

*Review Article*

## **Bacterial Growth Rate and Growth Yield: Is There A Relationship?**

UTTARA N LELE<sup>1</sup> and MILIND G WATVE<sup>2\*</sup>

<sup>1</sup>*Department of Microbiology, Abasaheb Garware College, Pune, India*

<sup>2</sup>*Indian Institute of Science Education and Research, Pune, India*

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In microbial ecology and physiology, growth rate and growth yield are among the most fundamental parameters. The question whether the two are independent of each other or correlated in some way has been addressed by two schools of microbiologists with contradicting outcomes. After the classical Monodian assumption of constant growth yield was found unsupported, microbial physiologists predicted a positive rate-yield correlation. This was based on the assumption of constant maintenance energy. On the other hand evolutionary biologists predicted a rate-yield trade-off subject to differential selective pressures in different environments. Such a trade-off can explain the wide variation in growth rates and growth yields across the microbial world. However, empirical approaches to the question are plagued by methodological problems and inconsistencies across studies. We critically evaluate the alternative ways of thinking highlighting on selective forces, mechanisms shaping the relationship and appropriate experimental approaches.

**Key Words: Microbial Physiology; Ecophysiology; R and K Selection; Life History Strategies**

### **Introduction**

The study of growth of bacterial cultures was considered by Jacques Monod as ‘the basic method of microbiology’ (Monod, 1949). Although the concept remains fundamental to microbiology even today, studies focusing on understanding microbial growth have become rarer in recent decades in spite of many fundamental questions remaining unanswered. Monod’s classical work in the 1950s formed the basis of bacterial growth physiology and was a consistent line of research for a few decades to come. Although the work was of a fundamental nature, it had an underlying applied component. Understanding of bacterial physiology was viewed as a useful tool in optimising industrial processes such as fermentations. In later decades, after the advent of molecular biology and genetic engineering, new approaches were found successful in increasing the product yields in microbial processes. As a result, research related to physiology of growth took a back

seat and barring a few isolated studies almost vanished in the 2000s. This is consistent with what Salvatore Luria noted in his presidential address to the general meeting of American Society for Microbiology in 1968. ‘Microbiologists seem to have suffered themselves at times from a mild case of anti-intellectualism or at least of hyper-practicality almost as though research without immediate practical goals were something of a sinful luxury’ (Luria, 1968). Even after four decades, Luria’s criticism seems to hold true and though much better tools are now available, research on some of the basic concepts of ecophysiology of bacteria remain underexplored.

Two of the main pillars of ‘study of growth’ recognised from early on were growth rate and growth yield. Microbial physiologists of earlier decades shared an intuition that growth parameters are constant or follow certain simple rules (Declaux, 1898; Herenci, 1928; Hinshelwood, 1946; Marr, 1991). Several attempts were made to discover these

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\*Author for Correspondence: milind@iiserpune.ac.in

rules, experimentally and theoretically. Monod showed experimentally that growth rate depended upon energy and carbon substrate. It is well known that growth rate varies in response with environmental factors such as temperature, pH and nutrient availability (Hempfling and Mainzer, 1975; Shehata and Marr, 1971; Schaechter *et al.*, 1958). Monod (1949) theorised and seems to have observed a saturation relationship between substrate concentration and growth rate while growth yield remained constant over a wide range of nutrient concentrations. Theoretically, if growth yield is constant and growth rate varies according to nutritional environment then there cannot be a relationship between growth rate and growth yield. However, there have been many contradicting claims later towards a positive or a negative relationship between the two.

### Growth Yield and Related Confusions

There are more than one expressions in use for bacterial growth yield. The simplest term is biomass yield ( $Y$ ) described as biomass produced per unit of substrate consumed. Biomass produced can be determined by measuring dry weight of cells or estimating their total protein content in a known volume. The amount of substrate consumed is the difference between the initial concentration and the concentration of substrate left after the growth period. Another popularly used expression of bacterial yield is  $Y_{ATP}$  defined as the biomass produced per mole of ATP. ATP yield is calculated in the following manner (Bauchop and Elsdén, 1960). The ATP production from the substrate can be calculated if the substrate and its catabolic pathway are known. The biomass produced is estimated by gravimetry. With these estimates, one can calculate the amount of biomass that is produced per mole of ATP. Thus the yield can be expressed as mg of biomass per mg or mole of substrate consumed or mg of biomass formed per mole of ATP generated.

