Recursive relation leading to first passage times of BD process.

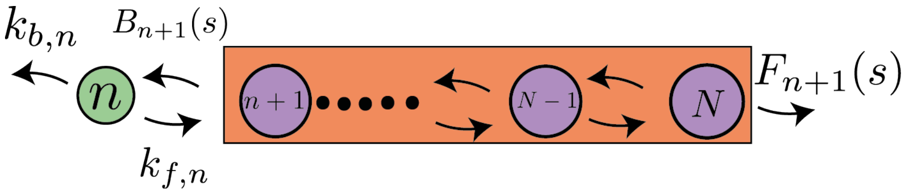

FIG. 1: ***If you are currently at state*** n***, the (moment generating function of) the event that you eventually make it to*** $n-1$ ***is given by*** $B_n(s)$***. This, in turn, is set by the probability to walk from*** n ***to*** $n+1$ ***and*** $B_{n+1}(s)$***.***

The RGN is described as either being unbound, bound to the PAM (in case of CRISPR systems), having formed an R-loop of length $n = 1, \ldots, N$ or having cleaved its target substrate. Let us label these states as $n \in [-1, N+1]$, with N being the total length of the guide (target) sequence. Each state $n \in [0, N]$ has rates $k_{\mathrm{f,n}}$ and $k_{\mathrm{b,n}}$ associated with it for transitioning to $n+1$ and $n-1$ respectively. Originally, we calulated the probability to cleave a sequence, staring from the PAM bound state. To do so, we wrote down a recursive relation for $P_{\mathrm{clv},n}$, the probability to cleave (visit $N+1$ before visiting $n = -1$) starting from state n.
Here, I attempted to take the same approach, but now using moment generating functions in order to (hopefully) get the expected time of the event:

Let $F_n(s)$ / $B_n(s)$ represent the Laplace transform of the first passage time distribution to make it to $n-1/N+1$ starting from state n (see figure). I think the following properly denotes it the way Frank would:

$$F_n(s) \equiv \mathcal{L}\left\{P(n_t = n-1|n_0 = n)\right\} \equiv \mathbb{E}[e^{sT_{n-1}}|T_{n-1} < T_{N+1}] \tag{1}$$
$$B_n(s) \equiv \mathcal{L}\left\{P(n_t = N+1|n_0 = n)\right\} \equiv \mathbb{E}[e^{sT_{N+1}}|T_{N+1} < T_{n-1}] \tag{2}$$

Eventually, we are seeking $B_0(s)$, as this gives us all events that started at state 0 (PAM bound) and eventually made it to state -1 (solution state/ dissociated) before making it to state N+1 (post cleavage). More precisely, $B_0(s)$ is the moment generating function. Initially, we seek the first moment:

$$\begin{aligned} T_{\mathrm{ub}} &= \frac{\int P(n_t = -1|n_0 = 0) \times t\mathrm{d}t}{\int P(n_t = -1|n_0 = 0)\mathrm{d}t} \\ &= -\frac{1}{B_0(s=0)} \left.\frac{\partial B_0}{\partial s}\right|_{s=0} \\ &= -\left.\frac{\partial \log(B_0(s))}{\partial x}\right|_{s=0} \end{aligned} \tag{3}$$

Additionally, we seek $F_0(s)$, to obtain the time needed to cleave, knowing you did not dissociate.
By counting all paths that start at state n and make it to $n-1$ for the first time before it has visited $N+1$ (eventually walk **B**ackwards):

$$B_n(s) = \sum_{m=0}^{\infty} \left(\frac{k_{\mathrm{f,n}}}{k_{\mathrm{f,n}} + k_{\mathrm{b,n}} + s}\right)^m \frac{k_{\mathrm{b,n}}}{k_{\mathrm{f,n}} + k_{\mathrm{b,n}} + s} \tag{4}$$

Similarly, counting any path that ends up in $N+1$ in stead (eventually walk **F**orward):

$$F_n(s) = \sum_{m=0}^{\infty} \left(\frac{k_{\mathrm{f,n}}}{k_{\mathrm{f,n}} + k_{\mathrm{b,n}} + s}\right)^m \frac{k_{\mathrm{f,n}}}{k_{\mathrm{f,n}} + k_{\mathrm{b,n}} + s} \times F_{n+1}(s) \tag{5}$$

Recognizing the geometric series:

$$B_n = \frac{k_{\mathrm{b,n}}}{s + k_{\mathrm{b,n}} + k_{\mathrm{f,n}}(1 - B_{n+1}(s))} \tag{6}$$

$$F_n = \frac{k_{\mathrm{f,n}} F_{n+1}(s)}{s + k_{\mathrm{b,n}} + k_{\mathrm{f,n}}(1 - B_{n+1}(s))} \tag{7}$$

If $\gamma_n = {}^{k_{\mathrm{b,n}}}/_{k_{\mathrm{f,n}}}$, then we also see:

$$\frac{1}{\gamma_n} B_n(s) = \frac{F_n(s)}{F_{n+1}(s)} \tag{8}$$

this might help.

BOUNDARY CONDITIONS

$$B_N(s) = \frac{k_{\mathrm{b,N}}}{k_{\mathrm{f,n}} + k_{\mathrm{b,n}} + s} \tag{9}$$

$$F_N(s) = \frac{k_{\mathrm{f,N}}}{k_{\mathrm{f,n}} + k_{\mathrm{b,n}} + s} \tag{10}$$

Typical guidelengths (N) are 20-30 (the most known system, spCas9, has a 20nt guide sequence).