**TFK1, a new basal body transition fibre protein that is essential for cytokinesis in Trypanosoma brucei**

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**1. TFK1, the first Transition Fibres protein**

*Kinetoplastid-specific*

- Trypanosoma brucei is an unicellular flagellated parasite with a single flagellum emerging from a flagellar pocket
- Responsible for Human and Animal African Trypanosomiasis
- Two forms: the procyclic insect forms (PCF) in the vector insect midgut and the infectious bloodstream forms (BSF) upon transmission to the vertebrate host.

**2. TFK1 is a transition fibre protein**

The TFK1 CC1 domain is sufficient to target to the transition fibre zone

**3. TFK1 displays nine circularly arranged dots located on the mature basal body**

(A) U-ExM with anti-TFK1 in PCF whole cells (green), with BL10 (red) (a), or CEPI614C (red) (b), or RP22 (red) (c) or BLBD1 (red) (d) in G1 stage cells. Scale bars: 5 μm, inset 1 μm. (B) Immunofluorescence (a, c, e) and U-ExM (b, d, f) on detergent-extracted co-labeling with anti-TFK1 (green) with BL10 (red) or BB and pro-BB maturation (gbBN) (a, b), or RP22 (red) (c, d), or BLBD1 (red) during pro-BB maturation in SK14s cells stage (Early kinetoplast 5 phase) (a, f). The labeling at the mature pro-BB is indicated by an asterisk. Scale bars: 2 μm. Expansion factor: 4.8 fold.

**4. Bloodstream form TFK1 knockdown induces cell death**

RNA interference knockdown of TFK1 in bloodstream form. (A) Cumulative growth curve of control (WT); RNA cell line before induction (+Tet) and after induction (+Tet). Western blot to monitor the protein level of endogenously tagged +Tet TFK1 with enolase as loading control. (B) Quantification of DNA state in control cells (WT) and RNAi induced cells (+Tet 3h, 6h, 24h and 48h).

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**New insight into the organisation of transition fibres**

TFK1 is a kinetoplastid specific protein and a mature and maturing BB marker, localized on the transitional fibre. TFK1 is the third component of the transition fibres region, with TURF2 and CEPI614C, in T. brucei. Our high-resolution ultrastructure expansion microscopy data demonstrate that TFK1 is displayed in a typical radial arrangement in the distal appendage matrix, as nine dense points between the molecules of CEPI614C.

**What does TFK1 do?**

Our results support an essential role of TFK1 in the segregation and positioning of BBs to ensure the shape of subsequent daughter cells. The role of TFK1 could be extended to other stages of the trypanosomiasis life cycle, which exhibit different morphologies (tryomastigote and epimastigote) as well as a different positioning of the nucleus and basal bodies along the axis of the cell.

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