Monod (1949) assumed growth yield to be constant. Further, Bauchop and Elsdén (1960) proposed that  $Y_{ATP}$ , that is the growth yield per mole of ATP turnover, is almost a universal biological

constant and stays around 10.5 g/mole of ATP across species. However, in 1970 Vries *et al.* (1970) observed different and very high values of  $Y_{ATP}$  reaching 20.9 gm/mole ATP. In 1973, Stouthamer systematically calculated the amount of energy in the form of ATP required for the formation of microbial cell mass under various growth conditions. These calculations were based on the assumption of a standard cellular composition reported by Morowitz (1968) which showed that from preformed monomers/ precursors 31.9 g cells can be formed from each mole of ATP. This value was called as  $Y_{ATP}^{MAX}$ . If the cell composition is constant, the  $Y_{ATP}^{MAX}$  can be expected to be a constant. However, cell composition can also be substantially different in different growth conditions (Schaechter *et al.*, 1958). Thus even  $Y_{ATP}^{MAX}$  does not appear to be a biological constant.

The calculations of  $Y_{ATP}^{MAX}$  are based on the assumption that the substrate for generation of ATP and that for biomass synthesis is different. More commonly in bacterial cultures, a single substrate may serve as an energy generating substrate and a source of carbon for biomass synthesis. In such cases, maximum biomass yield is possible when the division between the channelization of substrate for ATP generation and biomass production is optimum. The optimum point is where the amount of ATP generated by catabolising part of the substrate is just enough to convert the remaining amount of substrate into cellular components (Watve *et al.* 2000). i. e. when

$$C.A.Y'_{ATP} = (1 - C) \times \text{Molwt. of substrate} \\ \times \frac{\text{carbon fraction in substrate}}{\text{carbon fraction in dry biomass}}$$

Where  $C$  is the fraction of glucose catabolised and  $A$  is the moles of ATP produced per mole of substrate catabolised. Proportion of carbon in dry biomass is generally around 50% wt/wt whereas percentage of carbon in glucose is 40%. Therefore, the equation can be written for glucose as a substrate,

The maximum biomass yield at this optimum

will be  $144 \cdot (1-C)$ . If  $Y'_{ATP}$  is assumed to be the theoretical maximum i.e.  $Y_{ATP}^{MAX}$ , the optimum division  $C$  can be calculated to be close to 0.13. Only 13% of glucose needs to be catabolised. If the rest is available for biomass carbon the maximum yield from glucose is 125.17 g/mole or approximately 70% wt/wt. However, the empirical yields are reported to be substantially smaller (Hempfling and Mainzer, 1975; Vries *et al.*, 1970) under culture conditions commonly used in the laboratory. Growth yields close to the theoretical maximum are nevertheless reported in oligotrophic environments (Watve *et al.*, 2000). The causes of such a wide difference in growth yields are not clearly known.

### Is The Relationship Between Growth Rate and Growth Yield Positive?

In Monod's model, the maintenance energy of the growing culture was more or less neglected. However, maintenance energy soon became a widely discussed concept. Maintenance energy is the fraction of the energy consumed by cells for purposes other than growth i.e. the presumed 'endogenous metabolism' (Herbert, 1958) needed for cell maintenance and repair operations (Marr *et al.*, 1963; Pirt, 1965). For a long time, maintenance energy was considered as a constant defined as an amount of energy needed for maintenance per unit weight of organism per unit time. If maintenance energy is constant per unit time it follows that in a faster growing culture in unit time the biomass produced is larger but maintenance energy remains the same. Therefore the proportion of energy needed in maintenance becomes smaller as compared to the energy invested in growth. This theory predicts that growth rate has a positive relationship with growth yield. This prediction was also supported with a few observations (Hobson, 1965; Russell and Baldwin, 1979)

Pirt (1965) proposed an equation defining a relationship between theoretical growth yield ( $Y_G$ ), observed growth yield ( $Y$ ), specific growth rate ( $r$ ) and 'maintenance coefficient ( $m$ )'.

$$\frac{1}{Y} = \frac{m}{r} + \frac{1}{Y_G}$$

If this equation correctly describes the relationship, the double reciprocal plot of growth rates and observed growth yields should give a straight line with a positive slope. Pirt plotted double reciprocal graphs of rate and yields observed in chemostat experiments to calculate the maintenance energy as the slope of the double reciprocal plot. They found that this relationship was linear and positive implying that the maintenance energy was constant (Fig. 1). Similar patterns are also reported by others (Hempfling and Mainzer, 1975; Hobson, 1965).

However, not all experimenters reported constant maintenance energy. For example Russell and Baldwin (1979) estimated the maintenance coefficients for five rumen bacteria by growing them in continuous cultures. In two of the experimental organisms, they found a non-monotonic relationship between growth rate and growth yield (Fig. 2).

