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7 Reachability, Persistence, and
8 Constructive Chemical Reaction Networks

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11 **Abstract**

12 A positive dynamical system in \mathbb{R}^n is persistent if each component of
13 any solution trajectory originating in $\mathbb{R}_{>0}^n$ does not approach zero in
14 discrete time. Persistence is inherently important and has implications
15 for global asymptotic stability. We investigate the more stringent vac-
16 uous persistence for reaction networks: the asymptotic behavior is the
17 same, but we require it of trajectories originating in $\mathbb{R}_{\geq 0}^n$, barring ob-
18 vious degeneracies. Vacuous persistence covers situations with initial
19 states where all species are present implicitly but possibly not explic-
20 itly. Our main tools are the work of A. I. Vol’pert on the nullity and
21 positivity of species concentrations, and the enabling notion of reacha-
22 bility. We show that a reaction network is vacuously persistent if and
23 only if only the entire set of all species is both reach-closed and stoichio-
24 metrically admissible. We develop a theory of constructive networks
25 and use it to apply this characterization of vacuous persistence. We
26 obtain that a widely applicable reversibility condition along with the
27 absence of isomerism among elementary species assure vacuous persis-
28 tence. Also, we formally define binary enzymatic networks and show
29 that those that are futile and cascaded are vacuously persistent.

30 **Keywords.** Vacuous Persistence; Chemical Reaction Network; Species Composition; Construc-
31 tive Network; Reachability; A. I. Vol’pert’s Theorem; Futile Cascaded Enzymatic Network.

32 **Mathematics Subject Classification (2010).** Primary: 92C42. Secondary: 92C45, 34D05.

33 **1 Introduction**

34 This paper arose largely from the intuition that persistence should apply to
35 chemical reaction networks in which species are made of building blocks that

36 are conserved and processes are fundamentally reversible. For a trajectory
 37 $\mathbb{R}_{\geq 0} \rightarrow \mathbb{R}_{\geq 0}^n$, persistence is the property that there are no ω -limit points
 38 on the boundary $\partial\mathbb{R}_{\geq 0}^n = \mathbb{R}_{\geq 0}^n \setminus \mathbb{R}_{> 0}^n$, i.e. each of the n trajectory compo-
 39 nents $\mathbb{R}_{\geq 0} \rightarrow \mathbb{R}_{\geq 0}$ does not approach zero in discrete time. Many dynamical
 40 systems, including those that model mass-action reaction networks, are po-
 41 sitive: solution trajectories originating in $\mathbb{R}_{\geq 0}^n$ range in $\mathbb{R}_{\geq 0}^n$. Such a system
 42 is persistent if all solution trajectories originating in $\mathbb{R}_{> 0}^n$ are persistent.

43

44 Two results of Angeli, De Leenheer and Sontag [3] provide considerable in-
 45 sight into the persistence of conservative reaction networks with reasonable
 46 kinetics. Theorem 1 in this reference says that if such a network is persis-
 47 tent, then a fairly broad reversibility condition holds, namely the reaction
 48 vectors are positively dependent, i.e. there exists a vanishing linear combi-
 49 nation of the reaction vectors in which all coefficients are positive. Theorem
 50 2 in the same reference says that such a network is persistent provided every
 51 nonempty siphon contains the support of a nonzero nonnegative conserved
 52 vector of species. This latter result is applied to obtain examples of per-
 53 sistent networks among enzymatic networks presenting the common futility
 54 motif of molecular systems biology. As noted in the paper, the systematic
 55 determination of minimal siphons presents a challenge for a wider use of
 56 the result. We circumvent this difficulty by resorting to the concept of rea-
 57 chability and the results of A. I. Vol’pert on the nullity and positivity of
 58 trajectory components. One of our main results states the following.

59 **Theorem 1.1 (Theorem 4.6).** *Consider a mass-action reaction network*
 60 *for which all concentration trajectories are bounded. The following are equi-*
 61 *valent:*

- 62 • *The reaction network is vacuously persistent.*
- 63 • *Among the subsets of the set \mathcal{S} of all species, only the full set \mathcal{S} is*
 64 *both reach-closed and stoichiometrically admissible. \square*

65 Vacuous persistence (Definition 4.1) is the property that there are no oppor-
 66 tunities for non-persistence; specifically there are no ω -limit points on the
 67 boundary of any stoichiometric compatibility class with nonempty interior,
 68 including for trajectories originating on the boundary. Ordinary persistence
 69 allows the existence of boundary points that are ω -limit points, as long as
 70 they are so for trajectories originating on (and confined to) the boundary.
 71 Boundary initial states are quite common in experimental settings.

72

73 The siphon and reachability perspective are in fact somewhat dual. (See
74 Propositions 3.3 and 3.4.) So, in order to truly circumvent algorithmic
75 hurdles, we need mathematical tools that enable the use of Theorem 1.1
76 on large and relevant classes of reaction networks. We fulfill this need by
77 developing a theory of species composition and constructive networks. This
78 effort formalizes ideas on conservativeness as ubiquitously used in chemical
79 reaction network theory. Following is a notable outcome.

80 **Theorem 1.2 (Part of Theorem 7.7).** *Suppose that a mass-action reac-*
81 *tion network is explicitly-reversibly constructive. If there is no isomerism*
82 *among the elementary species, then the network is vacuously persistent. \square*

83 It is hence not coincidental that instances of non-persistence and of non-
84 obvious persistence in the literature always involve networks with isomerism
85 among the building blocks, e.g. isomerism among substrates and products
86 in enzymatic networks. (Theorem 7.7 also addresses global attraction.) The
87 absence of isomerism among elementary species is however not required for
88 vacuous persistence. In fact, we obtain another important result as the
89 culmination of a dedicated probing of enzymatic networks.

90 **Theorem 1.3 (Theorem 8.12).** *If a binary enzymatic network is futile*
91 *and cascaded, then it is vacuously persistent. \square*

92 The three networks of Angeli, De Leenheer and Sontag [3, Sections 6.1-6.3],
93 which were found to be persistent, are instances of binary enzymatic net-
94 works that are futile and cascaded.

95
96 Following is the organization of the paper. We assemble basic notions of
97 chemical reaction network theory in Section 2; most are from common lite-
98 rature but some are particular to our needs. Section 3 presents Vol’pert’s
99 reachability approach to understanding the positivity and nullity of species
100 concentrations, as well as certain immediate consequences of general inter-
101 est. Because the main theorem is instrumental and much more general than
102 the version we use, we include a specialized proof that we expect to be in-
103 formative to readers unfamiliar with Vol’pert’s work. We study persistence
104 in Section 4 with the material of Section 3 as a foundation. The develop-
105 ments on species composition and constructive networks begin in Section 5
106 with definitions and basic results, continue in Section 6 with results that
107 enable the use of the concepts of Section 5, and conclude in Section 7 with
108 ensuing results on reach-closures and persistence. Finally, in Section 8, we

109 propose definitions for and study binary enzymatic networks and several re-
 110 lated concepts, e.g. initial substrates, terminal products, reversing enzymes,
 111 futile networks, and cascaded networks.

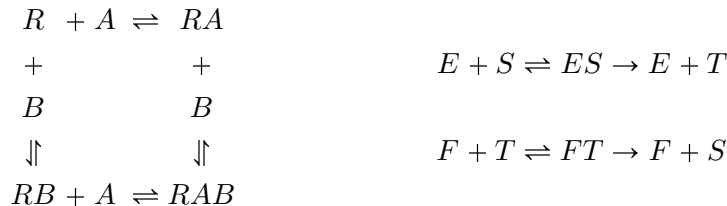
112

113 The following notations will be used throughout the paper.

114 **Notation 1.4.** For $m, n \in \mathbb{Z}$, $[m..n] = \{k \in \mathbb{Z} : m \leq k \leq n\}$. For $n \in \mathbb{Z}$
 115 and $i \in [1..n]$, $e_{n,i}$ will denote the n -tuple having 1 in position i and 0
 116 elsewhere. 0_n and 1_n will denote the n -tuples whose components all equal 0
 117 and 1 respectively. For $x = (x_1, \dots, x_n) \in \mathbb{R}^n$ and $\alpha = (\alpha_1, \dots, \alpha_n) \in \mathbb{R}^n$, if
 118 $x_i^{\alpha_i}$ is defined for all $i \in [1..n]$, then $x^\alpha = x_1^{\alpha_1} \dots x_n^{\alpha_n}$. The length of x is
 119 $|x| = |x_1| + \dots + |x_n| = \|x\|_{\ell^1}$. Its support is $\text{Supp}(x) = \{i \in [1..n] : x_i \neq 0\}$.
 120 For $x, y \in \mathbb{R}^n$, we write $x \leq y$ or $y \geq x$ (resp. $x < y$ or $y > x$) to mean that
 121 $x_i \leq y_i$ (resp. $x_i < y_i$) for all $i \in [1..n]$. The canonical inner product on \mathbb{R}^n
 122 will be denoted $\langle -, - \rangle$. For a finite set E , we denote $|E|$ the cardinality
 123 of E ; and when addition makes sense, we write $\text{sum}(E)$ for the sum of its
 124 elements.

125 2 Reaction Networks

126 This section gathers certain basic notions of Chemical Reaction Network
 127 Theory. Most can be found in the lecture notes of Feinberg [4] and the
 128 more recent tutorial of Gunawardena [7]. A system of chemical reactions
 129 is usually presented as a diagram that shows the interactions of chemical
 130 species; see for instance Figure 2.1. Following is a rendition of the formal
 131 definition that enables mathematical investigations.



(a) A receptor and two ligands in
allosteric interaction.

(b) Two enzymes and two sub-
strates in a futile cycle.

Figure 2.1: Examples of chemical reaction networks.

132 **Definition 2.1.** A *reaction network* is a triple $\mathcal{N} = (\mathcal{S}, \mathcal{C}, \mathcal{R})$ where

- 133 • \mathcal{S} , the set of *species*, is a nonempty finite set;
- 134 • \mathcal{C} , the set of *complexes*, is a nonempty finite subset of the nonnegative
135 orthant $\mathbb{Z}_{\geq 0}\mathcal{S}$ of $\mathbb{Z}\mathcal{S}$; and
- 136 • \mathcal{R} , the set of *reactions*, is a relation on \mathcal{C} , i.e. a subset of the Cartesian
137 square $\mathcal{C} \times \mathcal{C}$. □

138 Consistently with customary chemical notation, a reaction $R = (Q', Q'')$ is
139 also denoted $Q' \rightarrow Q''$. The complexes $\text{src}(R) := Q'$ and $\text{tgt}(R) := Q''$ are
140 respectively the *source* and the *target* of the reaction R , and we say that Q'
141 *reacts to* Q'' . The vector $\text{tgt}(R) - \text{src}(R) \in \mathbb{Z}\mathcal{S}$ is the *reaction vector* of R .
142 We set

$$143 \quad \underline{\text{src}}(R) := \text{Supp}(\text{src}(R)) \quad \text{and} \quad \underline{\text{tgt}}(R) := \text{Supp}(\text{tgt}(R)).$$

144 For $Q \in \mathcal{C}$, we set

$$145 \quad \mathcal{R}_Q^- := \{R \in \mathcal{R} : \text{src}(R) = Q\} \quad \text{and} \quad \mathcal{R}_Q^+ := \{R \in \mathcal{R} : \text{tgt}(R) = Q\}.$$

146 The *reaction graph* is the directed graph $(\mathcal{C}, \mathcal{R})$ with \mathcal{C} as the set of ver-
147 tices and \mathcal{R} as the set of edges. Given complexes $Q', Q'' \in \mathcal{C}$, we say that
148 Q' *ultimately reacts to* Q'' if there is a path from Q' to Q'' in the reaction
149 graph. The connected components of the associated undirected graph are
150 the *linkage classes* of the network \mathcal{N} . The network \mathcal{N} is *weakly reversible*
151 if every path in the reaction graph has a reverse path, i.e. if whenever Q'
152 ultimately reacts to Q'' , it also holds that Q'' ultimately reacts to Q' .

153
154 The *stoichiometric space* of \mathcal{N} is the subspace S of $\mathbb{R}\mathcal{S}$ spanned by the
155 reaction vectors. The *rank* of \mathcal{N} is the dimension of S . A *stoichiome-*
156 *tric compatibility class* is any $P = (u + S) \cap \mathbb{R}_{\geq 0}\mathcal{S}$ where $u \in \mathbb{R}_{\geq 0}\mathcal{S}$. We
157 have $P = P_{>0} \sqcup P_{\not>0}$, where $P_{>0} = (u + S) \cap \mathbb{R}_{>0}\mathcal{S}$ is the interior of P and
158 $P_{\not>0} = (u + S) \cap ((\mathbb{R}_{\geq 0}\mathcal{S}) \setminus (\mathbb{R}_{>0}\mathcal{S}))$ is the boundary of P , both relative to
159 the affine space $u + S$. We will say that the class P is *degenerate* if $P_{>0} = \emptyset$,
160 or equivalently if $P_{\not>0} = P$. For the following geometric properties of P , we
161 refer the reader to Rockafellar [10] for general background and to Ander-
162 son and Shiu [2, Section 2.3] for a directly relevant discussion. The class
163 P is a (convex, possibly unbounded) polyhedron. Let A be a face of P . If
164 $0 \in P$, we suppose that $A \neq \{0\}$. There is an (unique, nonempty) inclusion-
165 minimal set $\mathcal{Z} \subseteq \mathcal{S}$ of species such that $A = (u + S) \cap \mathbb{R}_{\geq 0}\mathcal{Z}$. The relative
166 interior of A in the sense of polyhedral geometry, which we shall herein call
167 the *intrinsic interior*, is $\text{int}(A) = (u + S) \cap \mathbb{R}_{>0}\mathcal{Z}$. We define the *support*

168 of the face A to be $\text{Supp}(A) = \mathcal{L}$. The intrinsic interior $\text{int}(A)$ consists of
 169 the points of P whose support is $\text{Supp}(A)$. We remark for the particular
 170 case $A = P$ that the intrinsic interior $\text{int}(P)$ and the interior $P_{>0}$ relative
 171 to $u + S$ coincide only if P is nondegenerate. Also, if A is a vertex, and in
 172 particular if $0 \in P$ and $A = \{0\}$, then $\text{int}(A) = A$. The class P is the disjoint
 173 union of the intrinsic interiors of its faces.

174

175 We augment Definition 2.1 with the following customary restrictions:

- 176 (i) Every species is in the support of at least one complex;
 177 (ii) No complex reacts to itself; and
 178 (iii) Every complex is the source or the target of at least one reaction.

179 Furthermore, $\mathbb{R}\mathcal{S}$ is equipped with the Euclidean structure with respect to
 180 which \mathcal{S} is an orthonormal basis. The inner product is denoted $\langle -, - \rangle$.

181

182 A kinetics on the reaction network $\mathcal{N} = (\mathcal{S}, \mathcal{C}, \mathcal{R})$ is any family $K = (K_R)_{R \in \mathcal{R}}$
 183 in which for each $R \in \mathcal{R}$, K_R is a continuous function $\mathbb{R}\mathcal{S} \rightarrow \mathbb{R}\mathcal{S}$. Associ-
 184 ated with a kinetics K is the species formation function
 185 $F(K, -) = (F_X(K, -))_{X \in \mathcal{S}} : \mathbb{R}\mathcal{S} \rightarrow \mathbb{R}\mathcal{S}$ given for $u \in \mathbb{R}\mathcal{S}$ by

$$186 \quad F(K, u) = \sum_{R \in \mathcal{R}} K_R(u) (\text{tgt}(R) - \text{src}(R)) . \quad (2.1)$$

187 The most common kinetics is the mass-action kinetics. It is given by a fam-
 188 ily $k = (k_R)_{R \in \mathcal{R}} \in \mathbb{R}_{>0}^{\mathcal{R}}$ of reaction rate constants and $K_R(u) = k_R u^{\text{src}(R)}$;
 189 we write $F(K, -) = F(k, -)$.

190

191 To say that the network \mathcal{N} is governed by the kinetics K is to say that
 192 there is a function $c = (c_X)_{X \in \mathcal{S}} : \mathbb{R}_{\geq 0} \rightarrow \mathbb{R}\mathcal{S}$ of time representing the con-
 193 centration of species, and that the function c obeys the dynamical system

$$194 \quad \dot{c}(t) = F(K, c(t)) . \quad (2.2)$$

195 The function $F(K, -)$ ranges into the stoichiometric space S , so the solu-
 196 tions of equation (2.2) are confined to affine subspaces of $\mathbb{R}\mathcal{S}$ parallel to
 197 S . Under certain conditions on the kinetics K , which are satisfied if K is
 198 a mass-action kinetics, every solution that originates at a nonnegative state
 199 remains nonnegative, and hence is in fact confined to a stoichiometric com-
 200 patibility class.

