Thermodynamics of switching in a multistable system

Jacob Cook$^{1,2}$ and Robert G. Endres$^{1,2,*}$

$^1$Department of Life Sciences, Imperial College, London SW7 2AZ, United Kingdom
$^2$Centre for Integrative Systems Biology and Bioinformatics, Imperial College, London SW7 2AZ, United Kingdom

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Multistable nonequilibrium systems are abundant outcomes of nonlinear dynamics with feedback but still relatively little is known about what determines the stability of the steady states and their switching rates in terms of entropy and entropy production. Here, we will link fluctuation theorems for the entropy production along trajectories with the action obtainable from the Freidlin–Wentzell theorem to elucidate the thermodynamics of switching between states in the large volume limit of multistable systems. We find that the entropy production at steady state plays no role, but the entropy production during switching is key. Additional stabilising and destabilising effects arise from the steady-state entropy and diffusive noise, respectively. The relevance to biology, ecological, and climate models is apparent.

When Niels Bohr and Erwin Schrödinger asked decades ago whether new physical principles are needed to explain living systems, the answer seemed No [1, 2]. More recently, however, the field of stochastic thermodynamics with its temporal violations of macroscopic thermodynamic laws at the microscopic scale have provided a new physical perspective on life. Most remarkable corner stones of far-from-equilibrium thermodynamics are the fluctuation theorems and Seifert’s thermodynamic uncertainty relation, stressing the important role of entropy fluctuation theorems and Seifert’s thermodynamic uncertainties for time intervals [3–6]. At equilibrium, detailed balance prohibits any entropy production on average, but far from equilibrium such entropy production is a characteristic feature [7] and determines the flow of time [8]. In particular, the fluctuation theorem by Evans and Searles allows the exact calculation of the entropy production along a trajectory from the time-forward and time-reversed path (corresponding to a movie played backwards), where paths can be calculated from e.g. Gillespie simulations of the underlying chemical master equation [4]. However, due to its intrinsic connection with the time-reversed path, it cannot be used to calculate the probability of a path simply from its entropy production. The situation is different when using the least-action principle, which allows the prediction of the most likely path between two points in a stochastic system from minimising the action (integral over the Lagrangian) [9, 10]. This is often done with a Langevin approximation of the master equation, such as using stochastic differential equations incorporating noise terms [11, 12]. However, now the link to thermodynamics is less clear as the role of the entropy production is obscured by the action functional.

In this letter, we combine the two to address the stability of steady states in non-equilibrium systems. In particular, we will elucidate the roles of steady-state entropy and fluctuations, as well as steady-state and path entropy production in state switching. For this purpose, we use two different low-dimensional minimal models shown in Fig. 1, the Schlögl [13] and the toggle switch [14] models, for different far-from-equilibrium constraints — concentration clamping and flux constraints.

To investigate the thermodynamics of state switching we shall study bistable systems with macrostates denoted $A$ and $B$, where both macrostates correspond to sets of microstates in the discrete space of molecule numbers $X$, which is a vector for multiple chemical species. The assumption is made that no significant amount of time is spent outside these macrostates. In the large volume limit, the process of switching between states can be assumed to be a Poisson process (with exponentially distributed waiting times, see Fig. 1B inset). Thus, $\langle \tau_A \rangle = \frac{k_{A\rightarrow B}}{k_{B\rightarrow A}} \int_0^{\infty} t \exp(-k_{A\rightarrow B}t) dt = k_{A\rightarrow B}^{-1}$ where $k_{A\rightarrow B}$ is the switching rate from $A$ to $B$, and similarly for the $B$ state. The occupation probability of the $A$ state is then given by $p_A = \frac{\langle \tau_A \rangle}{\langle \tau_A \rangle + \langle \tau_B \rangle} = 1/(1+k_{A\rightarrow B}/k_{B\rightarrow A})$. Hence, such a two-state system is completely described by the ratio of the switching rates. How do we calculate these for actual molecular systems?

For non-equilibrium systems the dynamics can be described by a path $\Gamma$, e.g. $X_0, X_1, X_2, \ldots, X_N$, obtainable from simulations of the chemical master equation [15]. For this time-forward path, there also exists a time-reversed path $\overline{\Gamma}, X_N, X_{N-1}, \ldots, X_0$. The probability of observing a particular path, e.g. the above time-forward path, is given by the path probability $P_\Gamma = \mathcal{N} P(X_0) P(\tau_{0,1}) W(X_0|X_1) \cdots P(\tau_{N-1,N}) W(X_{N-1}|X_N)$ assuming a memory-less Markov process with transition rates $W(X_i|X_{i+1})$. Furthermore, $P(\tau_{i,i+1})$ are probabilities for time intervals $\tau_{i,i+1}$ and $\mathcal{N}$ is a normalisation factor, ensuring $\sum_\Gamma P_\Gamma = 1$. As we are considering a non-equilibrium steady state (NESS) probability distribution, the total change in entropy can be found from the steady-state fluctuation theorem (FT) [16] as

$$\Delta S_\Gamma = \ln \left( \frac{P(X_0)}{P(X_N)} \right) + \ln (W_\Gamma/W_\overline{\Gamma}), \quad (1)$$

with $W_\Gamma = W(X_0|X_1) \cdots W(X_{N-1}|X_N)$ and $W_\overline{\Gamma} = W(X_N|X_{N-1}) \cdots W(X_1|X_0)$.

Restricting our consideration to paths that
The chemical Langevin equation can be expressed as \( \dot{x}_i = f_i(x) + \Omega^{-1/2}g_{ij}(x)\xi_j(t) \) with \( \xi_j(t) \) uncorrelated white Gaussian noises of zero mean and autocorrelation \( \langle \xi_i(t)\xi_j(t') \rangle = \delta_{ij}\delta(t-t') \). The deterministic force in direction \( i \) is given by \( f_i \), and \( g_{ij} \) determines the propagation of noise from direction \( j \) to \( i \). For the models considered in this paper \( g_{ij} \) is always diagonal.

When the probability of escape from a macrostate is sufficiently improbable the stochastic transition will be expected to concentrate along a single path \( x^* \), with paths significantly diverging having probabilities so low (for large \( \Omega \)) as to have negligible impact on overall escape probability [22]. The Wentzel-Kramers-Brillouin (WKB) approximation can then be used to obtain the probability of this path as \( P_{A\rightarrow B} \sim \exp(-\Omega A(x^*)) \), where \( P_{A\rightarrow B} \) is the probability of transition from macro-state \( A \) to \( B \), and \( A \) is the action, as derived in [23]. The path \( x^* \) will thus minimize the action \( A(x^*) = \min A[x] = A_{A\rightarrow B} \).

The relevant action for a path with duration \( \tau \) is the Freidlin–Wentzell (FW) action [23]

\[
A[x] = \frac{1}{2} \int_0^\tau (\dot{x} - f)^T D^{-1} (\dot{x} - f) dt, \tag{3}
\]

with the diffusion matrix given by \( D_{ij} = g_{ik}g_{kj}^T \). This action can be considered a first order truncation of a more complete action, and once more terms are included switching paths no longer pass through the same saddle point as their converse [17]. The mean-first passage time (MFPT) is given by \( T_{A\rightarrow B} \sim P_{A\rightarrow B}^{-1} = Q_{A\rightarrow B} \exp(\Omega A_{A\rightarrow B}) \) or \( \ln(T_{A\rightarrow B}) = \ln(Q_{A\rightarrow B}) + \Omega A_{A\rightarrow B} \), so that as \( \Omega \) grows the contribution from the prefactor \( Q_{A\rightarrow B} \) becomes less important and the second term on the RHS describes the MFPT to logarithmic precision.