Varying outcomes of different studies suggest that different microorganisms may have different maintenance needs and the maintenance energy may also scale with the nutritional environment. Theoretically, if the maintenance energy is constant, at varying substrate concentrations the graph of substrate consumed and biomass produced should be a straight line with a positive X intercept, which is equal to the maintenance energy. But in some studies,

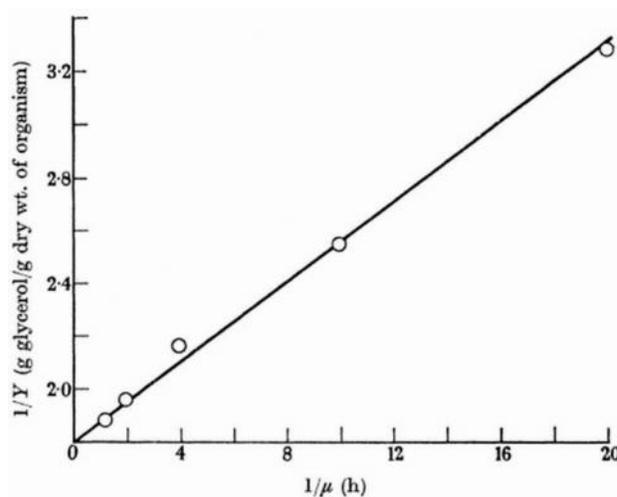


Fig. 1: Double reciprocal graph of yield and dilution rate of *Aerobacter aerogenes* grown in glycerol-NH<sub>3</sub> salt medium. (Reproduced with permission from Pirt, 1965 and Herbert, 1958)

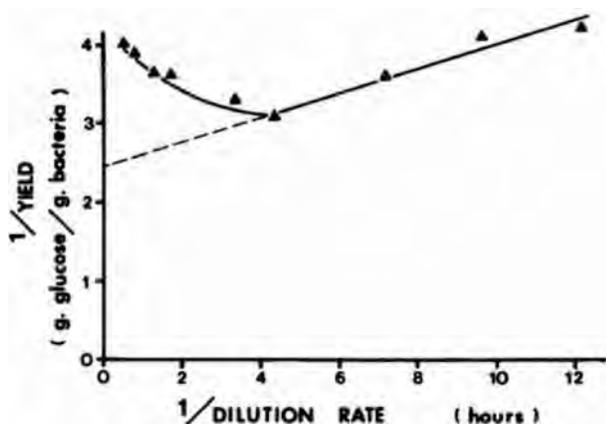


Fig. 2: Growth yield of *Streptococcus bovis* in a chemostat on glucose containing medium is a non-monotonic function of the dilution rate and thereby substrate concentration. (Reproduced with permission from Russell and Baldwin, 1979)

the X intercept becomes negative (Bauchop and Elsdén, 1960) (Fig. 3). Since negative maintenance energy is absurd, the alternative interpretation is that the slope of the line could be steeper at lower substrate concentrations. If this is true, for these organisms the relationship between growth rate and growth yield must be negative at the lower range of substrate concentration. Thus, the expectation of a positive growth rate-growth yield relationship is faced with some contradictions.

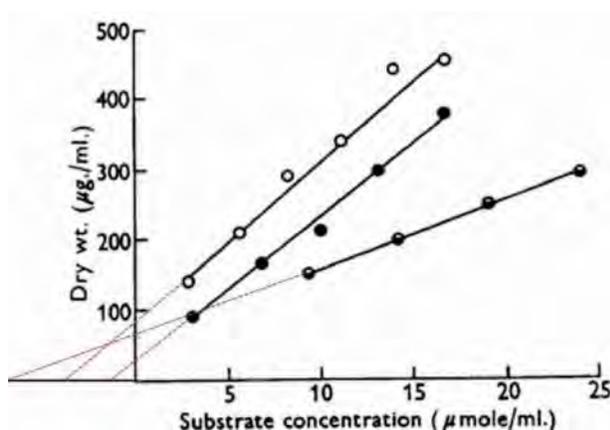


Fig. 3: Growth of *Streptococcus faecalis* on glucose, ribose and arginine. (Based on Bauchop and Elsdén, 1960). We interpolated the trends (dotted lines) to show that maintenance energy, if any is paradoxically negative

