H_2 Threshold Concentrations and f_e Values as Indicators of Halorespiration

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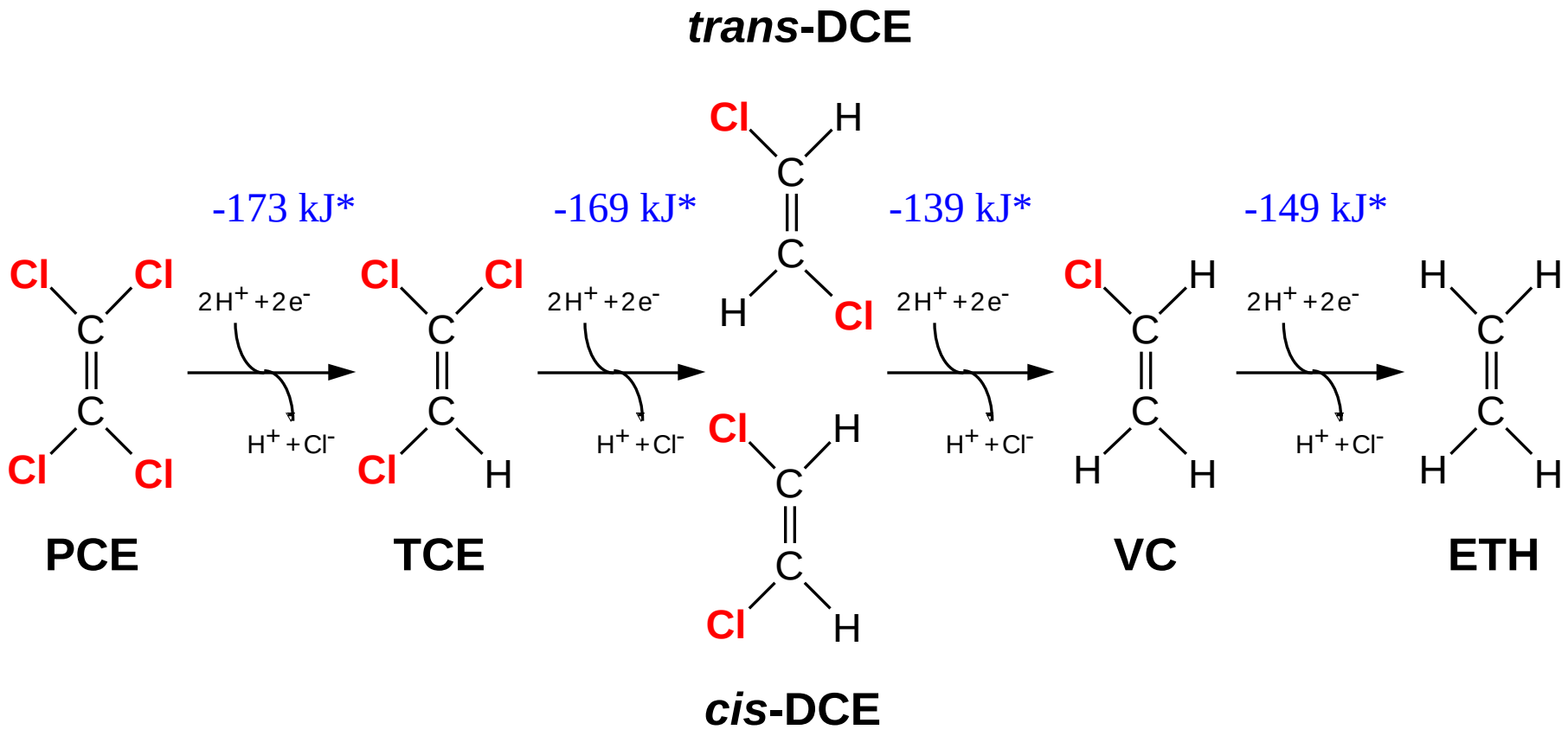
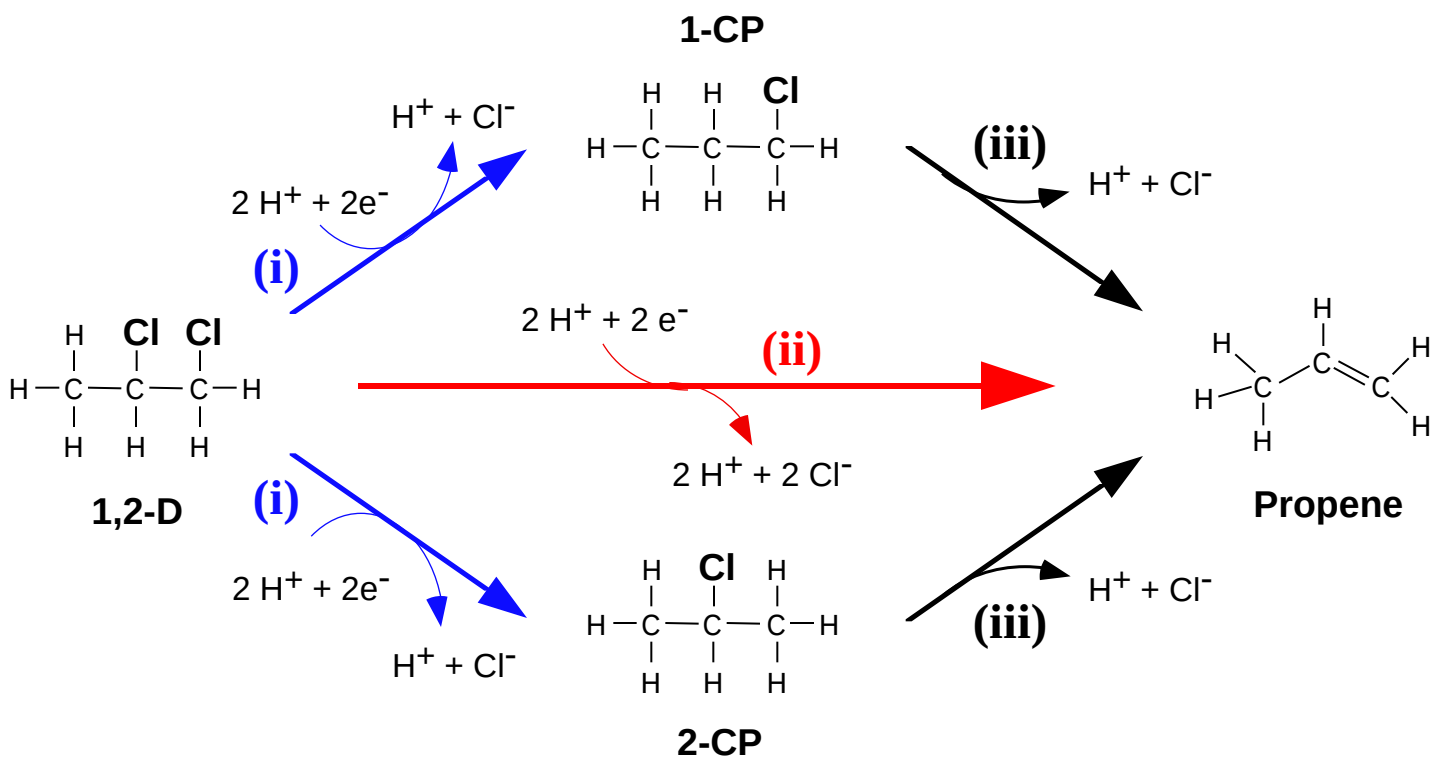
Reductive Dechlorination of PCE



