Supplemental Figure Legends

Supplemental Figure 1. Wild-type FLI, Δ22 and FLI ETS domain binding to GGAA repeats. EMSA was performed using DNA duplexes harboring 7 consecutive GGAA motifs and 3xFlag wild-type FLI nuclear extract (A), recombinant Δ22 protein (B), or recombinant FLI ETS domain protein (C). Unlabeled DNA duplexes (I) and (II) were used as positive or negative competitors respectively, while α-FLAG (F1804; Sigma-Aldrich, St. Louis, MO) and α-FLI antibodies (sc356x; Santa Cruz Biotechnology, Santa Cruz, CA) were used for supershift. A non-specific band is designated by 'ns.'

Supplemental Figure 2. Significant DNA conformational changes are not involved in EWS/FLI microsatellite binding. Electrophoresis of DNA probes harboring 4 consecutive GGAA motifs located at different linear positions along the DNA strand demonstrate identical mobilities in the unbound state. The addition of recombinant Δ22 protein demonstrates nearly identical protein-bound DNA mobilities, indicating the lack of significant DNA bending or other conformational changes. Sequences of the DNA duplexes used are given in Supplemental Table.

Supplemental Figure 3. Titration of Δ22 or FLI ETS domain proteins in EMSA. EMSA with probe containing 4 consecutive GGAA motifs and increasing amounts of recombinant Δ22 protein (A) or recombinant FLI ETS domain protein (B). Protein concentration varied from 0-1 μM from left to right.

Supplemental Figure 4. Affinity of Δ22 for microsatellite sequences is lower than for high affinity ETS binding site. EMSA was performed using DNA duplexes harboring 4 consecutive