Supplementary Material

Supplementary Figure 1

RN22 schwannoma cells are able to proliferate in the absence of serum. The number of RN22 schwannoma cells maintained either in the presence (circles) or absence (diamonds) of serum was estimated at different time points. Observe the exponential increase of the cells under both conditions. The duplication time was estimated to be 24 h in the presence of serum and 88 h in its absence. Mean ± s.e.m. is shown (n=3).
Supplementary Figure 2

Mitochondria from serum deprived RN22 schwannoma cells undergo swelling but do not release cytochrome-c. RN22 schwannoma cells were grown in medium containing serum (+FCS), or in serum depleted medium (-FCS) for 32 h before fixation. The distribution of cytochrome-c (Cyt-c) was determined using a specific antibody and double labeling with the mitochondrial-specific marker FITC-streptavidin (Strep.). Observe how cytochrome-c was retained by swollen mitochondria from serum-deprived RN22 schwannoma cells. Representative images are shown. Bar: 2 µm.
Supplementary Figure 3

RN22 schwannoma cells were cultured either in the presence (+FCS) or absence (-FCS) of serum for 18 h, and heavy membrane fractions (HM) or soluble fractions (Sol.) from these cultures were prepared and subjected to western blotting using antibodies specific for JNK, the active form of JNK (P-JNK) and the β subunit of the electron transfer chain complex V (β-V), a known mitochondrial marker. A. Note how JNK becomes enriched in the heavy membrane fraction of RN22 schwannoma cells grown in the absence of serum. It can be detected as a doublet corresponding to p46 JNK and p54 JNK. A representative western blot is shown (n=2). B. Active JNK was detected in the heavy membrane fraction of RN22 schwannoma cells grown in the absence of serum, mainly observed in the band corresponding to p54 JNK (arrows), in contrast to the soluble fraction where it was barely visible. A representative western blot is shown (n=3).
Supplementary Figure 4

The JNK specific inhibitor SP600125 was able to prevent cell death, as measured with nuclear labeling with bisbenzimide (Bisbenz.), in RN22 schwannoma cells 30 min after UV irradiation. Observe how the presence of condensed nuclei labeled with bisbenzimide (blue) was absent in the cultures previously treated with SP600125 (JNK Inh.) as compared to cells treated with vehicle. Bottom panels show phase contrast pictures of the same fields. Representative images are shown. Bar: 20 µm.
**Supplementary Figure 5**

Reduced viability of serum-deprived RN22 schwannoma cells exposed to JNK inhibitors other than SP600125. A. The ATP-competitive JNK inhibitor AS601245 was able to reduce cell viability in RN22 schwannoma cells in a dose dependent manner. B. The presence of 10 µM JNK inhibitor III (JNK Inh-III), a cell-permeable peptide that prevents the interaction of JNK with its substrates, reduced cell viability in RN22 schwannoma cells by ~25 %. Note that this peptide cannot be used at higher concentrations without compromising cell viability due to the intrinsic toxicity of the vehicle (dimethyl formamide), which was present in these cultures (in both control and treated cells) at a concentration of 4.4 %. *p<0.05; ***p<0.005.
**Supplementary Figure 6**

Inhibition of JNK does not alter the mitochondrial fragmentation process in serum-deprived RN22 schwannoma cells. **A.** RN22 schwannoma cells were maintained in serum depleted medium (-FCS) for 18 h or 24 h in the presence (JNK Inh.) or absence (-FCS) of SP600125 (9 µM) prior to labeling the cells with the mitochondria specific dye Mitotracker (Mit). In both cases, mitochondria fragmentation occurred in a similar manner. Representative images are shown. Bar: 5 µm.