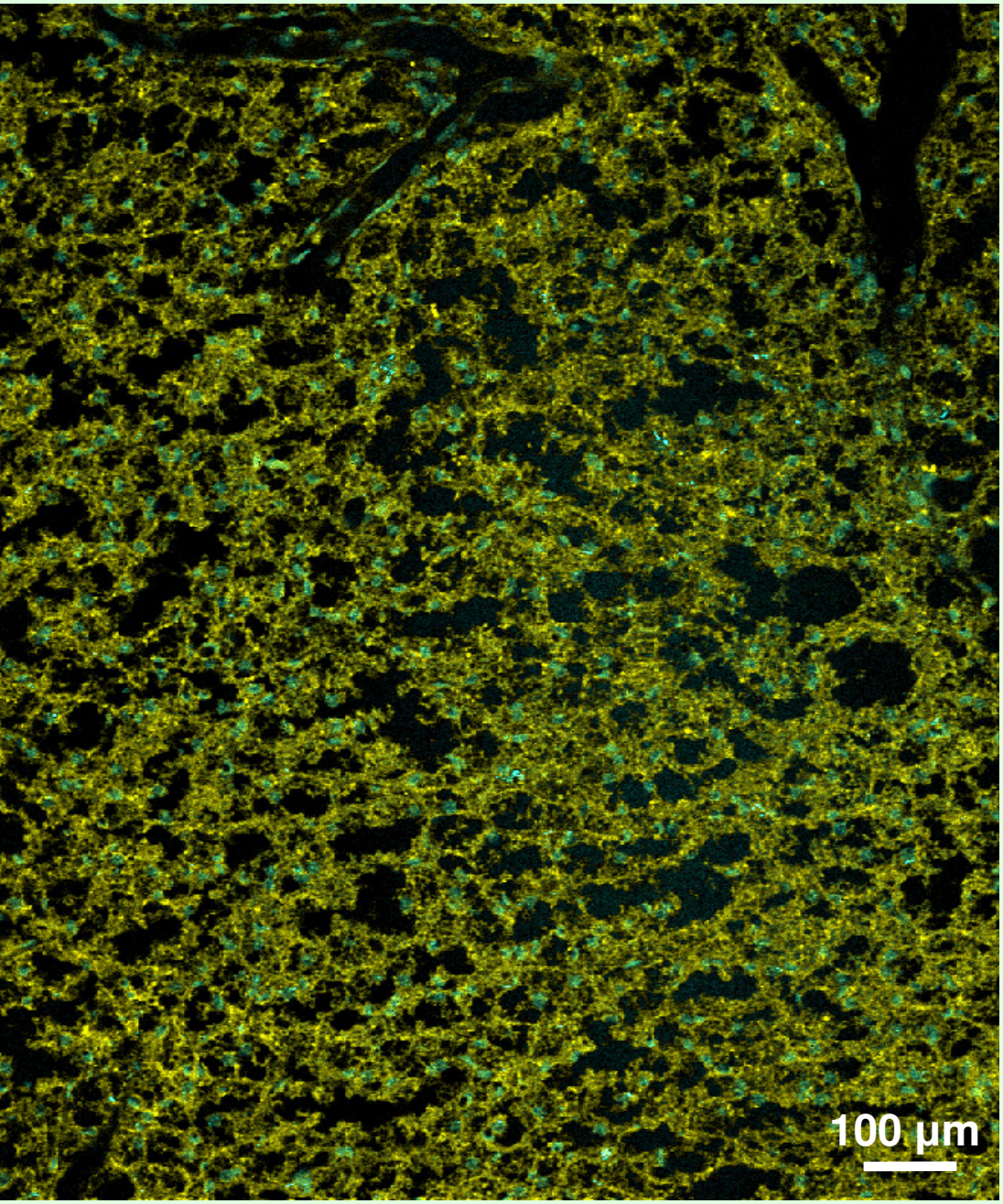
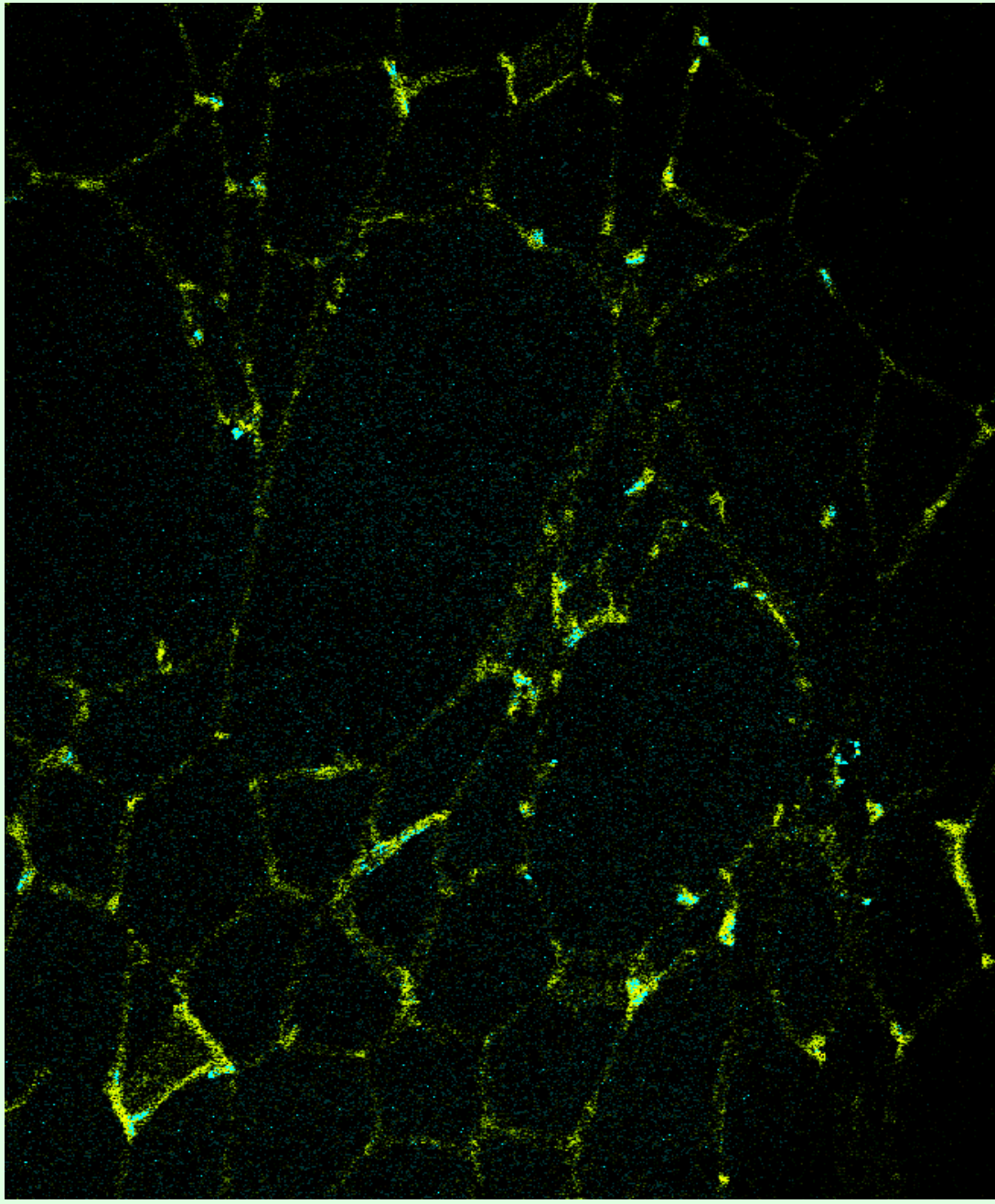

cerebrum
cerebellum
hypothamalus
hypophysis
heart
lung
liver
spleen
kidney
pancreas
adrenal gland
stomach
jejunum
ileum
large intestine
testis
gastrocnemius
epiWAT
sub WAT
BAT
Organs
0
20
40
60
80
100
120
140
160
GALP mRNA expression level (GALP/18S rRNA)/(GALP/18SrRNA from Htpothalamus)(%)

mouse carrying GALP-CreER/CAG-LNL-Venus gene

BAT



epididymal WAT



Blue: