Glycolytic Oscillations

Biological rhythms

Biological rhythm	Period
Neural rhythms*	0.001 s to 10 s
Cardiac rhythm*	1 s
Calcium oscillations*	sec to min
Biochemical oscillations*	30 s to 20 min
Mitotic oscillator*	10 min to 24 h
Hormonal rhythms*	10 min to 3-5 h (24 h)
Circadian rhythms*	24 h
Ovarian cycle	28 days (human)
Annual rhythms	1 year
Rhythms in ecology and epidemiology	years

*Cellular rhythms

Goldbeter, A. (1996). *Biochemical Oscillations and Biological Rhythms*. Cambridge University Press.

Biological rhythms

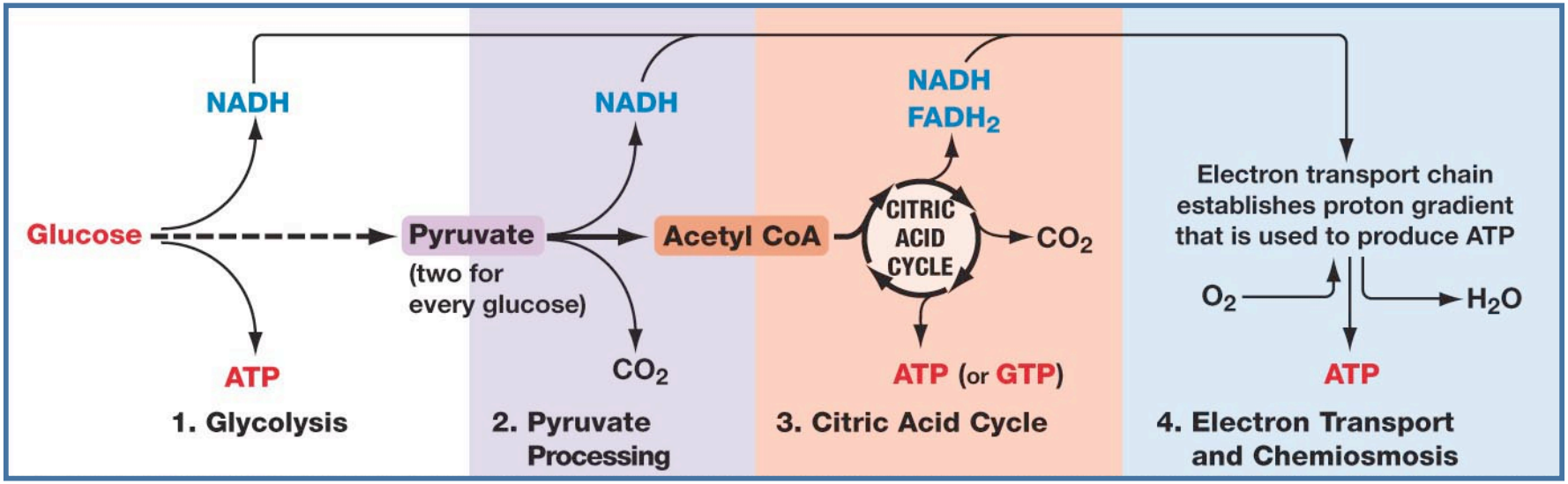
Biological rhythm	Period
Neural rhythms*	0.001 s to 10 s
Cardiac rhythm*	1 s
Calcium oscillations*	sec to min
Biochemical oscillations*	30 s to 20 min
Mitotic oscillator*	10 min to 24 h
Hormonal rhythms*	10 min to 3-5 h (24 h)
Circadian rhythms*	24 h
Ovarian cycle	28 days (human)
Annual rhythms	1 year
Rhythms in ecology and epidemiology	years

*Cellular rhythms

glycolytic oscillations

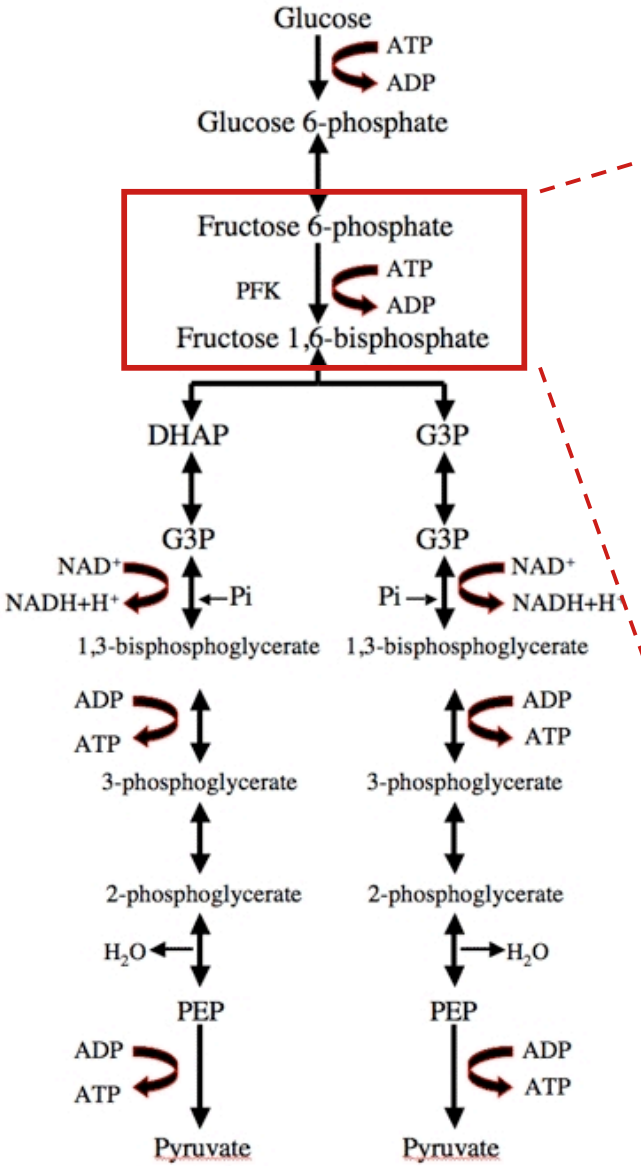
Glycolysis

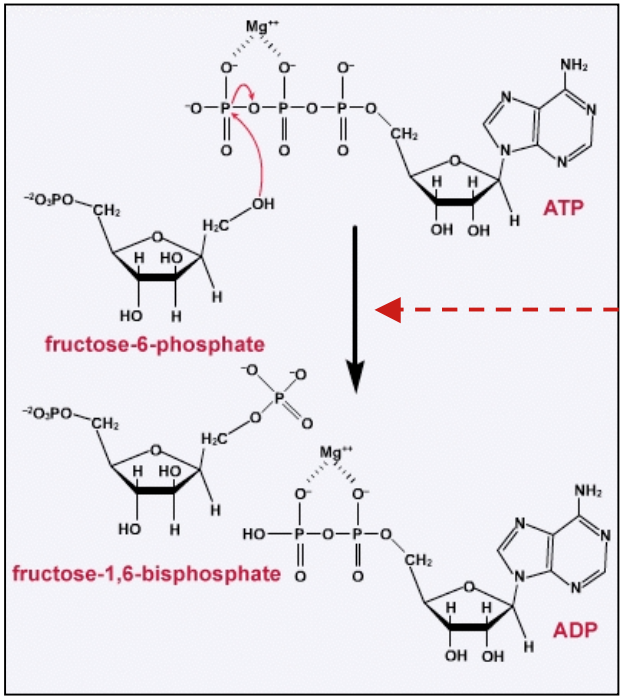
Glycolysis



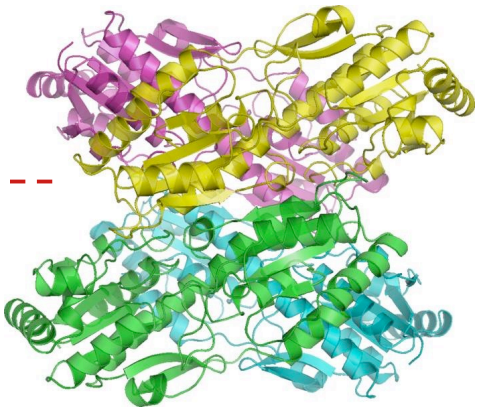
- Glycolysis literally means "splitting sugars."
- Glycolysis is the metabolic pathway that converts glucose into pyruvate.
- During glycolysis, two molecules of pyruvate are formed for every molecule of glucose.
- Pyruvate is then used in the Kreb cycle.
- Glycolysis also yields to 2 molecules of ATP and 2 molecules of NADH.
- Glycolysis takes place in the cytoplasm.

Glycolysis & PFK

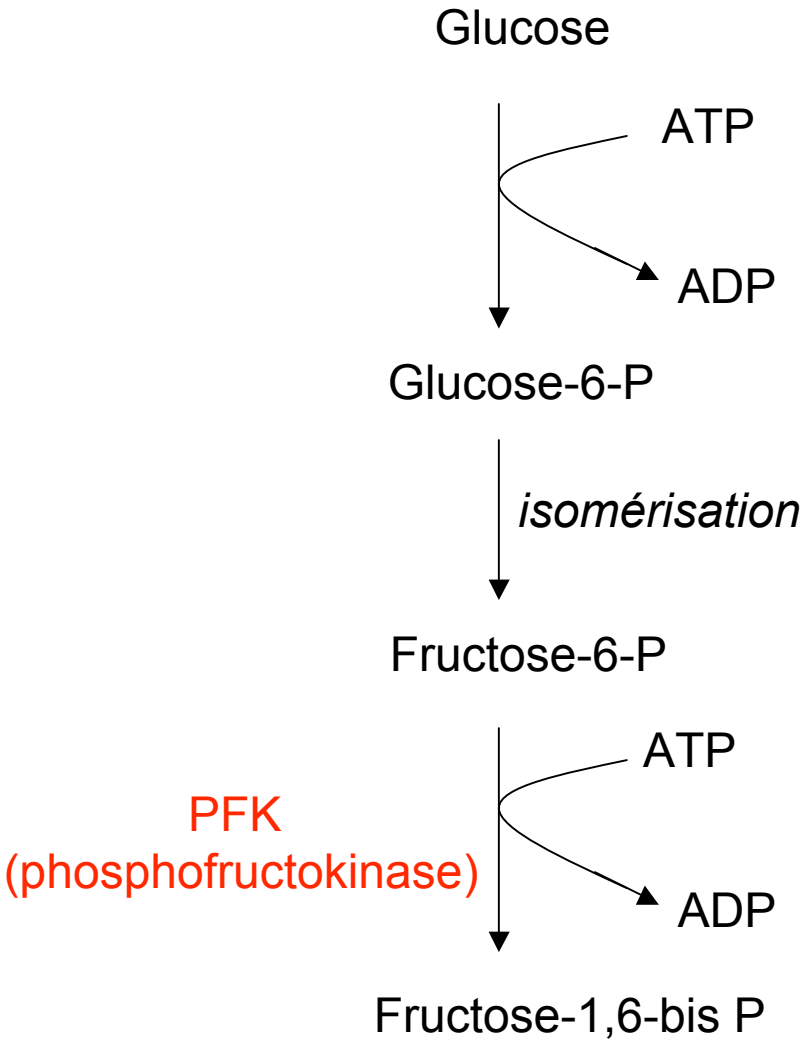


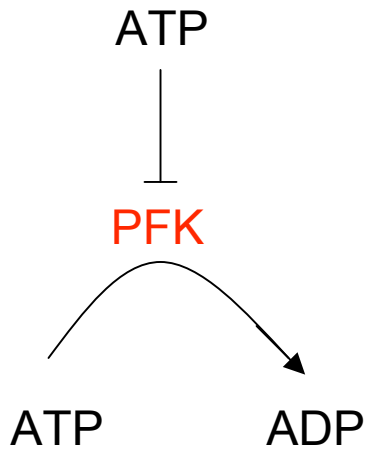


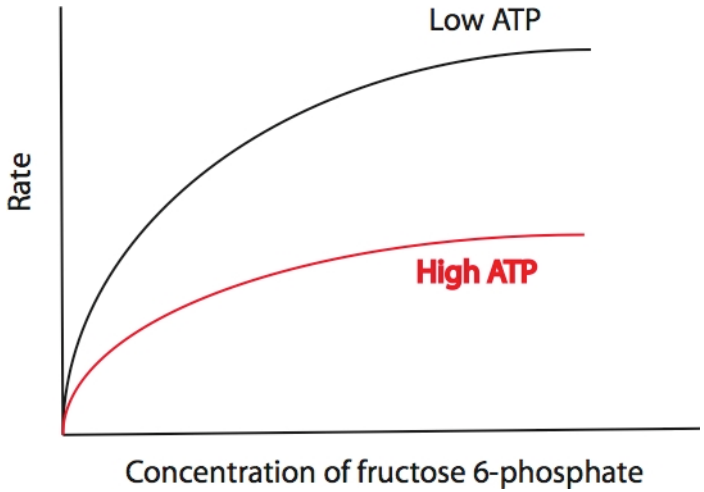
phosphofructokinase (PFK)



