Integrity of the Blood Brain Barrier and number of CA1 Neurons after chronic GSM and UMTS radiation

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Objective - Hypothesis

- The combined work and the inter-disciplinary competence of several scientific institutes of two Munich universities suggest that the analysis of behavioural, immunological, and blood-brain-barrier effects in three generations of rats should be capable of detecting even subtle functional consequences of long term exposure to low-level EMFs typical for mobile communication.
- The results of this study are expected to help discussing the issue whether non-thermal EMFs of mobile communication do present a health hazard to man or not

Study design

Detection of

Cognitive,

Immunological,

Stress and

Blood-brain-barrier effects

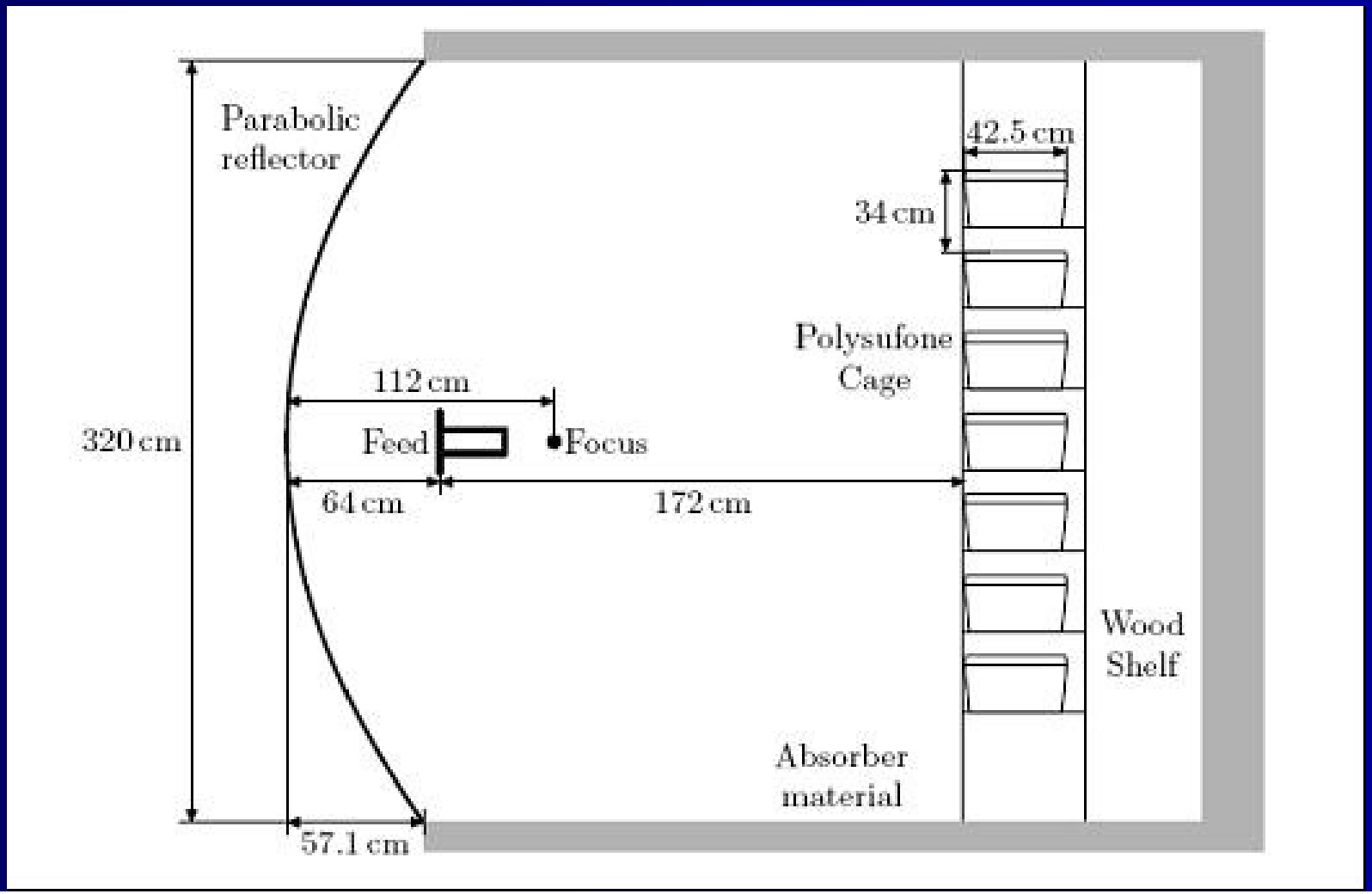
in three generations of rats induced by chronic exposure to GSM 900 MHz or UMTS 1800 MHz electromagnetic fields 0.4 W/kg SAR of mobile communication.

Radiation exposure

we exposed simultaneously groups of 120 WISTAR rats each in three specially designed shielded chambers (370 x 370 x 320 cm^3) for either GSM 900 MHz, UTMS 1800 MHz, or SHAM-exposure (SAR 0.4 W/kg)

a volume of about 2.5 m^3 exposure zones with plane-wave conditions: maximum field deviation +/- 15% in magnitude and +/- 5° in phase

Side view of the exposure setup



Exposure setup

Polycarbonate animal cages

Rats

WISTAR rats obtained from Charles River, Germany.

Animal care, 1:1 mating, culling of litters, rearing, etc. followed GLP rules.

Adult subjects were housed in groups of two or three in standard polycarbonate animal cages (425x266x240 mm^3) with non-metallic covers.

Double-blind study logistics require animal coding by subcutaneous implantation of HF transponders. In order to minimize inter-individual variability, animal groups of the F0 generation will consist of previously selected subjects. Selection parameters are determined by operant-behavior performance. Only animal performances corresponding to group performance means were chosen.

Rat Generations

Juli	Parentes	1 week habituation, selection, radiation, breeding
Jan Feb	F_0-Gen.Gr.1	BBB: 75 animals, after 4 month radiation
Sept Okt	F_0-Gen.Gr.2	BBB: 72 animals, after 11 month radiation
	F_1-Gen.	
Dez Jan	F_2-Gen.	BBB: 82 animals, after 4 month radiation

Assessment of blood-brain-barrier (BBB) function

- A BBB opening after EMF-exposure was described in numerous investigations. On the other hand, there are studies that could not find any BBB effects.
- Since the degree of a possible EMF-induced BBB opening is most probably minimal and of short duration, these effects should have only minor neuropathological relevance.
- In order to differentiate more clearly between marginal disturbances and eventual persisting EMF-induced BBB changes, we enhanced the neuropathological relevance of our experimental results by BBB labilization challenges and a quantitative method of detecting radioactive-labeled substance transfer rates.

Measurement of BBB integrity as unidirectional influx constant K_{in}

$$K_{in} = \frac{C_{br} - C_i}{\int_0^T C_{pl}(t)\,dt} \quad \left[\frac{\mu l}{g * \min}\right]$$