### Negative Relationship Between Growth Rate and Growth Yield

As on the one hand microbial physiologists prior to the 1980s expected and some of them observed a positive relationship, ecologists and evolutionary biologists on the other hand talked about a negative relationship or a trade-off between growth rates and growth yields (Pfeiffer and Bonhoeffer, 2002; Pfeiffer *et al.*, 2001). The notion appears to have arisen from theories of life history strategies. By naive thinking, traits with higher growth rates should always be selected. However, natural habitats have an abundance of slow growing organisms. This would not be possible unless the slow growers had some advantage that the fast growers could not harvest. In other words there ought to be some trade-offs associated with fast growth which enable the coexistence of organisms with widely differing growth rates. The trade-off predicted here is a physiological one and said to arise because of the decision of distribution of limited resources between two or more processes that compete with each other for the same resource (Stearns, 1976; Stearns, 1992). If growth yields positively correlated with growth rates, it would be even more unlikely to find any slow growing organisms in nature. This gave rise to the notion of growth rate-growth yield trade-off and this has been a dominant line of thinking post 1990s.

The growth rate-growth yield trade-off theory parallels the 'r and K selection' theory proposed by MacArthur and Wilson (1967). This theory proposes that the standing population density relative to the carrying capacity of the environment alters the selective forces. The theory mainly explains the life history traits and their evolution in higher animals and plants. The name 'r and K selection' comes from the two constants of the logistic growth equation.

$$\frac{dN}{dt} = rN \left[ 1 - \left( \frac{N}{K} \right) \right]$$

If we imagine a population having low population density and thereby near non-limiting resources, the best strategy will be to maximize the reproduction and offspring number since the

resources are in plenty. This life history strategy (*r* strategy) expects traits such as early reproduction, large value of *r*, many offspring with poor survivorship and small adult body size. On the other hand, *K*-selection is expected when an organism is growing in a crowded environment. In saturated environments, the *r*-strategy would not work since the resources are severely limiting. Here the best strategy would be to produce fewer but high-quality offspring. Thus, under resource limitation the *K* selection should favour late reproduction, low growth rate, fewer offspring with good survivorship and large adult body size.

A prediction that a parallel trade-off should be seen in heterotrophic bacteria has probably arisen from this ecological theory itself. Parameters like number and size of offspring, survivorship and reproductive age are clearly not applicable to unicellular organisms like bacteria that do not have age structure or a specific reproductive age and that divide by simple fission. In bacteria, growth rate and biomass conversion efficiency are two parameters that can be considered as main life history traits. Thus, this theory proposes a trade-off between reproduction and biomass, i. e. growth rate and growth yield. Even though, two extreme sets of life history traits are explained in this theory, a continuum between these two strategies in nature, between species and within species is seen. Watve *et al.* (2000) showed that bacteria growing in oligotrophic environments show the traits of *K* selection i. e. lower growth rate and higher biomass production.

Some evolutionary biologists and microbial ecologists suggested the rate-yield trade-off as a way of organisms to cope with changing environments (Pfeiffer and Bonhoeffer, 2002; Pfeiffer *et al.*, 2001). Different ecological strategies need to be supported by different sets of cellular functions. A corollary to this theory can be that a specific physiological trait can evolve as a characteristic of an organism if it is subjected to a frequently encountered ecological condition to which the trait is advantageous.

#### **Possible Proximate Mechanisms of the Trade-Off**

In addition to the evolutionary and ecological

arguments, a trade-off is suggested at the level of different cellular and biochemical mechanisms.

#### ***The ATP Paradox and the Growth Rate-Growth Yield Trade-Off***

Glucose acts as carbon as well as energy source for many heterotrophic organisms. It can follow two routes through catabolism – a respiratory route, where it is oxidised to CO<sub>2</sub> and a fermentative route, where it is partially oxidised to organic compounds like ethanol and organic acids. These catabolic reactions are coupled with ATP synthesis by oxidative or substrate level phosphorylation. One expects an increase in internal ATP concentration when concentration of extracellular glucose is increased. However, it has been observed in experiments performed on yeast and bacteria that a sudden transition from glucose limited to glucose sufficient state leads to a new steady state that has increased glycolytic flux but relatively or unexpectedly lower ATP/ADT ratio (Westerhoff *et al.*, 1983; Somsen *et al.*, 2000; Aledo *et al.*, 2004). This phenomenon has been named as the ‘ATP paradox’ (Westerhoff *et al.*, 1983). High glycolytic flux is responsible for higher growth rate but low ATP/ADP ratio does not permit high biomass conversion efficiency. Westerhoff *et al.* (1983) predicted a negative relationship between growth rate and growth yield caused by the ATP paradox.