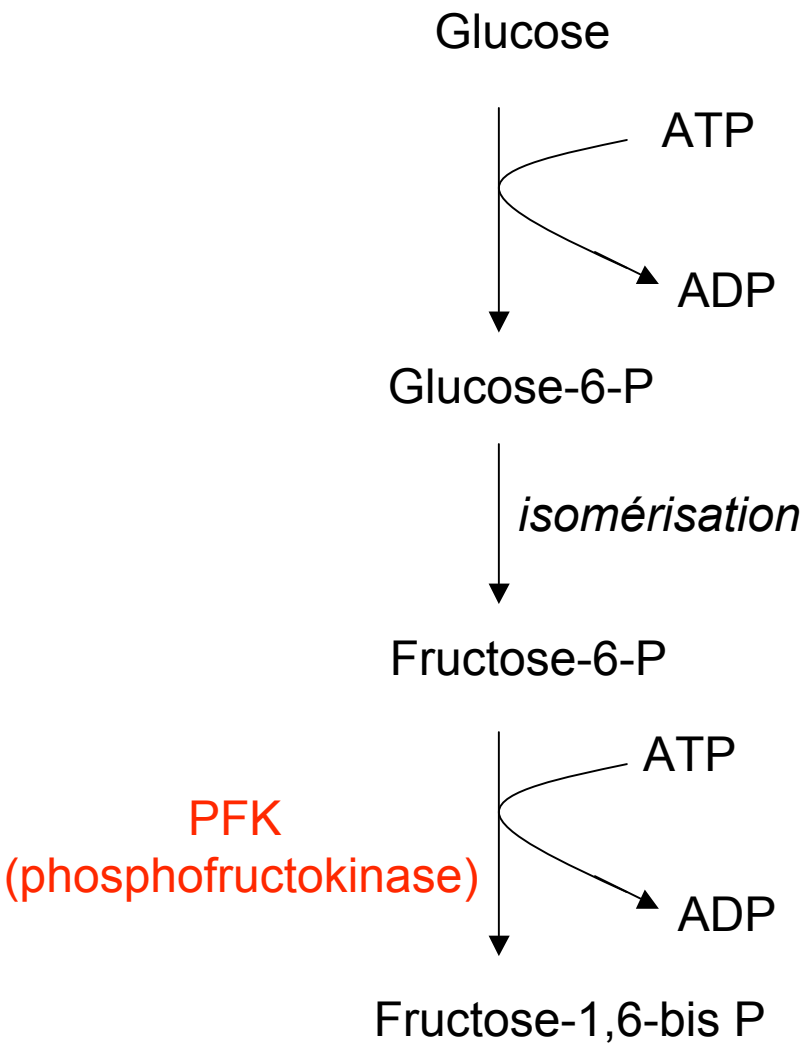
Glycolysis & PFK

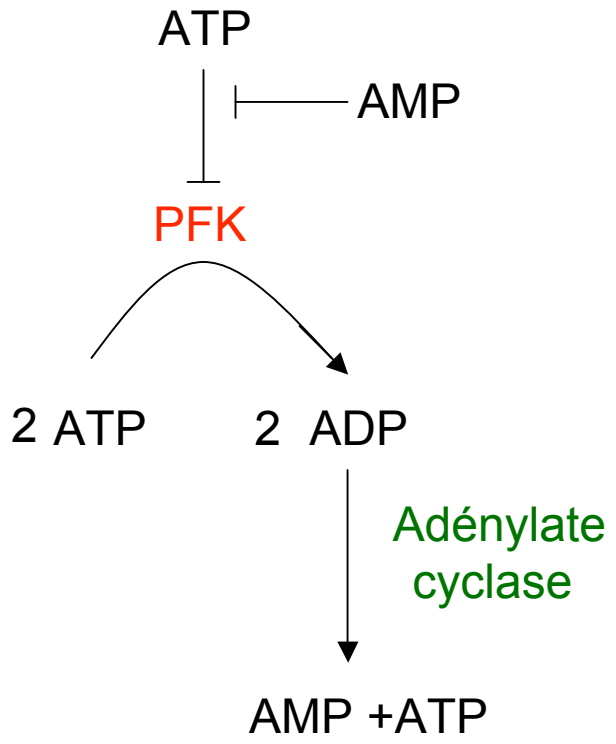






Glycolysis & PFK

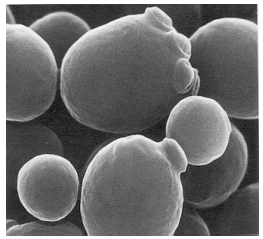




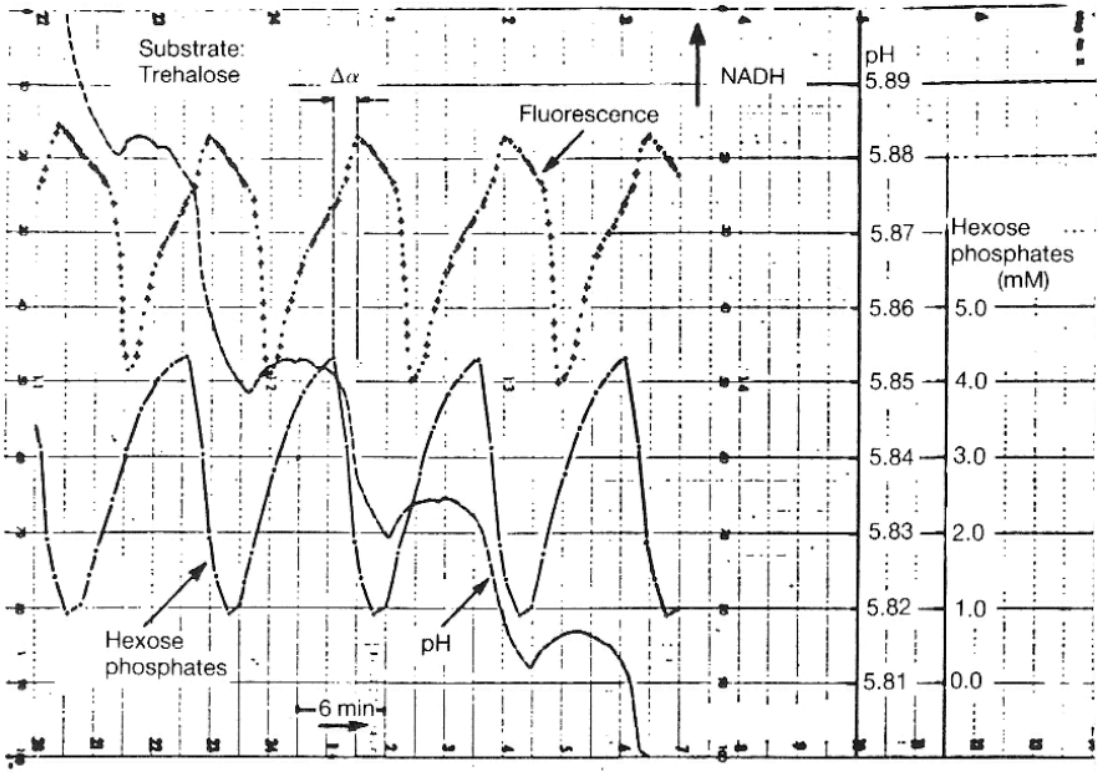
Donc, si PFK est active, le rapport [ATP]/[AMP] diminue. En conséquence, son activité augmente ----->
BOUCLE DE RETRO-ACTION POSITIVE

Glycolytic oscillations in yeast

Glycolytic oscillations in *Saccharomyces cerevisiae*



S. cerevisiae **(yeast)**



Glycolytic oscillations in a yeast extract subjected to constant injection of the substrate (trehalose). Chemical analyses show that the various hexoses oscillate with the same frequency as NADH.

Hess & Boiteux (1968) In Regulatory Functions of Biological Membranes. Ed. J. Jarnefelt, Elsevier.
Hess B, Boiteux A, Krüger J (1969) Cooperation of glycolytic enzymes. Adv Enzyme Regul. 7:149-67

Glycolytic oscillations in yeast

TABLE 1

RANGE OF GLYCOLYTIC OSCILLATION IN YEAST EXTRACT

Input rate* mM/hr	Period min	Amplitude in mM NADH	Damping	Waveform
< 20	—	steady high level of NADH	—	—
20	8.6	0.2–0.4	—	double periodicities, nonsinusoidal
40	6.5	0.6	—	nonsinus-sinus
60–80	5.0	0.3	—	stabile sinus
120	3.5	0.2	—	stabile sinus
> 160	—	steady low level of NADH	+++	

* Fructose or glucose serve as substrates. Cell-free extract of ~60 mg/ml.

Hess B, Boiteux A, Krüger J (1969) Cooperation of glycolytic enzymes. Adv Enzyme Regul. 7:149-67

Glycolytic oscillations in yeast

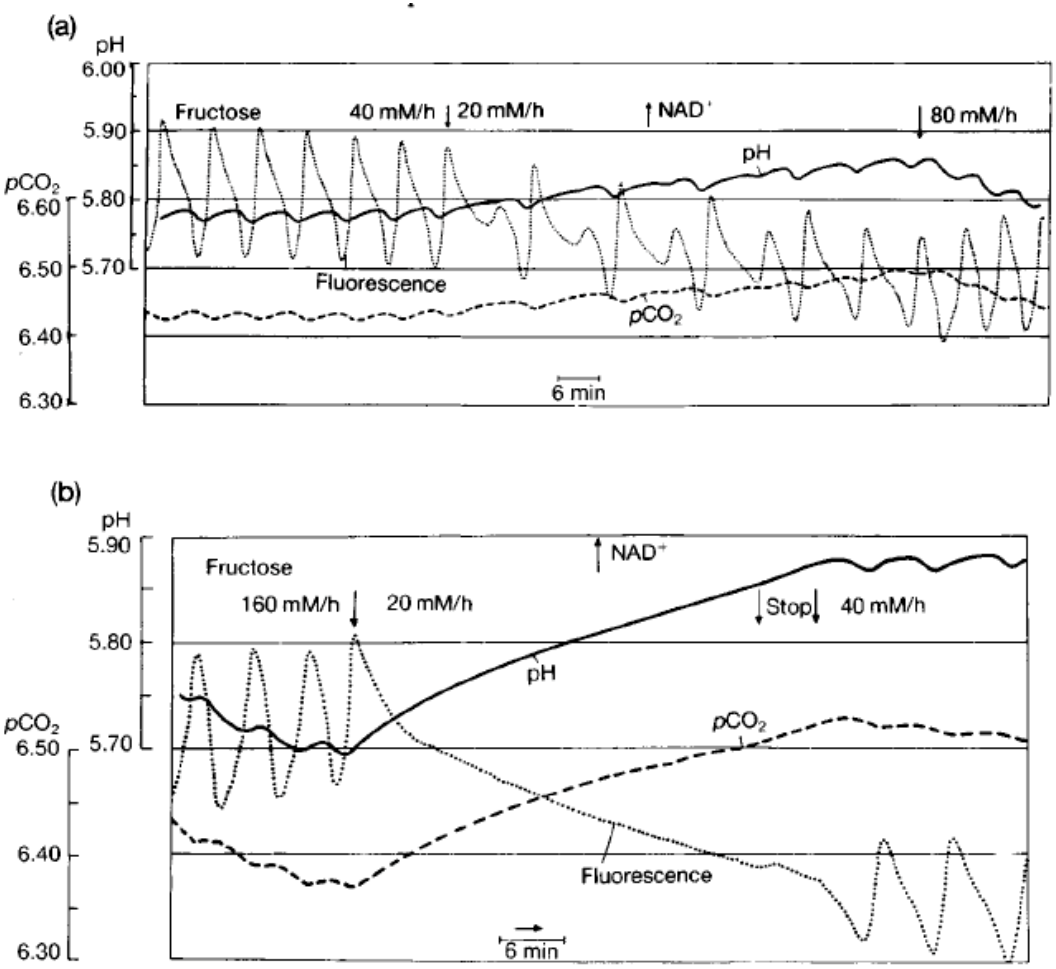


Fig. 2.4. Control of glycolytic oscillations in yeast extracts by the substrate injection rate. (a) The diminution of the rate of injection of fructose from 40 to 20 mM/h causes a lengthening of the period as well as a change in the waveform of oscillations; this change is reversible. (b) Decreasing the injection rate below 20 mM/h causes the reversible suppression of the oscillations (Hess & Boiteux, 1968b).

Control of glycolytic oscillations by the substrate injection rate:

(a) The diminution of the injection rate causes a lengthening of the period

(b) Decreasing the injection rate below a certain threshold causes the reversible suppression of the oscillations

Hess B, Boiteux A (1968) Hoppe Seylers Z Physiol Chem. 349:1567-74.

Glycolytic oscillations in yeast

Control of glycolytic oscillations by the substrate injection rate:

The successive decreases in the glucose injection rate cause the progressive lengthening of the period of the oscillations

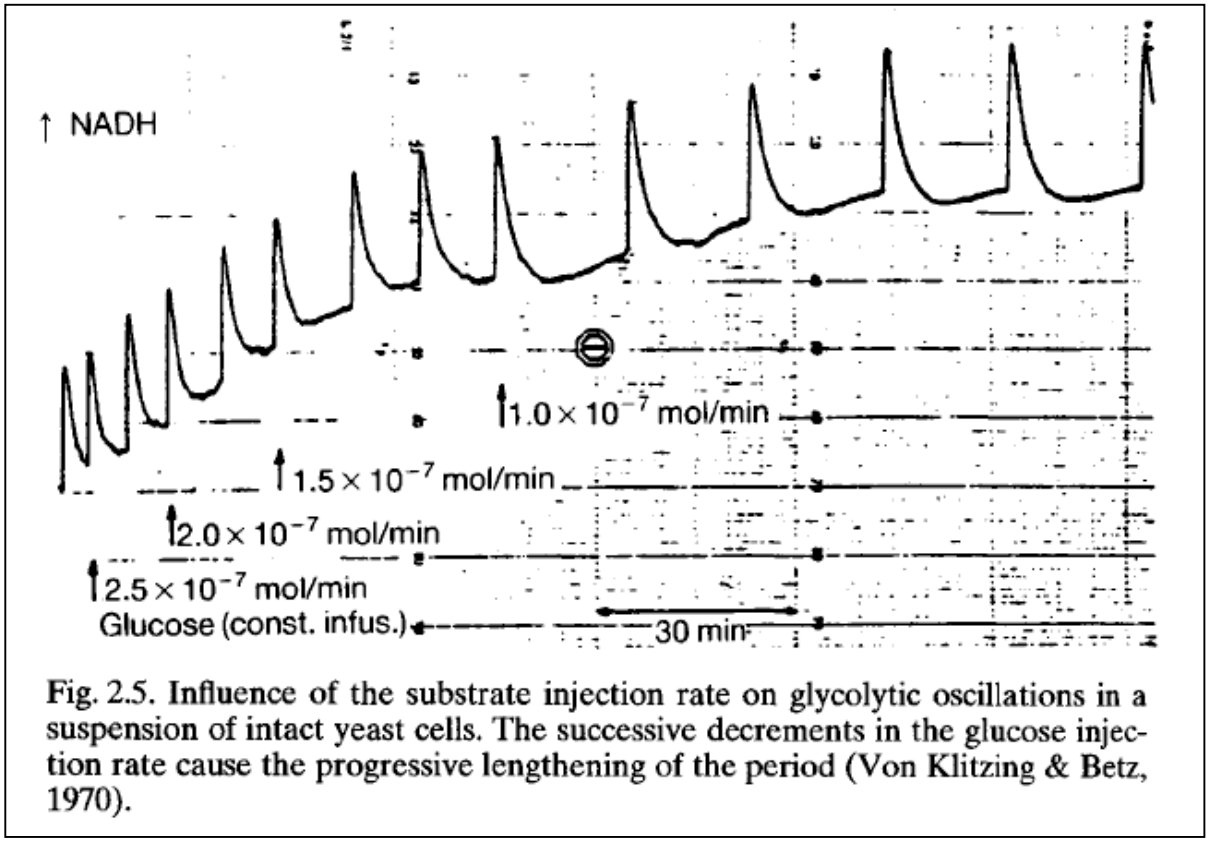


Fig. 2.5. Influence of the substrate injection rate on glycolytic oscillations in a suspension of intact yeast cells. The successive decrements in the glucose injection rate cause the progressive lengthening of the period (Von Klitzing & Betz, 1970).

von Klitzing L, Betz A (1970) Arch Mikrobiol. 71:220-5.

Glycolytic oscillations in yeast

PFK is responsible for the glycolytic oscillations.

Indeed:

- If glucose-6-phosphate or fuctcose-6-phosphate is taken as substrate, oscillations are still observed.
- If fructose 1,6-bis-phosphate is used as substrate, there is no oscillations.
- Ion NH_4^+, which actives PFK, inhibits the oscillations
- Citrate, which inhibits PFK, inhibits the oscillations
- Amplitude and frequency of the oscillations can be varied by adding purified PFK to the cultured yeast cells.

Origin of the glycolytic oscillations?

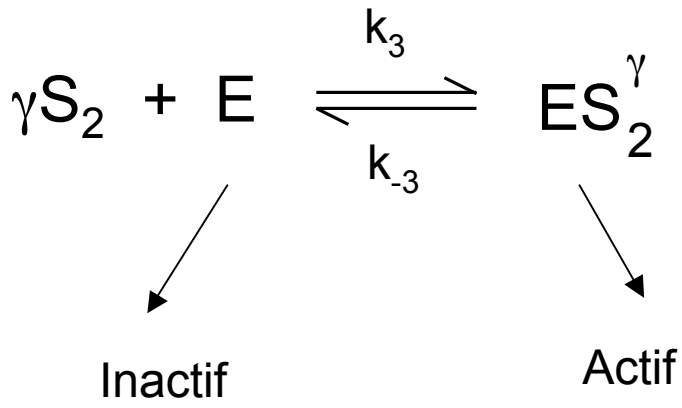
Can we explain the existence of glycolytic oscillations from our knowledge of PFK regulation?

=> Insights from mathematical models

Selkov model (1968)

First model : Selkov (1968)

Hypothesis: activity of PFK is stimulated by 1 or several ADP molecules



Sel'kov (1968) Self-oscillations in Glycolysis, *Eur J Biochem* 4: 79-86.

Selkov model (1968)

$\xrightarrow{v_1} S_1$

$\gamma S_2 + E \underset{k_{-3}}{\overset{k_3}{\rightleftharpoons}} ES_2^{\gamma}$

$S_1 + ES_2^{\gamma} \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} S_1ES_2^{\gamma} \xrightarrow{k_2} ES_2^{\gamma} + S_2$

$S_2 \xrightarrow{v_2}$

S_1 = ATP
S_2 = ADP
E = PFK

Evolution equations (s_1 = [ATP] ; s_2 = [ADP] ; e = [E] ; x_1 = [ES_2^{γ}] ; x_2 = [$S_1ES_2^{\gamma}$])

$$\begin{cases} \frac{ds_1}{dt} &= v_1 - k_1 s_1 x_1 + k_{-1} x_2 \\ \frac{ds_2}{dt} &= k_2 x_2 - k_3 s_2^{\gamma} e + k_{-3} x_1 - v_2 s_2 \\ \frac{dx_1}{dt} &= -k_1 s_1 x_1 + (k_{-1} + k_2) x_2 + k_3 s_2^{\gamma} e - k_{-3} x_1 \\ \frac{dx_2}{dt} &= k_1 s_1 x_1 - (k_{-1} + k_2) x_2 \end{cases}$$

with:

$$e_0 = e + x_1 + x_2$$

(total concentration in PFK)

Selkov model (1968)

Adimensionalisation

$$u_1 = \frac{x_1}{e_0} \qquad u_2 = \frac{x_2}{e_0} \qquad \sigma_1 = \frac{k_1 s_1}{k_{-1} + k_2} \qquad \sigma_2 = \left(\frac{k_3}{k_{-3}}\right)^{1/\gamma} s_2$$

$$\tau = \frac{e_0 k_1 k_2}{k_{-1} + k_2} t \qquad u_1 + u_2 + \frac{e}{e_0} = 1 \text{ (enzyme conservation)}$$

$$\begin{cases} \dfrac{d\sigma_1}{dt} &= v - \dfrac{k_{-1} + k_2}{k_2} u_1 \sigma_1 + \dfrac{k_{-1}}{k_2} u_2 \\ \dfrac{d\sigma_2}{dt} &= \alpha \left(u_2 - \dfrac{k_{-3}}{k_2} \sigma_2^\gamma (1 - u_1 - u_2) + \dfrac{k_{-3}}{k_2} u_1 \right) - \eta \sigma_2 \\ \epsilon \dfrac{du_1}{dt} &= u_2 - \sigma_1 u_1 + \dfrac{k_{-3}}{k_{-1} + k_2} \left(\sigma_2^\gamma (1 - u_1 - u_2) - u_1 \right) \\ \epsilon \dfrac{du_2}{dt} &= \sigma_1 u_1 - u_2 \end{cases}$$

where $\epsilon = \dfrac{e_0 k_1 k_2}{(k_2 + k_{-1})^2}$ $\quad v = \dfrac{v_1}{k_2 e_0}$ $\quad \eta = \dfrac{v_2 (k_{-1} + k_2)}{e_0 k_1 k_2}$ $\quad \alpha = \dfrac{k_{-1} + k_2}{k_1} \left(\dfrac{k_3}{k_{-3}} \right)^{1/\gamma}$

Selkov model (1968)

Quasi steady state hypothesis on u_1 and u_2 (ε small):

$$\varepsilon \frac{du_1}{d\tau} = u_2 - \sigma_1 u_1 + \frac{k_{-3}}{k_2 + k_{-1}} \left\{ \sigma_2^{\gamma} (1 - u_1 - u_2) - u_1 \right\} \approx 0$$

$$\varepsilon \frac{du_2}{dt} = \sigma_1 u_1 - u_2 \approx 0$$

$$\Rightarrow u_2 = \sigma_1 u_1 \quad ; \qquad u_2 = \frac{\sigma_1 \sigma_2^{\gamma}}{\sigma_1 \sigma_2^{\gamma} + \sigma_2^{\gamma} + 1} = f(\sigma_1, \sigma_2) \leq 1$$

Hence:

$$\begin{cases} \dfrac{d\sigma_1}{d\tau} = v - f(\sigma_1, \sigma_2) \\ \dfrac{d\sigma_2}{d\tau} = \alpha f(\sigma_1, \sigma_2) - \eta \sigma_2 \end{cases}$$

NB: Since f<1, to avoid explosion of σ_1, we impose 0<v<1

Selkov model (1968)

Analysis of the 2-equation system (for σ_1 et σ_2) in the phase space

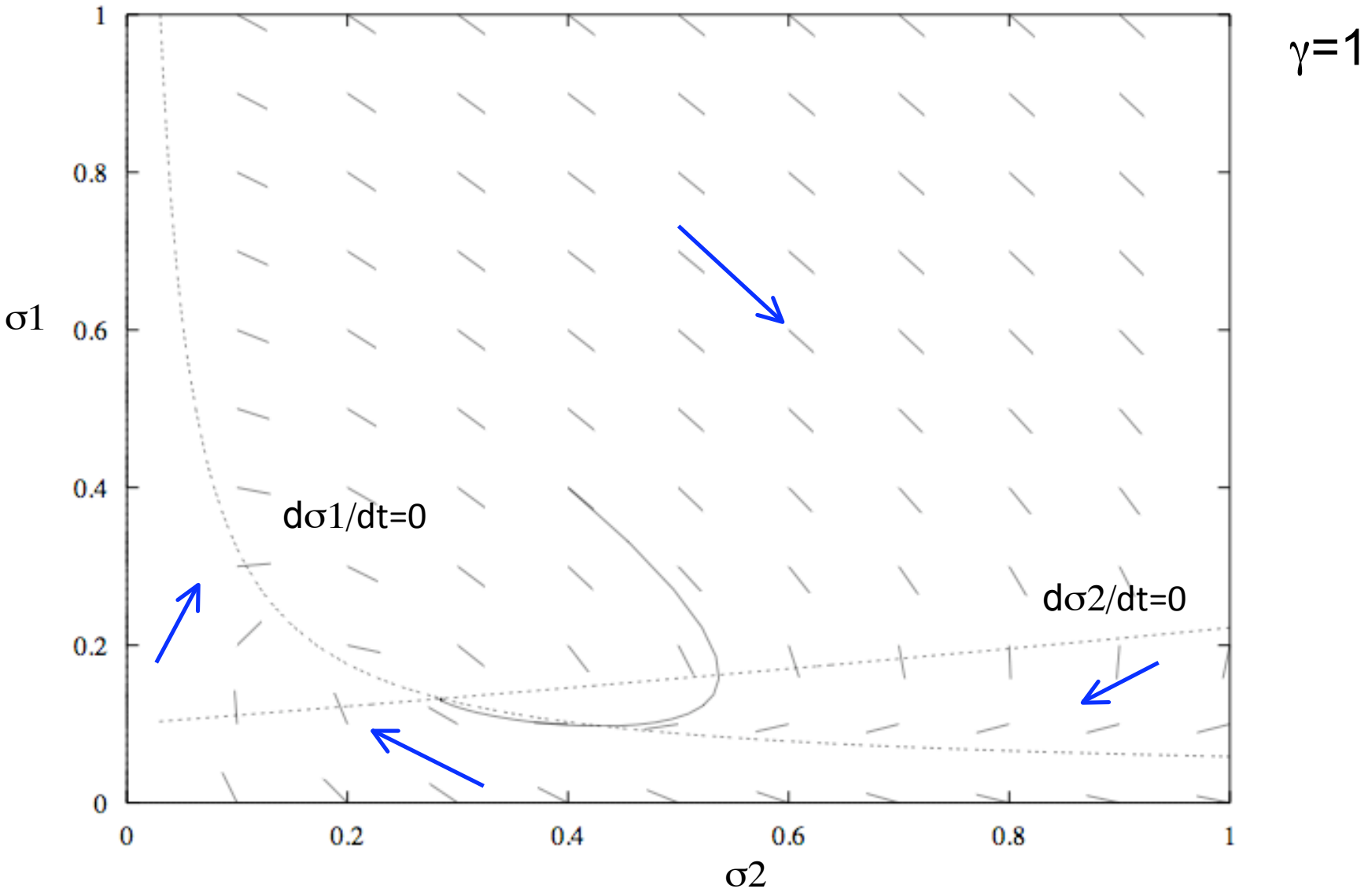
Nullclines:

(1) $v = f(\sigma_1, \sigma_2)$ Hence: $\sigma_1 = \frac{v}{1-v} \frac{1+\sigma_2^\gamma}{\sigma_2^\gamma}$

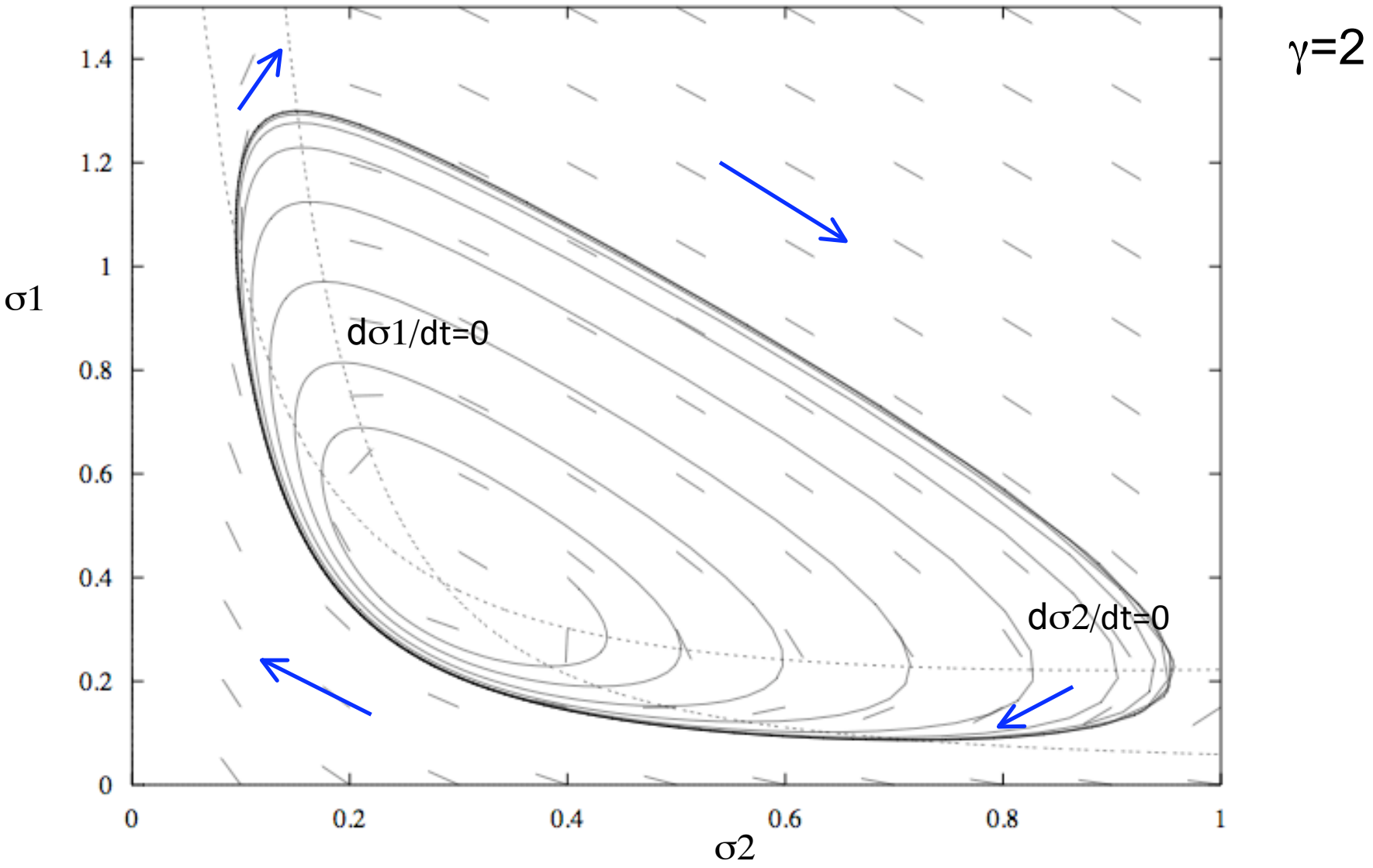
(2) $\alpha f(\sigma_1, \sigma_2) = \eta \sigma_2$ Hence: $\sigma_1 = \frac{1+\sigma_2^\gamma}{\sigma_2^{\gamma-1}(p-\sigma_2)}$ where $p = \frac{\alpha}{\eta}$

The intersection of the nullclines defines the steady state: $\sigma_1^{SS}, \sigma_2^{SS}$