C_{br} = marker-radioactivity in brain tissue dpm/g

C_{pl} = marker-radioactivity in blood plasma dpm/µl

C_i = marker-radioactivity in cerebral blood plasma dpm/µl

T = time point of euthanasia

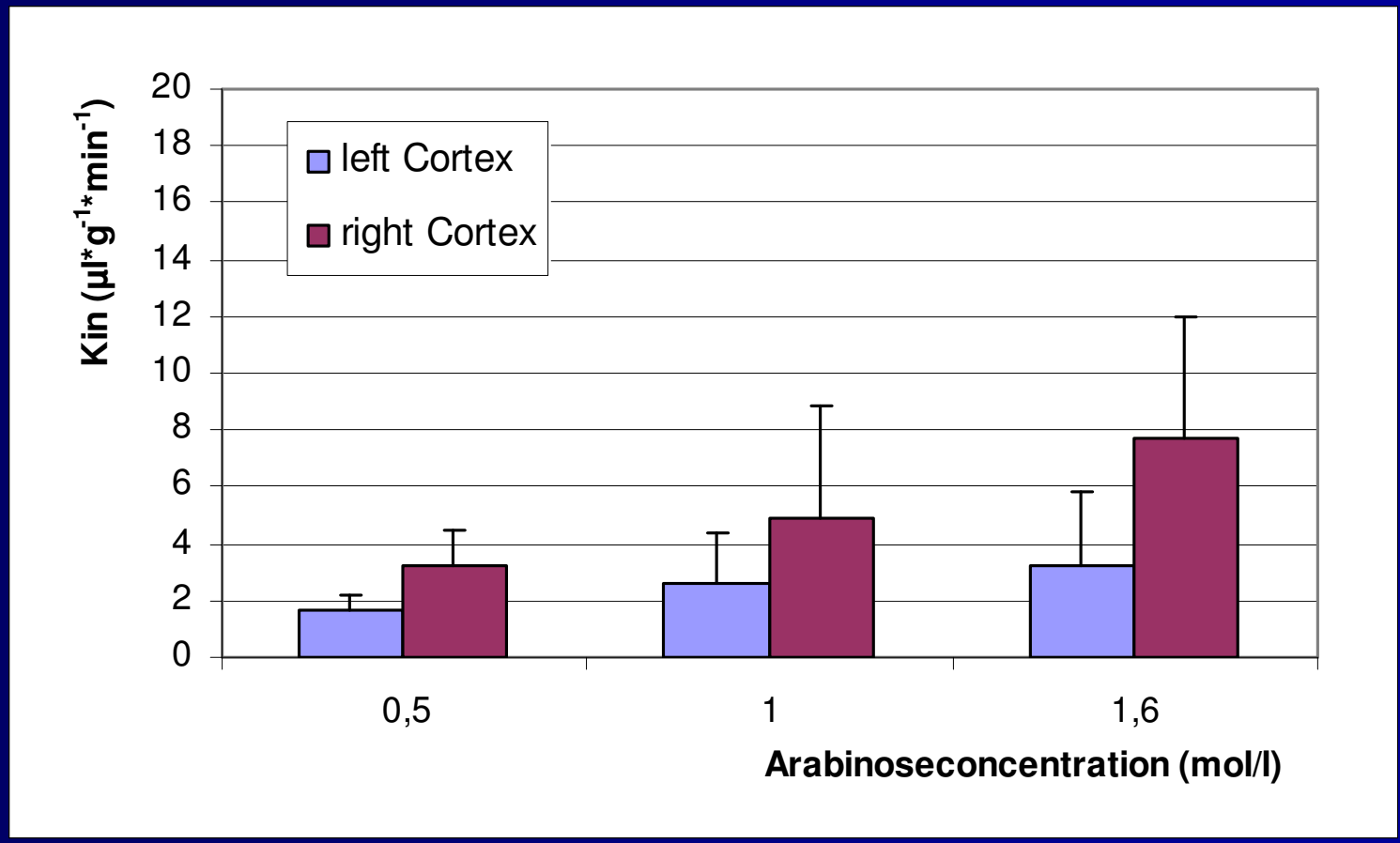
Vascular Space

	^{14}C-Sucrose		^{3}H-Inulin	
	Distribution volume (µl/g) after 1 minute	Extrapolated volume (µl/g)	Distribution volume (µl/g) after 1 minute	Extrapolated volume (µl/g)
Cortex	12.5 ± 1.3	11.7	10.4 ± 2.1	7.9
Dienecephalon	13.1 ± 2.9	15.2	12.7 ± 2.9	12.8
Mesencephalon	14.7 ± 5.0	13.8	12.4 ± 2.7	8.3
Pons	20.5 ± 5.7	17.6	14.3 ± 5.1	13.3
Medulla	21.9 ± 4.5	19.2	17.1 ± 6.5	17.4
Cerebellum	20.8 ± 4.5	19.5	18.4 ± 7.5	13.8
Bulbus	29.4 ± 6.3	24.9	19.6 ± 6.0	13.1

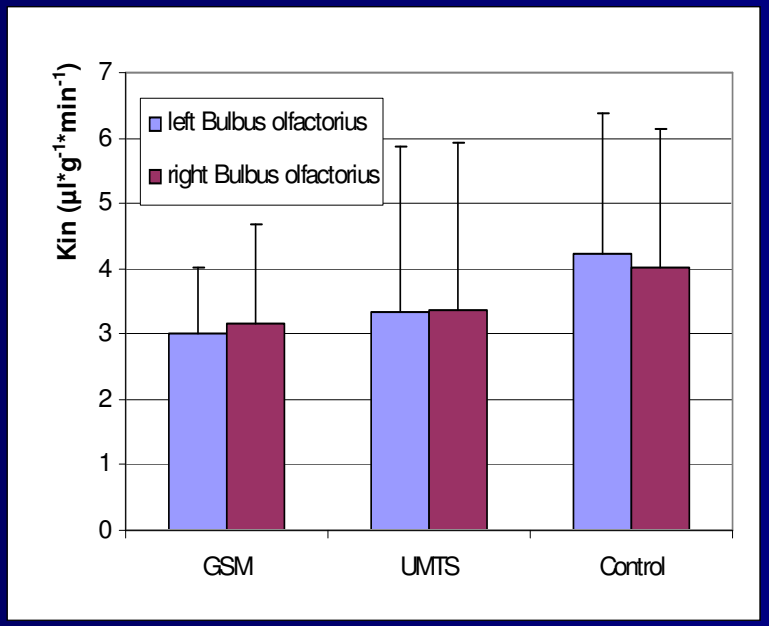
Goal of Challenge conditions

- Challenge conditions were established in order to increase the sensitivity of the method

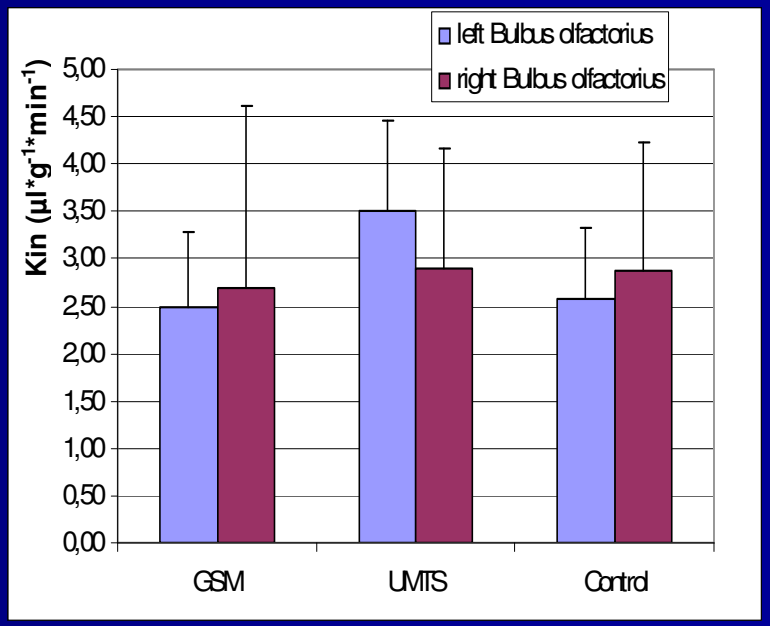
Successful Challenge: Arabinose i.v.