An expression for entropy production based on the time-reversal of Langevin paths can be obtained by noting that the probability of the most probable switching path \( A \rightarrow B \) is given by \( P_{A\rightarrow B} = \exp(-\Omega A_{A\rightarrow B})/Q_{A\rightarrow B} \). The probability of the corresponding time-reversed path \( B \rightarrow A \) is then found to be \( P_{B\rightarrow A} = \exp(-\Omega A_{B\rightarrow A})/Q_{A\rightarrow B} \), where \( A_{B\rightarrow A} \) is the action of the time-reversed path. Combining the above two expressions as in Eq. 1 generates an expression for entropy production for Langevin paths as \( \Delta S_{A\rightarrow B} = \Omega (A_{B\rightarrow A} - A_{A\rightarrow B}) \) [24]. Substituting Eq. 3 into this relation leads to

\[
\Delta S_{A\rightarrow B}^f = 2\Omega \int_0^\tau \dot{x}_i D_{ij}^{-1} f_j dt. \tag{4}
\]

Under the assumption that the ratios \( k_{A\rightarrow B}/k_{B\rightarrow A} \) and \( P_{A\rightarrow B}/P_{B\rightarrow A} \) approximately match, \( \Delta S_{A\rightarrow B} \) and
\[ \Delta S^L_{A \rightarrow B} \] should be expected to return identical entropy production. However, the apparent entropy production from Eq. 4 disappears along with the action at steady state (\( \dot{x} = f_i(x) = 0 \) in the small noise limit). This suggests that only entropy production along the path matters, and that the Langevin formalism within the steady state is equivalent to a quasi-equilibrium [21]. Despite the obvious difference between the master equation entropy production (Eq. 1) and the Langevin path entropy production (Eq. 4), there conceivably remains explanatory potential in the later. How does this entropy production vary along the path, and how do diffusion strength and steady-state entropies matter? Furthermore, does it matter how the non-equilibrium constraint is implemented? To answer these questions we introduce explicit minimal models.

The two models considered are the Schlögl and toggle switch models (Fig. 1). For both models concentration-constraints are used to make the models non-equilibrium. Flux constrained models are not considered as the flux-constrained Schlögl model is not meaningfully bistable [21], and the flux-constrained toggle switch model is four-dimensional, and thus too computationally intensive for simulation of switches (mathematical details in [21]).

Figs. 2A,B show exemplar minimum action paths for switching in the Schlögl and the toggle switch model, respectively, calculated using the geometric minimum action method [21, 25–27]. The Schlögl model is sufficiently simple that the time-reversed switching paths correspond to the switching paths for the contrary direction. Fig. 2C shows how this leads to equal and opposite entropy productions (orange lines) along the paths. For the more complicated toggle switch model this simple relation between paths is lost, but Fig. 2D shows a linear relation between the difference in minimum action (purple line) and the difference in path entropy productions (gold line). Despite the systems’ quasi-equilibrium behavior, non-equilibrium processes still occur. Figs. 2E,F show plots of the derived entropy production (EP, blue lines) and flow (EF, red lines), demonstrating non-zero contributions at the steady states (for full derivation see [21]).

In order to investigate links between occupation probabilities, entropy, and entropy production, 100 random parameters sets were created for each model (generated with constraints to avoid pathological cases, e.g., negative concentrations, degradation processes on average increasing copy numbers, etc.). Fig. 3A shows a weak correlation between state occupation probability and steady-state entropy production, which provides some evidence for the maximum entropy production principle (MaxEPP) [24]. This extremal principle proposes that states with higher entropy production are more dynamically stable (subject to other dynamical constraints) [28]. We then approximate the log ratio of state occupation probabilities via the Freidlin–Wentzell theorem as,

\[
\ln \left( \frac{p_A}{p_B} \right) \approx \ln \left( \frac{P_{B \rightarrow A}}{P_{A \rightarrow B}} \right) = \ln \left( \frac{Q_{B \rightarrow A}}{Q_{A \rightarrow B}} \right) + \Omega \left( \mathcal{A}_{A \rightarrow B} - \mathcal{A}_{B \rightarrow A} \right).
\]

For large \( \Omega \) only the second term would be expected to contribute but this limit is difficult to simulate. Simulated occupation probabilities match well with this approximation (see Fig. 3B), demonstrating the validity of our use of the FW action. Fig. 3C shows a weak correlation between difference in action and difference in state entropy, as expected from equilibrium theory where higher entropy states are more stable. However, state entropy increases sublinearly with \( \Omega \) so for large \( \Omega \) it has no effect on the stability. Fig. 3D shows a comparison of the difference in action and the difference in path entropy production, showing that the linear relation observed in Fig. 2C,D holds generally across parameter sets surveyed. The effect of diffusion strength

**FIG. 2. Dependence of actions on path entropy productions.** (a) Minimum action paths for Schlögl model; aside direction, the only difference between paths is the amount of time spent at the fixed points where there is no contribution to the action. (b) Minimum action paths for toggle switch model, now showing clear differences. (c) Action (\( A \)), entropy production (\( \Delta S^L \)), kinetic (KE) and potential energy (PE) of Schlögl paths, clearly showing that the entropy production of one path is the opposite of the other. In this and the remaining panels a solid line corresponds to the path from \( A \rightarrow B \) and a dashed line to \( B \rightarrow A \). Magnifications are meant to explain line styles. (d) Action, differences in action and entropy production, KE and PE along the toggle switch paths. The difference in action is proportional to the difference in entropy produced along the path (this linear relationship is further discussed below). (e) Entropy production (EP) and entropy flow (EF) terms along the Schlögl paths. The difference (EP-EF) is equal to the entropy production of time-reversed Langevin paths. (Inset) Plot showing how \( f \) and \( \dot{x} \) vary along an exemplar minimum action path (see [21] for details). (f) Equivalent plot for the toggle switch paths.
was found to be minimal, and is therefore provided in [21].

Our results suggest a limited form of the MaxEPP, which applies to the rate of switching between macrostates. We shall proceed with our derivation by noting that the action can be split into two parts as \( \Omega A_{A \rightarrow B} = C_{A \rightarrow B} - \frac{1}{2} \Delta S^L_{A \rightarrow B} \), where \( C_{A \rightarrow B} \) is the conservative action along the path \( A \rightarrow B \) and \( \Delta S^L_{A \rightarrow B} \) is the Langevin path entropy production (Eq. 4) [24]. The conservative action can be expressed in a similar form to Eq. 4 as

\[
C_{A \rightarrow B} = \frac{\Omega}{2} \int_0^\infty (\dot{x}_i D_{ij}^{-1} \dot{x}_j + f_i D_{ij}^{-1} f_j) \, dt,
\]

where the two terms resemble kinetic and potential energy contributions, respectively. By substituting the expanded form of the action into the expression for switching path probability, a reduced form of MaxEPP can be obtained

\[
P_{A \rightarrow B} = \exp \left( \frac{1}{2} \Delta S^L_{A \rightarrow B} - C_{A \rightarrow B} \right) / Q_{A \rightarrow B},
\]

where \( P_{A \rightarrow B} \) is the probability of the (most probable) switching path along \( A \rightarrow B \), and \( Q_{A \rightarrow B} \) is a constant. This equation shows that there is a trade-off between minimization of the conservative action (i.e. fulfilling the equation of motion) and maximization of the path entropy production (i.e. being as dissipative as possible).

Generally, the switching path and its contrary path are travelled with the same speed and pass through similar regions of space. Thus, they are expected to have similar conservative actions (i.e. \( C_{A \rightarrow B} \approx C_{B \rightarrow A} \)). This relation is exact in the limit of matching time-reversed and converse switching paths (i.e. \( A \rightarrow B = B \rightarrow A \)), which is the case for the Schlögl model. This expectation does not extend to the dissipative (path entropy production) component Eq. 4, as this depends on the cross terms of velocity \( \dot{x} \) and deterministic force \( f \), and so the velocity with which a region of space is passed matters. Hence, the entropy production components are not expected to cancel upon subtraction. An analytic relation can then be obtained as

\[
\frac{1}{2} (\Delta S^L_{B \rightarrow A} - \Delta S^L_{A \rightarrow B}) \approx \Omega (A_{A \rightarrow B} - A_{B \rightarrow A})
\]

in line with expectation from the FT (Eq. 2, see [21] for details). In the toggle switch model where \( A \rightarrow B \neq B \rightarrow A \) this relation still holds for the majority of parameterizations, as can be seen in Fig. 3D. Significant divergence from the relation was generally observed in cases where the saddle point occurred at a low copy number compared to the steady states, due to the substantially faster variation of the force in low-copy-number regions (see [21]).

