Online Supplementary Figures

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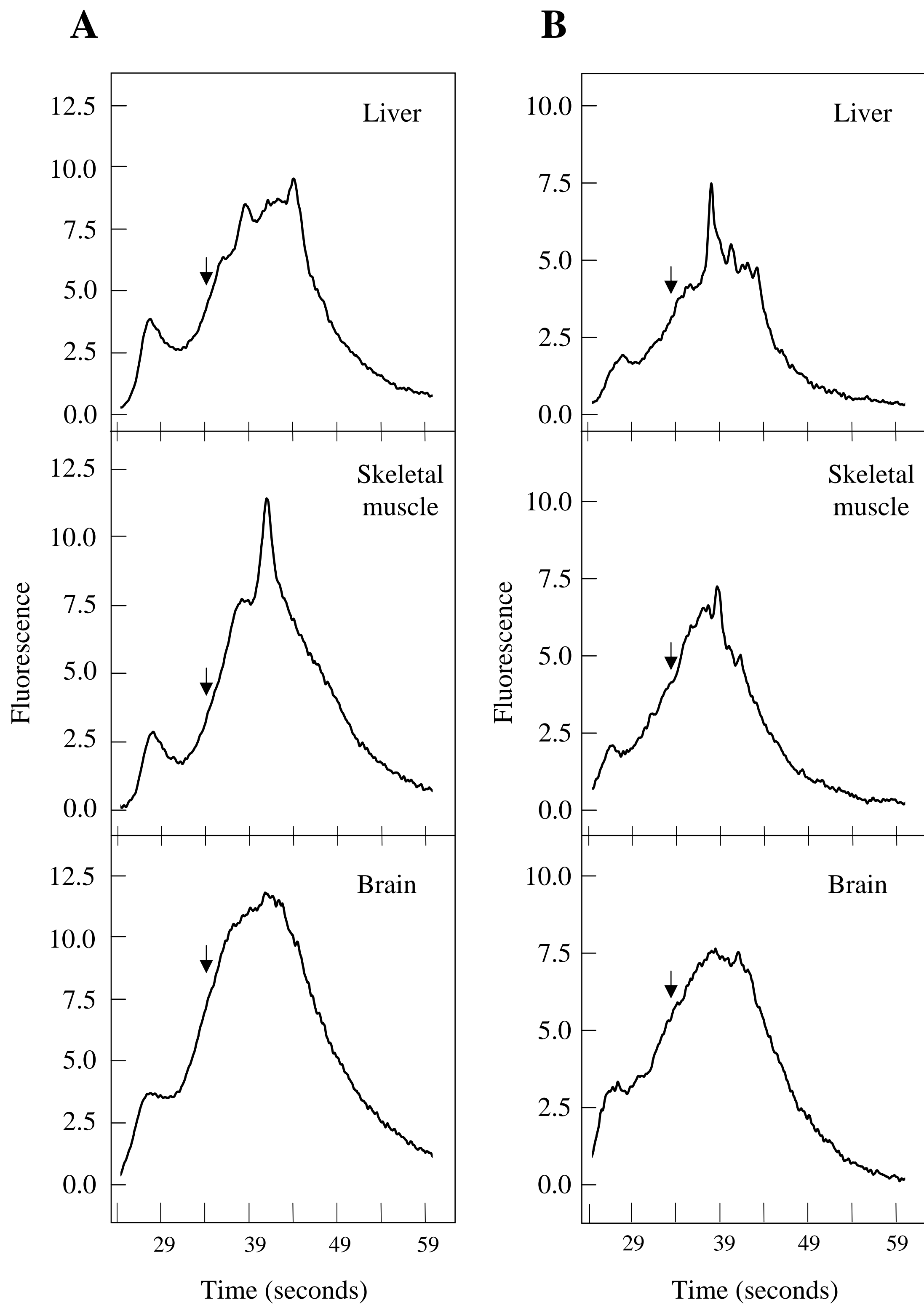
Probe-independent and direct quantification of insulin mRNA and growth hormone mRNA in enriched cell preparations

Supplementary Figure 1 Tissue-specific *ex vivo* liver, muscle and brain cRNA profiles after microchannel electrophoresis. Representative experiments are shown from cRNA prepared from liver, skeletal muscle and brain from male Wistar rats (**a**) and C57Bl6 mice (**b**). The elution position of the major cRNA species in islets and purified beta cells is indicated by the arrows.

Supplementary Figure 2 Microchannel electrophoresis of polyA mRNA extracted from mouse seminal vesicles and brain. Total RNA extracts from both tissues are shown in (panels **a** and **d**) with distinct 18S and 28S rRNA peaks. Profiles of mRNA enriched from total RNA using oligo-dT columns exhibit a tissue-specific electrophoresis profile (**b** and **e**) that can be compared to the profiles of cRNA prepared as explained in Research Design and Methods (panels c and f), except that contaminating 18S and 28S rRNA is still present in the mRNA profiles. Thus a major cRNA species is present seminal vesicles but not in brain. Based upon known abundant transcripts in this tissue, predicted length and Northern blots (not shown) we propose that this mRNA encoding seminal vesicle secretion 5 (svs5) (NM_009301 - length without poly A-tail = 638 nt). There was a difference in the estimated length of the cRNA (630 nt ; AUC = 10.4±1% of total cRNA in the profile) and

mRNA (876±10 nt). We attribute this difference to the artificial 20 nt poly A-tail in the cDNA-cRNA and a longer natural poly A-tail. This was confirmed by Northern blots using the prepared cRNA and extracted mRNA from seminal vesicles using a svs5 probe since that the mRNA was about 0.2 kb longer (data not shown). This indicates that the poly-A tail of svs5 mRNA is between 200 and 250 nt long which seems not unreasonable and perhaps expected for a transcript which is so abundantly present in the cell. As a control for the proposed experiment, we extracted mRNA from mouse brain, a tissue in which no major cRNA species was detected; apart from the rRNA contamination, the mRNA and cRNA Bioanalyzer profiles were found to be comparable (panels **2 d-f**).


A
B
Liver
Skeletal muscle
Brain
Fluorescence
Time (seconds)
0.0
2.5
5.0
7.5
10.0
12.5
29
39
49
59

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