Effects of GSM and UMTS on Kin in Bulbus Olfactorius

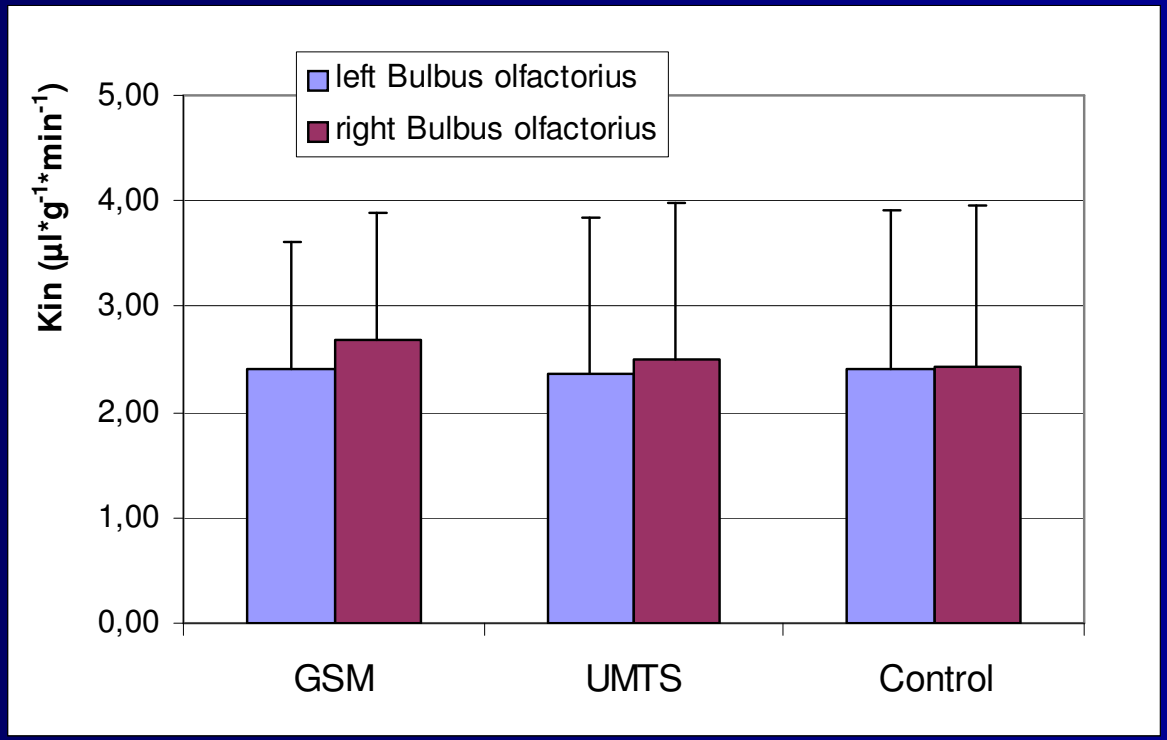


F0 after 4 months



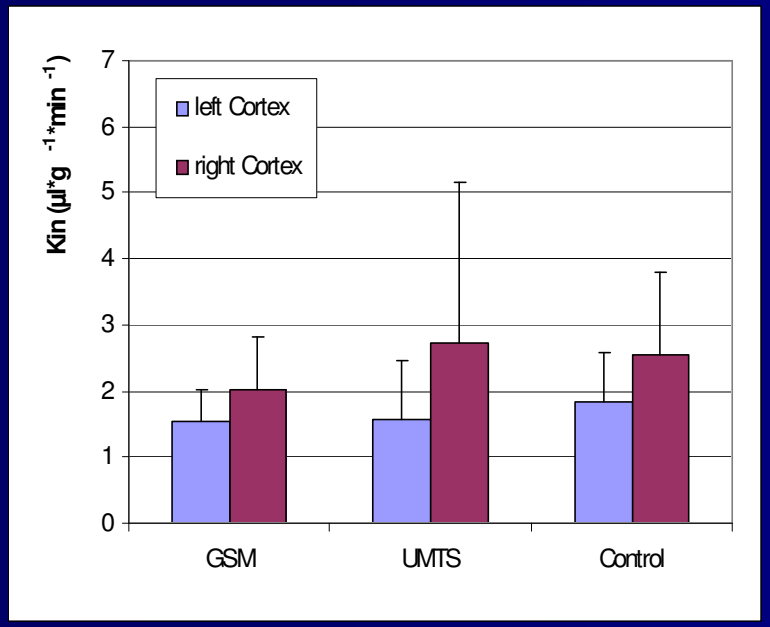
F0 after 11 months

Effects of GSM and UMTS on Kin in Bulbus Olfactorius

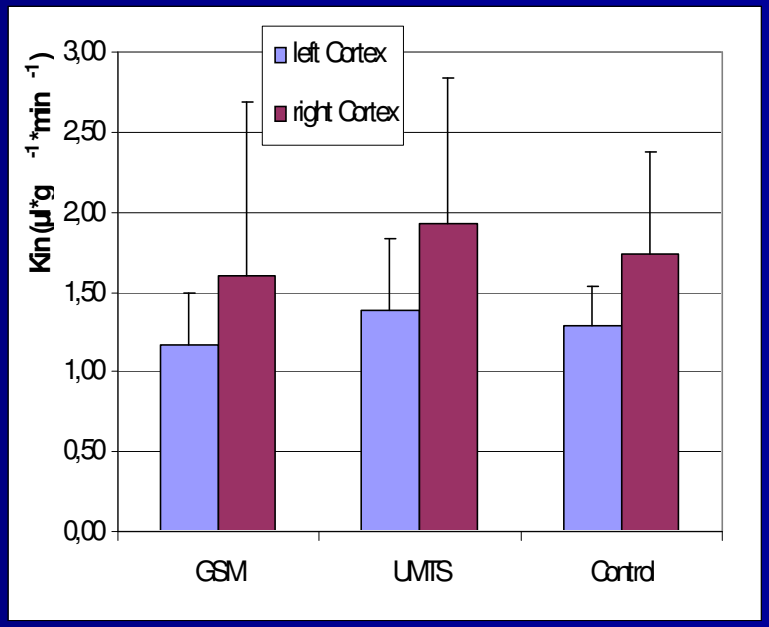


F2 after 4 months

Effects of GSM and UMTS on Kin in Cortex

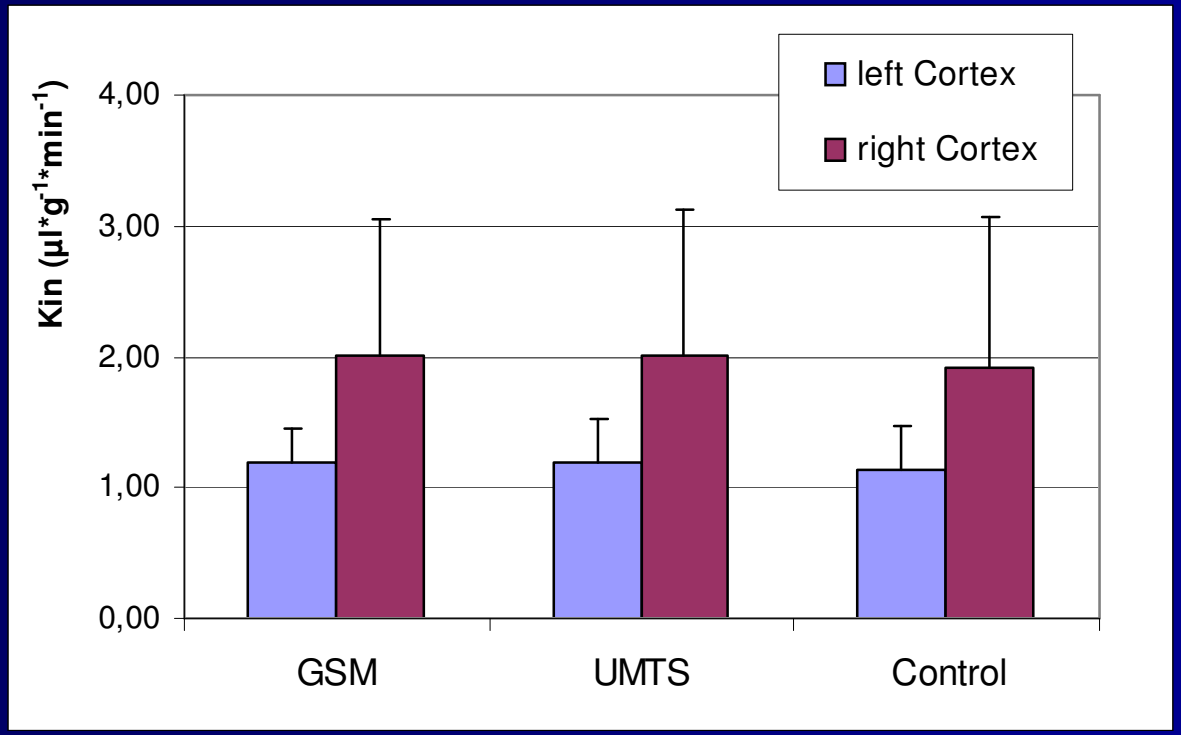


F0 after 4 months



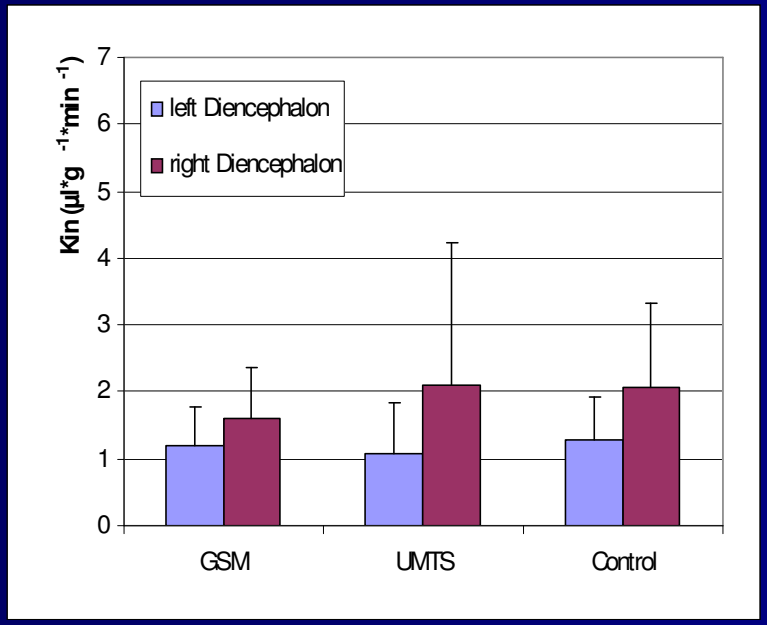
F0 after 11 months

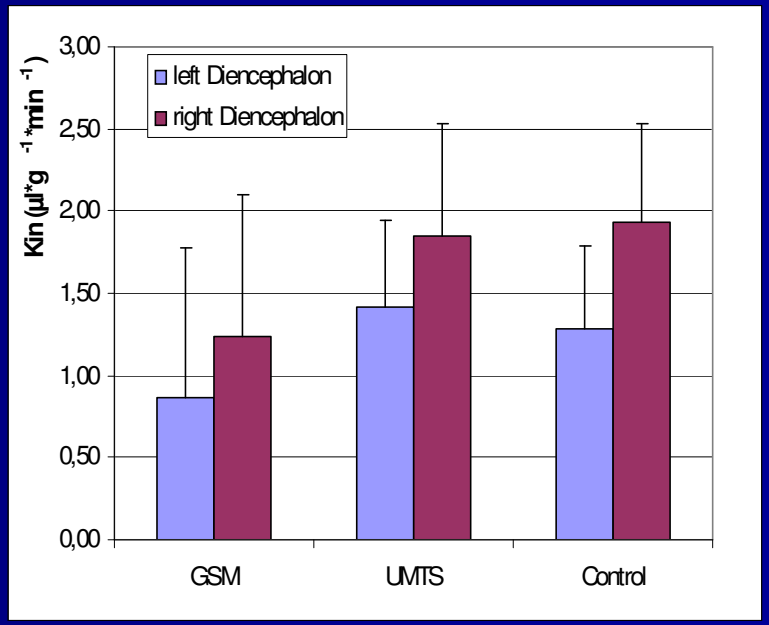
Effects of GSM and UMTS on Kin in Cortex