201

202 Consider the function $\hat{F}(K, -) = \left(\hat{F}_Q(K, -) \right)_{Q \in \mathcal{C}} : \mathbb{R}^{\mathcal{S}} \rightarrow \mathbb{R}^{\mathcal{C}}$ given for $Q \in \mathcal{C}$
 203 and $u \in \mathbb{R}^{\mathcal{S}}$ by

$$204 \quad \hat{F}_Q(K, u) = \sum_{R \in \mathcal{R}_Q^+} K_R(u) - \sum_{R \in \mathcal{R}_Q^-} K_R(u).$$

205 Then we have $F(K, u) = \sum_{Q \in \mathcal{C}} \hat{F}_Q(K, u) Q$. An *equilibrium state* of the net-
 206 work \mathcal{N} for the kinetics K is any $u \in \mathbb{R}^{\mathcal{S}}$ such that $F(K, u) = 0$. An
 207 equilibrium state $u \in \mathbb{R}^{\mathcal{S}}$ that satisfies the stronger condition $\hat{F}(K, u) = 0$
 208 is a *complex-balanced state*. The network is said to be *complex-balancing* if
 209 it admits a positive complex-balanced state. It is known that a mass-action
 210 complex-balancing network must be weakly reversible.

211 **Definition 2.2.**

- 212 • A *binding* or *association reaction* is a reaction $Q' \rightarrow Q''$
 213 such that $|Q'| \geq 2$ and $|Q''| = 1$. (Q'' is a species, Q' is not.)
- 214 • An *unbinding* or *dissociation reaction* is a reaction $Q' \rightarrow Q''$ such that
 215 $|Q'| = 1$ and $|Q''| \geq 2$. (Q' is a species, Q'' is not.)
- 216 • An *isomerization reaction* is a reaction $Q' \rightarrow Q''$ such that
 217 $|Q'| = |Q''| = 1$. (Both Q' and Q'' are species.)
- 218 • Two species X' and X'' are *stoichiometrically isomeric* if $X' - X'' \in S$.
- 219 • A *bound species* is a species that is the target of a binding reaction or
 220 the source of a dissociation reaction (or both). \square

221 The notion of stoichiometric isomerism is intended to account for sequences
 222 of reactions whose net effect is to transform one species into another, as is
 223 the case for instance in enzymatic reactions. Stoichiometric isomerism gives
 224 rise to an equivalence relation on the set \mathcal{S} of species.

225
 226 For illustration, the network of Figure 2.1(a) consists of four reversible bin-
 227 ding reactions and the bound species are RA , RB , and RAB . It should be
 228 noted that it is not because these species are denoted with expressions of
 229 more than one letter that they are bound species. The network of Figure
 230 2.1(b) consists of two binding reactions and four dissociation reactions, and
 231 the bound species are ES and FT . The species S and T form a stoichiome-
 232 tric isomerism class.

233 3 On the Positivity and Nullity of Species Concentrations

234 A. I. Vol’pert [20] has studied the question of when the solutions of certain
 235 dynamical systems have components that remain zero while the other com-
 236 ponents become and remain positive. The theory is developed for differential
 237 equations on graphs, of which mass-action kinetics dynamical systems are
 238 instances. The work is revisited in Vasil’ev, Vol’pert and Khudyaev [18]
 239 (with an erratum in [19]). The more recent book of Vol’pert and Hudjaev
 240 [21] covers the topic in Chapter 12. In this section, we present this work
 241 in a form specialized to chemical reaction networks and with a view to-
 242 ward showing in Section 4 how it contributes to results on persistence and
 243 asymptotic stability. A reaction network $\mathcal{N} = (\mathcal{S}, \mathcal{C}, \mathcal{R})$ and a set $\mathcal{Z} \subseteq \mathcal{S}$
 244 of species are fixed throughout this section.

245
 246 We define the following sets:

$$\begin{aligned}
 247 \quad \text{React}(\mathcal{Z}) &:= \{R \in \mathcal{R} : \text{src}(R) \subseteq \mathcal{Z}\}; \\
 248 \quad \text{Prod}(\mathcal{Z}) &:= \left[\bigcup_{R \in \text{React}(\mathcal{Z})} \text{tgt}(R) \right] \setminus \mathcal{Z}; \\
 249 \quad \text{Reach}_0(\mathcal{Z}) &:= \mathcal{Z}; \\
 250 \quad \text{Reach}_{\leq r}(\mathcal{Z}) &:= \bigcup_{\rho=0}^r \text{Reach}_{\rho}(\mathcal{Z}), \text{ for } r \in \mathbb{Z}_{\geq 0}; \\
 251 \quad \text{Reach}_r(\mathcal{Z}) &:= \text{Prod}(\text{Reach}_{\leq r-1}(\mathcal{Z})), \text{ for } r \in \mathbb{Z}_{\geq 1}.
 \end{aligned}$$

253 The sets $\text{Reach}_r(\mathcal{Z})$ are pairwise disjoint. Also, if $\text{Reach}_{r_0}(\mathcal{Z}) = \emptyset$, then
 254 $\text{Reach}_r(\mathcal{Z}) = \emptyset$ for all $r \geq r_0$. And because \mathcal{S} is finite, there does exist
 255 $r_0 \geq 0$ such that $\text{Reach}_{r_0}(\mathcal{Z}) = \emptyset$. We pose:

$$\begin{aligned}
 256 \quad \text{Reach}(\mathcal{Z}) &:= \bigsqcup_{r=0}^{\infty} \text{Reach}_r(\mathcal{Z}); \\
 257 \quad \text{NonReach}(\mathcal{Z}) &:= \mathcal{S} \setminus \text{Reach}(\mathcal{Z}).
 \end{aligned}$$

259 Definition 3.1.

- 260 • The species in $\text{Reach}(\mathcal{Z})$ and in $\text{NonReach}(\mathcal{Z})$ are said to be *reachable*
 261 from \mathcal{Z} and *non-reachable* from \mathcal{Z} respectively.
- 262 • The species in $\text{Reach}_r(\mathcal{Z})$ are said to have *reachability index* r with
 263 respect to \mathcal{Z} .

- 264 • The set $\text{Reach}(\mathcal{Z})$ is the *reach-closure* of the set \mathcal{Z} .
- 265 • The set \mathcal{Z} is said to be *reach-closed* if $\text{Reach}(\mathcal{Z}) = \mathcal{Z}$. □

266 Vol’pert’s reachability is an instance of the notion of reachability in the
 267 theory of Petri nets. There is abundant literature on Petri nets and a stan-
 268 dard comprehensive reference is Murata [9]. Another notion from Petri net
 269 theory, that of siphon, is the subject of growing use in reaction network
 270 theory; see for example Angeli, De Leenheer and Sontag [3], Anderson [1],
 271 Shiu and Sturfels [12], and Anderson and Shiu [2]. We define siphons
 272 in Definition 3.2 and examples can be found in the literature just cited.
 273 Propositions 3.3 and 3.4 show that there is a certain duality relationship
 274 between siphons and reach-closures. The reachability perspective has been
 275 used advantageously in Siegel and Chen [13] and Siegel and MacLean [14].

276 **Definition 3.2.** The set \mathcal{Z} is a *siphon* provided
 277 $(R \in \mathcal{R} \text{ and } \mathcal{Z} \cap \underline{\text{tgt}}(R) \neq \emptyset) \Rightarrow (\mathcal{Z} \cap \underline{\text{src}}(R) \neq \emptyset)$. □

278 **Proposition 3.3.** *The complement $\text{NonReach}(\mathcal{Z})$ of the reach-closure of*
 279 *\mathcal{Z} is a siphon.*

280 *Proof.* For any reaction $R \in \mathcal{R}$, we have:

$$\begin{aligned}
 281 \quad \text{NonReach}(\mathcal{Z}) \cap \underline{\text{src}}(R) = \emptyset &\Leftrightarrow \underline{\text{src}}(R) \subseteq \text{Reach}(\mathcal{Z}) \\
 282 &\Rightarrow \underline{\text{tgt}}(R) \subseteq \text{Reach}(\mathcal{Z}) \\
 283 &\Leftrightarrow \text{NonReach}(\mathcal{Z}) \cap \underline{\text{tgt}}(R) = \emptyset. \quad \square
 \end{aligned}$$

284 **Proposition 3.4.** *The set \mathcal{Z} is reach-closed if and only if its complement*
 285 *$\mathcal{S} \setminus \mathcal{Z}$ is a siphon.*

Proof.

$$\begin{aligned}
 286 \quad &(\mathcal{S} \setminus \mathcal{Z} \text{ is a siphon}) \\
 287 \quad &\Leftrightarrow \left((R \in \mathcal{R} \text{ and } (\mathcal{S} \setminus \mathcal{Z}) \cap \underline{\text{src}}(R) = \emptyset) \Rightarrow ((\mathcal{S} \setminus \mathcal{Z}) \cap \underline{\text{tgt}}(R) = \emptyset) \right) \\
 288 \quad &\Leftrightarrow \left((R \in \mathcal{R} \text{ and } \underline{\text{src}}(R) \subseteq \mathcal{Z}) \Rightarrow (\underline{\text{tgt}}(R) \subseteq \mathcal{Z}) \right) \\
 289 \quad &\Leftrightarrow \bigcup_{R \in \text{React}(\mathcal{Z})} \underline{\text{tgt}}(R) \subseteq \mathcal{Z} \\
 290 \quad &\Leftrightarrow \text{Prod}(\mathcal{Z}) = \emptyset \\
 291 \quad &\Leftrightarrow \text{Reach}(\mathcal{Z}) = \mathcal{Z}. \quad \square
 \end{aligned}$$

292 Propositions 3.3 and 3.4 immediately imply the following result, which is
293 already intuitively clear.

294 **Proposition 3.5.** *The reach-closure $\text{Reach}(\mathcal{Z})$ of \mathcal{Z} is reach-closed;*
295 $\text{Reach}(\text{Reach}(\mathcal{Z})) = \text{Reach}(\mathcal{Z})$. □

296 Moreover, $\text{Reach}(\mathcal{Z})$ is the inclusion-minimal subset of \mathcal{S} that contains \mathcal{Z}
297 and is reach-closed. In particular, if $\mathcal{Z}' \subseteq \text{Reach}(\mathcal{Z})$, then
298 $\text{Reach}(\mathcal{Z}') \subseteq \text{Reach}(\mathcal{Z})$. This expresses a transitivity feature of reachabi-
299 lity: For $X \in \mathcal{S}$ and $\mathcal{Z}' \subseteq \mathcal{S}$, if X is reachable from \mathcal{Z}' , and if all elements
300 of \mathcal{Z}' are reachable from \mathcal{Z} , then X is reachable from \mathcal{Z} .

301

302 For $u \in \mathbb{R}_{\geq 0}\mathcal{S}$, we set

$$\begin{aligned} 303 \quad \text{Reach}(u) &:= \text{Reach}(\text{Supp}(u)), \\ 304 \quad \text{NonReach}(u) &:= \text{NonReach}(\text{Supp}(u)). \end{aligned}$$

306 For the rest of this section, the reaction network $\mathcal{N} = (\mathcal{S}, \mathcal{C}, \mathcal{R})$ is as-
307 sumed to be governed by mass-action kinetics. The family of reaction rate
308 constants is $k = (k_R)_{R \in \mathcal{R}}$ and we set $F = F(k, -)$ for the species formation
309 function. Associated with the dynamical system $\dot{c}(t) = F(c(t))$ is the evo-
310 lution semigroup $C : \mathbb{R}_{\geq 0}\mathcal{S} \times \mathbb{R}_{\geq 0} \rightarrow \mathbb{R}_{\geq 0}\mathcal{S}$. Thus, C is continuous; for
311 every $u \in \mathbb{R}_{\geq 0}\mathcal{S}$, the map $\mathbb{R}_{\geq 0} \rightarrow \mathbb{R}_{\geq 0}\mathcal{S}, t \mapsto C(u, t)$ is the concentration
312 trajectory originating at u ; and for all $u \in \mathbb{R}_{\geq 0}\mathcal{S}$ and $t, t' \in \mathbb{R}_{\geq 0}$, we have
313 $C(u, t + t') = C(C(u, t), t')$.

314 **Theorem 3.6 (A. I. Vol'pert).** *Let $c = (c_X)_{X \in \mathcal{S}} : \mathbb{R}_{\geq 0} \rightarrow \mathbb{R}\mathcal{S}$ be a con-*
315 *centration trajectory. Let $X \in \mathcal{S}$ be a species.*

- 316 • *If $X \in \text{NonReach}(c(0))$, then $c_X(t) = 0$ for all $t \geq 0$; and*
- 317 • *If $X \in \text{Reach}(c(0))$, then $c_X(t) > 0$ for all $t > 0$.*

318 *Hence, we have $\text{Supp}(c(t)) = \text{Reach}(c(0))$ for all $t > 0$.*

319 We present a proof of Theorem 3.6 at the end of this section. We focus for
320 now on its implications. A stoichiometric compatibility class P is fixed for
321 the rest of this section. We define

$$322 \quad \Phi(P) := \{u \in P : \text{Supp}(u) \text{ is reach-closed}\}. \quad (3.1)$$

323 Note that

$$324 \quad P_{>0} \subseteq \Phi(P) \subseteq P. \quad (3.2)$$

325 The combination of Theorem 3.6 and Proposition 3.5 readily gives:

326 **Proposition 3.7.** *For any concentration trajectory $c : \mathbb{R}_{\geq 0} \rightarrow P$, we have*
 327 *$c(t) \in \Phi(P)$ for all $t > 0$; only the initial point $c(0)$ may (but need not) be in*
 328 *the subset $P \setminus \Phi(P)$ of the boundary $P_{\succ 0}$. \square*

329 Vol'pert's Theorem also leads to certain properties of the faces of stoichio-
 330 metric compatibility classes.

331 **Proposition 3.8.** *Let A be a face of P . The following are equivalent:*

332 (i) *The intrinsic interior $\text{int}(A)$ of A contains a trajectory.*

333 (ii) *The support $\text{Supp}(A)$ of A is reach-closed.*

334 (iii) *The intrinsic interior $\text{int}(A)$ of A is forward-invariant.*

335 (iv) *The face A is forward-invariant.*

336 *If these conditions are satisfied and if A is bounded, then A contains an*
 337 *equilibrium state.*

338 Refer to the paragraph introducing stoichiometric compatibility classes in
 339 Section 2 for the notions of intrinsic interior and support of a face. The im-
 340 plication (i) \Rightarrow (iv) can be obtained by using an unpublished result of Sontag
 341 [16] in the more general context of dynamical systems on manifolds with
 342 boundary.

343 *Proof.* If a trajectory c ranges in $\text{int}(A)$, then all the points along c have
 344 $\text{Supp}(A)$ as their support. So we get the implication (i) \Rightarrow (ii) from Propo-
 345 sition 3.7. We have (ii) \Rightarrow (iii) by Theorem 3.6. The implication (iii) \Rightarrow (i)
 346 is trivial. We have (iii) \Rightarrow (iv) by the continuity of the evolution semi-group
 347 C and because A is the topological closure of $\text{int}(A)$. We have (iv) \Rightarrow (iii)
 348 because Theorem 3.6 shows that the support along a trajectory cannot get
 349 inclusion-smaller. Now suppose that A is forward-invariant and bounded.
 350 Then A is convex, compact and forward-invariant. As a result, A contains
 351 an equilibrium state. \square

352 Recall that a point $z \in \mathbb{R}_{\geq 0}\mathcal{S}$ is an equilibrium state if $F(z) = 0$. This is
 353 equivalent to the condition that $C(z, t) = z$ for all $t \in \mathbb{R}_{\geq 0}$. We denote $\Omega(P)$
 354 the set of equilibrium states in P . A point $z \in \mathbb{R}_{\geq 0}\mathcal{S}$ is an ω -limit point
 355 of a point $u \in \mathbb{R}_{\geq 0}\mathcal{S}$ provided there exists a sequence $(t_n)_{n \geq 0}$ in $\mathbb{R}_{\geq 0}$ such
 356 that $\lim_{n \rightarrow \infty} t_n = \infty$ and $\lim_{n \rightarrow \infty} C(u, t_n) = z$. We denote $\omega(u)$ the set of ω -limit
 357 points of u . And for $U \subseteq \mathbb{R}_{\geq 0}\mathcal{S}$, we set $\omega(U) = \bigcup_{u \in U} \omega(u)$. Note that, while

358 $\omega(u)$ is the ω -limit set of the point u , $\omega(U)$ is a (possibly proper) subset of
 359 the ω -limit set of the set U . Evidently,

$$360 \quad \Omega(P) \subseteq \omega(P) \subseteq P.$$

361 It is possible to have $\Omega(P) = \omega(P)$. Sontag [15, Theorem 1] has proved
 362 that this holds when the network is weakly reversible and the number of
 363 complexes exceeds the rank by precisely one. Also, this is trivially the case
 364 if the class P contains a point which is a global attractor.