There has been significant interest in the thermodynamics of the transition between different steady-state probability distributions when controlled by an external parameter [29, 30]. This is fundamentally different to our work, which is about the thermodynamics of switching between metastable states. In our two-state systems, we would naively expect a net zero entropy production through switching as the entropy produced by a switch in one direction would be cancelled by the eventual switch back. Only in cases where the switching path differs from the converse switching path are there net fluxes of probability through the system and thus entropy production. Consistently, for the Schlögl model we find no net entropy production (i.e. \( \Delta S^L_{A \rightarrow B} = -\Delta S^L_{B \rightarrow A} \)). In [31], bounds on the ratio of transition rates between two metastable states based on relative entropy \( \Delta H \) and path entropy production are found. This ratio is determined as \( \pi(B \rightarrow A) / \pi(A \rightarrow B) \geq \exp (-\Delta S^L_{A \rightarrow B}) \), where \( \pi(A \rightarrow B) \) is the sum of the rates of switching \( A \) to \( B \).

**FIG. 3. Comparison of states and switching paths.** In all panels, red and blue dots denote specific parameterizations of Schlögl and toggle switch models, respectively. In each plot the lines and shaded regions indicate best fits and 95% confidence intervals for the particular data sets, respectively. (a) Comparison of log ratios of occupation probabilities vs difference in entropy productions at steady states from Gillespie simulations. The Schlögl data has a Pearson correlation of 0.5172 and the toggle switch has 0.3476. (b) Log ratio of occupation probabilities obtained from Gillespie simulation vs difference in minimum action. The dashed line indicates a perfect correspondence. The toggle switch and Schlögl data have correlations of 0.9515 and 0.9738, respectively. The results shown are coarsely discretized due to the low \( \Omega \) used to save computational time. The discretization will effect the Schlögl \( B \) state disproportionately as it is formed of significantly fewer microstates than \( A \). This represents a potential explanation for the downwards shift of the Schlögl data. (c) Comparison of difference in entropy of steady states vs difference in action. Entropies were found by Gillespie simulation with \( \Omega = 1 \). Both sets of data show weak correlations of 0.2892 and 0.4622 for the Schlögl and toggle switch models, respectively. (d) Difference in action vs difference in entropy produced along paths. Both models display a strong linear relationship, with correlations of 0.9445 and 1.0000 for the toggle switch and Schlögl models, respectively.
over all possible switching channels. With the expectation that a single most probable path will dominate, consideration of the two contrary switching paths will be sufficient. Combing the main result of their paper with our analytic relation (Eq. 7) leads to a bound on the path entropy produced as $\Delta S^L_{A\rightarrow B} + \Delta S^L_{B\rightarrow A} \geq 0$, which becomes an equality in the limit of time-reversed switching paths (e.g. Schlögl). Every parameter set used in Fig. 3 was found to satisfy this condition. Beyond the physics literature, related frameworks to ours have been used in evolutionary science [32] where cumulative fitness flux is maximized (like entropy production) subject to the trade-off that speed of allele change and magnitude of selective forces are minimized (like the conservative action). Our results therefore suggest, that states in evolutionary systems that require greater cumulative fitness fluxes to reach should be expected to be more stable.

Our primary conclusion is that a MaxEPP for switching paths can be obtained within the Langevin approximation (Eq. 6), extending the rule that “exerogenic reactions occur spontaneously” to switching in multistable systems. In a system with a large number of potential macrostates our relation predicts that for sufficiently large volumes switches that produce more entropy will be favoured. If regions of state space with greater entropy productions also require greater path entropy productions to reach, then this could form a basis for a more extensive maximum entropy production principle. Our secondary conclusion is that there exists a relationship between the difference in action of minimum action paths and difference in entropy produced along these paths (Eq. 7), valid for all paths that do not pass through regions of rapidly varying force. Exploring the application of our theory in ecology and evolutionary science with multiple stable states will be an interesting way forward [33].

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* E-mail: r.endres@imperial.ac.uk

[21] See Supplemental Material at [URL will be inserted by publisher] for extended description of models, details of numerical methods used and extended derivation of results.
Supplemental material

Jacob Cook\textsuperscript{1,2} and Robert G. Endres\textsuperscript{1,2}

\textsuperscript{1}Department of Life Sciences, Imperial College, London SW7 2AZ, United Kingdom
\textsuperscript{2}Centre for Integrative Systems Biology and Bioinformatics, Imperial College, London SW7 2AZ, United Kingdom

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S1 Brief summary of methods

The Gillespie algorithm was used to directly simulate the chemical master equation in Fig. 3A-C to obtain state entropies and probabilities of state occupations. In Fig. 3A steady state entropy productions were calculated using the Schnakenberg method (product of flux and reaction affinity) [1]. To construct the parameter sets, random numbers were drawn in a range, some of these were used as parameters values and the rest were used as ratios of parameters. These sets were accepted if they resulted in multiple non-zero steady states. Our choice of diffusion matrix (D) was constructed following the process of [2], this involves assuming no off-diagonal terms and taking the diffusion strength in a given dimension to be equal to the sum of the magnitudes of all force contributions in that dimension. The geometric minimum action method was then used to find minimum action paths [3–5]. These paths could then be used in Eq. 2 in order to generate the corresponding minimum actions. Path entropy productions were then calculated using Eq. 3, which was originally developed in [6].

S2 Concentration-constrained Schlögl model

In the Schlögl model [7] a chemical species A is converted to chemical species B via an intermediary species X. These chemical dynamics are described by the following equations

\[ A \xrightarrow{k_{+1}} X \]
\[ A \xrightarrow{k_{-1}} X \]
\[ 3X \xrightarrow{k_{+2}} 2X + B. \]

The deterministic dynamics of the copy number \( x \) in the external species constrained Schlögl model are therefore described by

\[ \frac{dx}{dt} = k_{-2}bx^2 - k_{+2}x^3 - k_{-1}x + k_{+1}a, \]  
(S2)

where \( a \) and \( b \) are externally clamped copy numbers and \( (k_{\pm 1}, k_{\pm 2}) \) are rate constants. Under certain parametrizations Eq. (S2) will have 3 real positive solutions and thus the system will display bistability [8–10]. The external clamping of the copy numbers makes these states non-equilibrium steady states rather than equilibria.

S3 Flux-constrained Schlögl model

When moving from the concentration-constrained to the flux-constrained model, a system of three deterministic equations needs to be considered [6]. The equations for describing the rate of change of real-valued species numbers \( a, b, \) and \( x \) are given by

\[ \frac{da}{dt} = k_{-1}x - k_{+1}a + F \]  
(S3)

\[ \frac{db}{dt} = k_{+2}x^3 - k_{-2}bx^2 - F \]  
(S4)

\[ \frac{dx}{dt} = k_{-2}bx^2 - k_{+2}x^3 - k_{-1}x + k_{+1}a, \]  
(S5)

where \( F \) is the flux constraint on the system. In order to find the steady states of the system we
must look for solutions to $\frac{da}{dt} = \frac{db}{dt} = \frac{dx}{dt} = 0$. It can be seen by addition of Eqs. S3 and S4 that $\frac{da}{dt} + \frac{db}{dt} = -\frac{dx}{dt}$. This means that $\frac{da}{dt} = \frac{db}{dt} = 0$ ensures $\frac{dx}{dt} = 0$, we therefore cannot extract any additional information about the steady state from Eq. S5.

The equations we are thus interested in solving are

$$k_{-1}x - k_{+1}a + F = 0 \quad (S6)$$
$$k_{+2}x^3 - k_{-2}bx^2 - F = 0. \quad (S7)$$

There are three unknowns in this pair of equations ($a$, $b$, and $x$), therefore a constraint must be introduced in order to find the solution to this system. We make the assumption that as every process conserves the number of molecules in the system that total molecule number is conserved, i.e

$$a + b + x = n = \text{const.} \quad (S8)$$

With this constraint we can now solve the system for a particular total molecular species number $n$. Eq. S6 can be rearranged to,

$$a = \frac{k_{-1}x + F}{k_{+1}}, \quad (S9)$$

so that Eq. S8 becomes

$$b = n - x - a = n - x - \frac{k_{-1}x + F}{k_{+1}}. \quad (S10)$$

Substituting for $b$ in Eq. S7 and gathering terms leads to

$$\left( k_{+2} + k_{-2} + \frac{k_{-2}k_{-1}}{k_{+1}} \right) x^3 + \left( \frac{k_{-2}F}{k_{+1}} - Nk_{-2} \right) x^2 - F = 0. \quad (S11)$$

There is only one real positive solution to this equation regardless of parametrization. This can be demonstrated by considering a cubic equation with three real positive solutions $x_1$, $x_2$ and $x_2$.

$$(x - x_1)(x - x_2)(x - x_3) = 0 \quad (S12)$$

which can be expanded to,

$$x^3 - (x_1 + x_2 + x_3)x^2 + (x_1x_2 + x_2x_3 + x_3x_1)x - x_1x_2x_3 = 0. \quad (S13)$$

The lack of a first order term in $x$ in Eq. S11 thus implies $(x_1x_2 + x_2x_3 + x_3x_1) = 0$. This cannot be realized with $x_1$, $x_2$ and $x_3$ all being real and non-negative. Hence under the constraint of conservation of total molecular concentration the flux constrained Schlögl model will have at most one physically sensible (real, +ve) steady state.