F2 after 4 months

Effects of GSM and UMTS on Kin in Diencephalon

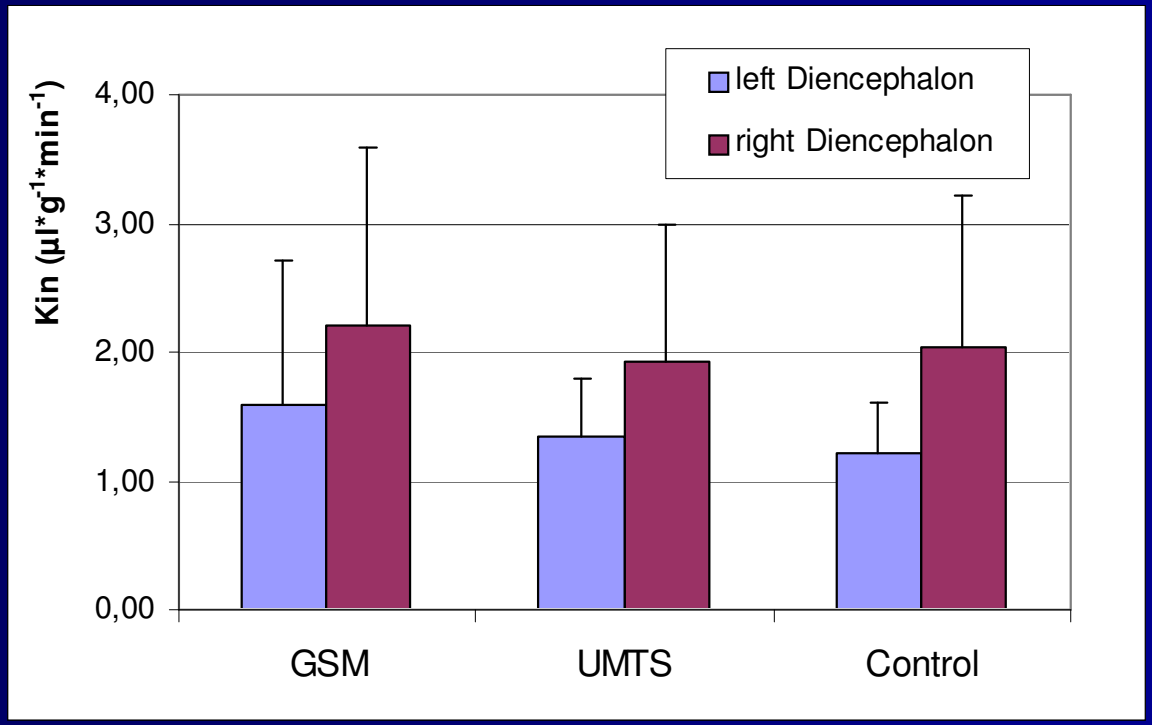




F0 after 4 months

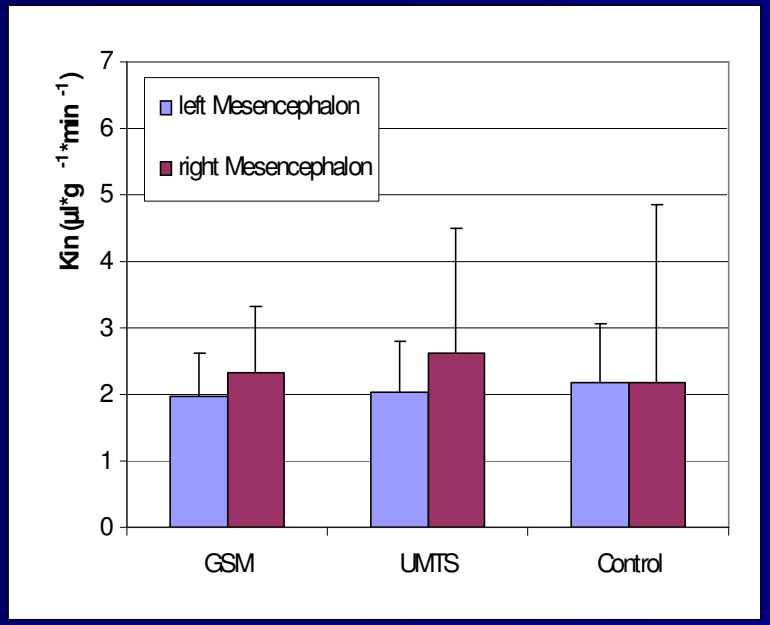
F0 after 11 months

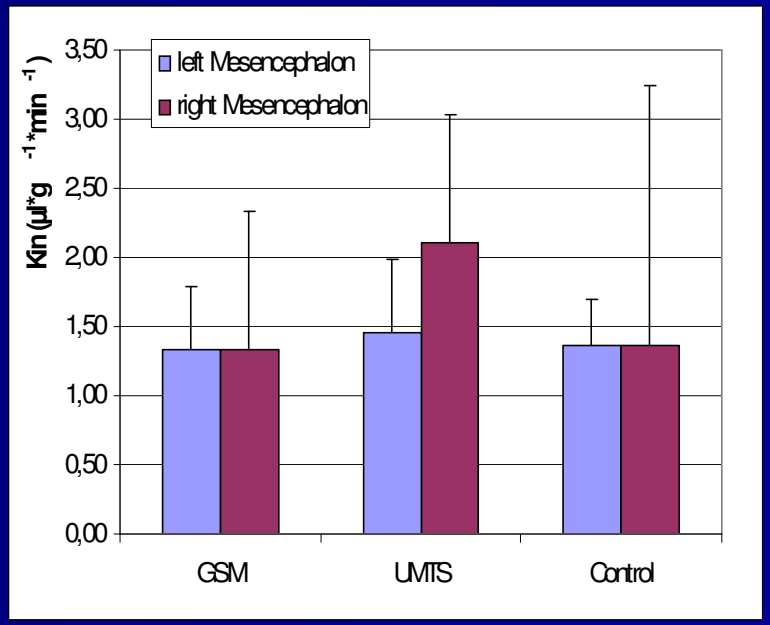
Effects of GSM and UMTS on Kin in Diencephalon