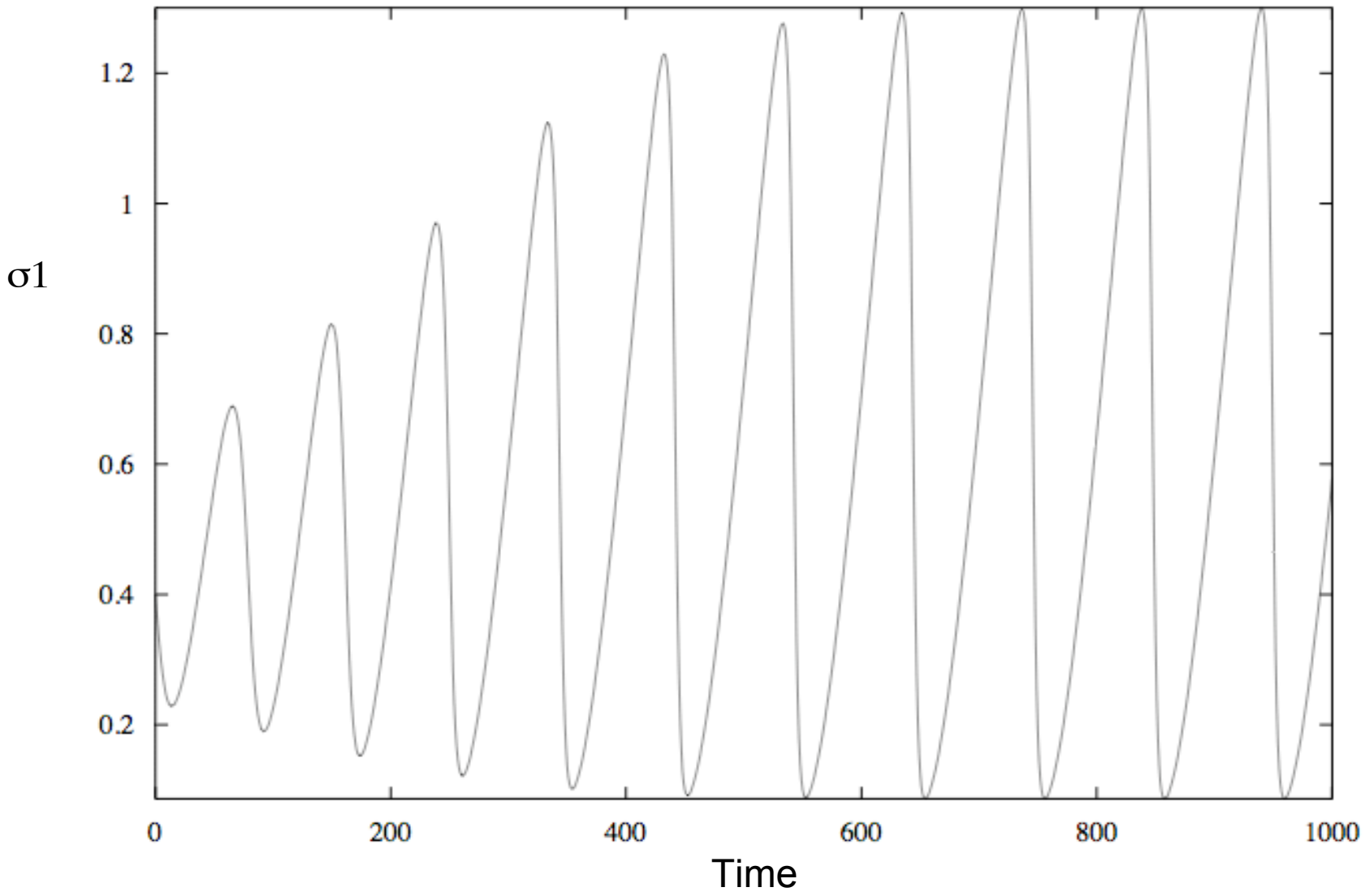
Selkov model (1968)



Selkov model (1968)



Selkov model (1968)



Selkov model (1968)

Characteristic equation

$$\omega^2 - T\omega + D = 0 \qquad \text{avec} \qquad T = -\frac{\partial f}{\partial \sigma_1} + \alpha \frac{\partial f}{\partial \sigma_2} - \eta \qquad \text{et} \qquad D = \eta \frac{\partial f}{\partial \sigma_1}$$

$$\Rightarrow \omega^2 + \left(\frac{\partial f}{\partial \sigma_1} - \alpha \frac{\partial f}{\partial \sigma_2} + \eta \right) \omega + \eta \frac{\partial f}{\partial \sigma_1} = 0$$

Hence, the instability condition reads:

$$\frac{\partial f}{\partial \sigma_1} - \alpha \frac{\partial f}{\partial \sigma_2} + \eta < 0 \qquad \text{ou} \qquad A = \frac{1 - v}{\left(1 + (pv)^\gamma\right)} \left(\eta\gamma + (v-1)(pv)^\gamma \right) - \eta > 0$$

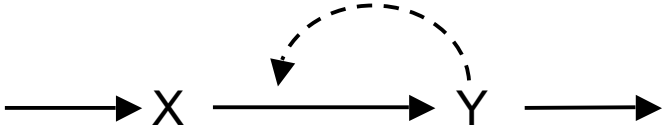
Si $v = 0 \rightarrow A = \eta(\gamma - 1)$ Si $v = 1 \rightarrow A = -\eta$

---->Il y a **possibilité d'instabilité si v petit et $\gamma > 1$**

Remarque : pour le modèle de Selkov, si l'état stationnaire est instable, il y aura toujours des oscillations.

Schnakenberg model (1979)

A variant of the Selkov model: the Schnakenberg model



$$\rightarrow \mathrm{X}$$

$$\mathrm{X} + 2\mathrm{Y} \rightarrow 3\mathrm{Y}$$

$$\mathrm{Y} \rightarrow$$

$$\frac{dx}{dt} = v - k_1 x y^2$$

$$\frac{dy}{dt} = k_1 x y^2 - k_2 y$$

It can be shown that under some conditions, the steady state can be unstable. However, using the Poincaré-Bendixson theorem, it is possible to show that the system does not display limit-cycle oscillations.

Schnakenberg (1979) Simple chemical reaction systems with limit cycle behaviour, *J. Theor Biol* 81:389-400

Schnakenberg model

A variant of the Selkov model: the Schnakenberg model

In order to allow limit-cycle oscillations, Schnakenberg introduces the following modification:

$$\rightarrow X$$

$$X + 2Y \rightarrow 3Y$$

$$Y \rightleftarrows B$$

Exercise

- *How should the kinetic equations be changed?*
- *Calculate the steady state and determine its stability.*
- *Draw the nullclines in the phase space and use the Poincaré-Bendixson theorem to show that the system can display limit cycle oscillations.*
- *Simulate the system and show the oscillations.*

Allosteric model (Goldbeter-Lefever)

Second model: Goldbeter-Lefever (1972)

L'enzyme étudiée par Monod, Wyman & Changeux pour établir leur modèle est la phosphofructokinase, enzyme tetramerique constituee de 4 sous-unites identiques.

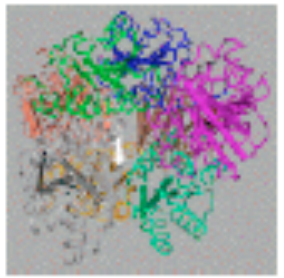
Les deux états conformationnels que peut adopter un protomère sont denommes respectivement :

- T (pour "*Tense*"), conformation "*tendue*", les sites de fixation ont une affinite faible pour un ligand donne ;
- R (pour "*Relax*"), conformation "*relâchée*", les sites de fixation ont une affinite forte pour un ligand donne;

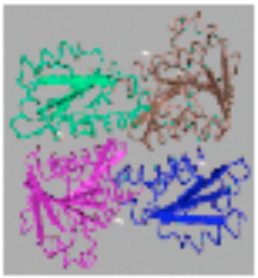
Ces deux conformations sont en équilibre (quelle que soit la concentration d'un ligand donné) et cet equilibre est regit par la constante allosterique, L :

$$T_0 \overset{L}{\Longleftrightarrow} R_0$$

structure phosphofructokinase dans l'état T



structure phosphofructokinase dans l'état R



Hypothèse supplémentaire: seule la forme R peut lier l'effecteur (ADP)

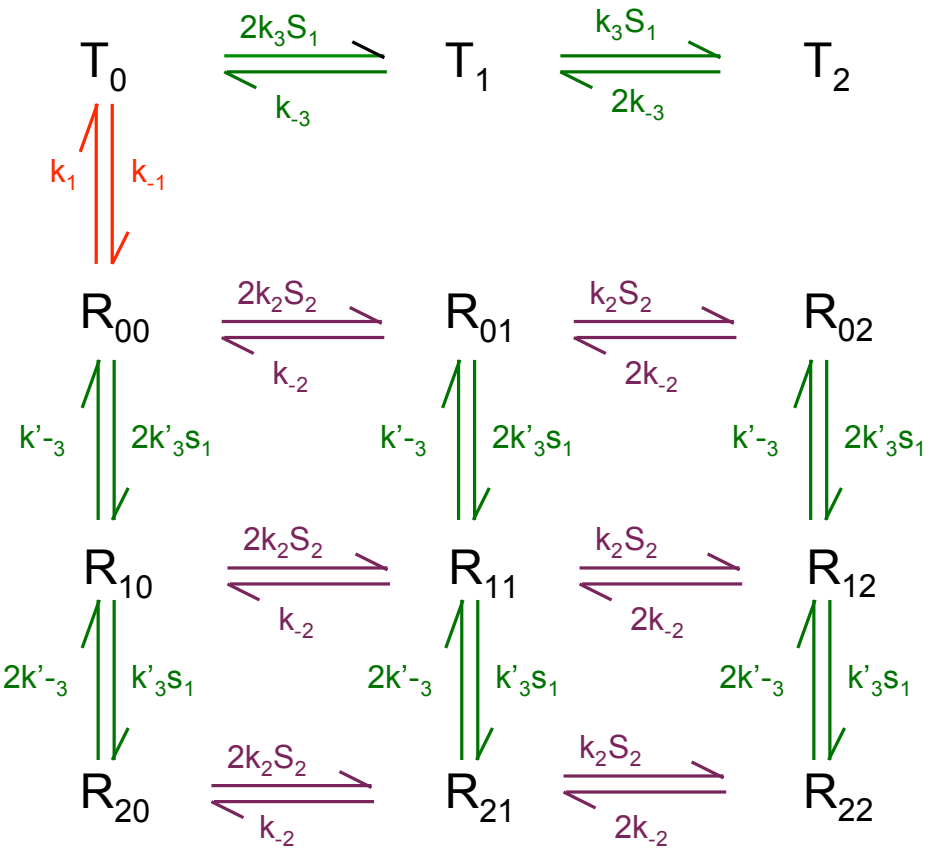
Allosteric model (Goldbeter-Lefever)

PFK : enzyme allostérique

Dimère R_{ij} ⇌ Dimère T_i

Actif
Peut fixer 2 ATP (S_1, index i)
Peut fixer 2ADP (S_2, index j)

Inactif
Peut fixer 2 ATP (S_1, index i)



Allosteric model for the PFK

An allosteric enzyme binds both the *substrate* to convert it into the product and an *effector* that affect the activity of the enzyme, either by activating or by inhibiting the reaction.

In the case of the PFK, the effector is the product of the reaction itself (ATP) and it acts as an activator. In addition, the PFK has several binding sites for the substrate and for the effector (product) that interact *cooperatively*.

Allosteric model (Goldbeter-Lefever)

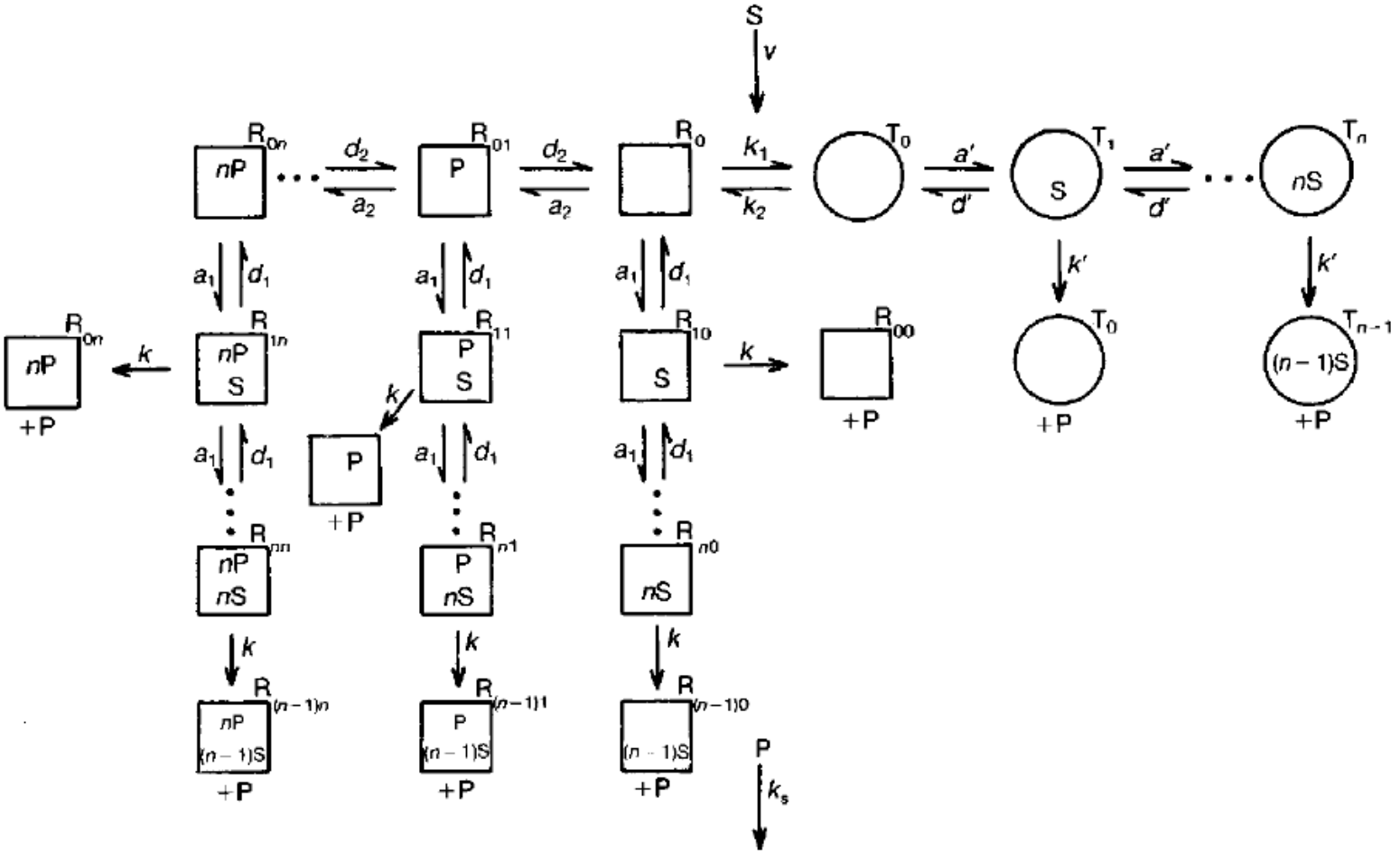


Fig. 2.11. Allosteric model for glycolytic oscillations. The enzyme is formed by n subunits existing in the states R and T. The substrate (S), injected at a constant rate, binds to the two forms of the enzyme with different affinities. The complexes thus formed in the two states decompose with different rates to yield the product (P). The latter binds in an exclusive manner to the the most active, R, form of the enzyme, and disappears from the reaction medium in an apparent first-order reaction (Goldbeter & Lefever, 1972; Venieratos & Goldbeter, 1979; Goldbeter, 1980).

Goldbeter & Lefever (1972) *Biophys. J.* 4: 79-86.