If the assumption of Eq. S8 is not made, then multi-stability is preserved. However, this is not an interesting multi-stability in that if one of $(x,a,b)$ is provided the other two can be determined uniquely by Eqs. S6 and S7. The apparent multi-stability here merely means that the system is stable for a wide range in $n$. 

3
S4 Concentration-constrained toggle switch

The chemical equations below describe the dynamics of a simple bistable switch \cite{11}, with promoters $\alpha$ and $\beta$ allowing the synthesis of “proteins” $A$ and $B$. Protein dimers can then repress the promoter of the opposite type:

\begin{align}
\frac{d\alpha}{dt} &= \frac{k}{k_-} \alpha + A, \\
A &= \frac{K}{K_-}, \\
2B + \alpha &= \frac{f}{r} \varnothing, \\
\beta &= \frac{q}{q_-} \beta + B, \\
B &= \frac{Q}{Q_-}, \\
2A + \beta &= \frac{f}{r} \varnothing.
\end{align}

These equations contain reverse reactions for thermodynamic consistency. The dynamics can be summarized as a system of deterministic differential equations,

\begin{align}
\frac{da}{dt} &= k\alpha - k_-a - Ka + K_- - 2\beta fa^2 + 2(1 - \beta)r, \\
\frac{db}{dt} &= q\beta - q_-b - Qb + Q_- - 2\alpha fb^2 + 2(1 - \alpha)r, \\
\frac{d\alpha}{dt} &= (1 - \alpha)r - \alpha fb^2, \\
\frac{d\beta}{dt} &= (1 - \beta)r - \beta fa^2.
\end{align}

We consider the fast switching limit $r, fa^2, fb^2 >> k, q, K, Q$, so that promoters $\alpha$ and $\beta$ can be treated as continuous variables that run between 0 and 1. This separation of time scales means that Eqs. S15 and S16 can be solved for the average promoter values, which can then be inserted into Eqs. S17 and S18 (quasi-steady state approximation). The average promoter values are given by

\begin{align}
\alpha^* &= \frac{r}{r + fb^2}, \\
\beta^* &= \frac{r}{r + fa^2}.
\end{align}

Substitution then gives,

\begin{align}
\frac{da}{dt} &= \frac{kr}{r + fb^2} - k_-a - Ka + K_- \\
\frac{db}{dt} &= \frac{qr}{r + fa^2} - q_-b - Qb + Q_-
\end{align}
These two equations for the approximate dynamics can be solved to find the multiple stationary points of the system. These stable states are non-equilibrium steady states as the Spontaneous generation and degradation of chemical copies ensures a constant flux through the system preventing equilibration. These spontaneous reactions can be thought of as equivalent to reactions with concentration-clamped chemical reservoirs.

S5 Flux-constrained toggle switch

In order to apply a flux constraint on our system we augment Eq. S14 by two additional chemical species, substrate S and waste W.

\[ a + S \xrightarrow{k_{-}} a + A \]  
\[ A \xrightarrow{K_{-}} W \]  
\[ 2B + a \xrightarrow{f_{r}} \varnothing \]  
\[ b + S \xrightarrow{q_{-}} b + B \]  
\[ B \xrightarrow{Q_{-}} W \]  
\[ 2A + b \xrightarrow{f_{r}} \varnothing \]  
\[ \varnothing \xrightarrow{F} S \]  
\[ W \xrightarrow{F} \varnothing. \]  

Making the same assumption of time scale separation as above, a new set of deterministic differential equations can be obtained as

\[ \frac{da}{dt} = \frac{ksr}{r + f b^2} - k_{-}a - ka + K_{-}w \]  
\[ \frac{db}{dt} = \frac{qs}{r + fa^2} - q_{-}b - Qb + Q_{-}w \]  
\[ \frac{ds}{dt} = - \frac{ksr}{r + f b^2} - \frac{qs}{r + fa^2} + k_{-}a + q_{-}b + F \]  
\[ \frac{dw}{dt} = ka + Qb - (Q_{-} + K_{-})w - F. \]

Once again as in the flux constrained Schlögl model this system of equations is not fully independent, e.g. \( \frac{da}{dt} + \frac{db}{dt} = -\frac{ds}{dt} - \frac{dw}{dt} \). At steady state, finding a solution to \( \frac{da}{dt} = \frac{db}{dt} = \frac{ds}{dt} = \frac{dw}{dt} = 0 \) guarantees that \( \frac{dw}{dt} = 0 \). So once again to find steady states an additional constraint must be introduced,

\[ a + b + s + w = n = \text{const.} \]  

Here meaningful bistability can be obtained in contrast to the flux constrained Schlögl model. This can be explained through the fact that generally, there are three mechanisms through which multi-stability can be maintained:
Figure S1: **Schematic of flux constrained models.** In the Schlögl model (A), there is no ability to partition flux, so if net flux $F$ and total number $n = a + x + b$ are constrained, then there exists on only one stable configuration for a given $F$ and $n$. In the bistable switch model (B), the flux $F$ can be partitioned down two separate pathways such that $F = F_1 + F_2$. This partitioning means that multiple stable states can exist for a given $F$ and $n$.

1. Net flux through the system can be changed.

2. Number of molecules in the system could change, leading to different reaction affinities for the same fluxes.

3. The way the flux is partitioned between reaction pathways could be changed.

In the flux-constrained Schlögl model there exists only one pathway through which flux can pass. Additionally, the assumption is made of conservation of particle number. This means that none of the three mechanisms are available to generate multi-stability. For the flux-constrained toggle switch model two distinct pathways for flux exist so multi-stability can be achieved by changing how flux is partitioned (Fig. S1). The continual flux of chemical copies through this model ensures that the multiple state states found here are non-equilibrium steady states.

### S6 Minimum action path

Consideration of any the above models via deterministic approximation can give information about the steady state behavior but will provide only limited information about state switching. In order to investigate state stability stochastic behavior must therefore be considered. In the large volume (small noise) limit the stochastic evolution of the system can be considered via the over-damped chemical Langevin equation

$$\dot{x}_i = f_i(x) + \Omega^{-1/2} g_{ia}(x) \xi_a.$$  

(S26)

In this equation $\dot{x}_i$ is the rate of change of species $i$, $f_i$ is the net deterministic force on species $i$,
\( \Omega \) is the volume of the system, \( \xi_a \) is the noise from species \( a \), and \( g_{ia} \) parametrizes the effect of the noise in species \( a \) on species \( i \). This is a continuous stochastic approximation to the underlying master equation, thus allowing a greater range of analytic techniques.