#### ***Thermodynamic Trade-Off Between Rate and Yield of ATP Production***

The thermodynamics of a reaction depends upon whether the reactants and products are close to equilibrium or away from it. Close to equilibrium, the energy transaction is small but the net rate of reaction is also slow. Exactly at equilibrium, both the net energy transaction and net reaction become zero. On the other hand, if a system works away from equilibrium, the net reaction would be faster and greater energy change would also be involved. Pfeiffer *et al.* (2001, 2002) suggest that this leads to a trade-off in heterotrophic organisms in the rate and yield of ATP formation. The maximum ATP yield can be achieved if all the free energy difference is converted to ATP. This should happen when the

degradation reaction is near thermodynamic equilibrium. In that case however the rates of degradation and ATP production will be very low. Farther the system works away from equilibrium, the yield of ATP production decreases and rate of degradation coupled with ATP production increases. Thus heterotrophic organisms may generally face a trade-off between rate and yield of ATP production.

### ***Futile Cycles and Low Growth Yields***

Some metabolic reactions are such that they act in antagonistic action to each other; that is, these pathways serve as both catabolic and anabolic pathways in the directions opposite to each other. e. g. glycolysis and gluconeogenesis (Koch, 1997; Tempest and Neijssel, 1992). Organisms need to control these reactions very carefully so as to avoid over-flux of certain reaction when they are not needed. If cells fail to control the wastage of energy through such futile cycles, either because of inevitability or for some other selective advantage, then it will be wastage of substrate carbon and will result into lower growth yields.

### ***Metabolic Shifts and Fermentation – Respiration Ratio***

Fermentative microorganisms have the capacity to change their end-products and alter their ATP production capacities. The shift from homo-fermentation to hetero-fermentation is shown to increase growth yield by 2-3 folds (Vries *et al.*, 1970; Russell and Baldwin, 1979; Russell and Cook, 1995). Cooper and Anderson (1970) showed that *E. coli* used a pathway involving methylglyoxal to convert dihydroxyacetone phosphate to D-lactate. Since this pathway does not involve phosphate transferases, the free energy change of glucose catabolism does not generate ATP. Such metabolic shifts are the shifts within the fermentative pathways. Enteric bacteria being facultative anaerobic micro-organism can utilise and degrade glucose and other substrates by two alternative pathways – respiration and fermentation. Out of the two processes, respiration is a slower and longer process of glucose degradation. But at the end of the cycle, the reward in the form of ATP is several fold than that of the fermentative

pathways. Fermentative pathway renders the cell an advantage of fast degradation, thus it can be a useful strategy to use when fast growth is advantageous. When the substrate available is scarce, the efficiency of using available substrate might become a priority over the rate of utilisation of substrate. In such a case, respiration would be more advantageous way of substrate utilisation. Thus, using respiration or fermentation for better growth yield or better growth rate respectively can be one of the mechanisms behind the rate-yield trade-off (Pfeiffer and Bonhoeffer, 2002; Pfeiffer *et al.*, 2001).

### ***Leakage of Metabolic Intermediates***

Metabolic intermediates often leak into culture media even during the exponential growth phase (Koch, 1997; Tempest and Neijssel, 1992; Russell and Cook, 1995). Tempest and Neijssel (1992, 1984) noted that *Klebsiella aerogenes* produced pyruvate, 2-oxoglutarate, gluconate, 2-ketogluconate, and succinate when energy substrate was high. Similarly, it was observed that rumen bacteria excrete more amount of lactate under high dilution rates in a chemostat than under low dilution rates (Russell and Baldwin, 1979). This might be caused by the high rates of metabolic reactions. At high nutrient concentration, because of a large difference in the standing concentrations of substrate inside and outside of cells, the rate of uptake may be high. Then even the next metabolic steps would be faster owing to the rules of reaction kinetics. And preceding a rate limiting step in the whole pathway, there would be inevitable stacking of the metabolic intermediates followed by their leakage or overflow out of the cells. This loss is actually a loss of organic content and ultimately biomass. At the expense of this loss, the cells are able to achieve higher growth rates.

### ***Symmetry in Cell Division***

Until the last decade it was believed that, as long as the environmental conditions are optimum for growth and division, bacteria that divide by simple fission are immune to aging. However Stewart *et al.* (2005) showed that *E. coli* cells that undergo apparently symmetrical morphological cell division show functional asymmetry in cell division i.e. one of the

sister cells shows decreased growth rate. They predicted that this asymmetry was seen because one of the daughter cells received larger proportion of old and damaged cellular components, whereas the other daughter cell received larger proportion of newly synthesised components.