F2 after 4 months

Effects of GSM and UMTS on Kin in Mesencephalon

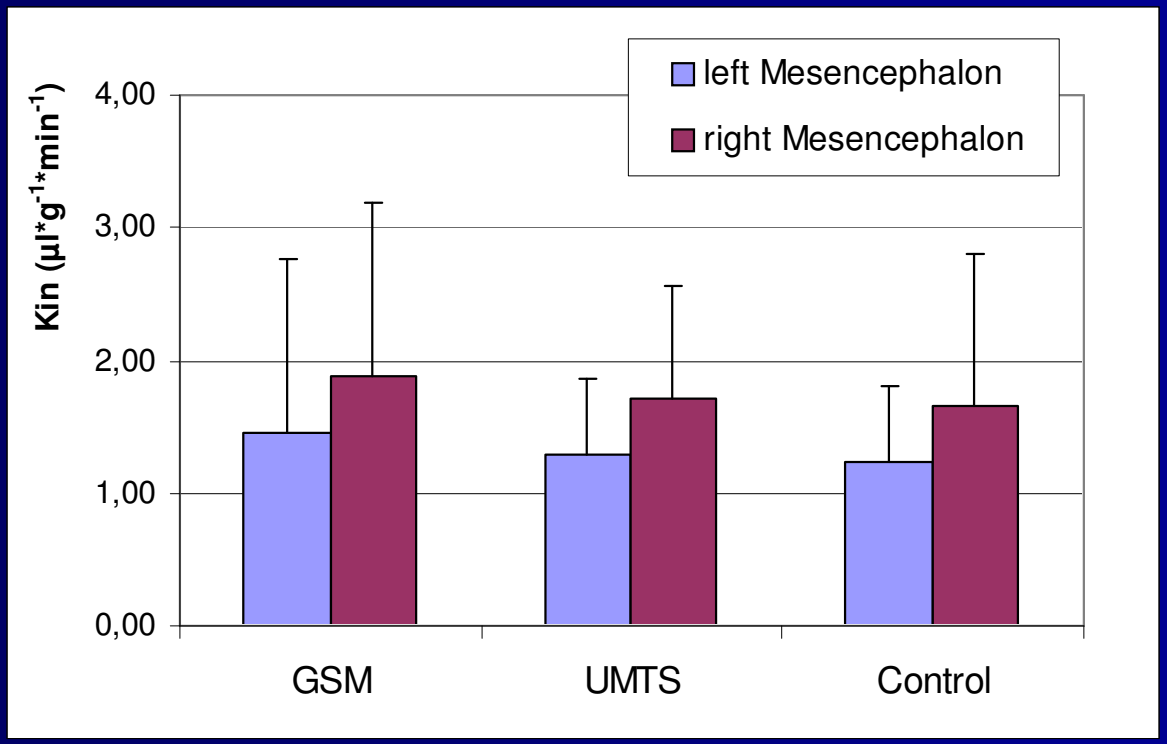




F0 after 4 months

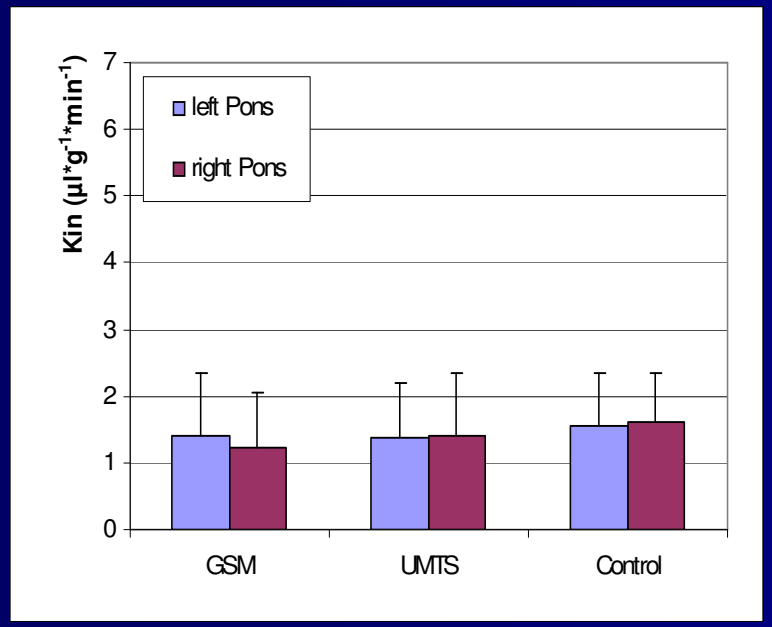
F0 after 11 months

Effects of GSM and UMTS on Kin in Mesencephalon

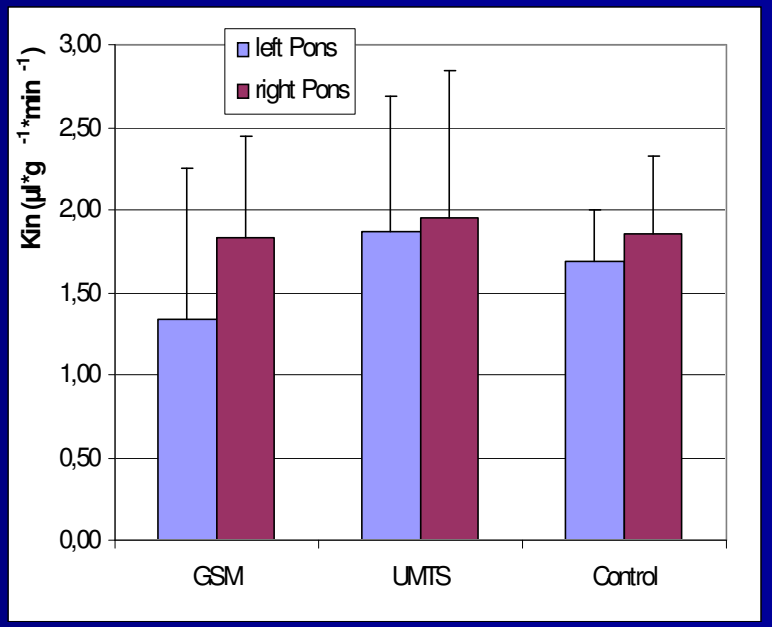


F2 after 4 months

Effects of GSM and UMTS on Kin in Pons

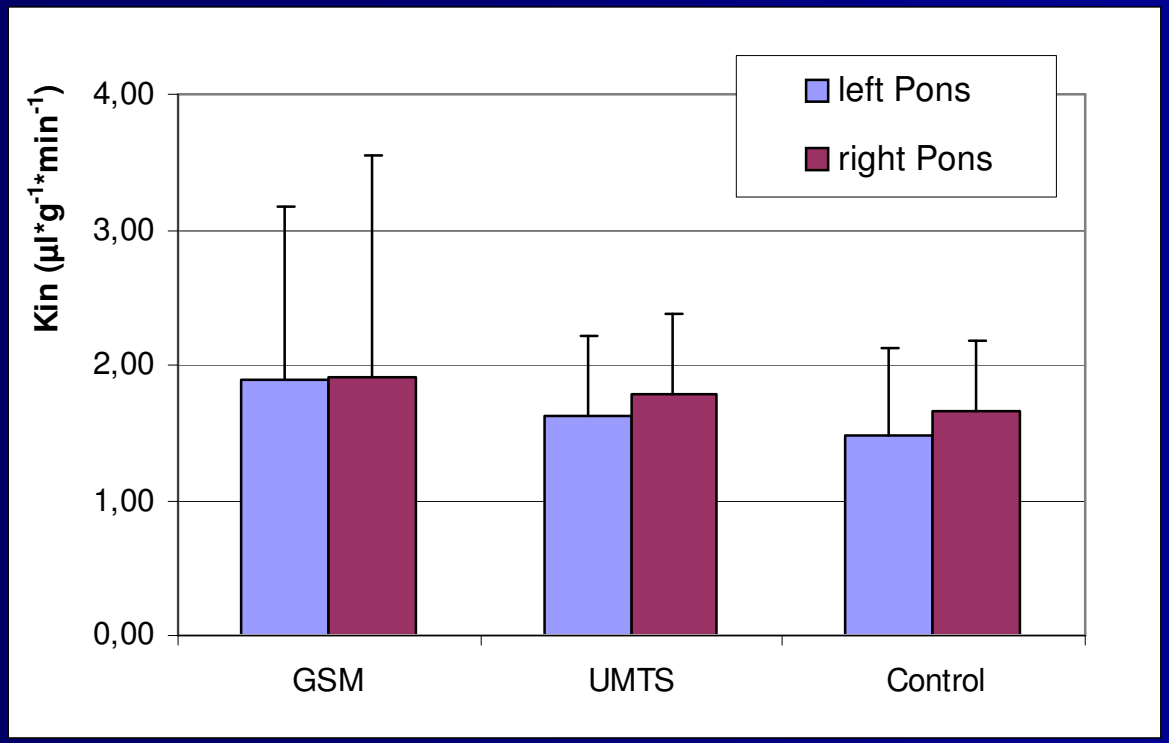


F0 after 4 months



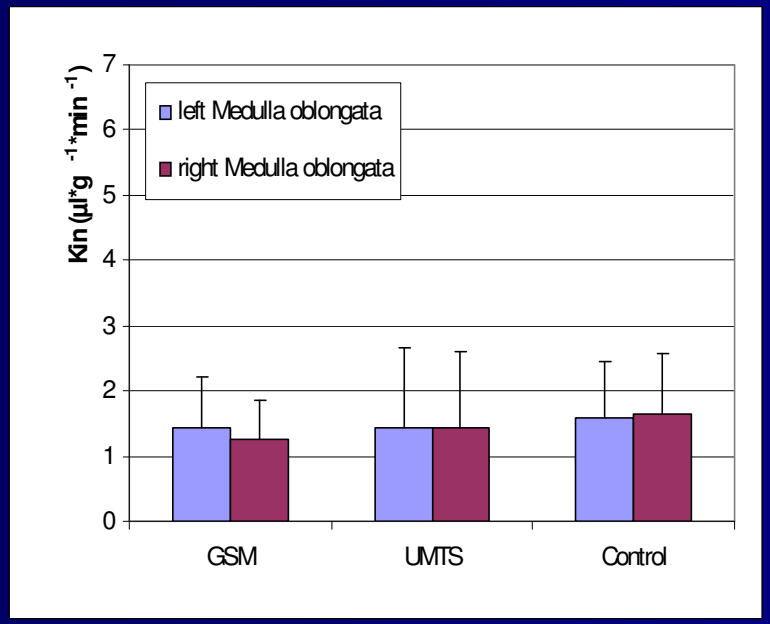
F0 after 11 months

Effects of GSM and UMTS on Kin in Pons

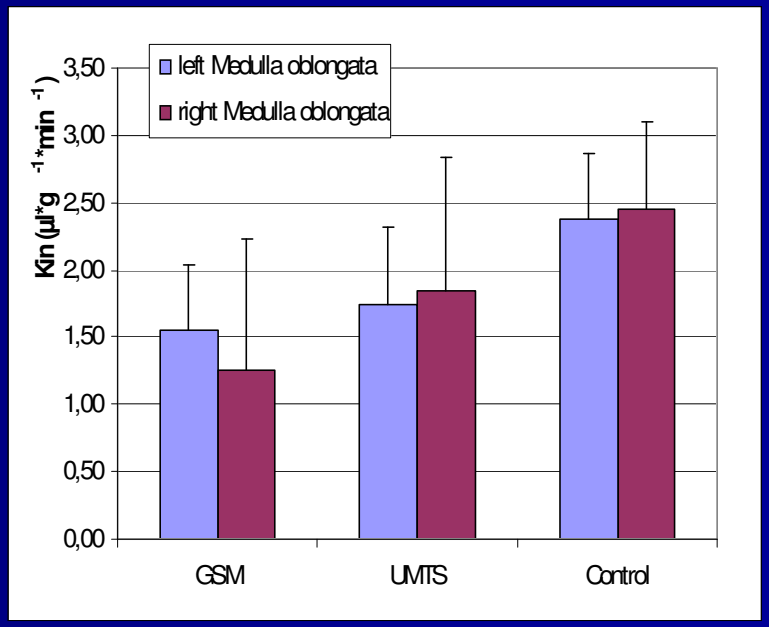


F2 after 4 months

Effects of GSM and UMTS on Kin in Medulla oblongata