One such technique is the minimization of the Freidlin–Wentzell Action \(^{12}\) in order to determine the most probable path between two points in parameter space. According to the large deviations theory this path will be exponentially more probable than paths substantially deviating from it \(^{13}\). This exponential fall off means that the mean first-passage time for state switching \( T_s \) can be found in an exponential form as

\[
T_s = Re^{\Omega A},
\]  

(S27)

where, due to \( \ln T_s = \ln R + \Omega A \), \( T_s \) is determined by \( \Omega A \) to logarithmic precision for large \( \Omega \). Here, \( A \) is the action associated with the minimum action path (MAP) for this stochastic transition and \( R \) is a pre-factor to the exponential behavior. Through this formula the mean rate of transition between two steady states in order to determine the relative state stability as described in \(^2\). Briefly, the path is split into \( N \) segments such that the points are separated by an equal amount of time. The action can be rewritten into an approximate form as

\[
A = \frac{1}{2} \int_0^t d\bar{t} (\dot{x}_i - f_i) D_{ij}^{-1} (\dot{x}_j - f_j) = \int_0^t d\bar{t} L(\bar{x}, \bar{x}),
\]  

(S28)

where

\[
D_{ij}^{-1} \equiv (g_{ia}^{-1} g_{ja}^{-1})
\]  

(S29)

is the diffusion matrix and parametrizes how easily noise in species \( i \) can propagate to species \( j \). \( L(\bar{q}, \bar{q}) \) is the Lagrangian for a stochastic differential equation with multiplicative white noise to leading order in \( \Omega \) \(^{14}\). The action in Eq. \(^{S28}\) can be minimized between two steady states in order to determine the relative state stability as described in \(^2\). Briefly, the path is split into \( N \) segments that generates the minimum action. The gradients to be used with this algorithm are given by

\[
\frac{\partial A}{\partial x_s^l} = \frac{\dot{x}_s^l - f_s^l}{D_s^l} - \frac{\dot{x}_{s+1}^l - f_{s+1}^l}{D_{s+1}^l} - \frac{\Delta t}{2} \sum_{j=1}^{N_s} \left[ \frac{\dot{x}_{s+1}^j - f_{s+1}^j}{D_{s+1}^j} f_{l,s}^{j} + \frac{\dot{x}_s^j - f_s^j}{D_s^j} f_{l,s}^{j} \right. \\
+ \left. \frac{(D_{l,s}^j)^2}{2(D_s^j)^2} (\dot{x}_{s+1}^j - f_{s+1}^j)^2 + \frac{(D_{l,s}^j)^2}{2(D_s^j)^2} (\dot{x}_s^j - f_s^j)^2 \right].
\]  

(S31)
These expressions are significantly more complicated when $D^{-1}$ ceases to be diagonal. Additionally, as the dimensionality (number of chemical species) increases the L-BFGS algorithm becomes increasingly numerically intensive. For these reasons an alternative method of computing the minimum action, termed the geometric minimum action path (gMAP) method, was also used.

S7 Geometric minimum action path method

The geometric minimum action path (gMAP) method involves discretising the curve by equal units of arc length $\alpha$ rather than by equal time spacing $[3]$. This eliminates the need for a gradient descent method to find optimal time of path, saving computational time. The new algorithm is more complicated than the previous algorithm, but through sensible consideration of geometry, avoids the creation of unphysical (non-smooth) paths, thus speeding up the algorithm considerably for higher dimensions.

Contrary to the prior method we now consider the Hamiltonian rather than the Lagrangian. This allows determination of the minima of the action via solution of Hamilton’s equations. Solution of these equations speeds the numerical minimization compared to e.g. hill-climbing optimization algorithms, as knowledge of the underlying physics has been integrated into the procedure. The Hamiltonian for a stochastic differential equation

\[ \dot{x} = f(x) + \Omega^{-1/2}g(x(t))\xi(t) \]  

is given as

\[ H(x, \theta) = \langle f(x), \theta \rangle + \frac{1}{2} \langle \theta, D(x)\theta \rangle, \]  

where $D = gg^T$, $\xi(t)$ is the vector of noises, $x$ is the vector of copy numbers, $f$ is the vector of deterministic forces, $\theta$ is the vector of conjugate momenta, and $\langle a, b \rangle$ means the dot product of vector $a$ and vector $b$ $[3]$. Through the rest of this text, $H_x \equiv \frac{\partial H}{\partial x}$, $H_\theta \equiv \frac{\partial H}{\partial \theta}$, $H_{xx} \equiv \frac{\partial^2 H}{\partial x^2}$ and $H_{\theta \theta} \equiv \frac{\partial^2 H}{\partial \theta^2}$. The conjugate momenta cannot simply be taken to be $m\dot{x}$ as would be the case when considering the motion of a particle in Cartesian coordinates with no noise. Here, the noise intensity will effect how readily the path can pass through a region, thus the conjugate momenta must be a function of the noise in the vicinity. Although conjugate momenta $\vartheta$ can be obtained via $p_i = \frac{\partial L}{\partial \dot{x}_i}$, this expression cannot be solved just with knowledge of position $x$ and arc-length “velocity” $y = \frac{\partial x}{\partial \alpha}$. There will also be a dependence on the undefined function $\lambda(x,y)$, which relates “velocity” in arc-length parametrization to the velocity in species space.

The only information available to the algorithm is the path in $q$ discretized in $\alpha$. Thus all functions must be definable in terms of $x$ and $y = \frac{\partial x}{\partial \alpha}$. In order to determine $\vartheta$ and $\lambda$ in form dependant only $x$ and $y$, a number of assumptions are made. The first is that $\lambda$ is always positive, i.e. the assumption that the movement along the arc is occurring forwards in time. The second is that Hamilton’s equations, specifically $\frac{\partial H}{\partial \theta} = \dot{x}_1$, are obeyed. Obeying Hamilton’s equations is a sufficient and necessary condition for action $S$ to be a minimum, so this will only give the “correct” momenta when $x$ is the path minimizing $S$, but as the algorithm proceeds this will be an increasingly good assumption. The final assumption is that the minimizing solution will be a zero energy path $E = H = 0$. This is a reasonable assumption as for all Hamiltonians without time dependence, $H = const$ along the minimizing path. All paths are started with zero velocity in a steady state, therefore the value of
this constant $E$ should be zero. The three assumptions are summarized as

$$\lambda \geq 0, \quad H_\theta(x, \vartheta) = \lambda y, \quad H(x, \vartheta) = 0. \quad (S34)$$

For Eq. S34, a unique solution $(\vartheta(x, y), \lambda(x, y))$ exists. In the case of a stochastic drift process, equations for both $\lambda$ and $\vartheta$ can be obtained as

$$\lambda(x, y) = \frac{|f(x)|_D}{|y|_D} \quad (S35)$$
and

$$\vartheta(q, y) = D^{-1}(x) \left( \frac{|f(x)|_D}{|y|_D} y - f(x) \right) \quad (S36)$$

where $|X|_D = \sqrt{X D^{-1} X}$. The gMAP algorithm selects points that are separated by an equal arc-length. This implies a rediscretization as

$$\varphi(\alpha) = \psi(t), \quad (S37)$$

where $\alpha \in [0, 1]$, and $t \in [0, T_f]$ where $T_f$ is the total time of the path. The two paths $\varphi$ and $\psi$ are identical, but the differing discretizations mean that the points that are selected to be used by the optimization algorithm differ. The relation of $t$ and $\alpha$ is done via the function,

$$t = G(\alpha) = \int_0^\alpha \frac{1}{\lambda(a)} da, \quad (S38)$$

leading to

$$\varphi(\alpha) = \psi(G(\alpha)) \quad (S39)$$

and

$$\frac{\partial}{\partial \alpha}(\varphi(\alpha)) = \varphi'(\alpha) = \dot{\psi}(t)G'(\alpha) = \frac{\dot{\psi}(t)}{\lambda(\alpha)}. \quad (S40)$$

Further differentiation of Eq. S40 by $\alpha$ leads to

$$\lambda^2 \varphi'' + \lambda \lambda' \varphi' = \ddot{\psi}. \quad (S41)$$

The necessary and sufficient condition for the path $\psi$ to minimize the action $S$ is that Hamilton’s equations are satisfied at every point along the path, i.e. $\ddot{\psi} = H_\theta(\psi, \theta)$ and $\dddot{\theta} = -H_x(\psi, \theta)$. Together with Eq. S41 this can be used to derive a condition for the solution $\varphi$ being stationary dependent only on $\varphi$ and $\alpha$

$$\ddot{\psi} = \frac{\partial}{\partial \psi}(H_\theta(\psi, \theta))$$
$$= \frac{\partial}{\partial \psi}(H_\theta)\dot{\psi} + \frac{\partial}{\partial \theta}(H_\theta)\dot{\theta}$$
$$\lambda^2 \varphi'' + \lambda \lambda' \varphi' = H_{\theta x} \dot{\psi} - H_{\theta \theta} H_x$$
$$\lambda^2 \varphi'' + \lambda \lambda' \varphi' = H_{\theta x} \lambda \varphi' - H_{\theta \theta} H_x$$
$$\lambda^2 \varphi'' + \lambda \lambda' \varphi' - H_{\theta x} \lambda \varphi' + H_{\theta \theta} H_x = 0.$$
This provides a condition that is satisfied when the curve minimizes $S$. At any stage in the algorithm, the deviation of the present curve from this condition can be used to determine the appropriate update for the curve. This deviation $\delta$ can be used iteratively in a simple relaxation method towards minima. A relaxation time step $\tau$, is artificially introduced in order to determine the step size the algorithm will use as it relaxes towards this minima. Use of $\dot{\varphi} = \frac{\partial \varphi}{\partial \tau}$ = $\delta$ can then determine $\varphi$ for the next step. Iterative use of this procedure represents an algorithm to minimize the action.