Allosteric model (Goldbeter-Lefever)

$$\frac{dR_0}{dt} = -k_1 R_0 + k_2 T_0 - n a_2 P R_0 + d_2 R_{01} - n a_1 S R_0 + (d_1 + k)\, R_{10}$$

$$\vdots$$

$$\frac{dR_{0n}}{dt} = a_2 P R_{0(n-1)} - n d_2 R_{0n} - n a_1 S R_{0n} + (d_1 + k)\, R_{1n}$$

$$\vdots$$

$$\frac{dR_{nn}}{dt} = a_1 S R_{(n-1)n} - n\,(d_1 + k)\, R_{nn}$$

$$\frac{dT_0}{dt} = k_1 R_0 - k_2 T_0 - n\, a' S T_0 + (d' + k')\, T_1$$

$$\vdots$$

$$\begin{aligned}\frac{dS}{dt} = {} & v_i - n\, a_1 S \Sigma_0 - (n-1)\, a_1 S \Sigma_1 - \ldots - a_1 S \Sigma_{n-1} + d_1 \Sigma_1 + 2 d_1 \Sigma_2 + \ldots \\ & + n\, d_1 \Sigma_n - n\, a' S T_0 - (n-1)\, a' S T_1 - \ldots \\ & - a' S T_{n-1} + d' T_1 + 2 d' T_2 + \ldots + n\, d' T_n\end{aligned}$$

$$\begin{aligned}\frac{dP}{dt} = {} & -n\, a_2 P R_0 - (n-1)\, a_2 P R_{01} - \ldots - a_2 P R_{0(n-1)} + d_2 R_{01} + 2 d_2 R_{02} + \ldots \\ & + n\, d_2 R_{0n} + k \Sigma_1 + 2\, k\, \Sigma_2 + \ldots + n\, k\, \Sigma_n + k' T_1 + 2\, k' T_2 + \ldots \\ & + n\, k' T_n - k_s P\end{aligned}$$

with the conservation relation:

$$R_0 + R_{ij} + T_0 + T_i = D_0 \,(i,j = 1, \ldots n)$$

12 evolution equations for the various enzyme forms (n=2)
+ 1 conservation relation

Hypothesis: enzyme forms vary faster than the concentrations in substrate and products
=> quasi-stationarity hypothesis

Normalized concentrations:

$$\alpha = S/K_R,\ \gamma = P/K_P$$

$$K_R = d_1/a_1,\ K_P = d_2/a_2$$

Analytical expression for R_0:

$$R_0 = \frac{D_0}{L\,(1 + \alpha c e')^n + (1 + \alpha e)^n\,(1 + \gamma)^n}$$

Allosteric model (Goldbeter-Lefever)

After some simplifications and rearrangements, we obtain:

$$\frac{d\alpha}{dt} = v - \sigma\phi = f(\alpha, \gamma)$$ (substrate)

$$\frac{d\gamma}{dt} = q\sigma\phi - k_s\gamma = g(\alpha, \gamma)$$ (product)

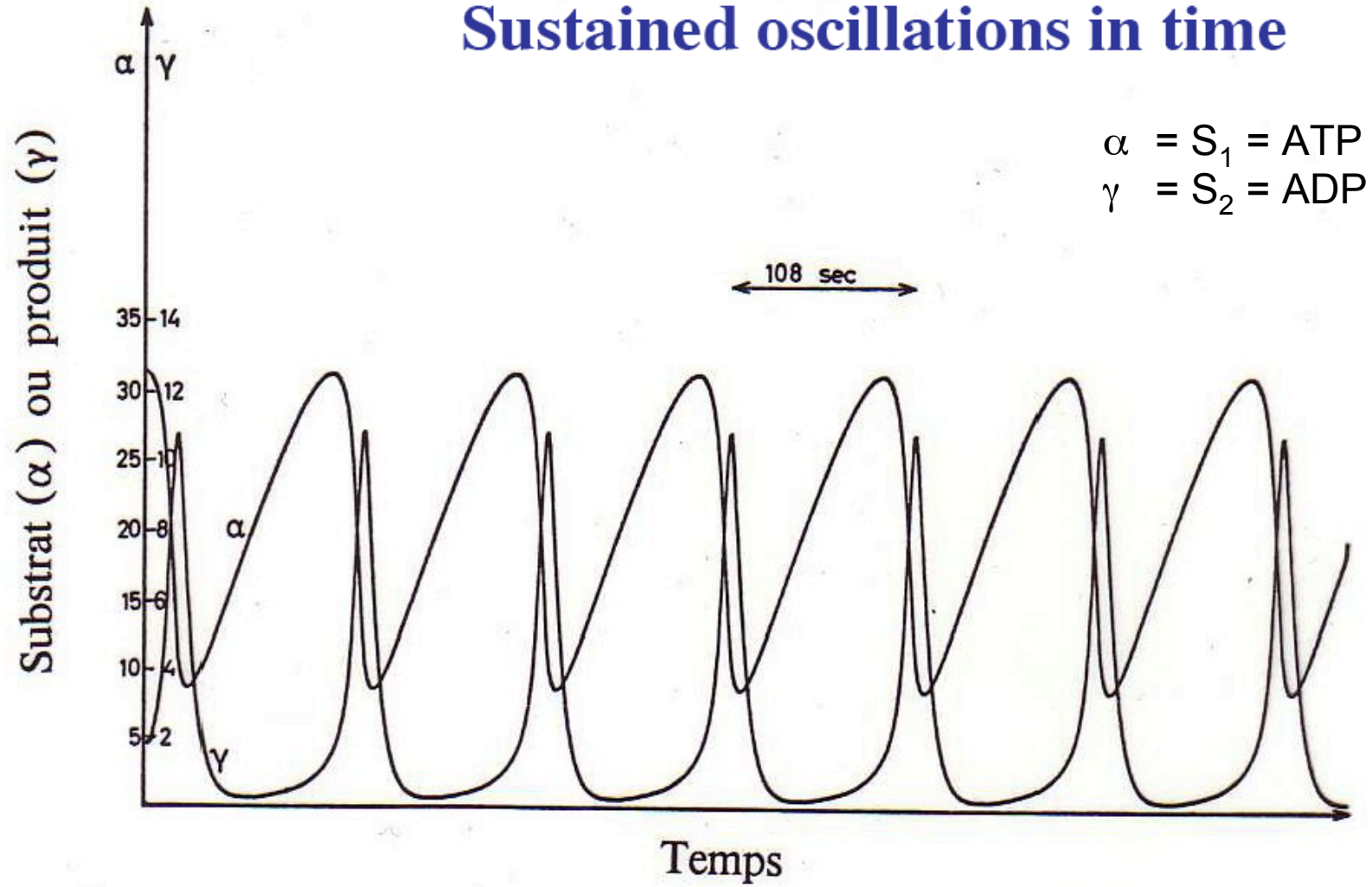
$$\phi = \frac{\alpha(1+\alpha)(1+\gamma)^2}{L + (1+\alpha)^2(1+\gamma)^2}$$

Exercise

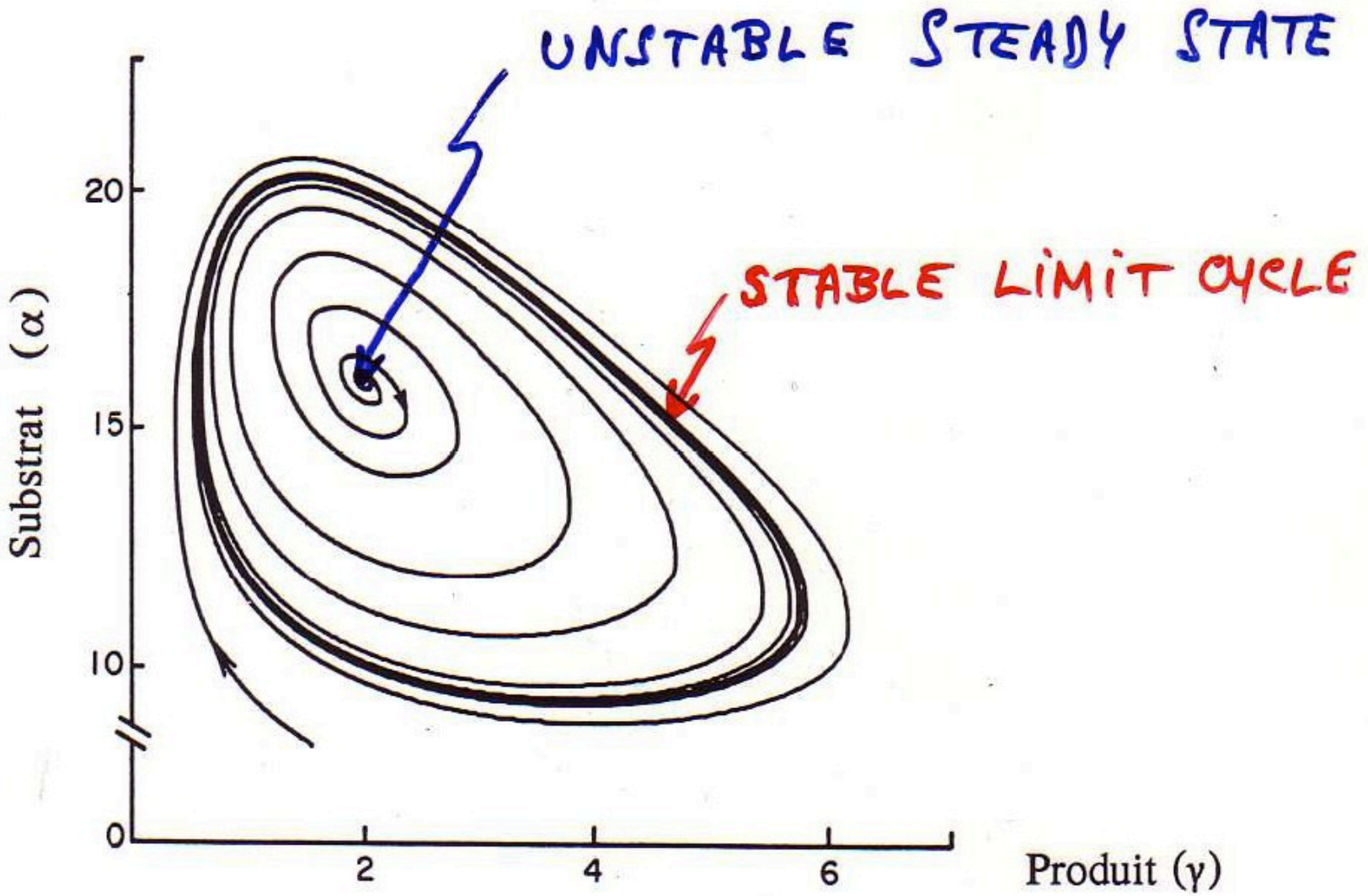
- *Simulate the system (e.g. with XPP) and show that the oscillations occurs for a bounded range of values of v.*
- *Show how the period and amplitude of the oscillations vary as a function of v.*

Goldbeter & Lefever (1972)
Biophys. J. 4: 79-86.

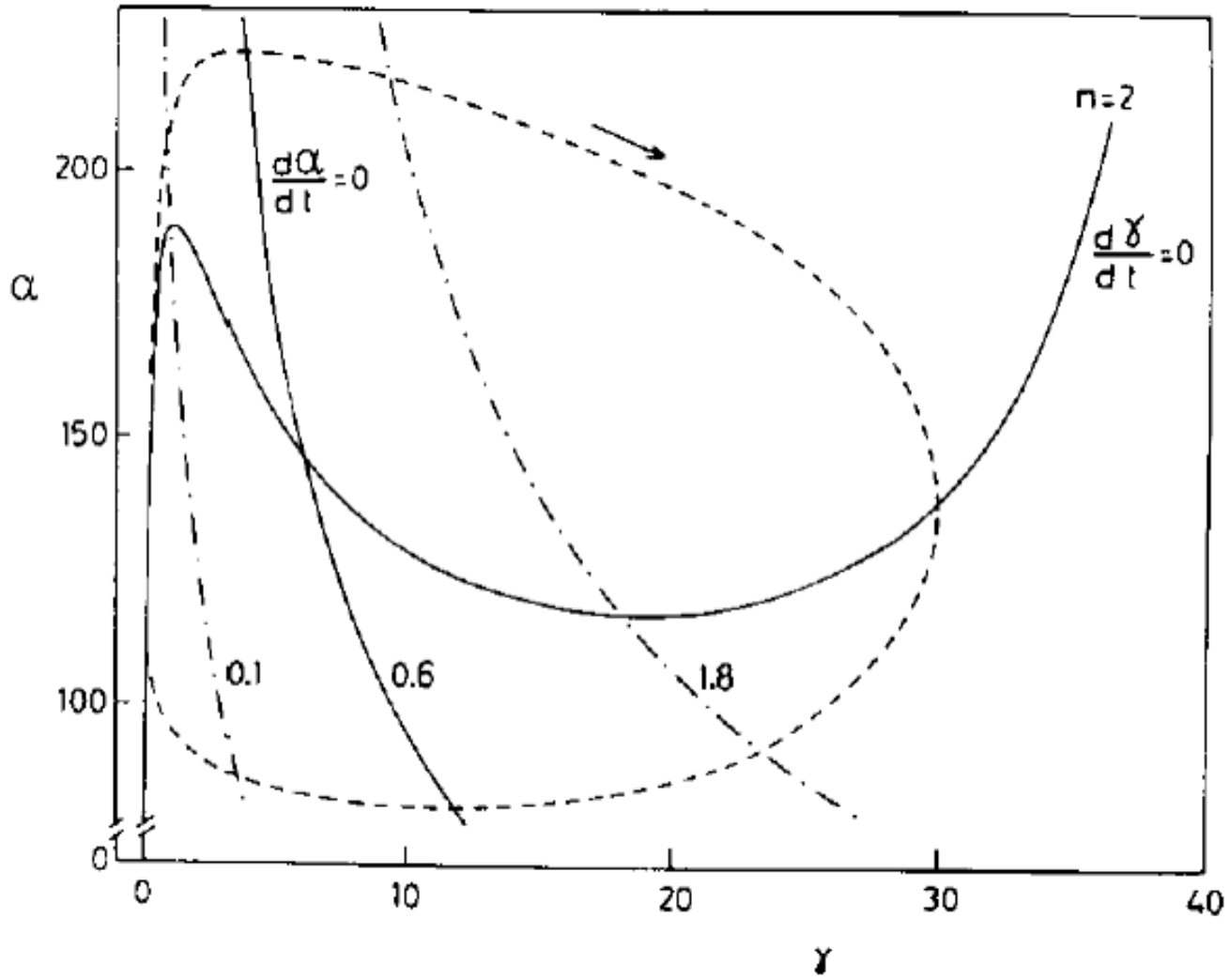
Allosteric model (Goldbeter-Lefever)



Allosteric model (Goldbeter-Lefever)