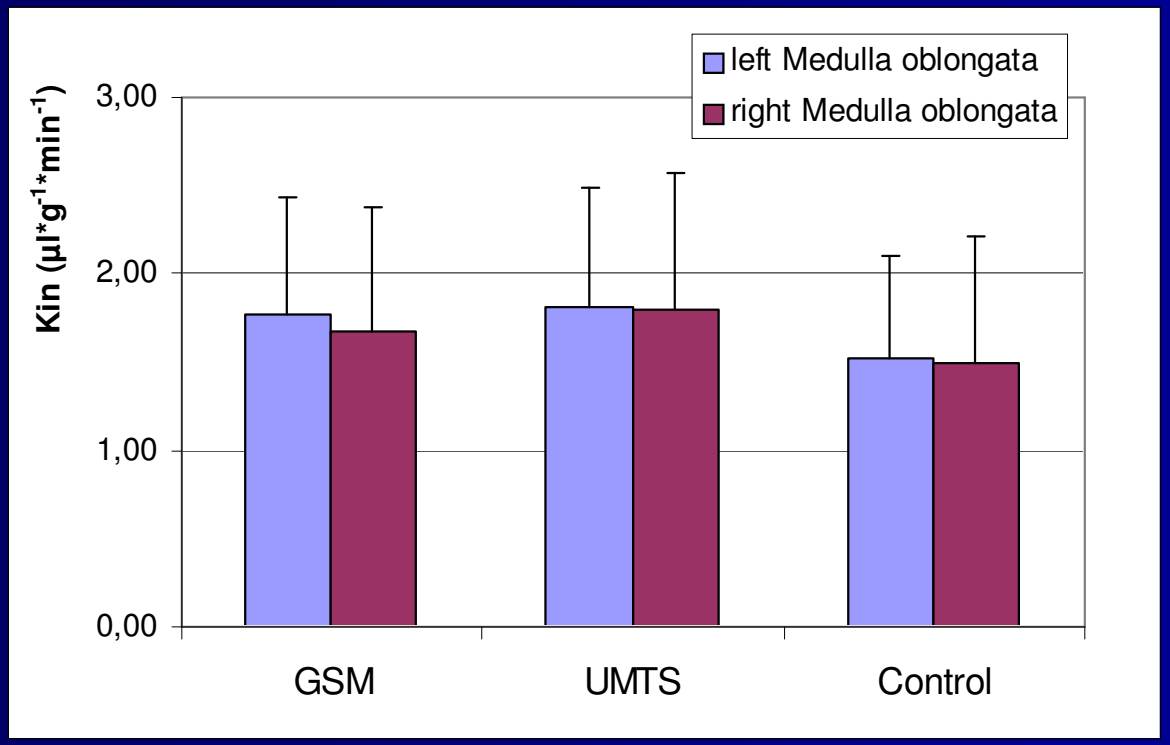


F0 after 4 months



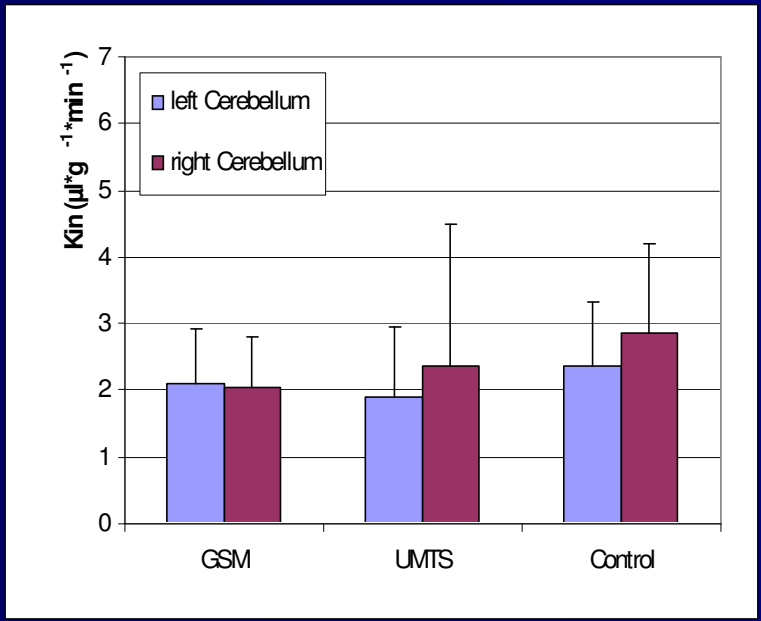
F0 after 11 months

Effects of GSM and UMTS on Kin in Medulla oblongata

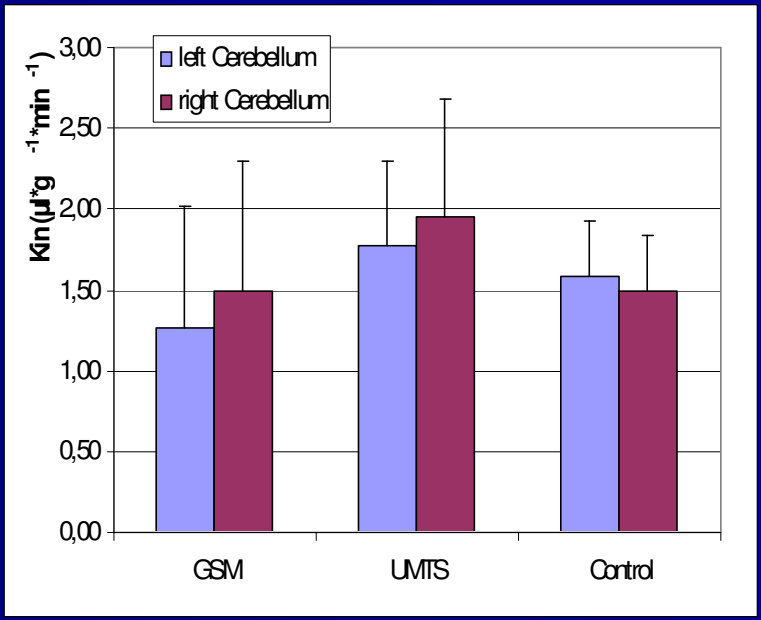


F2 after 4 months

Effects of GSM and UMTS on Kin in Cerebellum

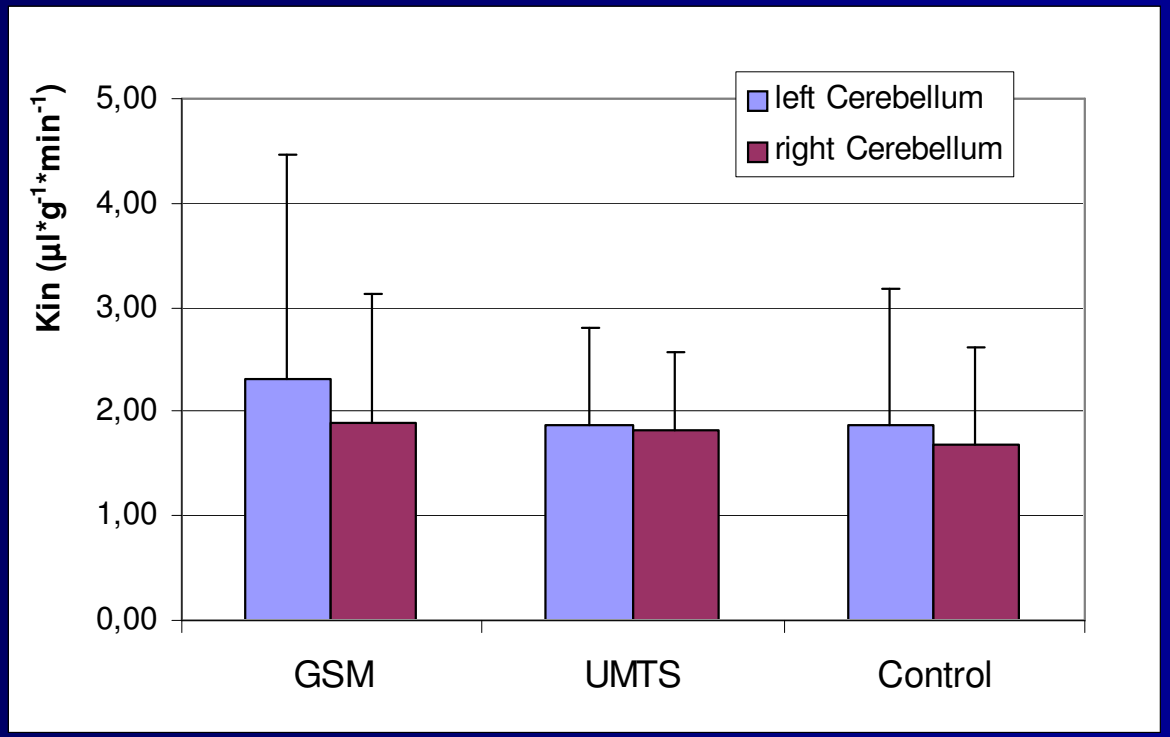


F0 after 4 months



F0 after 11 months

Effects of GSM and UMTS on Kin in Cerebellum



F2 after 4 months

BBB integrity and brain tissue heating

BBB integrity	normotherm = N hypertherm = H	author
+	N	Cosquer et al. 2005
+	N	Franke et al. 2005
+	N	Finnie et al. 2002
+	N	Finnie et al. 2001
+	N	Finnie et al. 2006
+	N	Tsurita et al. 2000
+	N	Gruenau et al. 1982
+	N	Ward et al. 1982
+	N	Preston et al. 1979
+ / -	SAR > 7,5W/kg	Fritze et al. 1997
+ / -	42,5 ℃ 60 min, 44,3 ℃ 30min	Moriyama et al. 1991
+ / -	> 43 ℃	Neilly and Lin 1986
+ / -	> 43 ℃ after 5 min, increasing ffects	Goldman et al. 1984
+ / -	H	Sutton and Carroll 1979
+ / -	42±2 ℃ 9 0min without radiation > 41,5 ℃ 30/90 min, SAR 13W/kg	Williams et al. 1984
+ / -	> 43 ℃	Lin and Lin 1982
-	N	Schirmacher et al 2000
-	N	Persson et al. 1992
-	N	Salford et al. 1994
-	N	Salford et al. 2003