### S7.1 Algorithmic procedure

Here we explain how the above gMAP algorithm can be implemented as a numerical optimization algorithm to find the gMAP for the case of a stochastic drift process. We then layout how the action and the time of the path can be found through this optimization. The steps are:

1) Discretize $\varphi(\tau, \alpha)$ in both $\tau$ and $\alpha$ and thus define $\varphi_i^k = \varphi(k\Delta\tau, i\Delta\alpha)$, $k$ being a natural number, $i = 0, ..., N$, $\Delta\tau$ is the “time” step, and $\Delta\alpha = \frac{1}{N}$. Here the curve is being discretized into $N + 1$ points.

2) From $\varphi_i^k$ calculate $\varphi_i^{k+1}$, then use Eqs. S35 and S36 to calculate $\lambda_i^k$ and $\vartheta_i^k$, the edge cases are then taken to be $\lambda_0^k = 3\lambda_1^k - 3\lambda_2^k + \lambda_3^k$ and $\lambda_N^k = 3\lambda_{N-1}^k - 3\lambda_{N-2}^k + \lambda_{N-3}^k$. Now the calculation of $\lambda_i^k = \frac{\lambda_{i+1}^k - \lambda_{i-1}^k}{2N}$ can be done.

3) Find $\{\tilde{\varphi}_i\}_{i=0, ..., N}$ the solution to the following system of equations

$$\frac{\tilde{\varphi}_i - \varphi_i^k}{\Delta\tau} = (\lambda_i^k) \frac{2\tilde{\varphi}_{i+1} - \tilde{\varphi}_i + \tilde{\varphi}_{i-1}}{1/N^2} + \lambda_i^k H_{\theta x} \varphi_i^k + H_{\theta \theta} H_x + \lambda_i^k \vartheta_i^k \varphi_i^k$$

(S43)

where $H_{\theta x}$, $H_{\theta \theta}$ and $H_x$ are evaluated at $(\varphi_i^k, \vartheta_i^k)$.

4) Now interpolate a curve across $\tilde{\varphi}$ and then rediscretize in order to obtain $\varphi^{k+1}$. This discretization ensures that points remain at equal spacing along the arc length.

5) Repeat steps 2-4 until change in path between steps becomes sufficiently small.

6) The final path can then be used to calculate the action by

$$A = \frac{1}{N} \left( \frac{3}{2} \langle \varphi_1, \vartheta_1 \rangle + \sum_{i=2}^{N-2} \langle \varphi_i^k, \vartheta_i^k \rangle + \frac{3}{2} \langle \varphi_{N-1}^k, \vartheta_{N-1}^k \rangle \right).$$

(S44)

7) The time to point $i$ can now be found by

$$t_i = \frac{1}{2\lambda_0^k} + \frac{1}{\lambda_1^k} + ... + \frac{1}{\lambda_{i-1}^k} + \frac{1}{2\lambda_i^k}.$$  

(S45)

8) Finally these times can be used to reverse the procedure followed in step 1) in order to generate a path that is discretized in time rather than along arc length.

### S8 Derivation of limited maximum entropy production principle

Large deviation theory [13] can be applied to systems where transitions between the states are sufficiently improbable. The ensemble of switching paths between two states is then predicted to
be dominated by paths infinitesimally deviating from a single most probable path. These most probable paths are defined by the fact that they minimize the Freidlin-Wentzell action \[12\], defined as

\[
\mathcal{A}[\mathbf{x}] = \frac{1}{2} \int_0^\tau (\mathbf{\dot{x}} - \mathbf{f})D^{-1}(\mathbf{\dot{x}} - \mathbf{f})^T dt, \tag{S46}
\]

where \(\mathbf{\dot{x}}\) is the velocity along the path, \(\mathbf{f}\) the deterministic force experienced along the path, and \(D^{-1}\) the inverse of the diffusion matrix along the path. The minimum of this action can then be used to determine the probability of a switch between two macro-states, as \(P_{A\rightarrow B} \sim \exp(-\Omega A_{A\rightarrow B})\). Thus, the ratio of switching rates can be obtained as

\[
\frac{P_{B\rightarrow A}}{P_{A\rightarrow B}} \approx \exp[\Omega(A_{A\rightarrow B} - A_{B\rightarrow A})]. \tag{S47}
\]

with the assumption that the multiplicative constants for the forward and backwards processes approximately cancel.

This description pertains to Langevin paths. In order to make a linkage with the fluctuation theorem (Eq. 1 in the main text), a relation to the path probabilities at the level of master equation paths must be obtained. The master equation has transition rates \(W(X_N|X_{N-1})\), which can be used to obtain a path probability as \(W_\Gamma = W(X_0|X_1)W(X_1|X_2)...W(X_{N-1}|X_N)\). In order to proceed, we make the assumption that the probability \(P_{\Gamma^*}\) of the minimum action path \(\Gamma^*\) (a Langevin path) is equal to the probability \(W_\Gamma\) of the corresponding master equation path. We shall now layout two methods of calculating the entropy production that should return equivalent results if this assumption holds.

The fluctuation theorem derived by Evan and Searles \[15\] allows the calculation of entropy production, via the ratio of probabilities of the time-forward and time-reversed paths

\[
\Delta S_\Gamma = \ln \left( \frac{P(X_0)}{P(X_N)} \right) + \ln \left( \frac{W_\Gamma}{W_\Gamma} \right), \tag{S48}
\]

where \(P(X_0)\) is the steady-state probability of finding the system in state \(X_0\), and \(W_\Gamma\) is the probability of the time-reversed path. For long trajectories the leading term can be neglected and a simplified form of the fluctuation theorem can be obtained as

\[
\Delta S_\Gamma = \ln \left( \frac{W_\Gamma}{W_\Gamma} \right), \tag{S49}
\]

with \(\Delta S_\Gamma\) as the entropy produced along the path \(\Gamma\). For the master equation the probability of the time-reversed path is easily calculable by

\[
W_\Gamma = W(X_N|X_{N-1})W(X_{N-1}|X_{N-2})...W(X_2|X_1), \tag{S50}
\]

and so the entropy production can immediately be calculated. For Langevin paths time-reversed probabilities are not so simple to calculate and so an expression must be derived.

From large deviations theory the probability of the path can be seen to depend on the Friedlen–Wentzell action Eq. \[S46\] by
\[ P_{A\rightarrow B} = \frac{\exp(-\Omega A_{A\rightarrow B})}{Q}, \]  
\[ (S51) \]

where Q is a constant. The probability of the time-reversed path can be obtained by time reversing all components of Eq.\( \text{[S51]} \) so
\[ P_{A\rightarrow B} = \frac{\exp(-\Omega A_{A\rightarrow B})}{Q}. \]  
\[ (S52) \]

The expression for entropy production for Langevin paths can then be found as
\[ \Delta S_{A\rightarrow B}^L = \ln \left( \frac{P_{A\rightarrow B}}{P_{A\rightarrow B}} \right) = \Omega (A_{A\rightarrow B} - A_{B\rightarrow A}). \]  
\[ (S53) \]

This expression can then be combined with the expression for the Friedlen–Wentzell action (Eq.\( \text{[S46]} \)) in order to generate an expression in terms of properties of a Langevin path
\[ \Delta S_{A\rightarrow B}^L = 2\Omega \int_0^\tau \hat{q}_i D_{ij}^{-1} f_j \, dt. \]  
\[ (S54) \]

This expression is expected to match the entropy produced along the corresponding master equation path when the assumption that \( P_{A\rightarrow B} \approx W_{A\rightarrow B} \) holds. This assumption implies that the macroscopic probability of a path \( P_{A\rightarrow B} \) is provided by the product of the microscopic transition rates along the most probable path \( W_{A\rightarrow B} \). Taking this assumption to be true a full expression linking large deviation theory with master equation trajectories can then be stated as
\[ \frac{W_{B\rightarrow A}}{W_{A\rightarrow B}} \approx \frac{P_{B\rightarrow A}}{P_{A\rightarrow B}} \approx \exp [\Omega (A_{A\rightarrow B} - A_{B\rightarrow A})]. \]  
\[ (S55) \]