365 **Proposition 3.9.**

- 366 • We have $\Omega(P) \subseteq \Phi(P)$; the support of any equilibrium state in P is
 367 reach-closed.
- 368 • If all trajectories in P are bounded, then $\omega(P) \subseteq \Phi(P)$; the support of
 369 any ω -limit point of any point of P is reach-closed.

370 The assertions in Proposition 3.9 have already been established through
 371 other means. The property that equilibrium states have reach-closed sup-
 372 port is an earlier result of Feinberg [5, Proposition 5.3.1]. The fact that the
 373 ω -limit points of any $u \in P$ have reach-closed supports is proved by Angeli,
 374 De Leenheer and Sontag [3, Proposition 5.4], and also by Anderson [1, Theo-
 375 rem 2.5] for $u \in P_{>0}$. The first of the two assertions in Lemma 2.8 of this
 376 latter reference is the containment relationship $P_{\neq 0} \cap \Omega(P) \subseteq P_{\neq 0} \cap \Phi(P)$
 377 for weakly reversible deficiency-zero networks.

378 *Proof.* Let $z \in P$ and $t > 0$. By Proposition 3.7, we have $C(z, t) \in \Phi(P)$. If
 379 $z \in \Omega(P)$, then $C(z, t) = z$, and so $z \in \Phi(P)$. The inclusion $\Omega(P) \subseteq \Phi(P)$
 380 is thus proved. For the inclusion $\omega(P) \subseteq \Phi(P)$, the key idea is that when
 381 trajectories in P are bounded, the ω -limit set $\omega(u)$ of any $u \in P$ is backward-
 382 invariant (in addition to being forward-invariant unconditionally). The de-
 383 tails are as follows. Suppose that trajectories in P are bounded and let
 384 $u \in P$ and $z \in \omega(u)$. Let $t_0 > 0$. There exists a sequence $(t_n)_{n \geq 0}$ in $\mathbb{R}_{\geq 0}$
 385 such that $t_n \geq t_0$ for all $n \geq 0$, $\lim_{n \rightarrow \infty} t_n = \infty$, and $\lim_{n \rightarrow \infty} C(u, t_n) = z$. We have
 386 in P the sequence $(C(u, t_n - t_0))_{n \geq 0}$. This sequence is bounded and P is a
 387 closed set, so there exists a sequence $(n_k)_{k \geq 0}$ in $\mathbb{Z}_{\geq 0}$ such that $\lim_{k \rightarrow \infty} n_k = \infty$
 388 and $z^0 = \lim_{k \rightarrow \infty} C(u, t_{n_k} - t_0)$ exists in P . We then have

$$389 \quad C(z^0, t_0) = \lim_{k \rightarrow \infty} C(C(u, t_{n_k} - t_0), t_0) = \lim_{k \rightarrow \infty} C(u, t_{n_k}) = z.$$

390 Then, by Proposition 3.7, $\text{Supp}(z)$ is reach-closed. □

391 The condition that all trajectories in P be bounded is of course satisfied
 392 if the class P is bounded. The condition is also satisfied if the network is
 393 complex-balanced and the class P is nondegenerate. Siegel and MacLean
 394 [14, Lemma 3.5] have derived this from the fact that the canonical Lyapunov
 395 function decreases along trajectories.

396

397 The remaining matter for this section is a proof of Vol’pert’s Theorem 3.6.
 398 We begin with a few preparatory steps. First, we note that if a trajectory
 399 originates at a nonnegative state, then it remains nonnegative. This is a
 400 well-known fact with several proofs in the literature, including the work of
 401 Vol’pert. Second, we record the following elementary fact of calculus for
 402 convenient subsequent reference.

403 **Lemma 3.10.** *Let $I \subseteq \mathbb{R}$ be an interval and let $\alpha : I \rightarrow \mathbb{R}$ and $\beta : I \rightarrow \mathbb{R}$
 404 be continuous functions. Consider the C^1 -function $A : I \times I \rightarrow \mathbb{R}$ given by*

405 $A(t_0, t) = \int_{t_0}^t \alpha(\tau) d\tau$. *A function $x : I \rightarrow \mathbb{R}$ satisfies $\dot{x}(t) = \alpha(t)x(t) + \beta(t)$*

406 *if and only if $x(t) = e^{A(t_0, t)}x(t_0) + \int_{t_0}^t e^{A(\tau, t)}\beta(\tau)d\tau$. \square*

407 The final step in preparation for the proof of Vol’pert’s Theorem 3.6 is to
 408 note certain alternate expressions of the species formation function. For any
 409 species $X \in \mathcal{S}$ and any reaction $R \in \mathcal{R}$, let $\sigma(R, X) = \langle X, \text{tgt}(R) - \text{src}(R) \rangle$.
 410 Then the species formation function F of Equation (2.1) is given by

$$411 \quad F_X(u) = \sum_{R \in \mathcal{R}, \text{src}(R) \cup \text{tgt}(R) \ni X} \sigma(R, X) k_R u^{\text{src}(R)}. \quad (3.3)$$

412 Let \mathcal{Z} be a siphon. For any reaction $R \in \mathcal{R}$ such that
 413 $\mathcal{Z} \cap (\text{src}(R) \cup \text{tgt}(R)) \neq \emptyset$, we in fact have $\mathcal{Z} \cap \text{src}(R) \neq \emptyset$. Consider
 414 then a selected species $\nu(R) \in \mathcal{Z} \cap \text{src}(R)$. Then there is a (unique) mono-
 415 mial function μ_R on $\mathbb{R}_{\mathcal{S}}$ such that $u^{\text{src}(R)} = \mu_R(u) u_{\nu(R)}$. It follows that for
 416 $X \in \mathcal{Z}$, Equation (3.3) becomes

$$417 \quad F_X(u) = \sum_{R \in \mathcal{R}, \text{src}(R) \cup \text{tgt}(R) \ni X} \sigma(R, X) k_R \mu_R(u) u_{\nu(R)}. \quad (3.4)$$

418 On another hand, consider for any species $X \in \mathcal{S}$ the polynomial functions

419 g_X and h_X on $\mathbb{R}\mathcal{S}$ given by

$$420 \quad g_X(u) u_X = \sum_{R \in \mathcal{R}, \sigma(R, X) < 0} |\sigma(R, X)| k_R u^{\text{src}(R)} ;$$

$$421 \quad h_X(u) = \sum_{R \in \mathcal{R}, \sigma(R, X) > 0} \sigma(R, X) k_R u^{\text{src}(R)} .$$

422
423 They give rise to another relevant form of Equation (3.3):

$$424 \quad F_X(u) = -g_X(u) u_X + h_X(u) . \quad (3.5)$$

425 We are now ready for the announced proof.

426 *Proof (Proof of Vol'pert's Theorem 3.6).* Recall from Proposition 3.3 that
427 $\mathcal{Z} := \text{NonReach}(c(0))$ is a siphon. Therefore, thanks to Equation (3.4), we
428 may consider the system of differential equations

$$429 \quad \dot{v}_X(t) = \sum_{R \in \mathcal{R}, \text{src}(R) \cup \text{tgt}(R) \ni X} \sigma(R, X) k_R \mu_R(c(t)) v_{\nu(R)}(t) \quad (3.6)$$

430 for $v = (v_X)_{X \in \mathcal{Z}} : \mathbb{R}_{\geq 0} \rightarrow \mathbb{R}^{\mathcal{Z}}$. We impose the initial condition $v(0) = 0$.
431 Then the zero function is a solution. It results from Equation (3.4) that the
432 function $t \mapsto (c_X(t))_{X \in \mathcal{Z}}$ is also a solution. Therefore c_X is identically zero
433 for all $X \in \mathcal{Z}$.

434

435 Now let X be any species and let $G_{X,c}(t_0, t) = \int_{t_0}^t g_X(c(\tau)) d\tau$. With Equa-
436 tion (3.5) and Lemma 3.10, we get

$$437 \quad c_X(t) = e^{-G_{X,c}(0,t)} c_X(0) + \int_0^t e^{-G_{X,c}(\tau,t)} h_X(c(\tau)) d\tau .$$

438 We have $c(\tau) \geq 0$ for all $\tau \geq 0$, so $c_X(t) \geq e^{-G_{X,c}(0,t)} c_X(0)$ for all $t \geq 0$.
439 Therefore, if $X \in \text{Supp}(c(0))$, then $c_X(t) > 0$ for all $t \geq 0$.

440

441 Let $r \in \mathbb{Z}_{\geq 1}$ and assume for induction that if X has reachability index $< r$,
442 then $c_X(t) > 0$ for all $t > 0$. Suppose that X has reachability index r .
443 Then there exists a reaction $R \in \mathcal{R}$ such that $X \in \text{tgt}(R)$ and all species
444 in $\text{src}(R)$ have reachability index $< r$; in particular, $X \notin \text{src}(R)$. Because
445 $X \notin \text{src}(R)$ and $X \in \text{tgt}(R)$, we have $\sigma(R, X) > 0$. Plus, as noted earlier, we
446 have $c(\tau) \geq 0$ for all $\tau \geq 0$. Therefore, $h_X(c(\tau)) \geq \sigma(R, X) k_R (c(\tau))^{\text{src}(R)}$
447 for all $\tau \geq 0$. Because all species in $\text{src}(R)$ have reachability index $< r$, the
448 induction hypothesis implies that $(c(\tau))^{\text{src}(R)} > 0$ for all $\tau > 0$. Therefore,
449 $h_X(c(\tau)) > 0$ for all $\tau > 0$. As a result, $c_X(t) > 0$ for all $t > 0$. \square

450 **4 Reachability Approach to Persistence**

451 We now study persistence using the reachability work of Vol’pert and its
452 consequences discussed in Section 3. Some of the results we obtain have
453 equivalent formulations in various references which we indicate in each case.
454 For these, our contribution is to show how they derive from Vol’pert’s Theo-
455 rem. The main result in this section is Theorem 4.6. It provides a necessary
456 and sufficient condition for vacuous persistence, which is essentially the ab-
457 sence of boundary ω -limit points. A reaction network $\mathcal{N} = (\mathcal{S}, \mathcal{C}, \mathcal{R})$ is
458 fixed for this section.

459 **Definition 4.1.** The reaction network is *persistent* (resp. *vacuously persis-*
460 *tent*) if we have $\omega(P_{>0}) \subseteq P_{>0}$ (resp. $\omega(P) \subseteq P_{>0}$) for every nondegenerate
461 stoichiometric compatibility class P . \square

462 Persistence is studied in other areas of mathematics; see for example the dis-
463 cussion in Angeli, De Leenheer and Sontag [3, Section 1.2]. In a persistent
464 reaction network, if all species are present at initial time, then no species
465 approach extinction, either in discrete or continuous time. In a vacuously
466 persistent reaction network, this asymptotic property holds even if not all
467 species are present at initial time, as long as the stoichiometric compatibility
468 class, by being nondegenerate, allows for the presence of all species.ordi-
469 nary persistence can occur with ‘opportunities for non-persistence’, whereby
470 we mean boundary points that are ω -limit points, but only for trajectories
471 confined to the boundary. The qualifier ‘vacuous’ is to indicate the absence
472 of such opportunities. Vacuous persistence is relevant to biochemical exper-
473 imental settings in which some species may not be initially present.

474
475 Persistence is not only important in and of itself, it also affects global asymp-
476 totic stability. For instance, if a reaction network is persistent and complex-
477 balancing, then each nondegenerate class P contains a unique equilibrium
478 state, which is complex-balanced and is an attractor of $P_{>0}$; see Siegel and
479 MacLean [14, Theorem 3.2]. Extending this fact, we note that with vacu-
480 ous persistence instead, the equilibrium state is an attractor of P . Efforts
481 to understand the persistence of complex-balanced networks with global
482 asymptotic stability as the motivation include Siegel and Chen [13], Siegel
483 and MacLean [14], Anderson [1], and Anderson and Shiu [2]. The work of
484 Angeli, De Leenheer and Sontag [3] is on persistence as an inherently im-
485 portant property and on approaches to study it through Petri nets. The
486 earlier work of Feinberg [5, Sections 5 and 6] discusses persistence and many
487 of the related ideas in a direct fashion. In particular, Remark 6.1.E in this

488 reference consolidates the topic and includes the conjecture that weakly re-
489 versible networks are persistent.

490

491 It is easy to see that vacuous persistence is equivalent to persistence together
492 with the non-existence of trajectories entirely contained in the boundary of
493 nondegenerate stoichiometric compatibility classes. Condition (iii) in Pro-
494 position 4.2 provides an even simpler characterization.

495 **Proposition 4.2 (Sontag [16]).** *Suppose that stoichiometric compatibility*
496 *classes are bounded. Then the following are equivalent:*

- 497 (i) *The reaction network is vacuously persistent.*
498 (ii) *The reaction network is persistent and there are no trajectories on the*
499 *boundary of nondegenerate stoichiometric compatibility classes.*
500 (iii) *The reaction network is persistent and there are no equilibrium points*
501 *on the boundary of nondegenerate stoichiometric compatibility classes.*

502 *Proof.* We already noted the equivalence of conditions (i) and (ii). We triv-
503 ially have (ii) \Rightarrow (iii). We obtain (iii) \Rightarrow (ii) by contraposition with Proposition
504 3.8. \square

505 We now proceed through the steps leading to the main theorem of this
506 section. Let P be a stoichiometric compatibility class.

507 **Lemma 4.3.** *The set $P_{\neq 0} \cap \Phi(P)$ of boundary points with reach-closed sup-*
508 *port is forward-invariant and $\omega(P_{\neq 0} \cap \Phi(P)) \subseteq P_{\neq 0}$.*

509 *Proof.* Let $u \in P_{\neq 0} \cap \Phi(P)$ and $t \geq 0$. The support of u is reach-closed, so
510 $\text{Supp}(C(u, t)) = \text{Supp}(u)$ by Vol’pert’s Theorem 3.6. Therefore, we have
511 $C(u, t) \in P_{\neq 0} \cap \Phi(P)$. Hence, $P_{\neq 0} \cap \Phi(P)$ is forward-invariant. It then fol-
512 lows that $\omega(P_{\neq 0} \cap \Phi(P)) \subseteq P_{\neq 0}$ because the boundary $P_{\neq 0}$ is a closed set. \square

513 **Definition 4.4.** The set \mathcal{Z} is P -admissible if there exists $u \in P$ such that
514 $\text{Supp}(u) = \mathcal{Z}$, i.e. if \mathcal{Z} is the support of a face of P . The set \mathcal{Z} is *stoichio-*
515 *metrically admissible* if it is P -admissible for some nondegenerate stoichio-
516 metric compatibility class P . \square

517 By Vol’pert’s Theorem 3.6, if a set is P -admissible, then so is its reach-
518 closure. Shiu and Sturmfels [12] have introduced the notion of relevant
519 siphons. Provided a slight extension of the definition of relevance, the re-
520 lation with admissible sets is that a siphon is P -relevant if and only if its
521 (reach-closed) complement contains a P -admissible set.

522 **Theorem 4.5.** *Suppose that all the trajectories in the stoichiometric com-*
 523 *patibility class P are bounded. Then the following conditions are equivalent:*

- 524 (i) $\omega(P) \subseteq P_{>0}$.
 525 (ii) $\Phi(P) = P_{>0}$.
 526 (iii) *Only the entire set \mathcal{S} of species is both reach-closed and P -admissible.*
 527 (iv) *We have $\text{Reach}(\mathcal{Z}) = \mathcal{S}$ for every P -admissible set $\mathcal{Z} \subseteq \mathcal{S}$.*

528 The second of the two assertions in Anderson [1, Lemma 2.8] is the impli-
 529 cation $(\Omega(P) \subseteq P_{>0}) \Rightarrow$ (iii) for weakly reversible deficiency-zero networks.

530 *Proof.* Because of the definition of $\Phi(P)$ in Equation (3.1) and the definition
 531 of admissibility in Definition 4.4, condition (iii) is just another formulation
 532 of condition (ii).