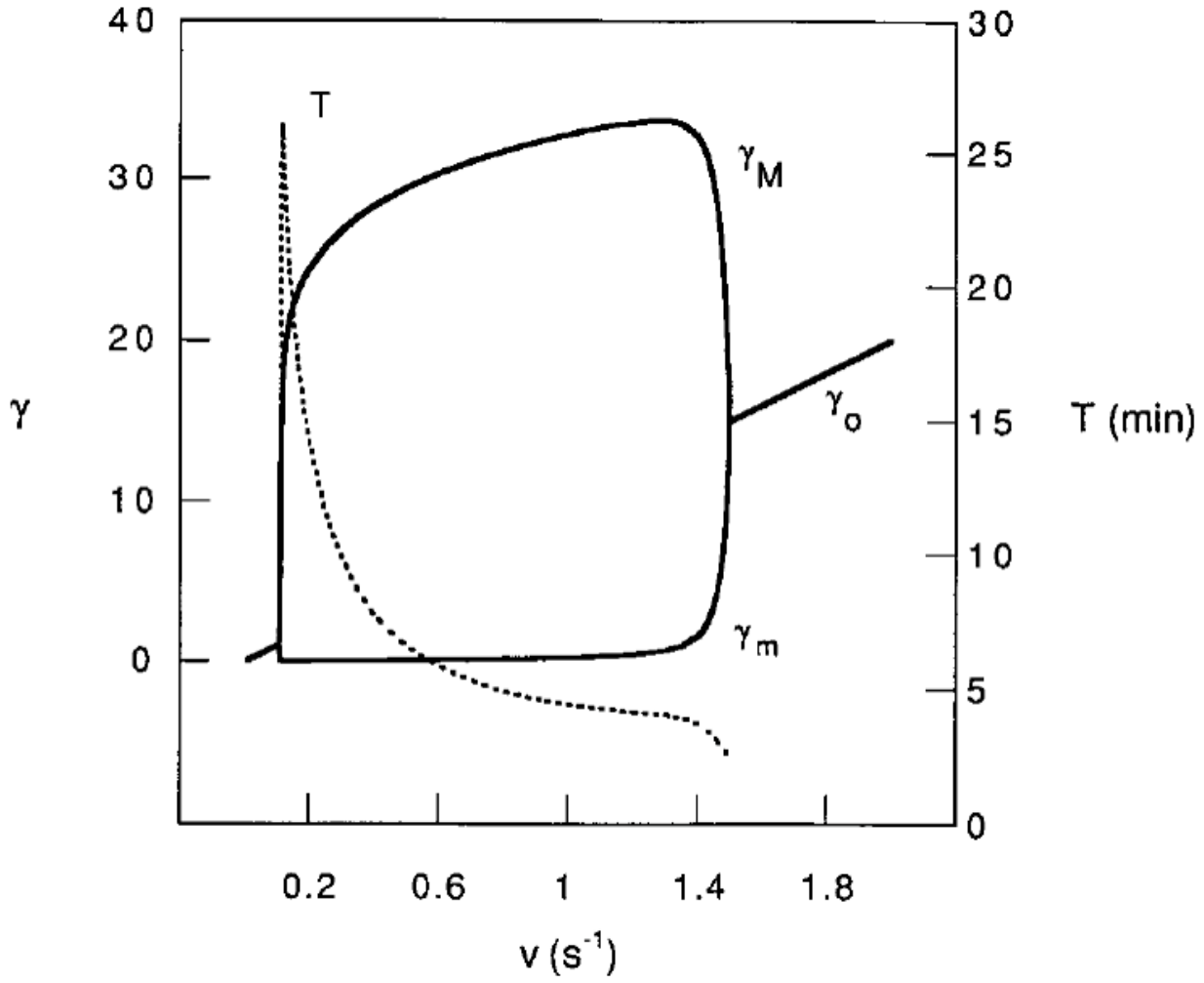


Allosteric model (Goldbeter-Lefever)



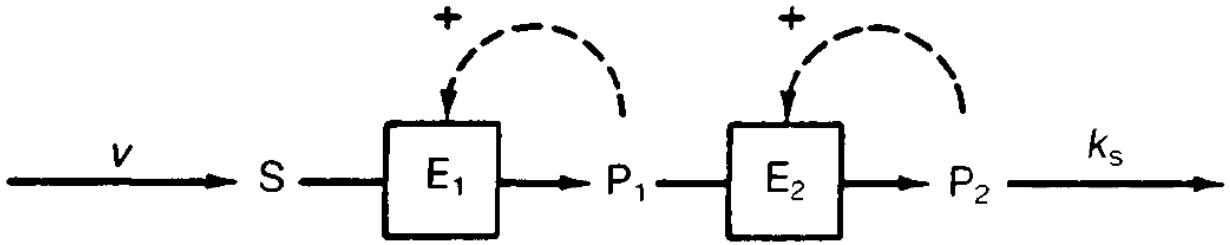
Allosteric model (Goldbeter-Lefever)

Bifurcation diagram for glycolytic oscillations as a function of the substrate input rate, v



Allosteric model (Decroly-Goldbeter)

A 3-variable biochemical model for the coupling in series of 2 enzyme reactions with autocatalytic regulation



$$\frac{d\alpha}{dt} = \nu - \sigma_1 \, \phi \, (\alpha, \beta)$$

$$\frac{d\beta}{dt} = q_1 \, \sigma_1 \, \phi \, (\alpha, \beta) - \sigma_2 \, \eta \, (\beta, \gamma)$$

$$\frac{d\gamma}{dt} = q_2 \, \sigma_2 \, \eta \, (\beta, \gamma) - k_s \, \gamma$$

with

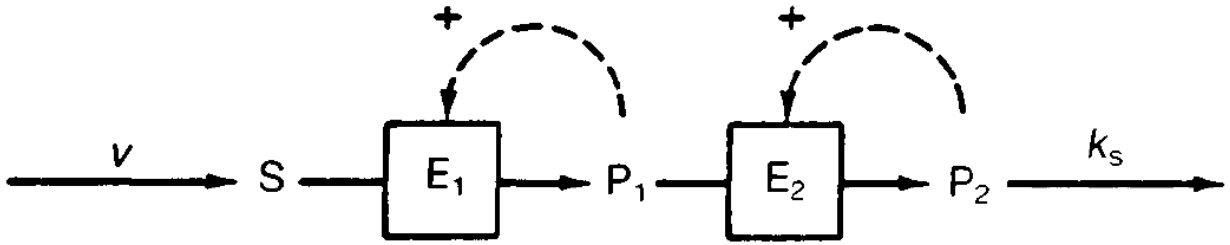
$$\phi \, (\alpha, \beta) = \frac{\alpha \, (1+\alpha) \, (1+\beta)^2}{L_1 + (1+\alpha)^2 \, (1+\beta)^2}$$

$$\eta \, (\beta, \gamma) = \frac{\beta \, (1+\gamma)^2}{L_2 + (1+\gamma)^2}$$

Decroly O, Goldbeter A (1982) Birhythmicity, chaos, and other patterns of temporal self-organization in a multiply regulated biochemical system. Proc Natl Acad Sci USA 9:6917-21.

Allosteric model (Decroly-Goldbeter)

A 3-variable biochemical model for the coupling in series of 2 enzyme reactions with autocatalytic regulation



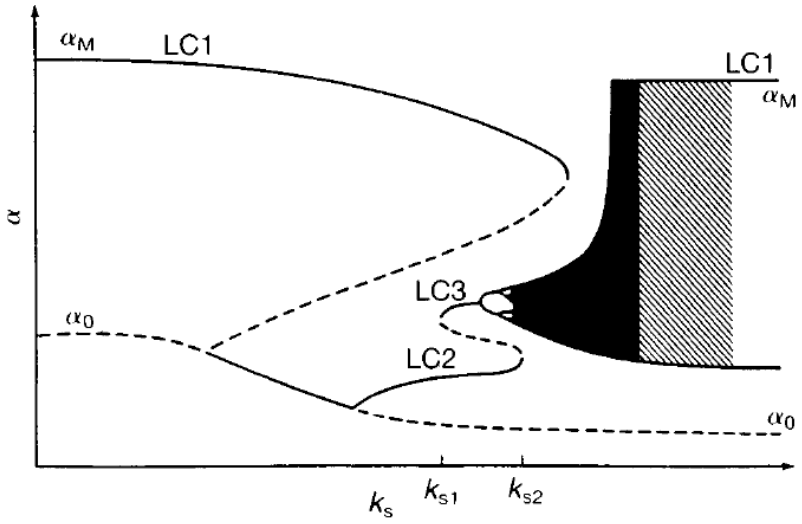
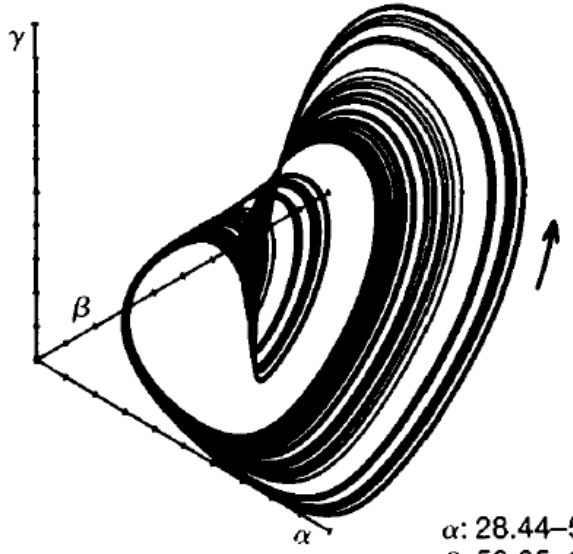


Fig. 4.6. Schematic bifurcation diagram showing the origin of trirhythmicity. For certain values of k_s, the three stable limit cycles LC1, LC2 and LC3 coexist; they are separated by unstable limit cycles. The black and the dashed domains correspond to chaos and bursting, respectively (Decroly & Goldbeter, 1985).



α: 28.44–50.6
β: 50.05–351.1
γ: 0.05–2.28

Physiological significance

What about the physiological significance of glycolytic oscillations ?

Physiological significance

- In mammals (incl human), insulin is secreted in a pulsatile manner by Beta cells of the pancreas.

  Lang et al (1979) *N. Engl. J. Med.* 301:1023–1027.
  Chou HF & Ipp E (1990) Diabetes 39:112–117
  Song et al (2002) J Clin Endocrinol Metab. 87:213-21.

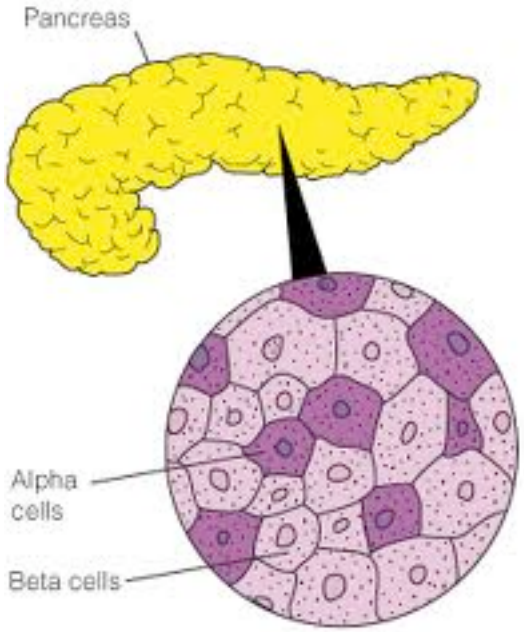
- The loss of oscillatory insulin secretion has been associated to some disease, such as diabete II.

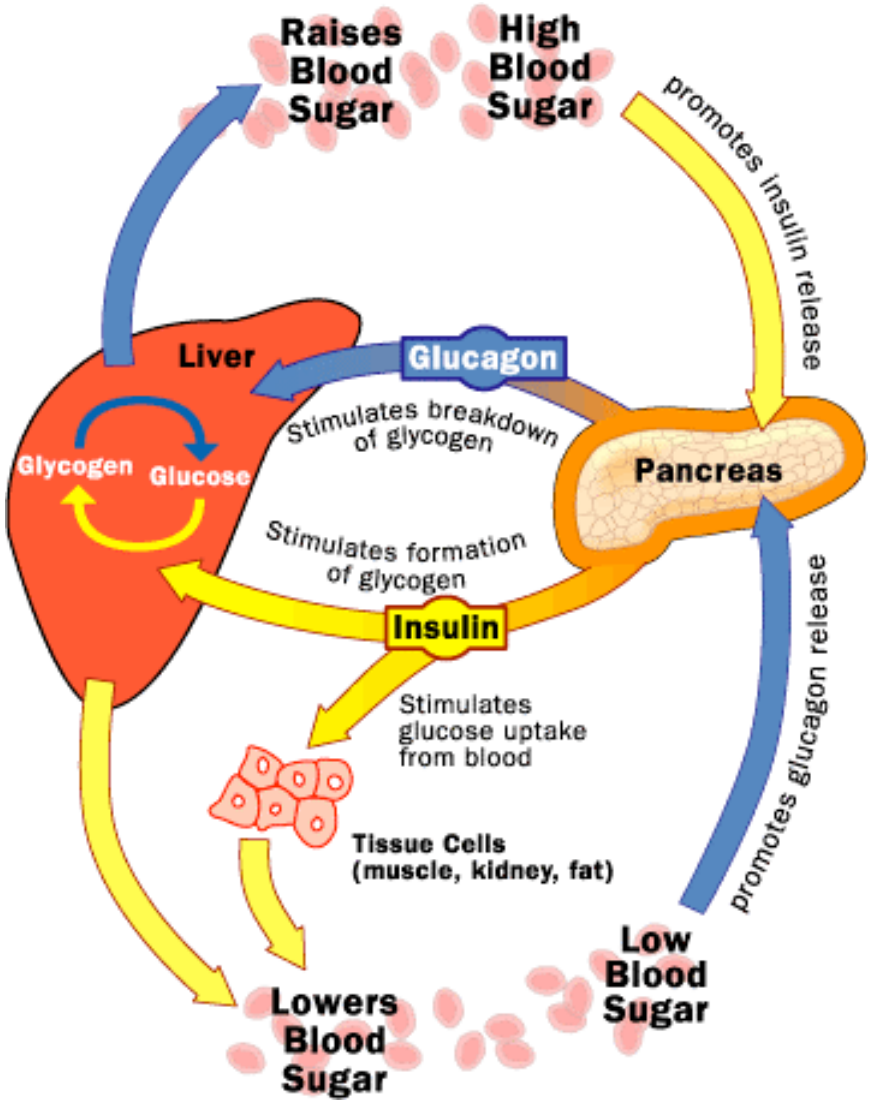
  Polonsky et al (1988) N. Engl. J. Med. 318:1231–1239.

- It has been proposed that oscillations in glycolysis may be the pulse generator for pulsatile insulin release by islets

  Tornheim K (1997) Are metabolic oscillations responsible for normal oscillatory insulin secretion? Diabetes 46:1375–1380
  Chou HF, Berman N, Ipp E (1992) Oscillations of lactate released from islets of Langerhans: evidence for oscillatory glycolysis in beta-cells, Am J Physiol. 262:E800-5

Physiological significance





Insulin, produced by beta cells of the pancreas, regulates the absorption of glucose (from the blood) in the liver

Physiological significance

Pulsatile insulin delivery has greater metabolic effects than continuous hormone administration in man: importance of pulse frequency.

Paolisso G, Scheen AJ, Giugliano D, Sgambato S, Albert A, Varricchio M, D'Onofrio F, Lefèbvre PJ.

J Clin Endocrinol Metab 72:607-15 (1991).

The aim of this study was to see if the greater effect of insulin on hepatic glucose output when insulin is given using 13-min pulses in man remains when the same amount of insulin is delivered using 26-min pulses. The study was performed on nine male healthy volunteers submitted to a 325 min glucose-controlled glucose iv infusion using the Biostator. The endogenous secretion of pancreatic hormones was inhibited by somatostatin. Three experiments were performed in each subject on different days and in random order. In all cases glucagon was replaced (58 ng min-1). The amounts of insulin infused were identical in all instances and were 0.2 mU kg-1 min-1 (continuous), 1.3 mU kg-1 min-1, 2 min on and 11 min off (13-min pulses) or 2.6 mU kg-1 min-1, 2 min on and 24 min off (26-min pulses). Blood glucose levels and glucose infusion rate were monitored continuously by the Biostator, and classic methodology using D-[3-3H] glucose infusion allowed to study glucose turnover. When compared with continuous insulin, 13-min insulin pulses induced a significantly greater inhibition of endogenous glucose production. This effect disappeared when insulin was delivered in 26-min pulses. We conclude that, in man, an adequate pulse frequency is required to allow the appearance of the greater inhibition of pulsatile insulin on endogenous glucose production.