For relatively simple models (e.g. the Schlögl model), each minimum action path is the microscopic time-reversal of the other, e.g. \( W_{A\rightarrow B} = W_{B\rightarrow A} \). Substituting this relation into Eq.\( \text{[S49]} \) gives an expression, relating the two formalisms for the case of reversible switching paths as
\[ \frac{W_{B\rightarrow A}}{W_{A\rightarrow B}} \approx \frac{P_{B\rightarrow A}}{P_{A\rightarrow B}} \approx \exp (\Delta S_{B\rightarrow A}) = \exp (-\Delta S_{A\rightarrow B}). \]  
\[ (S56) \]

For the case of more complicated models (e.g. the toggle switch model), the two minimum action paths are no longer expected to be time-reversals of each other. We shall therefore proceed by looking more carefully at the components of the action
\[ \Omega A_{A\rightarrow B} = \mathcal{C}_{A\rightarrow B} - \frac{1}{2} \Delta S_{A\rightarrow B}^L, \]  
\[ (S57) \]

where \( \mathcal{C} \) describes the time-reversible (conservative) contributions to the action and \( \Delta S_{A\rightarrow B}^L \) gives the entropy production (dissipative) contribution. From the above decomposition the components of \( \mathcal{C} \) can be written out in a similar form to Eq.\( \text{[S54]} \) as
\[ \mathcal{C}_{A\rightarrow B} = \frac{\Omega}{2} \int_0^\tau \left( \hat{q}_i D_{ij}^{-1} \hat{q}_j + f_i D_{ij}^{-1} f_j \right) \, dt, \]  
\[ (S58) \]

where the leading term may be considered the kinetic energy and second term the potential energy.
Substituting Eq. S57 into our expression for forward path probability (Eq. S51) a new relation can be obtained linking the entropy production and forward path probability,

\[ P_{A\rightarrow B} = \exp\left(\frac{1}{2} \Delta S_{A\rightarrow B} - C_{A\rightarrow B}\right) \]  

This expression can be thought of a restricted form of the maximum entropy production principle (MaxEPP) applicable to trajectories between steady states. Here, the probability of a switching path will be maximized when the conservative action is minimized and the entropy production is maximized. These two components are not independent and so a strict extremal principle will not apply and there will instead be a trade-off between minimization of the conservative action and entropy production maximization.

### S8.1 Derivation of semi-analytic result

We shall now attempt to derive a relation for the pattern that was noted in our data (see Fig. 3 main text). The assumption is made that very little time is spent by the system outside the macrostates \( A \) and \( B \). Probability of occupation of state \( A \) can be taken to be

\[ P_A = \frac{\langle \tau_A \rangle}{\langle \tau_A \rangle + \langle \tau_B \rangle} \]

where \( \langle \tau_A \rangle \) is the average time to exit state \( A \). The process of switching between states \( A \) and \( B \) is then assumed to be an exponentially distributed stochastic process with rate constant \( k_{A\rightarrow B} \). Thus,

\[ \langle \tau_A \rangle = k_{A\rightarrow B} \int_0^\infty t \exp(-k_{A\rightarrow B}t) dt = k_{A\rightarrow B}^{-1}, \]

and likewise for the \( B \) state. It then follows that \( P_A = 1/(1 + k_{A\rightarrow B}/k_{B\rightarrow A}) \). A full expression for the ratio of probabilities of the two minimum action paths is then found by substitution of Eq. S57 as

\[ \frac{P_{B\rightarrow A}}{P_{A\rightarrow B}} \approx \frac{k_{B\rightarrow A}}{k_{A\rightarrow B}} \approx \exp\left(C_{A\rightarrow B} - \frac{1}{2} \Delta S_{A\rightarrow B} - C_{B\rightarrow A} + \frac{1}{2} \Delta S_{B\rightarrow A}\right). \]  

(S60)

It is observed that though the two switching paths differ they generally have similar speeds and pass through proximate regions in space. When this is the case the two paths have similar reversible action components, i.e. \( C_{A\rightarrow B} \approx C_{B\rightarrow A} \). This assumption does not extend to the dissipative parts of the action (entropy production), as these depend on the cross terms of the deterministic force with the velocity. Thus, the velocity with which the path passes a region of space significantly alters the magnitude of the entropy production terms for that region. Comparison of Eqs. S47 and S60 under this assumption leads to the relation

\[ \Omega(A_{A\rightarrow B} - A_{B\rightarrow A}) = \frac{1}{2} \left(\Delta S_{B\rightarrow A} - \Delta S_{A\rightarrow B}\right), \]  

(S61)

which describes the empirically observed relationship shown in Fig. 3D of the main text. This relation can be extended to obtain a ratio of switching rates now uniquely determined by the difference in path entropy productions

\[ \frac{k_{B\rightarrow A}}{k_{A\rightarrow B}} \approx \exp\left[\frac{1}{2} (\Delta S_{B\rightarrow A} - \Delta S_{A\rightarrow B})\right], \]  

(S62)

which can then be re-expressed as a ratio of state occupation probabilities

\[ \frac{P_A}{P_B} \approx \exp\left[\frac{1}{2} (\Delta S_{B\rightarrow A} - \Delta S_{A\rightarrow B})\right]. \]  

(S63)
S9 Derivation of approximate entropy production and flow

It is possible to obtain a relation magnitude of velocity and magnitude of force along the minimum action path. This is done by following the method set out in [16], where the action (Eq. S28) is re-expressed in a compact notation as

\[
A = \frac{1}{2} \int_0^\tau (|\dot{x}|^2_d - 2|\dot{x}|_d|f|_d + |f|^2_d) \, dt + \int_0^\tau (|\dot{x}|_d f|_d - \dot{x} \cdot f) \, dt
\]

Equation S64

with \(|g|^2 = g_iD^{-1} g_j\) and \(g \cdot h = g_iD^{-1} h_j\). The first term in this action reaches its minimum value of zero when \(|\dot{x}|_d = |f|_d\). The second term depends solely on how the vector of forces \(f\) aligns with the velocity \(\dot{x}\). Once the first term has been minimized, changes that decrease the second term along with increasing the first term will always lead to an increase in the first term that is greater than the decrease in the second term. Therefore for all minimum action paths the following relation would be expected to hold

\[
\dot{x}_i D^{-1}_{ij} \dot{x}_j = f_i D^{-1}_{ij} f_j,
\]

Equation S65

where both sides are summed over \(i\) and \(j\). In the case of a uniform \(D_{ij}\) or the trivial case of a 1 dimensional model (e.g. concentration constrained Schlögl model) this relation reduces to

\[
|\dot{x}| = |f|.
\]

Equation S66

In the concentration constrained toggle switch model the inverse diffusion matrix \(D^{-1}\) is no longer uniform i.e. \(D_{11}^{-1} \neq D_{22}^{-1}\), but it does not possess cross terms i.e. \(D_{12}^{-1} = D_{21}^{-1} = 0\). This means that Eq. S66 remains a minimizer of the first part of the action in Eq. S64 but is no longer a unique one. We shall proceed by making the assumption that Eq. S66 still approximately holds. In order to obtain an expression linking velocity with deterministic force the additional assumption is then be made that the direction of motion opposes the force when travelling from steady state to saddle point and follows the force when travelling from saddle point to steady state. This can be symbolically expressed as

\[
\dot{x}_i \approx -f_i \quad \text{when} \quad t \leq t_s
\]

\[
\dot{x}_i \approx f_i \quad \text{when} \quad t > t_s,
\]

Equation S67

where \(t_s\) is the time it takes to reach the saddle point. This relation is shown in Fig. S2. Using these approximations along side a splitting of \(f\) into two parts \(f = f_1 - f_2\), a new approximate expression for \(\Delta S\) can be derived as

\[
\Delta S = 2\Omega \int_0^t \left(2f_{1,i}D^{-1}_{ij} f_{2,j} - f_{1,i}D^{-1}_{ij} f_{1,j} - f_{2,i}D^{-1}_{ij} f_{2,j}\right) \, dt \quad \text{when} \quad t \leq t_s
\]

\[
\Delta S = 2\Omega \int_{t_s}^t \left(f_{1,i}D^{-1}_{ij} f_{1,j} + f_{2,i}D^{-1}_{ij} f_{2,j} - 2f_{1,i}D^{-1}_{ij} f_{2,j}\right) \, dt \quad \text{when} \quad t > t_s.
\]