533 Proof that (i) \Rightarrow (ii). Assume $\omega(P) \subseteq P_{>0}$. Then in particular
 534 $\omega(P_{\neq 0} \cap \Phi(P)) \subseteq P_{>0}$. But by Lemma 4.3, $\omega(P_{\neq 0} \cap \Phi(P)) \subseteq P_{\neq 0}$. There-
 535 fore, $\omega(P_{\neq 0} \cap \Phi(P)) = \emptyset$. Trajectories in P have ω -limit points because
 536 they are bounded. So necessarily, $P_{\neq 0} \cap \Phi(P) = \emptyset$, i.e. $\Phi(P) \subseteq P_{>0}$. We
 537 noted in Equation (3.2) that $P_{>0} \subseteq \Phi(P)$. So $\Phi(P) = P_{>0}$.

538 Proof that (iii) \Rightarrow (i). Assume that property (iii) holds. Let $u \in \omega(P)$. Then
 539 $\text{Supp}(u)$ is P -admissible, and by Proposition 3.9, is also reach-closed. There-
 540 fore, $\text{Supp}(u) = \mathcal{S}$, i.e. $u \in P_{>0}$.

541 Proof that (iii) \Rightarrow (iv). Assume (iii). Suppose that $\mathcal{Z} \subseteq \mathcal{S}$ is P -admissible.
 542 Then, as noted immediately after Definition 4.4, $\text{Reach}(\mathcal{Z})$ is P -admissible.
 543 Furthermore, $\text{Reach}(\mathcal{Z})$ is reach-closed by Proposition 3.5. Therefore,
 544 $\text{Reach}(\mathcal{Z}) = \mathcal{S}$.

545 Proof that (iv) \Rightarrow (iii). Assume (iv). Suppose that \mathcal{Z} is both reach-closed
 546 and P -admissible. We have $\text{Reach}(\mathcal{Z}) = \mathcal{S}$ because \mathcal{Z} is P -admissible, and
 547 $\text{Reach}(\mathcal{Z}) = \mathcal{Z}$ because \mathcal{Z} is reach-closed. So $\mathcal{Z} = \mathcal{S}$. \square

548 We now obtain the final and main result of this section as an immediate
 549 corollary of Theorem 4.5.

550 **Theorem 4.6.** *Suppose that all trajectories are bounded. Then the follow-*
 551 *ing are equivalent:*

- 552 • *The reaction network is vacuously persistent.*
 553 • *Only the entire set \mathcal{S} of all species is both reach-closed and stoichio-*
 554 *metrically admissible.* \square

555 **5 Species Composition and Constructive Networks**

556 We propose and develop a formal notion of species composition to account
 557 for the idea that species are composed of elementary units or building blocks.
 558 A reaction network $\mathcal{N} = (\mathcal{S}, \mathcal{C}, \mathcal{R})$ is fixed throughout this section.

559 **Definition 5.1.** A *species composition map*, or simply a *composition* of \mathcal{N}
 560 is a map $\mathcal{E} : \mathcal{S} \rightarrow \mathbb{Z}_{\geq 0}^n \setminus \{0_n\}$, where n is a positive integer. \square

561 A composition map $\mathcal{E} = (\mathcal{E}_1, \dots, \mathcal{E}_n) : \mathcal{S} \rightarrow \mathbb{Z}_{\geq 0}^n \setminus \{0_n\}$ for the network \mathcal{N} is
 562 fixed for the rest of this section. The idea of a composition map will become
 563 more precise as we develop it, but the basic intuition is that the composition
 564 $\mathcal{E}(X)$ of a species X indicates how many instances of the species of reference
 565 are present in X . There could be species of reference that are not in the
 566 network \mathcal{N} .

567 **Definition 5.2.** Let $X \in \mathcal{S}$ be a species of \mathcal{N} .

- 568 • The species X is \mathcal{E} -*elementary* if $|\mathcal{E}(X)| = 1$, i.e. $\mathcal{E}(X) \in \{e_{n,1}, \dots, e_{n,n}\}$.
 - 569 • The species X is \mathcal{E} -*composite* if $|\mathcal{E}(X)| \geq 2$, i.e. $\mathcal{E}(X) \in \mathbb{Z}_{\geq 0}^n \setminus \{0_n, e_{n,1}, \dots, e_{n,n}\}$.
- 570 \square

571 **Definition 5.3.** Two species $X', X'' \in \mathcal{S}$ are \mathcal{E} -*isomeric* if $\mathcal{E}(X') = \mathcal{E}(X'')$.
 572 \square

573 \mathcal{E} -isomerism gives rise to an equivalence relation on the set \mathcal{S} of species.
 574 We denote \mathcal{S}/\mathcal{E} the set of \mathcal{E} -isomerism (equivalence) classes.

575 Let $\tilde{\mathcal{E}} = (\tilde{\mathcal{E}}_1, \dots, \tilde{\mathcal{E}}_n) : \mathbb{R}\mathcal{S} \rightarrow \mathbb{R}^n$ be the unique \mathbb{R} -linear extension of \mathcal{E} . The
 576 map $\tilde{\mathcal{E}}$ gives rise to a sensible notion of composition of complexes, which
 577 leads to a concept of conservation of composition.
 578

579 **Definition 5.4.**

- 580 • A reaction $Q' \rightarrow Q''$ is \mathcal{E} -*conservative* if $\tilde{\mathcal{E}}(Q') = \tilde{\mathcal{E}}(Q'')$.
 - 581 • The network \mathcal{N} is \mathcal{E} -*conservative* if all reactions are \mathcal{E} -*conservative*.
- 582 \square

583 We record a few obvious results.

584 **Proposition 5.5.** (*The network \mathcal{N} is \mathcal{E} -conservative*) $\Leftrightarrow (\text{Ker } \tilde{\mathcal{E}} \supseteq S)$. \square

585 **Proposition 5.6.** *If the network \mathcal{N} is \mathcal{E} -conservative, then stoichiometric*
 586 *isomerism implies \mathcal{E} -isomerism; i.e. if two species are stoichiometrically*
 587 *isomeric, then they are \mathcal{E} -isomeric. \square*

588 **Lemma 5.7.** *For a nonzero nonsingleton complex $Q \in \mathcal{C}$, and for a species*
 589 *$X \in \text{Supp}(Q)$, we have $\mathcal{E}(X) \not\subseteq \mathcal{E}(Q)$. \square*

590 **Proposition 5.8.** *Suppose the network \mathcal{N} is \mathcal{E} -conservative. Then every*
 591 *bound species is \mathcal{E} -composite. \square*

592 The canonical example of composition is of course the atomic composition
 593 of molecules. In this case, n could be the number of entries in the Periodic
 594 Table of the Elements and positions in composition n -tuples could represent
 595 atomic numbers. However, this example is cumbersome and impractical,
 596 especially if the species under consideration are macromolecules. For the
 597 network of Figure 2.1(a) for instance, we have an intuition that species
 598 RAB is composed of species R , A and B , which are elementary within the
 599 network, even though they may not be atoms. We formalize this observation
 600 with the notion of core composition.

601 **Definition 5.9.** The composition \mathcal{E} is a *core composition* for \mathcal{N} provided

602 • $e_{n,1}, \dots, e_{n,n} \in \mathcal{E}(\mathcal{S})$ and

603 • $\text{Ker} \tilde{\mathcal{E}} = S$. \square

604 The first condition in Definition 5.9 says that all n elementary compositions
 605 do occur in the network \mathcal{N} . The second condition says that the network
 606 is \mathcal{E} -conservative in a minimal fashion. The following two theorems justify
 607 the terminology of core composition.

608 **Theorem 5.10.** *Any core composition of \mathcal{N} is universal among all com-*
 609 *positions with respect to which \mathcal{N} is conservative. For elaboration, suppose*
 610 *that $\mathcal{E} : \mathcal{S} \rightarrow \mathbb{Z}_{\geq 0}^n \setminus \{0_n\}$ is a core composition of \mathcal{N} , and let $\mathcal{F} : \mathcal{S} \rightarrow \mathbb{Z}_{\geq 0}^N \setminus \{0_N\}$*
 611 *be a composition of \mathcal{N} such that \mathcal{N} is \mathcal{F} -conservative. Then there exists*
 612 *a unique \mathbb{Z} -linear map $\Phi : \mathbb{Z}^n \rightarrow \mathbb{Z}^N$ that makes the following diagram com-*
 613 *mute, and Φ maps $\mathbb{Z}_{\geq 0}^n$ into $\mathbb{Z}_{\geq 0}^N$.*

614

$$\begin{array}{ccc}
 \mathcal{S} & \xrightarrow{\mathcal{F}} & \mathbb{Z}^N \\
 \mathcal{E} \downarrow & \nearrow \Phi & \\
 \mathbb{Z}^n & &
 \end{array}$$

615 *Proof.* Let $\tilde{\mathcal{F}} : \mathbb{R}\mathcal{S} \rightarrow \mathbb{R}^N$ be the linear extension of \mathcal{F} . The map
 616 $\tilde{\mathcal{E}} : \mathbb{R}\mathcal{S} \rightarrow \mathbb{R}^n$ is surjective and $\text{Ker}\tilde{\mathcal{F}} \supseteq S = \text{Ker}\tilde{\mathcal{E}}$. Therefore, there exists
 617 a unique \mathbb{R} -linear map $\tilde{\Phi} : \mathbb{R}^n \rightarrow \mathbb{R}^N$ such that $\tilde{\mathcal{F}} = \tilde{\Phi} \circ \tilde{\mathcal{E}}$. Then, by restric-
 618 tion to \mathcal{S} , we have $\mathcal{F} = \tilde{\Phi} \circ \mathcal{E}$. As a result, $\tilde{\Phi}$ maps $e_{n,1}, \dots, e_{n,n}$ into $\mathbb{Z}_{\geq 0}^N$.
 619 Therefore, $\tilde{\Phi}$ is the extension of a \mathbb{Z} -linear map $\Phi : \mathbb{Z}^n \rightarrow \mathbb{Z}^N$ which maps
 620 $e_{n,1}, \dots, e_{n,n}$ into $\mathbb{Z}_{\geq 0}^N$. The map Φ satisfies $\mathcal{F} = \Phi \circ \mathcal{E}$. The uniqueness of
 621 $\tilde{\Phi}$ implies the uniqueness of Φ . \square

622 **Theorem 5.11.** *If a reaction network has a core composition, then all core*
 623 *compositions are equivalent up to indexing permutations in composition tu-*
 624 *ples. For elaboration, suppose $\mathcal{E} : \mathcal{S} \rightarrow \mathbb{Z}_{\geq 0}^n \setminus \{0_n\}$ and $\mathcal{E}' : \mathcal{S} \rightarrow \mathbb{Z}_{\geq 0}^{n'} \setminus \{0_{n'}\}$*
 625 *are core compositions of the network \mathcal{N} . Then $n = n'$ and the \mathbb{Z} -linear*
 626 *map Φ such that $\mathcal{E}' = \Phi \circ \mathcal{E}$ is an automorphism of \mathbb{Z}^n that permutes the*
 627 *canonical basis vectors $e_{n,1}, \dots, e_{n,n}$.*

628 *Proof.* Let $\Phi : \mathbb{Z}^n \rightarrow \mathbb{Z}^{n'}$ (resp. $\Phi' : \mathbb{Z}^{n'} \rightarrow \mathbb{Z}^n$) be the \mathbb{Z} -linear map such that
 629 $\mathcal{E}' = \Phi \circ \mathcal{E}$ (resp. $\mathcal{E} = \Phi' \circ \mathcal{E}'$). Then we have $\mathcal{E} = \varphi \circ \mathcal{E}$ (resp. $\mathcal{E}' = \varphi' \circ \mathcal{E}'$)
 630 both if $\varphi = \Phi' \circ \Phi$ and $\varphi = \text{Id}_{\mathbb{Z}^n}$ (resp. $\varphi' = \Phi \circ \Phi'$ and $\varphi' = \text{Id}_{\mathbb{Z}^{n'}}$). There-
 631 fore $\Phi' \circ \Phi = \text{Id}_{\mathbb{Z}^n}$ and $\Phi \circ \Phi' = \text{Id}_{\mathbb{Z}^{n'}}$. It follows that $n = n'$ and Φ is a \mathbb{Z} -
 632 linear automorphism of \mathbb{Z}^n with inverse $\Phi^{-1} = \Phi'$. Because Φ maps $\mathbb{Z}_{\geq 0}^n$ into
 633 itself, we have $|\Phi(x)| \geq |x|$ for all $x \in \mathbb{Z}_{\geq 0}^n$. Likewise, we have $|\Phi^{-1}(x)| \geq |x|$
 634 for all $x \in \mathbb{Z}_{\geq 0}^n$, whence $|x| \geq |\Phi(x)|$ for all $x \in \mathbb{Z}_{\geq 0}^n$. So $|\Phi(x)| = |x|$ for all
 635 $x \in \mathbb{Z}_{\geq 0}^n$, and it follows that $\Phi(\{e_{n,1}, \dots, e_{n,n}\}) = \{e_{n,1}, \dots, e_{n,n}\}$. \square

636 As a result of Theorem 5.11, if \mathcal{E} is a core composition of \mathcal{N} , then the no-
 637 tions of \mathcal{E} -elementary, \mathcal{E} -composite, and \mathcal{E} -isomeric species are independent
 638 of the choice of \mathcal{E} among core compositions, and we simply use the termi-
 639 nology of *elementary*, *composite*, and *isomeric* species. Also, stoichiometric
 640 isomerism and \mathcal{E} -isomerism coincide and we simply call isomerism classes
 641 the equivalence classes with respect to the two relations.

642

643 Results in this section will show that in the presence of a core composi-
 644 tion, one has explicit, fairly canonical information about the stoichiometric
 645 space and its orthogonal. Most of these properties do not require that the
 646 conservativeness of the network be minimal, whence the following definition.

647 **Definition 5.12.** The composition \mathcal{E} is a *near-core composition* of \mathcal{N} if

- 648 • $e_{n,1}, \dots, e_{n,n} \in \mathcal{E}(\mathcal{S})$, i.e. all \mathcal{E} -elementary compositions occur, and
- 649 • $\text{Ker}\tilde{\mathcal{E}} \supseteq S$, i.e. \mathcal{N} is \mathcal{E} -conservative. \square

650 The composition \mathcal{E} is assumed to be a near-core composition of the network
651 \mathcal{N} for the rest of this section. We set the following notations.

652 **Notation 5.13.**

- 653 • For $i \in [1..n]$, $\mathcal{X}_i := \mathcal{E}^{-1}(e_{n,i})$ is the \mathcal{E} -isomerism class of \mathcal{E} -elementary
654 species with \mathcal{E} -composition $e_{n,i}$, and $p_i := |\mathcal{X}_i|$ is the number of these
655 species.
- 656 • $I := \mathcal{E}(\mathcal{S}) \setminus \{e_{n,1}, \dots, e_{n,n}\}$ is the set of n -tuples (of length ≥ 2) that
657 occur as \mathcal{E} -composition of \mathcal{E} -composite species.
- 658 • For $\alpha \in I$, $\mathcal{Y}_\alpha := \mathcal{E}^{-1}(\alpha)$ is the \mathcal{E} -isomerism class of \mathcal{E} -composite
659 species with \mathcal{E} -composition α , and $q_\alpha := |\mathcal{Y}_\alpha|$ is the number of these
660 species.
- 661 • \mathcal{S}_e is the set of \mathcal{E} -elementary species.
662 \mathcal{S}_c is the set of \mathcal{E} -composite species.
663 For $i \in [1..n]$, $\mathcal{S}_{c,i} := \{Y \in \mathcal{S}_c : \mathcal{E}_i(Y) \geq 1\}$ is the set of composite
664 species whose \mathcal{E} -composition has a nonzero term of index i .
665 \mathcal{S}_e^1 is the set of \mathcal{E} -elementary species whose \mathcal{E} -isomerism classes are
666 singletons.
- 667 • $p := |\mathcal{S}_e| = \sum_{i=1}^n p_i$ is the total number of \mathcal{E} -elementary species.
668 $q := |\mathcal{S}_c| = \sum_{\alpha \in I} q_\alpha$ is the total number of \mathcal{E} -composite species.
669 $r := |\mathcal{S}| = p + q$ is the total number of species.
670 n already denotes the number of \mathcal{E} -isomerism classes of elementary
671 species.
672 $m := |I|$ is the number of \mathcal{E} -isomerism classes of composite species. \square

673 By the Rank-Nullity Theorem, we have:

Lemma 5.14.