To examine the effects of symmetric and asymmetric cell division on the dynamics of growth in bacteria, Watve *et al.* (2006) designed a model based on classical Leslie-Matrix model. They modelled the dynamics of cell components of varying age classes instead of the dynamics of individuals as in classical Leslie-Matrix model. The results of the model showed that asymmetric cell division leads to higher growth rate and symmetric cell division gave higher growth yields under optimum repair rates. They suggest, therefore, that symmetric and asymmetric division may be favoured in different ecological niches. As mentioned earlier, it was shown in some experiments (Watve *et al.*, 2000) that organisms with low growth rate and higher biomass yields are abundant in natural environments suggesting that selection must be favouring higher yielding slow growers under some set of environmental conditions. Thus, this model by Watve *et al.* (2006) suggested symmetry of cell division as another possible mechanism for the rate-yield trade-off.

### Plasticity in the Trade-Off Mechanisms

While certain species are selected for rapid or slow growth because of their genome, as cited above, there is much phenotypic plasticity within individual bacterial species, allowing major physiological changes in cellular composition during shifts between rapid and slow growth phases. These physiological changes are proposed to be behind the rate-yield relationship. The relationship between growth rate and growth yield has not been studied extensively enough from biochemical as well as ecological considerations simultaneously. The relationship is indeed intriguing, especially in response to the nutrient availability. The same relation has been found to be positive as well as negative with changing conditions. This observation itself suggests that the

inconsistent relationship must be the effect of the phenotypic plasticity of organism/s because of which it can adapt to the changing habitats that it has to face while spreading and multiplying in nature. Thus rate-yield relationship seems to be one of the ways by which bacteria can achieve selective advantages in their changing surroundings. Phenotypic plasticity plays role in shaping up physiological strategies in bacteria such as symmetry of cell division. Symmetry of cell division is shown to be correlated to the growth yield and predicted to be correlated to the growth rate. *Escherichia coli* cells were shown to shift from symmetric cell division to asymmetric cell division when transferred from low nutrient to high nutrient concentration (Lele *et al.*, 2011).

### Growth Rate and Growth Yield in a New Perspective

This new look at the physiology is at substantial conceptual deviation from classical bacterial physiology. The classical concept of constant growth yield is certainly turned down by many studies. The concept of constant maintenance energy is also faced with a serious challenge. This is consistent with Ferenci (1999) who reviewed work on bacterial growth physiology after 50 years of Monod's 1949 paper. Ferenci finds that the classical textbook equations and the presumed constants are no more consistently followed across studies. He asks, "...is it meaningful, given the now better understood regulatory plasticity of bacteria, to expect currently accepted textbook equations to accurately describe bacterial growth?" For example, Monod found a smooth saturating relationship between nutrient concentration and growth rate described by a Michaelis-Menten equation (Fig. 4 a). He also found a linear relationship between substrate and biomass produced, implying constant growth yield (Figure 4 b). Many other studies failed to reproduce this smooth saturating curve. The studies that attempted to fit the saturating curve to their data came up with widely differing estimates for the constant  $K_s$  ranging from <1 to 44  $\mu\text{M}$  (Ferenci 1999; Senn *et al.* 1994). This inconsistency across different studies on the same species raises a suspicion that Monod's original results were too good to be true.