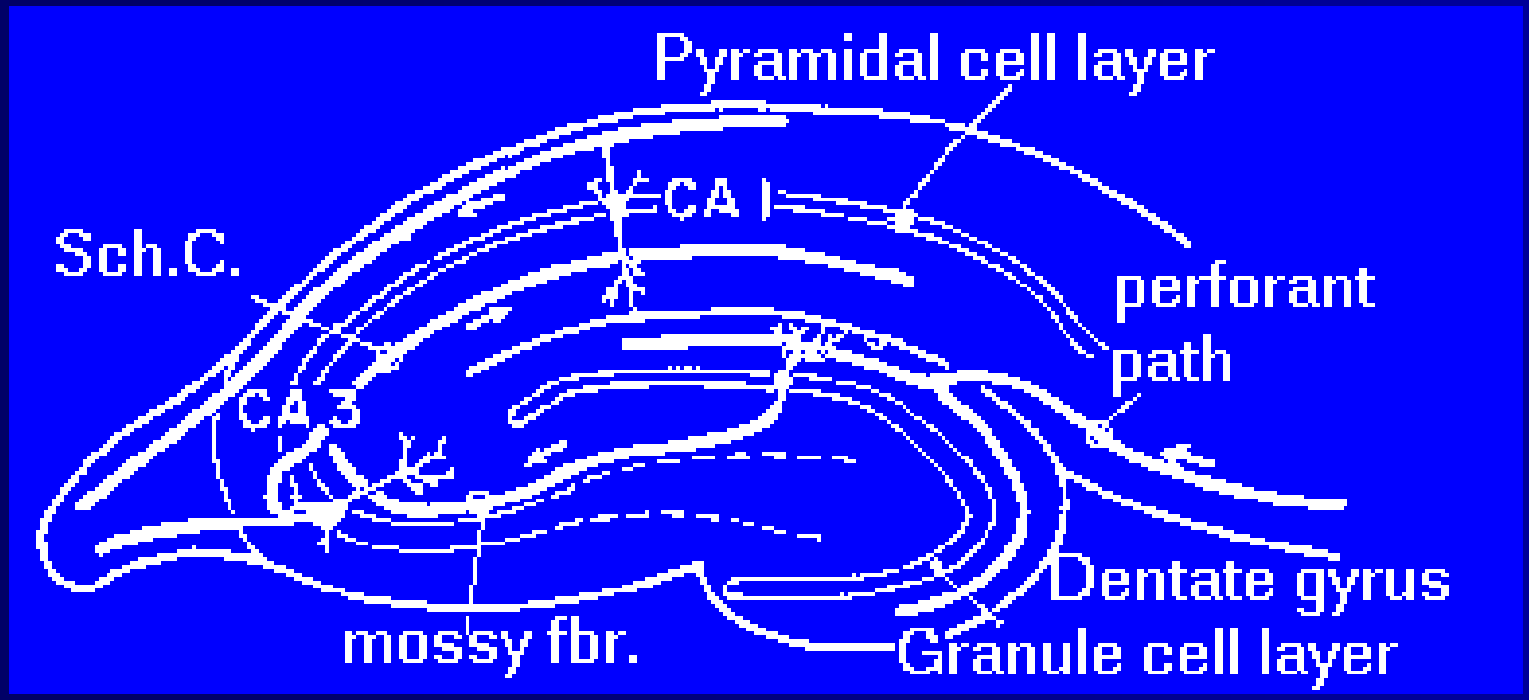
CA1 Neurons

- The investigation of the number of CA1 neurons in the hippocampus is particularly appropriate to detect potential radiation effects
- because minor short term cell loss would summarize during the long term study to a significant cell loss (e.g. 2% per week results in 32% after 4 months)

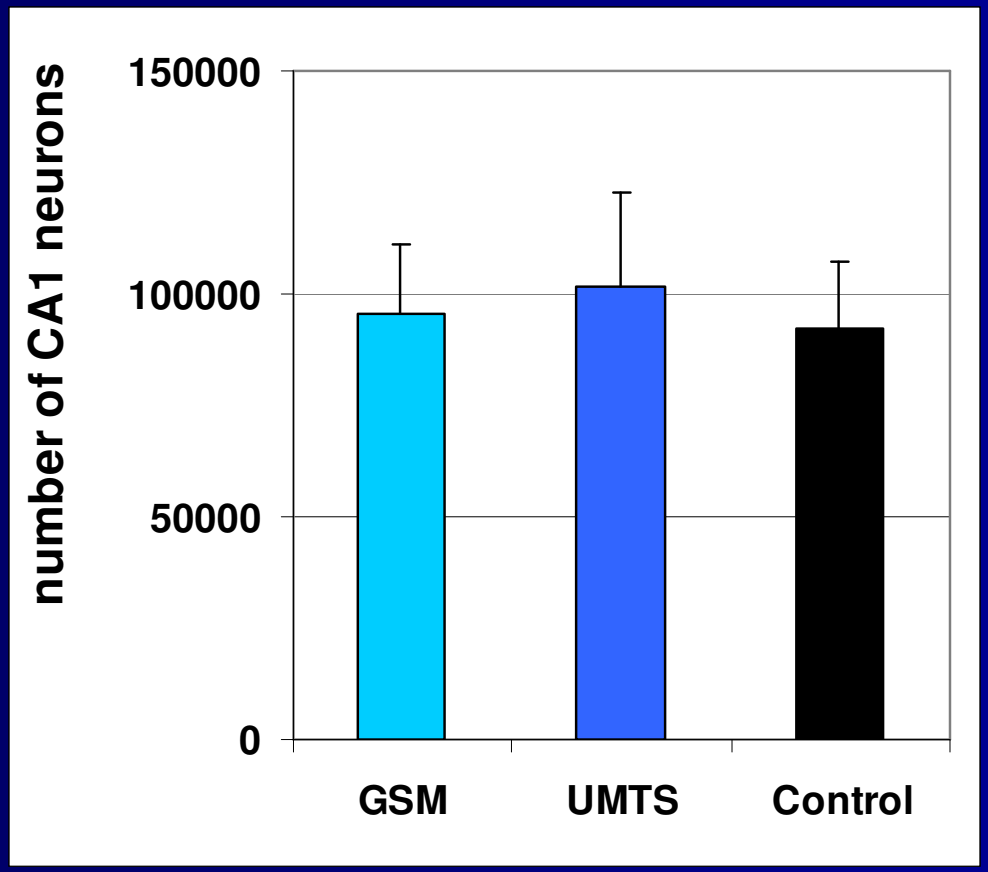
CA1 Neurons

- The optimal method to investigate the number of CA1 neurons is the physical dissector method.
- For this purpose 3000-4000 coronal sections (1,5 µm) are cut through the brain from rostral to caudal.
- The beginning and the end of the CA1 region were detected. After each 100th section, a pair of sections were stained and investigated for cell density and area of CA1 region.
- The CA1 region volume was calculated and multiplied with the average cell density to calculate the total CA1 cell number.

CA1 neurons in hippocampus



Number of CA1 neurons (F1, 7 months, n=10 per group)



Conclusions

- Arabinose-Infusion 0,5 mol/l (3,6 ml in 30 s) is the optimal challenge condition
- In the first generation of rats F_0 (age 4 months) was no influence of GSM and UMTS on BBB-integrity
- In the first generation of rats F_0 (age 11 months) was also no influence of GSM and UMTS on BBB-integrity
- In the third generation of rats F_2 (age 4 months) was no influence of GSM and UMTS on BBB-integrity
- One more 7 months radiated generation F_1 of rats was investigated for number of CA1 neurons – there was no effect of of GSM and UMTS on CA1 neurons