674
$$\text{nullity} \left(\tilde{\mathcal{E}} \right) = \dim \text{Ker} \tilde{\mathcal{E}} = r - n = p + q - n = q + \sum_{i=1}^n (p_i - 1).$$

675 *In particular, if (and only if) there is no \mathcal{E} -isomerism among the \mathcal{E} -elementary*
676 *species (i.e. $\mathcal{S}_e^1 = \mathcal{S}_e$), then the nullity of $\tilde{\mathcal{E}}$ equals the number of \mathcal{E} -*
677 *composite species. \square*

678 If \mathcal{E} is a core composition, i.e. if $\text{Ker}\tilde{\mathcal{E}} = S$, then Lemma 5.14 provides the
 679 rank of the network. This can be helpful in calculating the deficiency, an
 680 integer attribute of a reaction network of importance in Chemical Reaction
 681 Theory. See for instance Feinberg [5] for the definition and some implications
 682 of the deficiency. The rank of a network is the one ingredient that is usually
 683 the least readily available. The other ingredients, the number of complexes
 684 and the number of linkages classes, are usually obtained by observation. Of
 685 course, for this remark to be pertinent, we must have convenient ways to
 686 find core compositions. Section 6 will address this matter.

687

688 We define the linear map $\rho : \mathbb{R}\mathcal{S} \rightarrow \mathbb{R}\mathcal{S}$ by

$$689 \quad \rho(Z) = Z - \sum_{i=1}^n \frac{\mathcal{E}_i(Z)}{p_i} \text{sum}(\mathcal{X}_i) \quad \text{for } Z \in \mathcal{S}. \quad (5.1)$$

690 In particular,

$$691 \quad \rho(X) = X - \frac{1}{p_i} \text{sum}(\mathcal{X}_i) \quad \text{if } X \in \mathcal{X}_i, \text{ and} \quad (5.2)$$

$$692 \quad \rho(Y) = Y - \sum_{i=1}^n \frac{\alpha_i}{p_i} \text{sum}(\mathcal{X}_i) \quad \text{if } Y \in \mathcal{Y}_\alpha. \quad (5.3)$$

693

694 **Theorem 5.15.** For each $i \in [1..n]$, let \mathcal{B}_i be one of the p_i sets of $(p_i - 1)$
 695 elements obtained by excising one element from the set $\{\rho(X) : X \in \mathcal{X}_i\}$.
 696 Then let

$$697 \quad \mathcal{B} = \{\rho(Y) : Y \in \mathcal{S}_c\} \sqcup \bigsqcup_{i=1}^n \mathcal{B}_i = \{\rho(Y) : Y \in \mathcal{S}_c\} \sqcup \bigsqcup_{\substack{1 \leq i \leq n \\ p_i > 1}} \mathcal{B}_i.$$

698 The set \mathcal{B} is a basis of $\text{Ker}\tilde{\mathcal{E}}$.

699 *Proof.*

700 We see from Equations (5.2) and (5.3) that the vectors $\rho(Z)$ for $Z \in \mathcal{S} \setminus \mathcal{S}_e^1$
 701 are pairwise distinct. So the disjoint unions in the expression of \mathcal{B} are justi-
 702 fied and the cardinality of \mathcal{B} is equal to the dimension of $\text{Ker}\tilde{\mathcal{E}}$ as provided
 703 by Lemma 5.14.

704 From Equation (5.1), we get that $\tilde{\mathcal{E}}(\rho(Z)) = 0_n$ for all $Z \in \mathcal{S}$. Hence,
 705 $\mathcal{B} \subset \text{Ker}\tilde{\mathcal{E}}$.

706 One can verify that the set $\{\rho(Y) : Y \in \mathcal{S}_c\}$ is linearly independent, and that

707 so is the set \mathcal{B}_i for each $i \in [1..n]$ with $p_i > 1$. Furthermore, the sets \mathcal{B}_i are
 708 pairwise orthogonal because $\mathcal{B}_i \subset \mathbb{R}\mathcal{X}_i$. So the set $\bigsqcup_{i=1}^n \mathcal{B}_i$ is linearly inde-
 709 pendent. Now, intersecting the subspaces spanned by $\{\rho(Y) : Y \in \mathcal{S}_c\}$ and
 710 by $\bigsqcup_{i=1}^n \mathcal{B}_i$ yields the zero space. So the set \mathcal{B} is linearly independent. \square

711 Let the linear map $\tau : \mathbb{R}^n \rightarrow \mathbb{R}\mathcal{S}$ be defined by $\tau(e_{n,i}) = T_i$ for $i \in [1..n]$,
 712 where

$$713 \quad T_i := \text{sum}(\mathcal{X}_i) + \sum_{\alpha \in I} \alpha_i \text{sum}(\mathcal{Y}_\alpha) = \text{sum}(\mathcal{X}_i) + \sum_{Y \in \mathcal{S}_c} \mathcal{E}_i(Y) Y. \quad (5.4)$$

714 **Theorem 5.16.** *We have $(\text{Ker}\tilde{\mathcal{E}})^\perp = \text{Im}\tau$ and the set $\mathcal{B}' = \{T_1, \dots, T_n\}$
 715 is a basis of $(\text{Ker}\tilde{\mathcal{E}})^\perp$. In particular, \mathcal{B}' is a linearly independent subset of
 716 the orthogonal S^\perp of the stoichiometric space S .*

717 *Proof.* We obtain the adjunction property

$$718 \quad \langle \tau(x), Q \rangle = \langle x, \tilde{\mathcal{E}}(Q) \rangle, \quad \forall x \in \mathbb{R}^n, \forall Q \in \mathbb{R}\mathcal{S} \quad (5.5)$$

719 by verifying that each side of the equality equals $\mathcal{E}_i(Q)$ when $x \in \{e_{n,1}, \dots, e_{n,n}\}$
 720 and $Q \in \mathcal{S}_e$ or $Q \in \mathcal{S}_c$. As a result, we have $(\text{Im}\tau)^\perp = \text{Ker}\tilde{\mathcal{E}}$. Hence,
 721 $(\text{Ker}\tilde{\mathcal{E}})^\perp = \text{Im}\tau$, and the set $\{T_1, \dots, T_n\}$ spans $(\text{Ker}\tilde{\mathcal{E}})^\perp$. From Lemma
 722 5.14, $(\text{Ker}\tilde{\mathcal{E}})^\perp$ has dimension n , so the set $\{T_1, \dots, T_n\}$ is a basis of $(\text{Ker}\tilde{\mathcal{E}})^\perp$. \square

723 We combine in Theorem 5.17 what Theorems 5.15 and 5.16 say when \mathcal{E} is
 724 a core composition.

725 **Theorem 5.17.** *Suppose that \mathcal{E} is a core composition of \mathcal{N} . Then:*

- 726 • *The set \mathcal{B} from Theorem 5.15 is a basis of S .*
- 727 • *The set \mathcal{B}' from Theorem 5.16 is a basis of S^\perp that lies in $\mathbb{R}_{\geq 0}\mathcal{S}$. \square*

728 The basis \mathcal{B}' of S^\perp provides a canonical, comprehensive and non-redundant
 729 description of the conservativeness of the network. This result and its proof
 730 are rigorous statement and justification for the common practice of cata-
 731 logging conservation laws by visual inspection of reaction networks. Section
 732 6 will provide means of finding core compositions, thereby turning Theorem
 733 5.17 into a useful tool for applications.

734 **Definition 5.18.** A reaction network is *constructive* if it admits a core
735 composition. \square

736 This terminology is from Shinar, Alon and Feinberg [11, Definition 8.1].
737 Theorem 5.17 shows that the two usages are consistent. The notions in this
738 prior work that correspond to our elementary and composite species are
739 respectively the elements and the compounds. We note however that if an
740 element is one of many isomers, then the isomers other than the element are
741 compounds.

742 **Definition 5.19.** Consider a reaction network.

- 743 • A species Y is *explicitly constructible* (resp. *explicitly destructible*) if
744 there are isomerization reactions $Y_0 \rightarrow \cdots \rightarrow Y_\ell$ (resp. $Y_\ell \rightarrow \cdots \rightarrow Y_0$),
745 where $\ell \in \mathbb{Z}_{\geq 0}$, such that Y_0 is the target of a binding reaction (resp.
746 the source of a dissociation reaction) and $Y_\ell = Y$.
- 747 • A species X is *explicitly constructive* (resp. *explicitly destructive*)
748 if there is a binding reaction $Q \rightarrow Y$ (resp. a dissociation reaction
749 $Y \rightarrow Q$) such that $X \in \text{Supp}(Q)$.
- 750 • The reaction network is *explicitly constructive* provided
 - 751 – The network is constructive;
 - 752 – Each composite species is explicitly constructible or explicitly
753 destructible or both; and
 - 754 – Each elementary species is explicitly constructive or explicitly
755 destructive or both.
- 756 • The reaction network is *explicitly-reversibly constructive* provided
 - 757 – The network is constructive;
 - 758 – Each composite species is both explicitly constructible and ex-
759 plicitly destructible; and
 - 760 – Each elementary species is both explicitly constructive and ex-
761 plicitly destructive. \square

762 The pre-complete networks of reversible binding reactions we introduced in
763 Gnacadja [6] are explicitly-reversibly constructive. More generally, if a reac-
764 tion network consisting of binding and dissociation reactions is constructive
765 and weakly reversible, then it is explicitly-reversibly constructive. The futile

766 enzymatic network of Figure 2.1(b) is an example of an explicitly-reversibly
 767 constructive network that is not weakly reversible. More generally, binary
 768 enzymatic networks in which each enzyme both is and has a reversing en-
 769 zyme (see Section 8) are explicitly-reversibly constructive networks that are
 770 usually not weakly reversible.

771 6 Finding Core Compositions

772 Let $\mathcal{N} = (\mathcal{S}, \mathcal{C}, \mathcal{R})$ be a reaction network and let $\mathcal{E} : \mathcal{S} \rightarrow \mathbb{Z}_{\geq 0}^n \setminus \{0_n\}$ be
 773 a composition of \mathcal{N} . We devote this section to ways to prove that \mathcal{E} is a
 774 core composition of \mathcal{N} when it is known that \mathcal{E} is a near-core composition.
 775 We adopted this approach because the common practice of cataloging con-
 776 servation laws by visual inspection produces near-core compositions. The
 777 difficulty lies in the minimality aspect of a core composition, i.e. the fact
 778 that $\text{Ker} \tilde{\mathcal{E}} \subseteq S$. Because of Theorem 5.15, this containment relationship is
 779 equivalent to the property that $\rho(Z) \in S$ for all $Z \in \mathcal{S}$. But proving this
 780 from the expressions of Equations (5.1), (5.2) and (5.3) may not be obvious.
 781 The following result provides other expressions for these vectors.

782 **Lemma 6.1.** *Suppose that \mathcal{E} is a near-core composition of \mathcal{N} and refer to*
 783 *Notation 5.13.*

784 • For $i \in [1..n]$ and $X \in \mathcal{X}_i$, we have

$$785 \quad \rho(X) = \frac{1}{p_i} \sum_{W \in \mathcal{X}_i} (X - W). \quad (6.1)$$

786 • For $\alpha \in I$ and $Y \in \mathcal{Y}_\alpha$, we have

$$787 \quad \rho(Y) = \frac{1}{p_1 \cdots p_n} \sum_{(W_1, \dots, W_n) \in \mathcal{X}_1 \times \cdots \times \mathcal{X}_n} \left(Y - \sum_{i=1}^n \alpha_i W_i \right). \quad (6.2)$$

788 *Proof.* Equation (6.1) is an obvious reformulation of Equation (5.2). We

789 prove Equation (6.2). We have:

$$\begin{aligned}
790 & \sum_{(W_1, \dots, W_n) \in \mathcal{X}_1 \times \dots \times \mathcal{X}_n} \left(\sum_{i=1}^n \alpha_i W_i \right) \\
791 &= \sum_{i=1}^n \left(\sum_{(W_1, \dots, W_n) \in \mathcal{X}_1 \times \dots \times \mathcal{X}_n} \alpha_i W_i \right) \\
792 &= \sum_{i=1}^n \alpha_i \left(\sum_{(W_1, \dots, W_n) \in \mathcal{X}_1 \times \dots \times \mathcal{X}_n} W_i \right) \\
793 &= \sum_{i=1}^n \alpha_i \left(\sum_{(W_1, \dots, W_{i-1}, W_{i+1}, \dots, W_n) \in \mathcal{X}_1 \times \dots \times \mathcal{X}_{i-1} \times \mathcal{X}_{i+1} \times \dots \times \mathcal{X}_n} \left(\sum_{W_i \in \mathcal{X}_i} W_i \right) \right) \\
794 &= \sum_{i=1}^n \alpha_i \left(\sum_{(W_1, \dots, W_{i-1}, W_{i+1}, \dots, W_n) \in \mathcal{X}_1 \times \dots \times \mathcal{X}_{i-1} \times \mathcal{X}_{i+1} \times \dots \times \mathcal{X}_n} \text{sum}(\mathcal{X}_i) \right) \\
795 &= \sum_{i=1}^n \alpha_i p_1 \cdots p_{i-1} p_{i+1} \cdots p_n \text{sum}(\mathcal{X}_i) \\
796 &= p_1 \cdots p_n \sum_{i=1}^n \frac{\alpha_i}{p_i} \text{sum}(\mathcal{X}_i) \\
797 &= p_1 \cdots p_n (Y - \rho(Y)) . \\
798 &
\end{aligned}$$

799 It follows that:

$$\begin{aligned}
800 & p_1 \cdots p_n \rho(Y) = p_1 \cdots p_n Y - \sum_{(W_1, \dots, W_n) \in \mathcal{X}_1 \times \dots \times \mathcal{X}_n} \left(\sum_{i=1}^n \alpha_i W_i \right) \\
801 & = \sum_{(W_1, \dots, W_n) \in \mathcal{X}_1 \times \dots \times \mathcal{X}_n} \left(Y - \sum_{i=1}^n \alpha_i W_i \right) . \quad \square
\end{aligned}$$

802 **Theorem 6.2.** Refer to Notation 5.13 and suppose that:

- 803 (1) \mathcal{E} is a near-core composition of \mathcal{N} ;
- 804 (2) For every $i \in [1..n]$, all (\mathcal{E} -elementary) species of composition $e_{n,i}$ are
805 stoichiometrically isomeric, i.e. $(X, W \in \mathcal{X}_i) \Rightarrow (X - W \in S)$; and
- 806 (3) For every $\alpha \in I$ and $Y \in \mathcal{Y}_\alpha$, there exist $W_1 \in \mathcal{X}_1, \dots, W_n \in \mathcal{X}_n$ such
807 that $Y - \sum_{i=1}^n \alpha_i W_i \in S$.

808 Then \mathcal{E} is a core composition of \mathcal{N} .

809 *Proof.* We just need to show that $\rho(Z) \in S$ for all $Z \in \mathcal{S}$.

810 Condition (2) of the theorem along with Equation (6.1) of Lemma 6.1 imply
811 that $\rho(X) \in S$ if $X \in \mathcal{X}_i$ for some $i \in [1..n]$.

812 Let $\alpha \in I$ and $Y \in \mathcal{Y}_\alpha$. With Condition (3), we have $W_1^0 \in \mathcal{X}_1, \dots, W_n^0 \in \mathcal{X}_n$
813 such that $Y - \sum_{i=1}^n \alpha_i W_i^0 \in S$. Let $W_1 \in \mathcal{X}_1, \dots, W_n \in \mathcal{X}_n$. For every $i \in [1..n]$,
814 both W_i^0 and W_i are in \mathcal{X}_i , and so $W_i^0 - W_i \in S$ by Condition (2). Hence,

$$815 \quad Y - \sum_{i=1}^n \alpha_i W_i = Y - \sum_{i=1}^n \alpha_i W_i^0 + \sum_{i=1}^n \alpha_i (W_i^0 - W_i) \in S.$$

816 Then, Equation (6.2) of Lemma 6.1 implies that $\rho(Y) \in S$. \square

817 Theorem 6.2 is directly applicable if each composite species is explicitly con-
818 structible or explicitly destructible from a non-singleton complex in which
819 all species are elementary. This is the case for instance for the network of
820 Figure 2.1(b). When this is not the case, as in the network of Figure 2.1(a)
821 for instance, one can ascertain Condition (3) of Theorem 6.2 by induction,
822 or one can use the following theorem.