Physiological significance

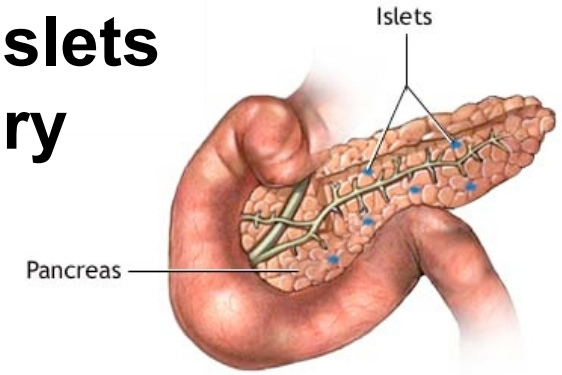
Are metabolic oscillations responsible for normal oscillatory insulin secretion?

Tornheim K.
Diabetes 46:1375-80 (1997).

Normal insulin secretion is oscillatory in vivo and in vitro, with a period of approximately 5-10 min. The mechanism of generating these oscillations is not yet established, but a metabolic basis seems most likely for glucose-stimulated secretion. The rationale is that 1) spontaneous oscillatory operation of glycolysis is a well-established phenomenon; 2) oscillatory behavior of glycolysis involves oscillations in the ATP/ADP ratio, which can cause alternating opening and closing of ATP-sensitive K+ channels, leading to the observed oscillations in membrane potential and Ca2+ influx in pancreatic beta-cells, and may also have downstream effects on exocytosis; 3) spontaneous Ca2+ oscillations are an unlikely basis in this case, since intracellular stores are not of primary importance in the stimulus-secretion coupling, and furthermore, insulin oscillations occur under conditions when intracellular Ca2+ levels are not changing; 4) a neural basis cannot account for insulin oscillations from perifused islets and clonal beta-cells or from transplanted islets or pancreas in vivo; 5) observed oscillations in metabolite levels and fluxes further support a metabolic basis, as does the presence in beta-cells of the oscillatory isoform of phosphofructokinase (PFK-M). The fact that normal oscillatory secretion is impaired in patients with NIDDM and in their near relatives suggests that such derangement may be involved in the development of the disease; furthermore, this probably reflects an early defect in the regulation and operation of the fuel metabolizing/sensing pathways of the pancreatic beta-cell.

Glycolytic oscillations in β-cells

Oscillations of lactate released from islets of Langerhans: evidence for oscillatory glycolysis in beta-cells



Chou HF, Berman N, and Ipp E
Department of Medicine, Harbor-University of California Los Angeles.
Am J Physiol 266: R1786-R1791 (1994).

Oscillations in the glycolytic process have been demonstrated in a number of different biological systems. However, their presence has never been demonstrated in insulin-secreting beta-cells. We used lactate as a marker for glycolysis and measured lactate and insulin concentrations in the effluent of isolated perifused rat islets of Langerhans. Sustained regular oscillations in lactate concentrations with an average period of 16-20 min were observed in islets that were perifused with medium containing 5.5 or 16.7 mM glucose. Sustained oscillations of insulin concentrations secreted by the islets were also observed in these experiments, and the average period of oscillation was 14.6 +/- 2.3 min at 16.7 mM glucose. Mean insulin concentrations at 5.5 mM glucose were too low to permit analysis of oscillations. Spectral analysis confirmed the regularity of the lactate and insulin oscillations and showed peaks that were consistent with the average periods obtained using the Clifton program. Moreover, spectral analysis demonstrated marked similarity between the patterns of lactate and insulin oscillation during perifusion with 16.7 mM glucose. Cross-correlation analysis found these oscillations not to be consistently in phase. In conclusion, sustained oscillations in lactate released from islets of Langerhans suggest that the glycolytic process in beta-cells also oscillates. The similarity of the periods of lactate and insulin raises the possibility that oscillations in glycolysis may provide a mechanism for pulsatile insulin secretion.

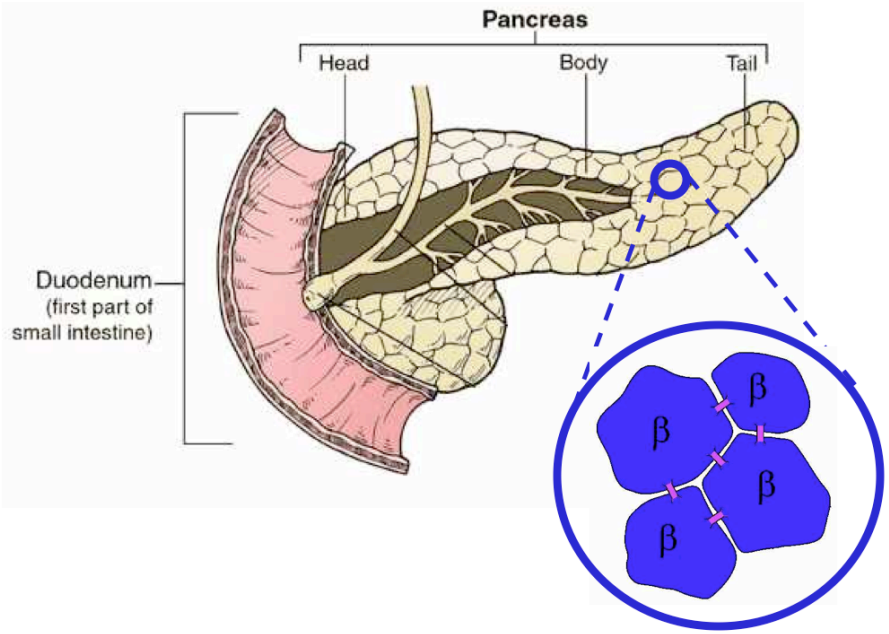
Glycolytic oscillations in β-cells

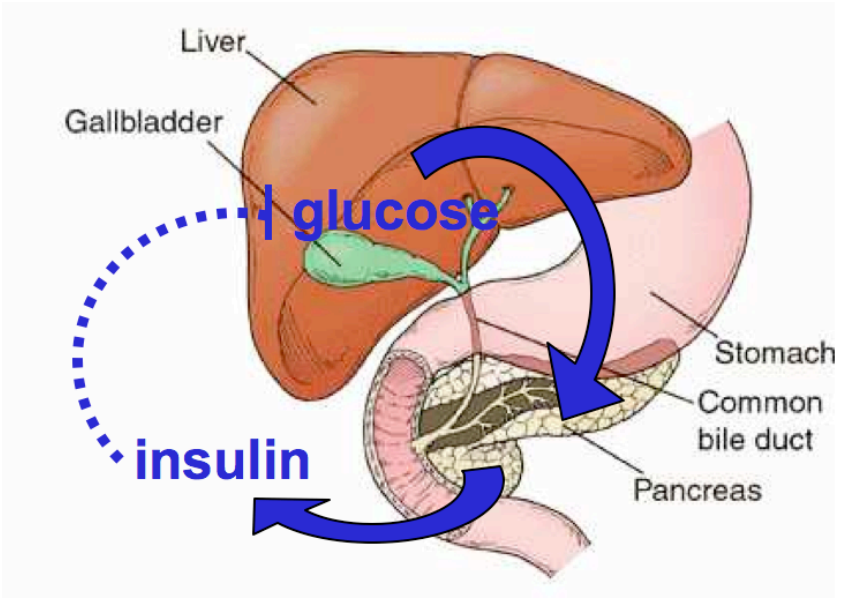
A model for the inter-cellular synchronization of glycolytic oscillations (via insulin secretion)

Glycolytic oscillations in β-cells

In the pancreas, oscillations are synchronized within a single Langherans islet because cells are coupled through gap junctions.

Inter-islets synchronization occurs through the modulation of glucose production in the liver by extracellular insulin.

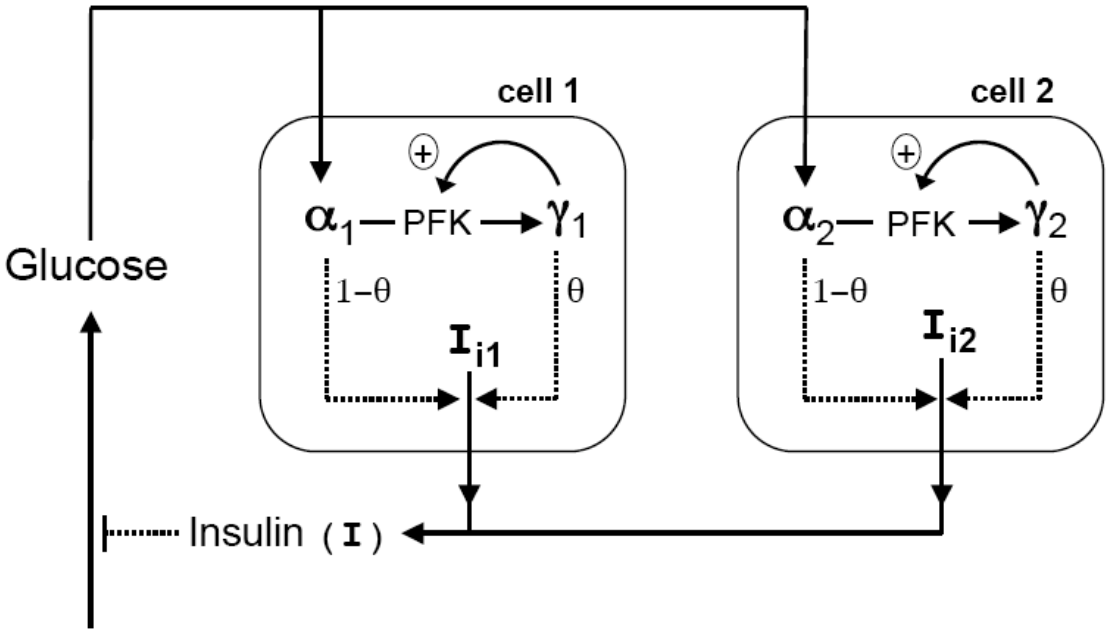




The model which is proposed here concerns inter-islets synchronization.

Glycolytic oscillations in β-cells: model

Model for the synchronization through global coupling of two β cells



The two oscillators represent two islets of Langerhans, each of which contain thousands of β cells synchronized through gap junctions. Each cell then represents the behavior of a single islet and the model applies to the synchronization of different islets. Intracellular glycolytic oscillations are produced by the PFK, which is activated by its reaction product. The release of insulin **(I)** is assumed to be coupled to variations in substrate (α) and product (γ). Extracellular insulin exerts a negative feedback on glucose production in the liver. Such a modulation of glucose production in the liver affects the input of substrate to the oscillatory PFK reaction within each cell.

Gonze D, Markadieu N, Goldbeter A (2008) Selection of in-phase or out-of-phase synchronization in a model based on global coupling of cells undergoing metabolic oscillations, *Chaos* 18:037127.

Glycolytic oscillations in β-cells: model

Glycolytic oscillations:

$$\frac{d\alpha_j}{dt} = v_i + v_e - \sigma_j \phi_j,$$ (glycolytic substrate)

$$\frac{d\gamma_j}{\mathrm{dt}} = \sigma_j \phi_j - k\gamma_j,$$ (glycolytic product)

$$\frac{\mathrm{d}I}{\mathrm{d}t} = v_{mr}\xi - k_d I$$ (insulin)

with

$$v_i = v_{\max}\frac{K_I^m}{K_I^m + I^m},$$ (K_I measures the coupling strength)

$$\phi_j = \frac{\alpha_j e(1+\alpha_j e)(1+\gamma_j)^2}{L(1+\alpha_j c)^2 + (1+\alpha_j e)^2(1+\gamma_j)^2}$$

Coupling:

- through the substrate α

$$\xi = \xi_a = \frac{1}{N}\sum_j \frac{\alpha_j^n}{K_a^n + \alpha_j^n}$$

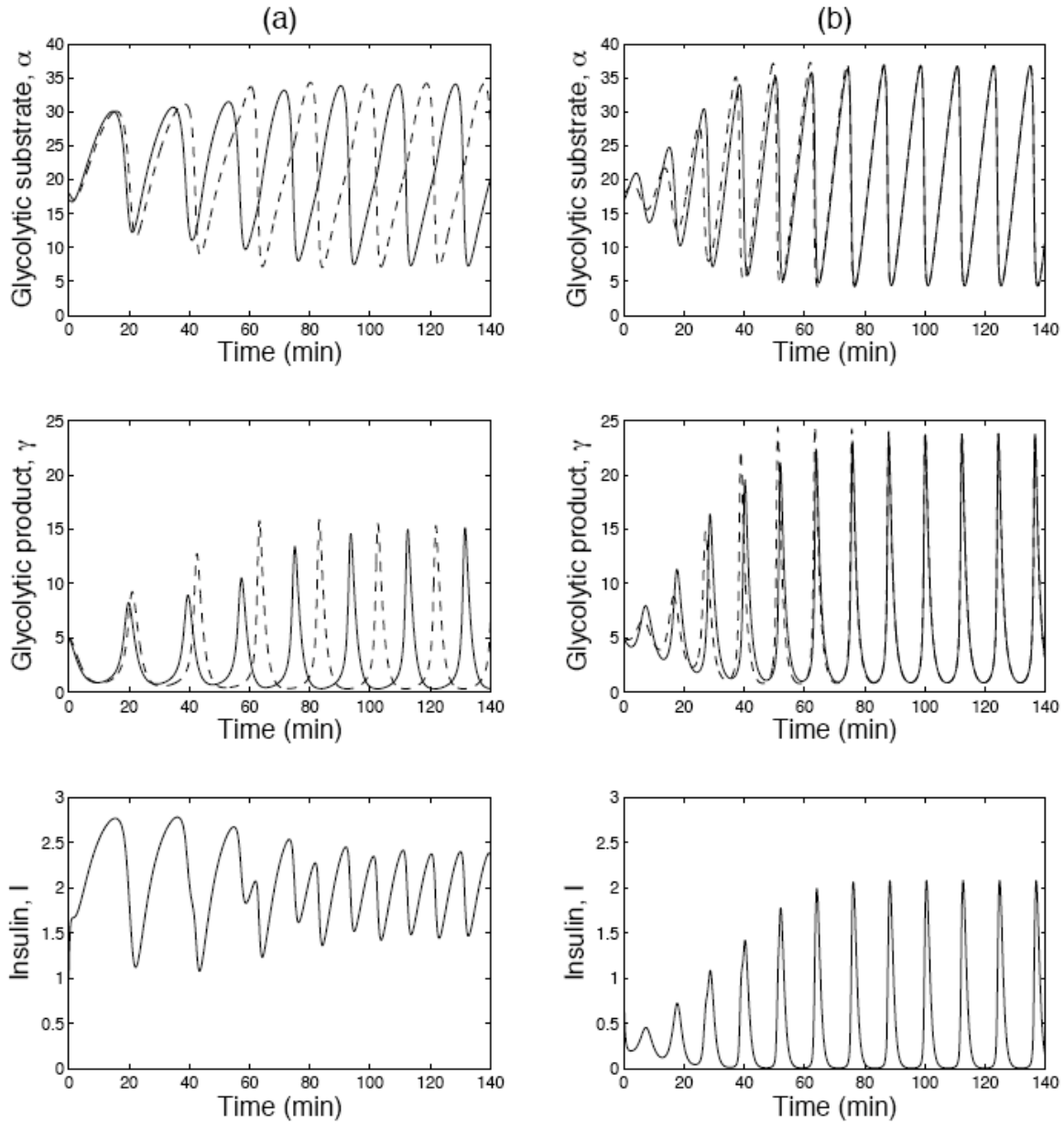
- through the product γ

$$\xi = \xi_b = \frac{1}{N}\sum_j \frac{\gamma_j^n}{K_a^n + \gamma_j^n}$$

- hybrid model

$$\xi = \xi_h = \frac{1}{N}\left((1-\theta)\sum_j \frac{\alpha_j^n}{K_a^n + \alpha_j^n} + \theta\sum_j \frac{\gamma_j^n}{K_a^n + \gamma_j^n}\right)$$