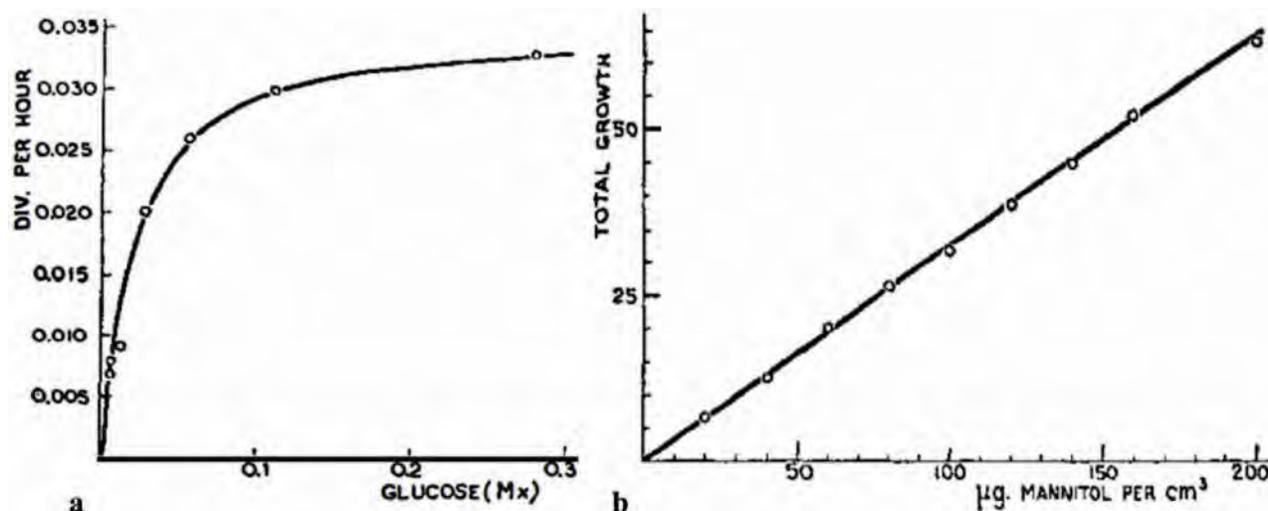


Fig. 4: (a) Saturating relationship between growth rate and substrate availability shown by Monod (1949), (b) Monod's observations showing growth yield as a constant. (Reproduced from Monod, 1949)

### Do We Observe a Clean Negative Correlation?

If a growth rate-growth yield trade-off is real, we should see significant negative correlations between the two growth parameters. However, there are no clear demonstrations of it so far. The two most likely problems in such a demonstration could be inadequate coverage of habitat types and the difficulty in playing on level grounds. Bacteria are adapted to widely varying nutritional environments and the relationship is likely to be detectable only in a cross section across the range of adaptations. Inclusion of extreme oligophiles and copiphiles is necessary to have a cross sectional look at the entire range. However, this creates another problem - that of determining the growth parameters under similar growth conditions. The range of organisms may fail to grow on a single growth medium. Even if they do grow, it is difficult to ensure that this is their naturally preferred substrate. Organisms may give different and unpredictable responses if they are grown in environments for which they have not evolved. It is therefore difficult to infer from cross sectional multispecies data. Growing the same strain in different nutrient concentration and comparison of different unrelated strains may give different types of and even contradictory correlations. Interestingly, most positive correlations have been shown by physiologists prior to the 1980s by growing

the same strain under different conditions (Hempfling and Mainzer, 1975; Hobson, 1965; Russell and Baldwin, 1979). The trade-offs literature by ecologists is mostly post 1990s (Pfeiffer *et al.*, 2001, Somsen *et al.* 2000). This is most likely to be a publication bias. Results that are expected and compatible with current paradigm are more likely to get published than the results that contradict current thinking.

An alternative promising approach would be to subject a given strain to contrasting selections under different selection regimes and see how selection for high growth rate affects growth yields and vice versa. Such inquiries have certainly begun, but are yet to yield conclusive pictures. A few studies and records in recent years show that a trade-off existed within a restricted group of observations, but it was not seen in the pooled data (Novak *et al.*, 2006; Lele, 2013). Therefore, the question whether there is a relationship between growth rates and growth yields in bacteria is muddled with paradoxes and contradictions. More carefully designed experimental approaches, sound theoretical models and a broad perspective vision is needed to resolve the problem. It is ironic that simple and fundamental phenotype level questions in microbial life have still remained unresolved while on the other hand, research is actively and aggressively unearthing the molecular details.