823 **Theorem 6.3.** *Refer to Notation 5.13 and suppose that:*

- 824 (1) \mathcal{E} is a near-core composition of \mathcal{N} ;
825 (2) \mathcal{E} -isomerism implies stoichiometric isomerism; and
826 (3) Every \mathcal{E} -composite species is stoichiometrically isomeric to a bound
827 species.

828 Then \mathcal{E} is a core composition of \mathcal{N} .

829 The proof of Theorem 6.3 will use the following lemma and will come after
830 the proof of the latter. For $\alpha \in I$, let $I_\alpha = \{\beta \in I : \beta \leq \alpha \text{ and } \beta \neq \alpha\}$.

831 **Lemma 6.4.** *Assume the hypotheses of Theorem 6.3 and refer to Notation*
832 *5.13. Let $Y \in \mathcal{S}_c$ and $\alpha = \mathcal{E}(Y)$. There exist $\sigma_i \in \mathbb{Z}_{\geq 0}$ for $i \in [1..n]$ and*
833 *$\sigma_\beta \in \mathbb{Z}_{\geq 0}$ for $\beta \in I_\alpha$ such that, for all $i \in [1..n]$ and $X_i \in \mathcal{X}_i$, and all $\beta \in I_\alpha$*
834 *and $Y_\beta \in \mathcal{Y}_\beta$, we have*

$$835 \quad Y - \left(\sum_{i=1}^n \sigma_i X_i + \sum_{\beta \in I_\alpha} \sigma_\beta Y_\beta \right) \in S.$$

836 We have the conservation relation $\alpha = (\sigma_1, \dots, \sigma_n) + \sum_{\beta \in I_\alpha} \sigma_\beta \beta$.

837 *Proof.* Condition (3) of Theorem 6.3 implies the existence of a complex
838 $Q = \sum_{Z \in \mathcal{S}} Q_Z Z \in \mathcal{C}$ of length $|Q| \geq 2$ such that $Y - Q \in S$. For each $i \in [1..n]$,

839 let $\sigma_i = \sum_{Z \in \mathcal{X}_i} Q_Z$ and $X_i \in \mathcal{X}_i$. For each $\beta \in I$, let $\sigma_\beta = \sum_{Z \in \mathcal{Y}_\beta} Q_Z$ and $Y_\beta \in \mathcal{Y}_\beta$.

840 One can verify that

$$841 \quad -Q = - \left(\sum_{i=1}^n \sigma_i X_i + \sum_{\beta \in I} \sigma_\beta Y_\beta \right)$$

$$842 \quad + \sum_{i=1}^n \sum_{Z \in \mathcal{X}_i} Q_Z (X_i - Z) + \sum_{\beta \in I} \sum_{Z \in \mathcal{Y}_\beta} Q_Z (Y_\beta - Z).$$

843 We know that $Y - Q \in S$, and with Condition (2) of Theorem 6.3, we get
844 that $X_i - Z \in S$ for $i \in [1..n]$ and $Z \in \mathcal{X}_i$, and $Y_\beta - Z \in S$ for $\beta \in I$ and
845 $Z \in \mathcal{Y}_\beta$. This leads to

$$846 \quad Y - \left(\sum_{i=1}^n \sigma_i X_i + \sum_{\beta \in I} \sigma_\beta Y_\beta \right) \in S.$$

847 And because $S \subseteq \text{Ker} \tilde{\mathcal{E}}$, it follows that $\alpha = (\sigma_1, \dots, \sigma_n) + \sum_{\beta \in I} \sigma_\beta \beta$.

848 We show that $\sigma_\beta = 0$ for $\beta \in I \setminus I_\alpha$. Let $\beta \in I$. We have $\sigma_\beta \beta \leq \alpha$, so if $\sigma_\beta \geq 1$,
849 then $\beta \leq \alpha$. By contraposition, if $\beta \notin I_\alpha$ and $\beta \neq \alpha$, then $\sigma_\beta = 0$. It remains
850 to show that $\sigma_\alpha = 0$. We in particular have $\sigma_\alpha \alpha \leq \alpha$, so $\sigma_\alpha \leq 1$. Suppose
851 $\sigma_\alpha = 1$. Then we successively have $0_n = (\sigma_1, \dots, \sigma_n) + \sum_{\beta \in I \setminus \{\alpha\}} \sigma_\beta \beta$; $\sigma_i = 0$
852 for $i \in [1..n]$ and $\sigma_\beta = 0$ for $\beta \in I \setminus \{\alpha\}$; and

853 $|Q| = \sum_{Z \in \mathcal{S}} Q_Z = \sum_{i=1}^n \sigma_i + \sum_{\beta \in I} \sigma_\beta = \sigma_\alpha = 1$. But $|Q| \geq 2$. So $\sigma_\alpha = 0$. \square

854 *Proof (Theorem 6.3).* Refer to Notation 5.13. Just as for Theorem 6.2, we
855 only need to prove that $\rho(Z) \in S$ for all $Z \in \mathcal{S}$. Condition (2) of Theorem
856 6.3 along with Equation (6.1) of Lemma 6.1 imply that $\rho(X) \in S$ if $X \in \mathcal{X}_i$
857 for some $i \in [1..n]$. For a species Y with $\mathcal{E}(Y) = \alpha \in I$, we show that

$$858 \quad \forall X_1 \in \mathcal{X}_1, \dots, \forall X_n \in \mathcal{X}_n, Y - \sum_{i=1}^n \alpha_i X_i \in S. \quad (6.3)$$

859 This together with Equation (6.2) of Lemma 6.1 will imply that $\rho(Y) \in S$.
 860 We proceed by induction on the length $|\alpha|$ of α to prove Property (6.3).

861 If $|\alpha| = 2$, then $I_\alpha = \emptyset$ and Property (6.3) holds by Lemma 6.4.

862 Let $\ell \geq 3$ and assume for induction that Property (6.3) holds if $|\alpha| < \ell$.
 863 Suppose $|\alpha| = \ell$ and let $Y \in \mathcal{Y}_\alpha$. Let σ_i for $i \in [1..n]$ and σ_β for $\beta \in I_\alpha$ be as
 864 in Lemma 6.4. Then let $X_i \in \mathcal{X}_i$ for $i \in [1..n]$ and $Y_\beta \in \mathcal{Y}_\beta$ for $\beta \in I_\alpha$. For
 865 each $\beta \in I_\alpha$, we have $|\beta| < \ell$, and so by the induction hypothesis, we have

866 $Y_\beta - \sum_{i=1}^n \beta_i X_i \in S$. Then:

$$\begin{aligned}
 867 \quad S &\ni Y - \sum_{i=1}^n \sigma_i X_i - \sum_{\beta \in I_\alpha} \sigma_\beta Y_\beta \\
 868 &= Y - \sum_{i=1}^n \sigma_i X_i - \sum_{i=1}^n \sum_{\beta \in I_\alpha} \sigma_\beta \beta_i X_i + \sum_{\beta \in I_\alpha} \sum_{i=1}^n \sigma_\beta \beta_i X_i - \sum_{\beta \in I_\alpha} \sigma_\beta Y_\beta \\
 869 &= Y - \sum_{i=1}^n \left(\sigma_i + \sum_{\beta \in I_\alpha} \sigma_\beta \beta_i \right) X_i - \sum_{\beta \in I_\alpha} \sigma_\beta \left(Y_\beta - \sum_{i=1}^n \beta_i X_i \right) \\
 870 &= Y - \sum_{i=1}^n \alpha_i X_i - \sum_{\beta \in I_\alpha} \sigma_\beta \left(Y_\beta - \sum_{i=1}^n \beta_i X_i \right).
 \end{aligned}$$

871 It follows that $Y - \sum_{i=1}^n \alpha_i X_i \in S$. Property (6.3) is thus proved. \square

872 7 Stoichiometry and Persistence in Constructive Networks

873 In this section, $\mathcal{N} = (\mathcal{S}, \mathcal{C}, \mathcal{R})$ is a constructive reaction network,
 874 $\mathcal{E} : \mathcal{S} \rightarrow \mathbb{Z}_{\geq 0}^n \setminus \{0_n\}$ is a core composition of \mathcal{N} , and Notation 5.13 is in ef-
 875 fect. We will describe the stoichiometric compatibility classes and discuss
 876 reachability and persistence.

877

878 For $b \in \mathbb{R}^n$, let

$$879 \quad P(b) := \tilde{\mathcal{E}}^{-1}(b) \cap (\mathbb{R}_{\geq 0} \mathcal{S}). \quad (7.1)$$

880 More explicitly, $P(b)$ consists of the points $c = (c_Z)_{Z \in \mathcal{S}} \in \mathbb{R} \mathcal{S}$ that satisfy:

$$\begin{cases} \forall Z \in \mathcal{S}, c_Z \geq 0 \\ \forall i \in [1..n], \sum_{X \in \mathcal{X}_i} c_X + \sum_{\alpha \in I} \alpha_i \sum_{Y \in \mathcal{Y}_\alpha} c_Y = b_i. \end{cases} \quad (7.2)$$

Proposition 7.1. *The correspondence $b \mapsto P(b)$ in which b ranges over $\mathbb{R}_{\geq 0}^n$ establishes a bijective parameterization of the stoichiometric compatibility classes of the network \mathcal{N} .*

Proof. The stoichiometric compatibility classes are the nonempty traces on $\mathbb{R}_{\geq 0} \mathcal{S}$ of the affine subspaces of $\mathbb{R} \mathcal{S}$ parallel to S . From Definition 5.9, we get that the map $\tilde{\mathcal{E}} : \mathbb{R} \mathcal{S} \rightarrow \mathbb{R}^n$ is surjective with kernel S . As a result, the affine spaces parallel to S are $\tilde{\mathcal{E}}^{-1}(b), b \in \mathbb{R}^n$. One can verify that $P(b) \neq \emptyset$ if and only if $b \in \mathbb{R}_{\geq 0}^n$, and that if $b, b' \in \mathbb{R}_{\geq 0}^n$ and $P(b) = P(b')$, then $b = b'$. \square

Let $b \in \mathbb{R}_{\geq 0}^n$. We consider the interior $P_{>0}(b)$ and the boundary $P_{\neq 0}(b)$ of $P(b)$, and also the subsets $P_e(b)$ and $P_e^1(b)$ of $P(b)$ consisting of the points $c = (c_Z)_{Z \in \mathcal{S}} \in P(b)$ that satisfy the following properties.

$$\begin{aligned}
P_e(b) &: \begin{cases} \forall i \in [1..n], \sum_{X \in \mathcal{X}_i} c_X = b_i \\ \forall Y \in \mathcal{S}_c, c_Y = 0. \end{cases} \\
P_e^1(b) &: \exists X_1 \in \mathcal{X}_1, \dots, \exists X_n \in \mathcal{X}_n : \begin{cases} (c_{X_1}, \dots, c_{X_n}) = b \\ \forall Z \in \mathcal{S} \setminus \{X_1, \dots, X_n\}, c_Z = 0. \end{cases}
\end{aligned}$$

One readily sees that $P_e^1(b) \subseteq P_e(b) = P(b) \cap \mathbb{R} \mathcal{S}_e \subseteq P_{\neq 0}(b)$, and that

$P_e^1(b)$ is a finite set of cardinality $|P_e^1(b)| = \prod_{i=1}^n p_i = \prod_{\substack{1 \leq i \leq n \\ p_i > 1}} p_i$. If there is

no isomerism among the elementary species, i.e. if $\mathcal{S}_e^1 = \mathcal{S}_e$, then

$$P_e^1(b) = P_e(b) = P(b) \cap \mathbb{R} \mathcal{S}_e = \{b\}.$$

The interpretation of the set $P(b)$ is that it consists of all concentration vectors such that for each $i \in [1..n]$, b_i is the total concentration of all the occurrences of the elementary composition $e_{n,i}$. The subset $P_e(b)$ consists of those concentration vectors for which only elementary species are present. Further restricting by allowing only one elementary species in each isomerism class yields $P_e^1(b)$. With reference to the discussion on polyhedral geometry topics in Section 2, we note that the stoichiometric compatibility class $P(b)$ is a compact convex polytope. The set $P_e(b)$ is a face of $P(b)$. The points

910 in $P_e^1(b)$ are the vertices of $P_e(b)$, or equivalently the vertices of $P(b)$ lying
 911 in the elementary species subspace $\mathbb{R}\mathcal{S}_e$.

912

913 The equations (7.2) that characterize the class $P(b)$ give rise to an obvious
 914 necessary condition for $P(b)$ -admissibility (Definition 4.4).

915 **Proposition 7.2.** *Suppose that a subset $\mathcal{Z} \subseteq \mathcal{S}$ is $P(b)$ -admissible. Then
 916 for every $i \in [1..n]$ such that $b_i > 0$, we have $\mathcal{Z} \cap (\mathcal{X}_i \sqcup \mathcal{S}_{c,i}) \neq \emptyset$, i.e. the
 917 elementary composition $e_{n,i}$ occurs in \mathcal{Z} , either explicitly from an elemen-
 918 tary species, or implicitly from a composite species, or both. Also, for every
 919 $i \in [1..n]$ such that $b_i = 0$, we have $\mathcal{Z} \cap (\mathcal{X}_i \sqcup \mathcal{S}_{c,i}) = \emptyset$. \square*

920 Next we have a series of properties of reach-closed sets and siphons which
 921 lead to the important results of Theorem 7.7. We begin with a rather obvious
 922 one.

923 **Lemma 7.3.** *Suppose that all composite species are explicitly destructible.
 924 Consider a reach-closed set $\mathcal{Z} \subseteq \mathcal{S}$ and let $i \in [1..n]$. If $\mathcal{Z} \cap \mathcal{S}_{c,i} \neq \emptyset$,
 925 then $\mathcal{Z} \cap \mathcal{X}_i \neq \emptyset$. \square*

926 **Lemma 7.4.** *Consider a siphon $\mathcal{Z} \subseteq \mathcal{S}$.*

927 (i) *Suppose that a species $Y \in \mathcal{Z}$ is explicitly constructible. Then there
 928 exists a species $Z \in \mathcal{Z}$ such that $\mathcal{E}(Z) \not\leq \mathcal{E}(Y)$.*

929 (ii) *Suppose that a species $X \in \mathcal{Z}$ is explicitly destructive. Then there
 930 exists a species $Z \in \mathcal{Z}$ such that $\mathcal{E}(Z) \not\geq \mathcal{E}(X)$.*

931 *Proof.*

932 We prove statement (i). Let $Y \in \mathcal{S}$ be explicitly constructible. Then let Q
 933 be a complex and Y_0, \dots, Y_ℓ be species such that we have the binding re-
 934 action $Q \rightarrow Y_0$ and the isomerization reactions $Y_0 \rightarrow \dots \rightarrow Y_\ell$, and $Y_\ell = Y$.
 935 By the conservation of composition, we have $\tilde{\mathcal{E}}(Q) = \mathcal{E}(Y)$. Then, because
 936 $|Q| \geq 2$, we have $\mathcal{E}(Z) \not\leq \mathcal{E}(Y)$ for all $Z \in \text{Supp}(Q)$ by Lemma 5.7. Now
 937 suppose that $Y \in \mathcal{Z}$. Because \mathcal{Z} is a siphon we obtain $Y_0 \in \mathcal{Z}$ by induc-
 938 tion, and therefore $Z \in \mathcal{Z}$ for some $Z \in \text{Supp}(Q)$.

939 We prove statement (ii). Let $X \in \mathcal{S}$ be explicitly destructive. Then let
 940 $Z \rightarrow Q$ be a dissociation reaction such that $X \in \text{Supp}(Q)$. By the conser-
 941 vation of composition, we have $\mathcal{E}(Z) = \tilde{\mathcal{E}}(Q)$. Then, because $|Q| \geq 2$ and
 942 $X \in \text{Supp}(Q)$, we have $\mathcal{E}(Z) \not\geq \mathcal{E}(X)$ by Lemma 5.7. Now suppose that
 943 $X \in \mathcal{Z}$. Because \mathcal{Z} is a siphon, we have $Z \in \mathcal{Z}$. \square

944 **Proposition 7.5.** *Consider a siphon $\mathcal{Z} \subseteq \mathcal{S}$.*

945 (i) *Suppose that all composite species are explicitly constructible.*

946 *If $\mathcal{S}_c \cap \mathcal{Z} \neq \emptyset$, then $\mathcal{S}_e \cap \mathcal{Z} \neq \emptyset$.*

947 (ii) *Suppose that all elementary species are explicitly destructive.*

948 *If $\mathcal{S}_e \cap \mathcal{Z} \neq \emptyset$, then $\mathcal{S}_c \cap \mathcal{Z} \neq \emptyset$.*

949 *Proof.* Statement (i) of Proposition 7.5 is proved by reasoning by induction
 950 with Statement (i) of Lemma 7.4. Statement (ii) of Proposition 7.5 is a
 951 direct consequence of Statement (ii) of Lemma 7.4. \square

952 Recalling that the reach-closed sets are the complements of siphons (Propo-
 953 sition 3.4), we immediately get the following result from Proposition 7.5.