Glycolytic oscillations in β-cells: model

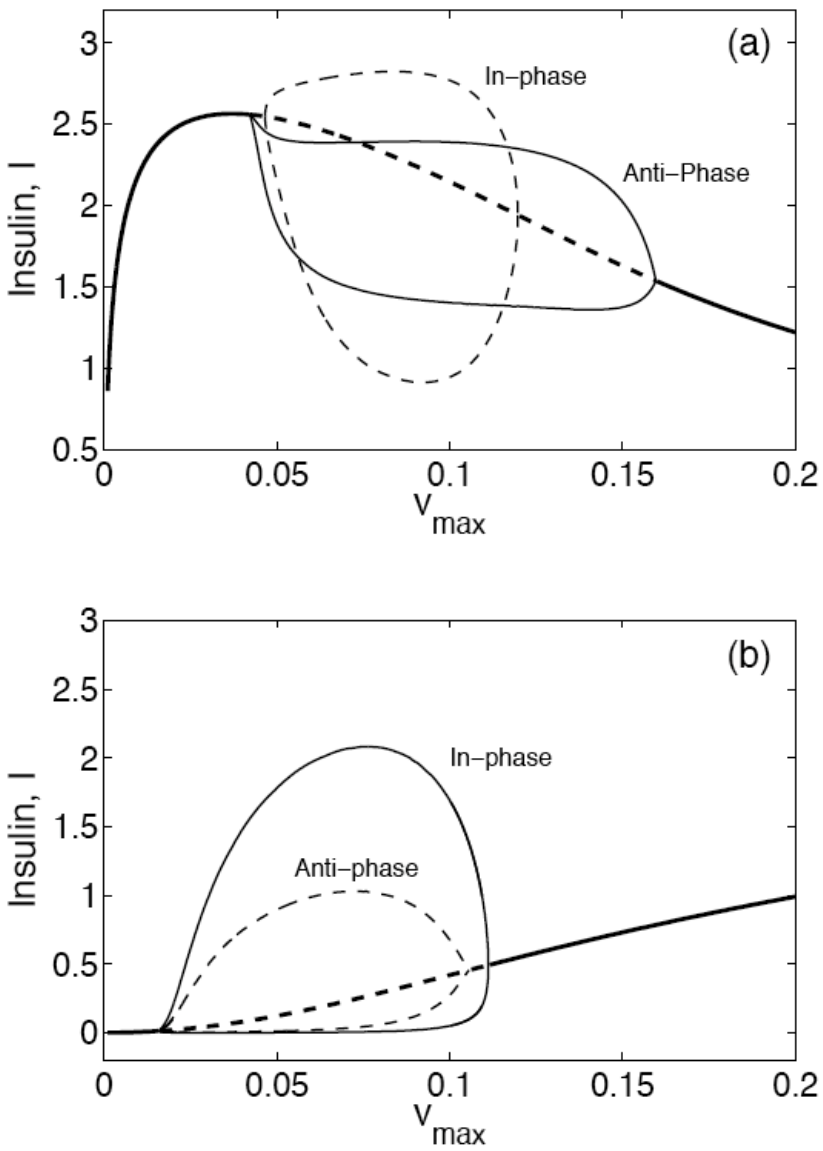


The mode of synchronization depends on the way the two oscillators are coupled:

(a) When insulin release is controlled by the glycolytic **substrate** (α), the oscillations are in anti-phase

(b) When insulin release is controlled by the glycolytic **product** (γ), the oscillations are in phase.

Glycolytic oscillations in β-cells: model



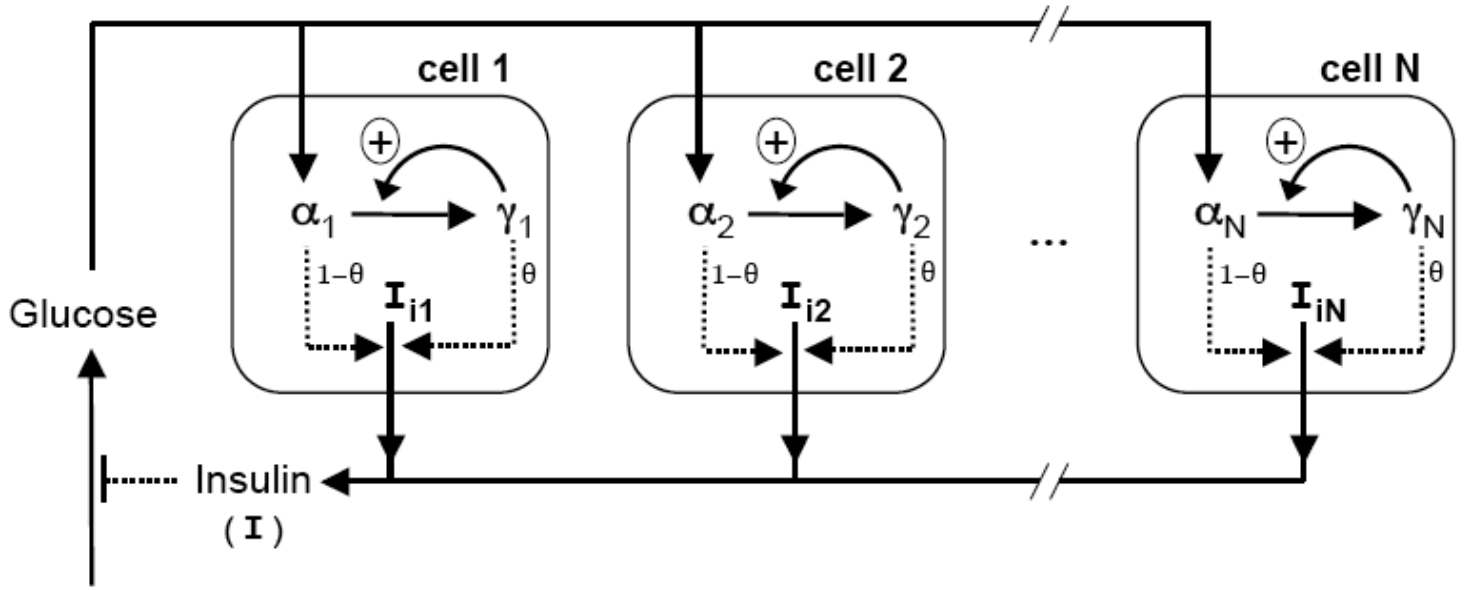
Bifurcation diagram as a function of the maximum rate of glucose input (v_{max}) into the cell.

(a) When insulin release is controlled by the glycolytic **substrate**, the stable limit cycle regime corresponds to anti-phase synchronization, while the unstable limit cycle regime corresponds to in-phase oscillations.

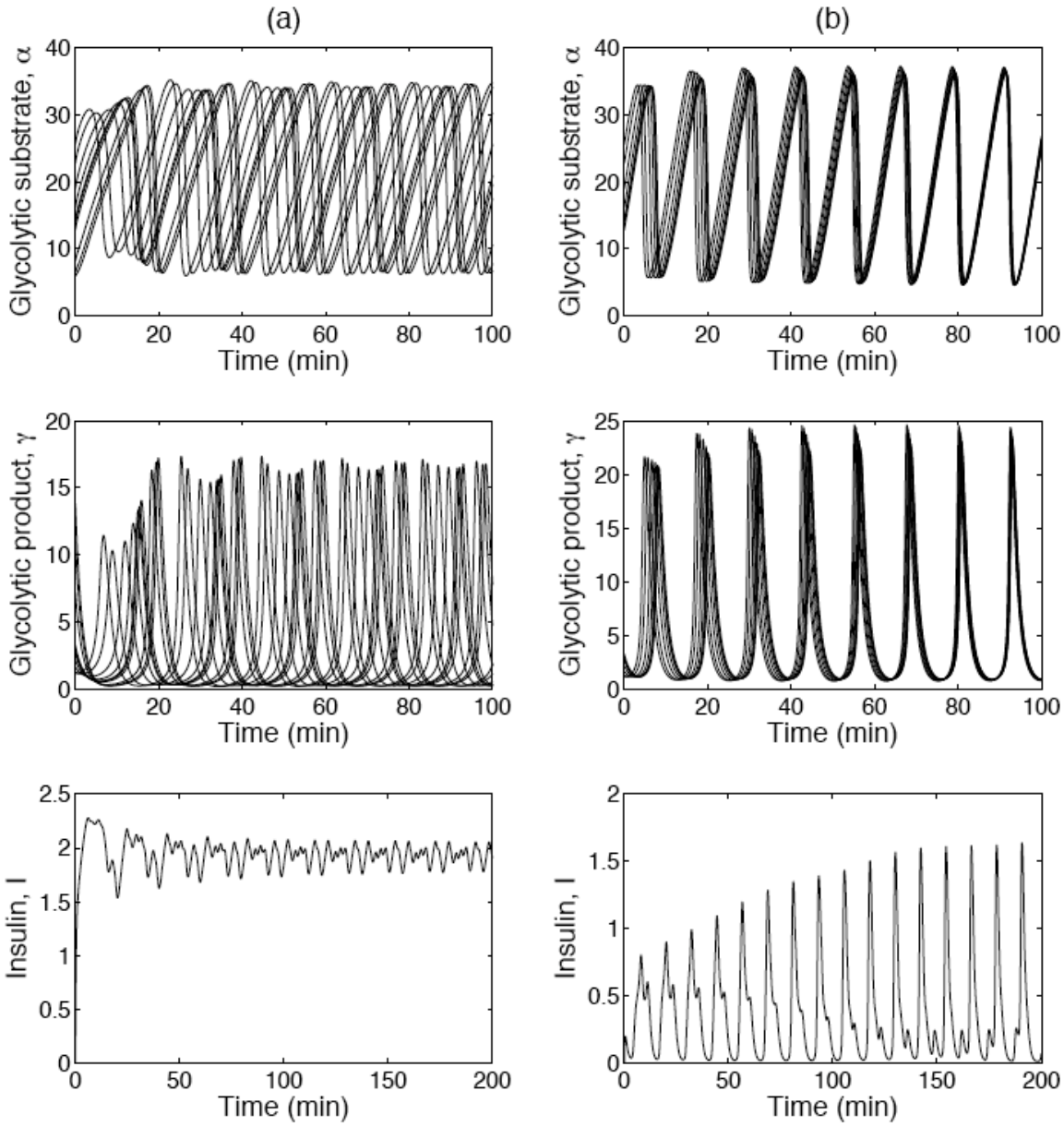
(b) When insulin release is controlled by the glycolytic **product**, the stable limit cycle regime corresponds to in-phase synchronization and the unstable limit cycle regime corresponds to antiphase oscillations.

Glycolytic oscillations in β-cells: model

From 2 to *N* coupled cells...



Glycolytic oscillations in β-cells: model



Oscillations obtained for the global coupling of N=10 oscillating cells

(a) When insulin release is controlled by the **substrate** oscillations are out-of-phase.

(b) When insulin release is controlled by the **product** the oscillations are in phase.

Glycolytic oscillations: Summary

- Self-sustained oscillations in the glycolytic oscillations have been measured in yeast and in mammalian beta cells, for a bounded range of substrate input rates.
- The enzyme responsible for these oscillations is the PFK (3rd step of the glycolysis)
- The regulation of the PFK is based on a positive retro-action.
- The Selkov model accounts for self-sustained oscillations in an autocatalytic biochemical system, but does not account for the bounded range of substrate input rates (single Hopf bifurcation).
- The allosteric model of Goldbeter-Lefever accounts for the occurrence of self-sustained (limit cycle) glycolytic oscillations, for the bounded range of substrate input leading to oscillations (2 Hopf bifurcations), and for the period dependency of the substrate input rate.
- An extension of the allosteric model accounting for an (hypothetical) allosteric regulation in 2 successive reaction step show the possibility to generate complex oscillations, including chaos and bursting.
- More detailed models, taking into consideration the full glycolysis, have also been proposed.
- Several models have been proposed to account for the inter-cellular synchronization of glycolytic oscillations in yeast and in mammals.
- A minimal model for the inter-islets synchronization of glycolytic oscillations, based on a global coupling between pancreatic β-cells via the secretion of insulin, suggests that the mode of synchronization (in-/out- of phase) obtained depends on the type of coupling.

References

- Higgins J (1964) A chemical mechanism for oscillation of glycolytic intermediates in yeast cells. Proc. Natl. Acad. Sci. USA 51: 989-994.
- Sel'kov EE (1968) Self-oscillations in glycolysis. 1. A simple kinetic model. Eur. J. Biochem. 4:79-86.
- Goldbeter A, Lefever R. Dissipative structures for an allosteric model. Application to glycolytic oscillations. Biophys J. (1972) 12:1302–1315.
- Decroly, O. & A. Goldbeter. (1982). Birhythmicity, chaos and other patterns of temporal self-organization in a multiply regulated biochemical system. Pro. Natl. Acad. Sci. USA, 79: 6917-6921.
- Wolf J, Heinrich R (2000) Effect of cellular interaction on glycolytic oscillations in yeast: a theoretical investigation. Biochem J. 345 Pt 2:321-34.
- Wolf J, Passarge J, Somsen OJ, Snoep JL, Heinrich R, Westerhoff HV (2000) Transduction of intracellular and intercellular dynamics in yeast glycolytic oscillations. Biophys J. 78:1145-53.
- Hynne F, Danø S, Sørensen PG (2001) Full-scale model of glycolysis in Saccharomyces cerevisiae. Biophys Chem. 94:121-63
- Danø S, Hynne F, De Monte S, d'Ovidio F, Sørensen PG, Westerhoff H. (2001) Synchronization of glycolytic oscillations in a yeast cell population. Faraday Discuss. 120:261-76.

See also: Goldbeter, A. (1996). Biochemical Oscillations and Biological Rhythms. Cambridge University Press (Chapters 2 & 4).