## References

- Aledo J C, Valle D and Esteban A (2004) The ATP paradox is the expression of an economizing fuel mechanism *J Biol Chem* **279** 55372-55375
- Bauchop T and Elsden S R (1960) The growth of micro-organisms in relation to their energy supply *J Gen Micro boil* **23** 457-469
- Cooper R A and Anderson A (1970) The formation and catabolism of methylglyoxal during glycolysis in *Escherichia coli* *Febs Lett* **11** 273-276
- Declaux E (1898) *Traite de microbiologie* Masson Paris
- Ferenci T (1999) 'Growth of bacterial cultures' 50 years on: towards an uncertainty principle instead of constants in bacterial growth kinetics *Res Microbiol* **150** 431-438
- Hempfling W P and Mainzer S E (1975) Effects of varying the carbon source limiting growth on yield and maintenance characteristics of *Escherichia coli* in continuous culture *J Bacteriol* **123** 1076-1087
- Herbert D (1958) Some principles of continuous culture *Recent Prog Microbiol* **3** 381-396
- Herenci A T (1928) *Morphologic variation and rate of growth of bacteria* Microbiology monographs Bailliere, Tindall, and Cox, London
- Hinshelwood C N (1946) *The chemical kinetics of the bacterial cell*. Clarendon Press Oxford
- Hobson P N (1965) Continuous culture of some anaerobic and facultatively anaerobic Rumen bacteria *J Gen Microbiol* **38** 167-180
- Koch A L (1997) Microbial physiology and ecology of slow growth *Microbiol Mol Biol Rev* **61** 305-318
- Lele U N (2013) Physiological adaptations in *Escherichia coli* under oligotrophic conditions PhD Thesis, University of Pune, pp 1-90
- Lele U N, Baig U I and Watve M G (2011) Phenotypic plasticity and effects of selection on cell division symmetry in *Escherichia coli* *Plos One* **6** e14516
- Luria S E (1968) The microbiologist and his times *Bacteriol Rev* **32** 401-403
- MacArthur R H and Wilson E O (1967) *Theory of Island Biogeography*. Princeton University Press
- Marr A G (1991) Growth rate of *Escherichia coli* *Microbiol Rev* **55** 316-333
- Marr A G, Nilson E H and Clark D J (1963) The maintenance requirement of *Escherichia coli* *Ann N Y Acad Sci* **102** 536-548
- Monod J (1949) The growth of bacterial cultures *Annu Rev Microbiol* **3** 371-394
- Morowitz H J (1968) *Energy flow in biology: biological organization as a problem in thermal physics*. Academic Press New York.
- Novak M, Pfeiffer T, Lenski R E, Sauer U and Bonhoeffer S (2006) Experimental tests for an evolutionary trade-off between growth rate and yield in *E coli* *Am Nat* **168** 242-251
- Pfeiffer T and Bonhoeffer S (2002) Evolutionary consequences of trade-offs between yield and rate of ATP production *Z Für Phys Chem* **216** 51-63
- Pfeiffer T, Schuster S and Bonhoeffer S (2001) Cooperation and competition in the evolution of ATP-producing pathways *Science* **292** 504-507
- Pirt S J (1965) The maintenance energy of bacteria in growing cultures *Proc RSocLond B Biol Sci* **163** 224-231
- Russell J B and Baldwin R L (1979) Comparison of maintenance energy expenditures and growth yields among several rumen bacteria grown on continuous culture *Appl Environ Microbiol* **37** 537-543
- Russell J B and Cook G M (1995) Energetics of bacterial growth: balance of anabolic and catabolic reactions *Microbiol Rev* **59** 48-62
- Schaechter M, Maaloe O and Kjeldgaard N O (1958) Dependency on medium and temperature of cell size and chemical composition during balanced growth of *Salmonella typhimurium* *J Gen Microbiol* **19** 592-606
- Senn H, Lendenmann U, Snozzi M, Hamer G and Egli T (1994) The growth of *Escherichia coli* in glucose-limited chemostat cultures: a re-examination of the kinetics *Biochim Biophys Acta Bba- Gen Subj* **1201** 424-436
- Shehata T E and Marr A G (1971) Effect of nutrient concentration on the growth of *Escherichia coli* *J Bacteriol* **107** 210-216
- Somsen O J, Hoeben M A, Esgalhado E, Snoep J L, Visser D, van der Heijden R T, Heijnen J J and Westerhoff H V (2000) Glucose and the ATP paradox in yeast *Biochem J* **352** 593-599
- Stearns S C (1976) Life-History Tactics: A Review of the Ideas *Q Rev Biol* **51** 3-47
- Stearns S C (1992) *The Evolution of Life Histories*. OUP Oxford
- Stewart E J, Madden R, Paul G and Taddei F (2005) Aging and death in an organism that reproduces by morphologically symmetric division *Plos Biol* **3** e45

- Stouthamer A (1973) A theoretical study on the amount of ATP required for synthesis of microbial cell material *Antonie Van Leeuwenhoek* **39** 545-565
- Tempest D W and Neijssel O M (1992) Physiological and energetic aspects of bacterial metabolite overproduction *Fems Microbiol Lett* **100** 169-176
- Tempest D W and Neijssel O M (1984) The Status of YATP and maintenance energy as biologically interpretable phenomena *Annu Rev Microbiol.* **38** 459-513
- Vries W D, Kapteijn W M C, Beek E G V D and Stouthamer A H (1970) Molar growth yields and fermentation balances of *Lactobacillus casei* L3 in batch cultures and in continuous cultures *J Gen Microbiol* **63** 333-345
- Watve M, Shejval V, Sonawane C, Rahalkar M, Matapurkar A, Shouche Y, Patole M, Phadnis N, Champhenkar A, Damle K, Karandikar S, Kshirsagar V and Jog M (2000) The 'K' selected oligophilic bacteria: A key to uncultured diversity? *Curr Sci* **78** 1535-1542
- Watve M, Parab S, Jogdand P and Keni S (2006) Aging may be a conditional strategic choice and not an inevitable outcome for bacteria *Proc Natl Acad Sci U S A* **103** 14831-14835
- Westerhoff H V, Hellingwerf K J and Dam K V (1983) Thermodynamic efficiency of microbial growth is low but optimal for maximal growth rate *Proc Natl Acad Sci* **80** 305-309.