954 **Theorem 7.6.** *Suppose that the reaction network \mathcal{N} is explicitly-reversibly
 955 constructive and let $\mathcal{Z} \subseteq \mathcal{S}$. If \mathcal{Z} is reach-closed, and if $\mathcal{S}_e \subseteq \mathcal{Z}$ or
 956 $\mathcal{S}_c \subseteq \mathcal{Z}$, then $\mathcal{Z} = \mathcal{S}$. \square*

957 **Theorem 7.7.** *Suppose that the reaction network \mathcal{N} is explicitly-reversibly
 958 constructive and that there is no isomerism among the elementary species.
 959 Then, only the entire set \mathcal{S} of species is both reach-closed and stoichiome-
 960 trically admissible. If the network is governed by mass-action kinetics, then
 961 it is vacuously persistent. If in addition the network is complex-balanced
 962 (and in particular if it is weakly reversible and has deficiency zero), then
 963 the (unique, positive, complex-balanced) equilibrium state in $P_{>0}(b)$ is an
 964 attractor of $P(b)$.*

965 *Proof.* Let $b \in \mathbb{R}_{>0}^n$ and let $\mathcal{Z} \subseteq \mathcal{S}$ be both reach-closed and $P(b)$ -admissible.
 966 Let $i \in [1..n]$. By Proposition 7.2, we have $\mathcal{Z} \cap (\mathcal{X}_i \sqcup \mathcal{S}_{c,i}) \neq \emptyset$. Then
 967 with Lemma 7.3, we have $\mathcal{Z} \cap \mathcal{X}_i \neq \emptyset$, i.e. $\mathcal{X}_i \subseteq \mathcal{Z}$ because \mathcal{X}_i is a sin-
 968 gleton. This holds for all $i \in [1..n]$, so $\mathcal{S}_e \subseteq \mathcal{Z}$. Finally, by Theorem 7.6,
 969 $\mathcal{Z} = \mathcal{S}$. We then obtain vacuous persistence with Theorem 4.6. We already
 970 noted the global attraction result in comments following Definition 4.1. \square

971 We record next some observations on the rank and the deficiency under
 972 structural conditions that are in effect in Theorem 7.7. They follow from
 973 Lemma 5.14.

974 **Remark 7.8.** Suppose that the reaction network \mathcal{N} is constructive and
 975 that there is no isomerism among the elementary species. Then the rank of
 976 \mathcal{N} equals the number of composite species and

978 $\text{Deficiency} = \#(\text{Complexes}) - \#(\text{Linkage classes}) - \#(\text{Composite species}).$

979 If in fact \mathcal{N} is explicitly constructive, then

$$980 \quad \text{Deficiency} = \#(\text{Complexes of length } \geq 2) - \#(\text{Linkage classes}),$$

982 and in particular, \mathcal{N} has deficiency zero if and only if each linkage class
983 contains precisely one complex of length ≥ 2 . \square

984 Theorem 7.7 shows that for a mass-action explicitly-reversibly constructive
985 network, the failure of vacuous persistence requires that there be isomerism
986 among the elementary species. (Isomerism among composite species does
987 not affect this feature.) This provides a theoretical explanation for the fact
988 that instances of non-persistence and of non-obvious persistence in the literature
989 always involve networks with isomerism among the building blocks.
990 Enzymatic networks are a natural consideration for satisfying the require-
991 ment of isomerism among elementary species. But, as we see in the next
992 section, even in that context, vacuous persistence is achieved for a subclass
993 of networks that are important in biochemistry.

994 8 Application: Binary Enzymatic Networks

995 *Half the battle in understanding is having the right representation.*

996 Attributed to Pierre-Simon Laplace.

997 We propose a definition of binary enzymatic networks with two intended
998 goals. We seek to cover the mechanisms in which one enzyme and one sub-
999 strate bind into an intermediate enzymatic complex which, possibly after
1000 isomerization, dissociates into the same enzyme and one product, which
1001 may be identical to or different than the substrate. At the same time, we
1002 want to facilitate the mathematical deduction of the properties of such net-
1003 works. The networks of Figures 2.1(b) and 8.1(a)-(c), which were found in
1004 Angeli, De Leenheer and Sontag [3] to be persistent, are examples of net-
1005 works that our definition will cover. These particular networks are examples
1006 of futile and cascaded networks as defined in this section. We prove that
1007 such networks are in fact vacuously persistent.

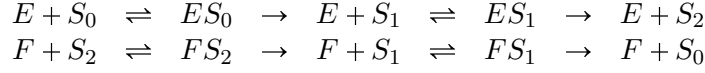
1008

1009 So as to focus the scope of the paper, we deliberately leave out a number
1010 of biochemically important mechanisms. These include the simultaneous or
1011 stepwise binding of several substrates onto one enzyme, and the simultane-
1012 ous or stepwise dissociation of several products from one enzyme.

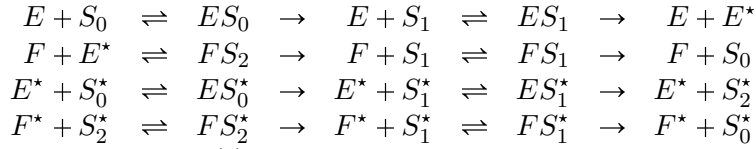
1013

1014 We fix a reaction network $\mathcal{N} = (\mathcal{S}, \mathcal{C}, \mathcal{R})$ and we proceed by formulating
 1015 a series of conditions which will lead to the definition.

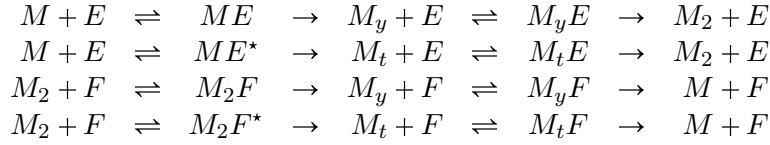
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(a) A futile cycle of two two-step pathways.



(b) Two futile cycles in a cascade.



(c) One futile cycle with two alternate pathways in each direction.

Figure 8.1: Examples of binary enzymatic networks.

1017 **Condition (Enz1).** Four proper and nonempty subsets Enz , Sub , Pro , Int
 1018 of \mathcal{S} are given and we have $\text{Enz} \cup \text{Sub} \cup \text{Pro} = \mathcal{S} \setminus \text{Int}$. \square

1019

1020 The species in Enz , Sub , Pro , Int are respectively the *enzymes*, the *substrates*,
 1021 the *products* and the *intermediates*. We collectively refer to the substrates
 1022 and the products as the *enzyme partners* or simply the *partners*. We set
 1023 $\text{Par} := \text{Sub} \cup \text{Pro}$ and $\text{Enz}_0 := \text{Enz} \setminus \text{Par}$, and we have $\mathcal{S} = \text{Enz}_0 \sqcup \text{Par} \sqcup \text{Int}$.

1024

1025 **Condition (Enz2).** A subset $\text{Cat} \subseteq \text{Enz} \times \text{Sub} \times \text{Pro}$ of *catalysis triples* is
 1026 given. Every enzyme occurs in some catalysis triple, so does every substrate
 1027 and so does every product. \square

1028

1029 A *substrate-product pair* is any $(A, B) \in \text{Sub} \times \text{Pro}$ such that (E, A, B) is
 1030 a catalysis triple for some enzyme E . We introduce the *partner graph*
 1031 ParGraph , the directed graph whose sets of vertices and edges are respec-
 1032 tively the set $\text{Par} = \text{Sub} \cup \text{Pro}$ of enzyme partners and the set of substrate-
 1033 product pairs. We call *undirected partner graph* and denote $\underline{\text{ParGraph}}$ the
 1034 corresponding undirected graph. We equip the set Par with the equiva-
 1035 lence relation whose equivalence classes are the connected components of

1036 the graph $\underline{\text{ParGraph}}$. The quotient map is $\text{cl} : \text{Par} \rightarrow \overline{\text{Par}}$.

1037

1038 **Condition (Enz3).** A surjective map $\text{par} : \text{Enz} \rightarrow \overline{\text{Par}}$ is given. We have
 1039 $E \notin \text{par}(E)$ for every enzyme $E \in \text{Enz}$ and $A, B \in \text{par}(E)$ for every catalysis
 1040 triple $(E, A, B) \in \text{Cat}$. \square

1041

1042 The requirement $E \notin \text{par}(E)$ expresses the absence of autocatalysis. The de-
 1043 velopments we carry out are actually possible without this restriction, but
 1044 at the cost of a longer discussion.

1045

1046 For $E \in \text{Enz}$, we set $\text{sub}(E) := \text{par}(E) \cap \text{Sub}$ and $\text{pro}(E) := \text{par}(E) \cap \text{Pro}$,
 1047 and we have $\text{par}(E) = \text{sub}(E) \cup \text{pro}(E)$, $\text{Sub} = \bigcup_{E \in \text{Enz}} \text{sub}(E)$, $\text{Pro} = \bigcup_{E \in \text{Enz}} \text{pro}(E)$.

1048

1049 **Condition (Enz4).** Given are an equivalence relation on Int with quotient
 1050 map $\text{cl} : \text{Int} \rightarrow \overline{\text{Int}}$ and two mutually inverse bijective maps $\text{int} : \text{Enz} \rightarrow \overline{\text{Int}}$
 1051 and $\text{enz} : \overline{\text{Int}} \rightarrow \text{Enz}$. \square

1052

1053 The map $\text{cl} : \text{Int} \rightarrow \overline{\text{Int}}$ groups intermediates by what enzyme they contain.
 1054 For $E \in \text{Enz}_0$, we set $\text{cl}(E) := E$, so $\text{cl}(X)$ is defined for every species X .

1055

1056 **Condition (Enz5).** For every catalysis triple $(E, A, B) \in \text{Cat}$, we are given
 1057 a nonempty set $\text{IntPath}(E, A, B)$ of *intermediates paths*, which are nonempty
 1058 finite tuples of intermediates in $\text{int}(E)$, such that with

$$1059 \mathcal{R}_{(Y_1, \dots, Y_\ell)}(E, A, B) := \{E + A \rightarrow Y_1, Y_\ell \rightarrow E + B\} \cup \{Y_{j-1} \rightarrow Y_j : j \in [2.. \ell]\}$$

1060 for each $(Y_1, \dots, Y_\ell) \in \text{IntPath}(E, A, B)$, and

$$1061 \mathcal{R}(E, A, B) := \bigcup_{(Y_1, \dots, Y_\ell) \in \text{IntPath}(E, A, B)} \mathcal{R}_{(Y_1, \dots, Y_\ell)}(E, A, B),$$

1062 we have for the set \mathcal{R} of all reactions,

$$1063 \mathcal{R} = \bigcup_{(E, A, B) \in \text{Cat}} \mathcal{R}(E, A, B).$$

1064 \square

1065 **Definition 8.1.** The reaction network $\mathcal{N} = (\mathcal{S}, \mathcal{E}, \mathcal{R})$ is a *binary enzy-*
 1066 *matic network* provided the five conditions (Enz1)-(Enz5) are satisfied. \square

1067 Enzyme-catalyzed mechanisms can be distributive or processive; see e.g.
 1068 Gunawardena [8]. With our terminology and notations, the action of an
 1069 enzyme E on a substrate $A \in \text{sub}(E)$ is distributive if there exists a unique
 1070 product $B \in \text{pro}(E) \setminus \{A\}$ such that (E, A, B) is a catalytic triple. Other-
 1071 wise the action is processive. It appears that with a distributive action,
 1072 we also have $\text{IntPath}(E, A, B) = \{(Y)\}$, where the intermediate species Y is
 1073 produced by the binding of E and A and dissociates either into E and A or
 1074 into E and B ; whereas a processive action may afford intermediates paths
 1075 of length two or more. Indeed, a long intermediates path presents the possi-
 1076 bility of several products being formed from dissociations of intermediates.
 1077 Definition 8.1 accommodates both distributive and processive mechanisms.
 1078 Also, because enzymes are allowed to be partners of other enzymes, Defini-
 1079 tion 8.1 accommodates cascaded mechanisms, as will be seen in more details
 1080 later on in this section. We expect that for a network of enzyme-catalyzed
 1081 conversions of substrates into products to not be covered by Definition 8.1,
 1082 there must be intermediate species that are at least ternary in terms of their
 1083 enzyme and substrate constituents.

1084

1085 For the rest of this section, \mathcal{N} is a binary enzymatic network. We define
 1086 the map $\mathcal{E} : \mathcal{S} \rightarrow (\mathbb{Z}_{\geq 0}(\text{Enz}_0 \sqcup \overline{\text{Par}})) \setminus \{0\}$ as follows.

$$\text{For } X \in \text{Enz} \cup \text{Sub} \cup \text{Pro}, \mathcal{E}(X) := \text{cl}(X).$$

1087

$$\text{For } Y \in \text{Int}, \mathcal{E}(Y) := \text{cl}(E) + \text{par}(E), \text{ where } E = \text{enz}(\text{cl}(Y)).$$

1088 The map \mathcal{E} is a composition map of the network \mathcal{N} , with the notable feature
 1089 that composition tuples are not indexed by $[1..n]$, but by $\text{Enz}_0 \sqcup \overline{\text{Par}}$. Let
 1090 \mathbb{E} be its extension $\mathbb{R}\mathcal{S} \rightarrow \mathbb{R}(\text{Enz}_0 \sqcup \overline{\text{Par}})$.

1091 **Proposition 8.2.** *The composition \mathcal{E} is a core composition of the network*
 1092 *\mathcal{N} . The elementary species are the enzymes, the substrates and the pro-*
 1093 *ducts, while the composite species are the intermediates. The network \mathcal{N} is*
 1094 *explicitly constructive.*

1095 The network may fail to be explicitly-reversibly constructive only if there are
 1096 substrates that are not also products or products that are not also substrates.

1097 *Proof.* We show that the three conditions of Theorem 6.2 are realized.

1098 We prove condition (1) of Theorem 6.2. We see from the definition of
 1099 \mathcal{E} that the \mathcal{E} -elementary species are the species $X \in \text{Enz} \cup \text{Sub} \cup \text{Pro}$ and
 1100 every \mathcal{E} -elementary composition occurs as $\text{cl}(X)$ for such a species X . We

1101 show that all reactions are \mathcal{E} -conservative. Let $(E, A, B) \in \text{Cat}$ and let
 1102 $(Y_1, \dots, Y_\ell) \in \text{IntPath}(E, A, B)$. We have $A, B \in \text{par}(E)$, which is equiva-
 1103 lent to $\text{cl}(A) = \text{cl}(B) = \text{par}(E)$, and we have $Y_1, \dots, Y_\ell \in \text{int}(E)$, which is
 1104 equivalent to $E = \text{enz}(\text{cl}(Y_1)) = \dots = \text{enz}(\text{cl}(Y_\ell))$. So by the definition of \mathcal{E} ,
 1105 we have $\mathcal{E}(Y_1) = \dots = \mathcal{E}(Y_\ell) = \text{cl}(E) + \text{cl}(A) = \text{cl}(E) + \text{cl}(B)$. On another
 1106 hand, we have $\tilde{\mathcal{E}}(E + X) = \mathcal{E}(E) + \mathcal{E}(X) = \text{cl}(E) + \text{cl}(X)$ for $X = A$ and
 1107 $X = B$. It results that all reactions in $\mathcal{R}_{(Y_1, \dots, Y_\ell)}(E, A, B)$ are \mathcal{E} -conservative.
 1108 Thus, \mathcal{E} is a near-core composition of \mathcal{N} .

1109 As a preparation for proving condition (2) of Theorem 6.2, we note that if
 1110 (A, B) is a substrate-product pair, then the species A and B are stoichio-
 1111 metrically isomeric. Indeed, let E be an enzyme such that (E, A, B) is a
 1112 catalysis triple, and let $(Y_1, \dots, Y_\ell) \in \text{IntPath}(E, A, B)$. We have
 1113 $B - A = (E + B - Y_\ell) + \sum_{j=2}^{\ell} (Y_j - Y_{j-1}) + (Y_1 - E - A)$, and therefore
 1114 $B - A$ lies in the stoichiometric space.