Equation S68

These entropy productions correspond to the entropy produced along a single segment. Thus, in order to determine entropy produced along the whole path they must be summed. We shall now consider the positive terms in the expression to approximate the entropy production (EP) and the
Figure S2: **Demonstration of relationship between f and \( \dot{x} \)**. Plot of \( f \) and \( \dot{x} \) along a minimum action path from high \( x \) to low \( x \) for the Schlögl model. It can be seen from the saddle point (black cross) to the final steady state (white dot) \( f \) and \( \dot{x} \) are identical. From the initial steady state (black dot) to the saddle point \( f \) and \( \dot{x} \) can be seen to be equal and opposite. This is in accordance with the relations obtained as Eq. S67.

negative terms to approximate the entropy flow (EF). Thus,

\[
\Delta S = \Omega \int_0^t (\text{EP} - \text{EF}) \, d\tilde{t}
\]  

(S69)

where

\[
\text{EP} = 4f_{1,i}D_{ij}^{-1}f_{2,j} \quad \text{when} \quad t \leq t_s
\]

(S70)

and

\[
\text{EP} = 2\left(f_{1,i}D_{ij}^{-1}f_{1,j} + f_{2,i}D_{ij}^{-1}f_{2,j}\right) \quad \text{when} \quad t > t_s,
\]

The subsections below will now lay out the definitions of \( f_1 \) and \( f_2 \) used for the various models considered in this paper.

**S9.1 \( f_1 \) and \( f_2 \) values for Schlögl model**

\[
f_1 = k_{+1}a - k_{-1}x
\]

\[
f_2 = k_{+2}x^3 - k_{-2}bx^2
\]  

(S72)
f_1 and f_2 values for toggle switch model

\[
f_1 = \left( \frac{k_r}{\tau + f^{r}_a} - \frac{k_- a}{\tau + f^{a}_r} \right) \\
f_2 = \left( \frac{K a - K_-}{Q b - Q_-} \right)
\]

(S73)

S10 Langevin path entropy production

The Langevin path entropy production is derived above (Eq. S54) to be

\[
\Delta S_{A\rightarrow B}^L = 2\Omega \int_0^t \dot{x}_i D^{-1}_{ij} f_j d\tilde{t},
\]

(S74)

which implies an entropy production rate of

\[
\frac{d(\Delta S^L)}{dt} = 2\Omega \dot{x}_i D^{-1}_{ij} f_j.
\]

(S75)

The paths found to satisfy the minimum action naturally have velocities (\(\dot{x}\)) that go to zero as they enter the steady state. Eq. S75 thus implies a zero rate of entropy production at steady state, which is clearly erroneous with respect to underlying master-equation dynamics. The application of constraints that stop the system from equilibrating leads to the expectation of broken detailed balance. Non-zero entropy production is a signature of broken detailed balance and therefore the absence of entropy production here indicates a significant issue in approximating to the Langevin formalism.

In Fig. S3 plots of Langevin path entropy production for the two species bistable gene switch both around the steady state and along switching paths are shown. It can be seen that this Langevin path entropy production approximates that of a reduced master-equation model, where the reactions creating a given species can be combined into a single “forward” reaction and likewise for the “reverse” reactions. Zero entropy production at steady state is now an expected result as this reduced model is an equilibrium model. Though this entropy production does not readily link to the master-equation entropy production of the full model, its consideration is still potentially illuminating as the probabilities of the Langevin paths derive from the ensemble of underlying master-equation trajectories. Thus, the Langevin path entropy production will not be able to provide the probability of a single trajectory, but should provide information on the probability of the ensemble of trajectories corresponding to a specific Langevin path.

S10.1 Generality of relation between Langevin and reduced master equation

The generality of the relationship discussed above could be questioned as it arises for a specific parameter set. Additionally, plots were only provided for one state and one switching direction. This subsection will provide additional figures in order to address these potential criticisms. Thus, Fig. S7 shows plots for the same parameter set as Fig. S3 but with plots that consider trajectories about the low A steady state and switching from high A to low A. Figs. S4 & S5 show the same plots for one alternative parameter set, and Figs. S6 & S8 show the same plots for another alternative parameter set. It can be seen that in all cases the entropy production from the Langevin formalism approximates that of consideration of the master equation of the reduced model. So, it seems reasonable to conclude that this is generally a good first approximation.
Figure S3: **Entropy production of steady states and switching paths.** (a) Entropy production of MAP calculated by reversal of Langevin level path as in [6]. Entropy production is taken as entropy produced between each pair of points in the $N = 600$ points the MAP was optimized over. (b) Histogram of entropy productions of 500 Gillespie simulations of full bistable switch model around the high A steady state, calculated via the same method. (c) Entropy production of above MAP when projected onto discrete chemical state space. Calculation was performed using transition probabilities from master equation corresponding to reduced model. (d) Histogram of entropy productions of 500 different Gillespie trajectories calculated via the same method. Inset shows schematic of reduced model. All 4 plots are produced using the following parameterization (given to 6 sf): $k = 11.9706$, $k_- = 0.502369$, $q = 5.07189$, $q_- = 0.207451$, $K = 1.74597$, $K_\infty = 0.0795083$, $Q = 0.439835$, $Q_- = 0.0286310$, $r = 2569.21$, $f = 2574.64$, $\Omega = 5000$. Appropriate volume (\(\Omega\)) rescaling of rates was used for the discrete simulations and calculations.
Figure S4: Same plots as in Fig. S3 but all 4 plots are now produced using the following alternative parameterization: $k = 26.0978$, $k_\perp = 2.17942$, $q = 25.7722$, $q_\perp = 1.41486$, $K = 8.24010$, $K_\perp = 0.600485$, $Q = 6.96377$, $Q_\perp = 0.165740$, $r = 3875.55$, $f = 9285.77$, $\Omega = 5000$. Appropriate volume ($\Omega$) rescaling of rates was used for the discrete simulations and calculations.

Figure S5: Same parameterization as Fig. S4 but now (a) & (c) are switches from high $A$ to low $A$, and (b) & (d) are simulated trajectories about the low $A$ steady state.
Figure S6: Same plots as in Fig. S3 but all 4 plots are now produced using the following alternative parameterization: \( k = 98.7494 \), \( k_- = 6.53603 \), \( q = 78.8964 \), \( q_- = 3.00145 \), \( K = 2.16553 \), \( K_- = 0.0566242 \), \( Q = 2.04786 \), \( Q_- = 0.135649 \), \( r = 3832.23 \), \( f = 9994.70 \), \( \Omega = 5000 \). Appropriate volume (\( \Omega \)) rescaling of rates was used for the discrete simulations and calculations.

Figure S7: Same parameterization as Fig. S3 but now (a) & (c) are switches from high \( A \) to low \( A \), and (b) & (d) are simulated trajectories about the low \( A \) steady state.
Figure S8: Same parameterization as Fig. S6, but now (a) & (c) are switches from high $A$ to low $A$, and (b) & (d) are simulated trajectories about the low $A$ steady state.
S11  Effect of diffusion and steady-state entropies

No consistent pattern was found for the effect of magnitude of diffusion matrix on action. The plot below considers ratios and differences as this provides a means of comparison of significantly different parameter regimes. Fig. S9 shows ratios of diffusion matrix traces against difference in minimum actions.

Figure S9: Red and blue points correspond to Schlögl and toggle switch model parameter sets, respectively. The link between the ratio of diffusion matrix traces and steady-state entropy production difference seems weak at best. The toggle switch data displays no apparent pattern. While the Schlögl data shows some limited pattern it is dominated by a few points of extremely large difference in action, other points of similarly small ratio of diffusion matrix traces do not show such large differences in action.

S12  Parameters for Figure 1

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Table S1: Parameter table for Schlögl model simulation shown in Fig. 1A (main text).

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Table S2: Parameter table for toggle switch model simulation shown in Fig. 1B (main text).
References