1115 We prove condition (2) of Theorem 6.2. From the definition of the composi-
 1116 tion map \mathcal{E} , we get that the \mathcal{E} -isomerism classes of \mathcal{E} -elementary species are
 1117 the singletons of elements of Enz_0 and the elements of $\overline{\text{Par}}$. So we need to
 1118 show that if $A, B \in \text{Par}$ and $\text{cl}(A) = \text{cl}(B)$, then A and B are stoichiometri-
 1119 cally isomeric. Consider such A and B . Then there exists $C_0, \dots, C_r \in \text{Par}$
 1120 such that $C_0 = A$, $C_r = B$, and for each $j \in [1..r]$, (C_{j-1}, C_j) or (C_j, C_{j-1}) is
 1121 a substrate-product pair. In either case, C_{j-1} and C_j are stoichiometrically
 1122 isomeric. Consequently, A and B are stoichiometrically isomeric.

1123 We prove condition (3) of Theorem 6.2. The \mathcal{E} -composite species are the
 1124 intermediates. Let $Y \in \text{Int}$. Because the species Y participates in at least
 1125 one reaction, there exists a catalysis triple (E, A, B) and an intermediates
 1126 path $(Y_1, \dots, Y_\ell) \in \text{IntPath}(E, A, B)$ such that Y is one of the intermediates
 1127 Y_1, \dots, Y_ℓ . We have $\mathcal{E}(Y) = \mathcal{E}(E) + \mathcal{E}(A)$ and $Y - E - A$ is in the stoichio-
 1128 metric space. \square

1129 We introduce the following sets of species for $E \in \text{Enz}_0$ and $\mathcal{X} \in \overline{\text{Par}}$.

$$\begin{aligned} 1130 \quad \mathcal{S}(E) &:= \{E\} \sqcup \text{int}(E), \\ 1131 \quad \mathcal{S}'(\mathcal{X}) &:= \{Y \in \text{Int} : \text{cl}(\text{enz}(\text{cl}(Y))) = \mathcal{X}\}, \\ 1132 \quad \mathcal{S}''(\mathcal{X}) &:= \{Y \in \text{Int} : \text{par}(\text{enz}(\text{cl}(Y))) = \mathcal{X}\}, \\ 1133 \quad \mathcal{S}(\mathcal{X}) &:= \mathcal{X} \sqcup \mathcal{S}'(\mathcal{X}) \sqcup \mathcal{S}''(\mathcal{X}). \end{aligned}$$

1135 We have $\mathcal{S}'(\mathcal{X}) \cap \mathcal{S}''(\mathcal{X}) = \emptyset$ because, by Condition (Enz3), we have

1136 $\text{enz}(\text{cl}(Y)) \notin \text{par}(\text{enz}(\text{cl}(Y)))$ for any $Y \in \text{Int}$. Now we set

$$1137 \quad T_E := \text{sum}(\mathcal{S}(E)) = E + \text{sum}(\text{int}(E)),$$

$$1138 \quad T_{\mathcal{X}} := \text{sum}(\mathcal{S}(\mathcal{X})) = \text{sum}(\mathcal{X}) + \text{sum}(\mathcal{S}'(\mathcal{X})) + \text{sum}(\mathcal{S}''(\mathcal{X})).$$

1140 With Theorem 5.17, we get:

1141 **Proposition 8.3.** *The vectors T_E for $E \in \text{Enz}_0$ and $T_{\mathcal{X}}$ for $\mathcal{X} \in \overline{\text{Par}}$ form*
 1142 *a basis of the orthogonal of the stoichiometric space.* \square

1143 A subset $\mathcal{Z} \subseteq \mathcal{S}$ is fixed for the rest of this section. The next two results
 1144 are applications of Proposition 7.2 and Lemma 7.3 respectively.

1145 **Lemma 8.4.** *If \mathcal{Z} is stoichiometrically admissible, then $\mathcal{Z} \cap \mathcal{S}(E) \neq \emptyset$*
 1146 *for all $E \in \text{Enz}_0$ and $\mathcal{Z} \cap \mathcal{S}(\mathcal{X}) \neq \emptyset$ for all $\mathcal{X} \in \overline{\text{Par}}$.* \square

1147 **Lemma 8.5.** *Suppose that \mathcal{Z} is reach-closed. Let $\mathcal{X} \in \overline{\text{Par}}$.*
 1148 *If $\mathcal{Z} \cap \mathcal{S}(\mathcal{X}) \neq \emptyset$, then $\mathcal{Z} \cap \mathcal{X} \neq \emptyset$.* \square

1149 For $E \in \text{Enz}$, let $\mathcal{C}(E) := \text{int}(E) \sqcup \{E + A : A \in \text{par}(E)\}$. By Condition (Enz5),
 1150 the set \mathcal{C} of complexes is given by $\mathcal{C} = \bigsqcup_{E \in \text{Enz}} \mathcal{C}(E)$. We define the sets
 1151 $\text{isub}(E) \subseteq \text{sub}(E)$ and $\text{tpro}(E) \subseteq \text{pro}(E)$ as follows.

1152 **Definition 8.6.**

- 1153 • For $A \in \text{sub}(E)$, we have $A \in \text{isub}(E)$ if and only if the complex $E + A$
 1154 ultimately reacts to every complex in $\mathcal{C}(E)$. The elements of $\text{isub}(E)$
 1155 are the *initial substrates* of E .
- 1156 • For $B \in \text{pro}(E)$, we have $B \in \text{tpro}(E)$ if and only if every complex in
 1157 $\mathcal{C}(E)$ ultimately reacts to the complex $E + B$. The elements of $\text{tpro}(E)$
 1158 are the *terminal products* of E . \square

1159 Following is a trivial but instrumental observation.

1160 **Remark 8.7.** Let $E \in \text{Enz} \cap \mathcal{Z}$. Suppose that \mathcal{Z} is reach-closed.

- 1161 • If $\mathcal{Z} \cap \text{isub}(E) \neq \emptyset$, then $\text{par}(E) \subseteq \mathcal{Z}$ and $\text{int}(E) \subseteq \mathcal{Z}$.
- 1162 • If $\mathcal{Z} \cap \text{par}(E) \neq \emptyset$ or if $\mathcal{Z} \cap \text{int}(E) \neq \emptyset$, then $\text{tpro}(E) \subseteq \mathcal{Z}$. \square

1163 **Definition 8.8.** An enzyme F is a *reversing enzyme* for an enzyme E if
 1164 $\emptyset \neq \text{tpro}(E) = \text{isub}(F)$. The network \mathcal{N} is *futile* if every enzyme is a re-
 1165 versing enzyme. \square

1166 Note that if F is a reversing enzyme for E , then $\text{par}(E) = \text{par}(F)$. Our
 1167 definition of a futile network is sufficient for the intended use. But it of-
 1168 ten also holds that every enzyme has a reversing enzyme. In fact, as is
 1169 the case for the networks of Figures 2.1(b) and 8.1(a)-(c), there often is a
 1170 *futility involution*, a map $\varphi : \text{Enz} \rightarrow \text{Enz}$ such that $\varphi^2 = \varphi \circ \varphi = \text{Id}_{\text{Enz}}$ and
 1171 for every enzyme E , $\varphi(E)$ is a reversing enzyme for E . With reference to
 1172 Proposition 8.2 and the subsequent comment, we note that if every enzyme
 1173 both is and has a reversing enzyme, then the network is explicitly-reversibly
 1174 constructive.

1175 **Theorem 8.9.** *Suppose that the network \mathcal{N} is futile. If \mathcal{Z} is stoichiome-*
 1176 *trically admissible and reach-closed, and if $\text{Enz} \subseteq \mathcal{Z}$, then $\mathcal{Z} = \mathcal{S}$.*

1177 *Proof.* Let $F \in \text{Enz}$. There exists $E \in \text{Enz}$ such that F is a reversing enzyme
 1178 for E . By Lemma 8.4, we have $\mathcal{Z} \cap \mathcal{S}(\text{par}(E)) \neq \emptyset$. Then by Lemma 8.5,
 1179 we have $\mathcal{Z} \cap \text{par}(E) \neq \emptyset$. Next, the second assertion of Remark 8.7 implies
 1180 that $\text{tpro}(E) \subseteq \mathcal{Z}$. Therefore, by Definition 8.8, $\emptyset \neq \text{isub}(F) \subseteq \mathcal{Z}$. Then,
 1181 with the first assertion of Remark 8.7, we have $\text{par}(F) \subseteq \mathcal{Z}$ and $\text{int}(F) \subseteq \mathcal{Z}$.
 1182 This holds for all $F \in \text{Enz}$, so $\mathcal{Z} = \mathcal{S}$. \square

1183 We see next that the condition $\text{Enz} \subseteq \mathcal{Z}$, which is required in Theorem 8.9,
 1184 can be satisfied if there are no enzymes that are also in the role of enzyme
 1185 partners (i.e. if $\text{Enz}_0 = \text{Enz}$), and more generally provided double roles occur
 1186 in an orchestrated scheme known as a cascade. We define the sets $\text{Enz}_{\leq m}$
 1187 for $m \in \mathbb{Z}_{\geq 0}$ and Enz_m for $m \in \mathbb{Z}_{\geq 1}$ as follows.

$$1188 \quad \text{Enz}_{\leq m} := \bigcup_{\mu=0}^m \text{Enz}_{\mu};$$

$$1189 \quad \text{Enz}_m := (\text{Enz} \setminus \text{Enz}_{\leq m-1}) \cap \bigcup_{E \in \text{Enz}_{m-1}} \text{tpro}(E).$$

1190

1191 The sets Enz_m for $m \in \mathbb{Z}_{\geq 0}$ are pairwise disjoint, so there exists $m_0 \in \mathbb{Z}_{\geq 0}$
 1192 such that $\text{Enz}_m = \emptyset$ for $m > m_0$.

1193 **Definition 8.10.** The network \mathcal{N} is *cascaded* if $\text{Enz} = \bigsqcup_{m=0}^{\infty} \text{Enz}_m$.

1194 An enzyme $E \in \text{Enz}_m$ is said to have *cascade index* $\gamma(E) = m$. \square

1195 Note that if \mathcal{N} is cascaded, then $\text{Enz}_0 \neq \emptyset$. This is because $\text{Enz} \neq \emptyset$ and
 1196 it holds that $\text{Enz}_m = \emptyset \Rightarrow \text{Enz}_{m+1} = \emptyset$.

1197 **Theorem 8.11.** *Suppose that the network \mathcal{N} is cascaded. If \mathcal{Z} is stoichio-*
 1198 *metrically admissible and reach-closed, then $\text{Enz} \subseteq \mathcal{Z}$.*

1199 *Proof.* Let $E \in \text{Enz}_0$. By Lemma 8.4, we have $\mathcal{Z} \cap \mathcal{S}(E) \neq \emptyset$, i.e. $E \in \mathcal{Z}$
 1200 or $\mathcal{Z} \cap \text{int}(E) \neq \emptyset$. But we have $\mathcal{Z} \cap \text{int}(E) \neq \emptyset \Rightarrow E \in \mathcal{Z}$ because \mathcal{Z} is
 1201 reach-closed. So $E \in \mathcal{Z}$. Hence, $\text{Enz}_0 \subseteq \mathcal{Z}$. Let $m \in \mathbb{Z}_{\geq 1}$ and assume for
 1202 induction that $\text{Enz}_{m-1} \subseteq \mathcal{Z}$. Then for every $E \in \text{Enz}_{m-1}$, we successively
 1203 have: $\mathcal{Z} \cap \mathcal{S}(\text{par}(E)) \neq \emptyset$ by Lemma 8.4; $\mathcal{Z} \cap \text{par}(E) \neq \emptyset$ by Lemma
 1204 8.5; and $\text{tpro}(E) \subseteq \mathcal{Z}$ by Remark 8.7. So $\bigcup_{E \in \text{Enz}_{m-1}} \text{tpro}(E) \subseteq \mathcal{Z}$, whence
 1205 in particular, $\text{Enz}_m \subseteq \mathcal{Z}$. \square

1206 By combining Theorems 8.9 and 8.11, and then using Theorem 4.6, we get:

1207 **Theorem 8.12.** *Suppose that the network \mathcal{N} is futile and cascaded. Then,*
 1208 *only the entire set \mathcal{S} of species is both reach-closed and stoichiometrically*
 1209 *admissible. The network \mathcal{N} is vacuously persistent. \square*

1210 The networks of Figures 2.1(b) and 8.1(a)-(c) are binary enzymatic net-
 1211 works that are futile and cascaded, and hence vacuously persistent. So are
 1212 the post-translational modification systems studied in Thomson and Gu-
 1213 nawardena [17], more specifically those which are futile, e.g. the network
 1214 of Figure 1e in the reference. This is stricter than Conditions 1 and 2 in
 1215 the reference but should not be restrictive biochemically. Four examples of
 1216 enzymatic mechanisms are studied in Siegel and MacLean [14, Section 4].
 1217 The mechanism with no inhibitor and the one with a competitive inhibitor
 1218 (respectively in Sections 4.1 and 4.3 in the reference) are futile, cascaded
 1219 binary enzymatic networks in our terminology. (The competitive inhibitor
 1220 is simultaneously a substrate and a product.) Hence, consistently with re-
 1221 sults in the reference (Theorems 4.2 and 4.3 in the no-inhibitor case and
 1222 Theorems 4.6 and 4.7 in the competitive inhibitor case), these networks are
 1223 vacuously persistent. The mechanism with a noncompetitive inhibitor and
 1224 the one with an uncompetitive inhibitor (respectively in Sections 4.2 and
 1225 4.4 in the reference) are not binary enzymatic networks because there are
 1226 ternary species.

1227

1228 Tables 8.1 and 8.2 illustrate some of the concepts discussed in this section.

1229

1230 9 Conclusion

1231 We have provided a necessary and sufficient condition for a mass-action reac-
 1232 tion network with bounded trajectories to have vacuous persistence, a strict
 1233 form of persistence which takes into account nondegenerate trajectories that

$X \in \text{Enz}$	$\text{par}(X)$	$\text{int}(X)$	$\text{isub}(X)$	$\text{tpro}(X)$	$\varphi(X)$	$\gamma(X)$
E	$\{S_0, S_1, E^*\}$	$\{ES_0, ES_1\}$	$\{S_0\}$	$\{E^*\}$	F	0
F	$\{S_0, S_1, E^*\}$	$\{FS_1, FS_2\}$	$\{E^*\}$	$\{S_0\}$	E	0
E^*	$\{S_0^*, S_1^*, S_2^*\}$	$\{ES_0^*, ES_1^*\}$	$\{S_0^*\}$	$\{S_2^*\}$	F^*	1
F^*	$\{S_0^*, S_1^*, S_2^*\}$	$\{FS_1^*, FS_2^*\}$	$\{S_2^*\}$	$\{S_0^*\}$	E^*	0

Table 8.1: Selected concepts of Section 8 illustrated for the network of Figure 8.1(b).

$X \in \text{Enz}$	$\text{par}(X)$	$\text{int}(X)$	$\text{isub}(X)$	$\text{tpro}(X)$	$\varphi(X)$	$\gamma(X)$
E	$\{M, M_y, M_t, M_2\}$	$\{ME, M_yE, M_tE\}$	$\{M\}$	$\{M_2\}$	F	0
F	$\{M, M_y, M_t, M_2\}$	$\{MF, M_yF, M_tF\}$	$\{M_2\}$	$\{M\}$	E	0

Table 8.2: Selected concepts of Section 8 illustrated for the network of Figure 8.1(c).

1234 are not considered in ordinary persistence. We employed the work of Vol’pert
 1235 which we hope will gain more awareness in the chemical reaction network
 1236 theory community. The machinery of species composition and constructive
 1237 networks facilitated our use of the characterization of vacuous persistence.
 1238 It is also interesting in its own right and should be useful in other areas of
 1239 chemical reaction network theory. We saw that isomerism among elemen-
 1240 tary species is necessary, but by no means not sufficient, for non-persistence.
 1241 It should be possible to bring these two findings closer with a generalization
 1242 of Theorem 7.7 that would affirm vacuous persistence in the presence of iso-
 1243 merism among elementary species. Indeed, to require that there be no such
 1244 isomerism is really an extreme way of imposing a condition which ensures
 1245 that membership of isomeric elementary species in reach-closed, stoichio-
 1246 metrically admissible sets occurs on an all-or-none basis. Essentially as a
 1247 consequence of Theorem 7.6, mass-action explicitly-reversibly constructive
 1248 networks that satisfy such condition would be vacuously persistent. The
 1249 problem is to formulate such condition in a nice and relevant fashion.

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