* $\Delta G^{o\prime}$ kJ/chlorine released with H_2 as the electron donor

Anaerobic Dechlorination of 1,2-D



(i) Hydrogenolysis

(iii) Dehydrochlorination

(ii) Dichloroelimination

Introduction

Chlorinated solvents, including chlorinated propanes, tetrachloroethene (PCE) and trichloroethene (TCE), are common ground water pollutants. The stimulation of halorespiring microorganisms is potentially the most promising and cost-effective technology for remediating sites contaminated with chlorinated compounds.

Chlororespiration (Chloridogenesis) is a metabolic process in which halogenated compounds serve as terminal electron acceptors. Organisms with this specialized process capture the energy released in the reductive dechlorination process, and exhibit dehalogenation rates several hundred times faster than previously known for organisms that co-metabolize chlorinated compounds.

Halorespiration - Breathing Halogens (Chloridogenesis)

Referring to the characteristic of coupling oxidation of electron donor to reductive dehalogenation via a respiratory process used for obtaining energy for growth

Central to the successful implementation of halorespiration as a bioremediation technology is to determine whether organisms which can use reductive dehalogenation as a terminal electron accepting process (TEAP) are present at the site.

Indicators of Halorespiration

Increased biomass yields and ATP production in the presence of the chlorinated electron acceptor have been taken as proof for an organism's ability to perform halorespiration.

Here we present evidence that determination of f_e and hydrogen threshold measurements are useful tools to determine whether an organism is capable of halorespiration or if the dechlorination reaction is not linked to the organism's energy metabolism.

- Increased growth yields in the presence of the halogenated electron acceptor
- Increased ATP levels in the presence of the halogenated electron acceptor

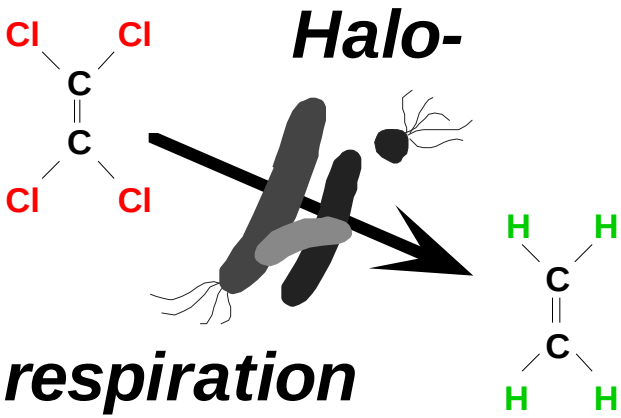
} Pure cultures
Defined consortia

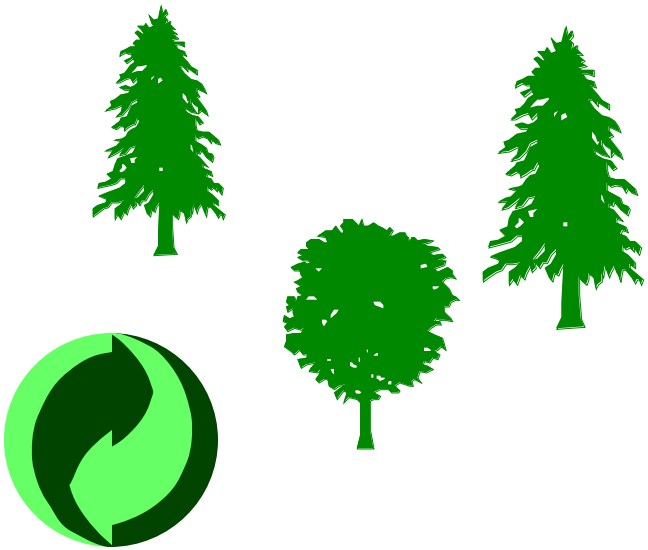
- Dechlorination rates
- Determination of f_e or f_s
- Hydrogen threshold measurements

} Pure cultures
Mixed cultures
Microcosms

Objectives

- Establish criteria useful to distinguish between co-metabolic and metabolic (halorespiring) dechlorination activities
- Establish parameters that are indicative of halorespiratory physiology



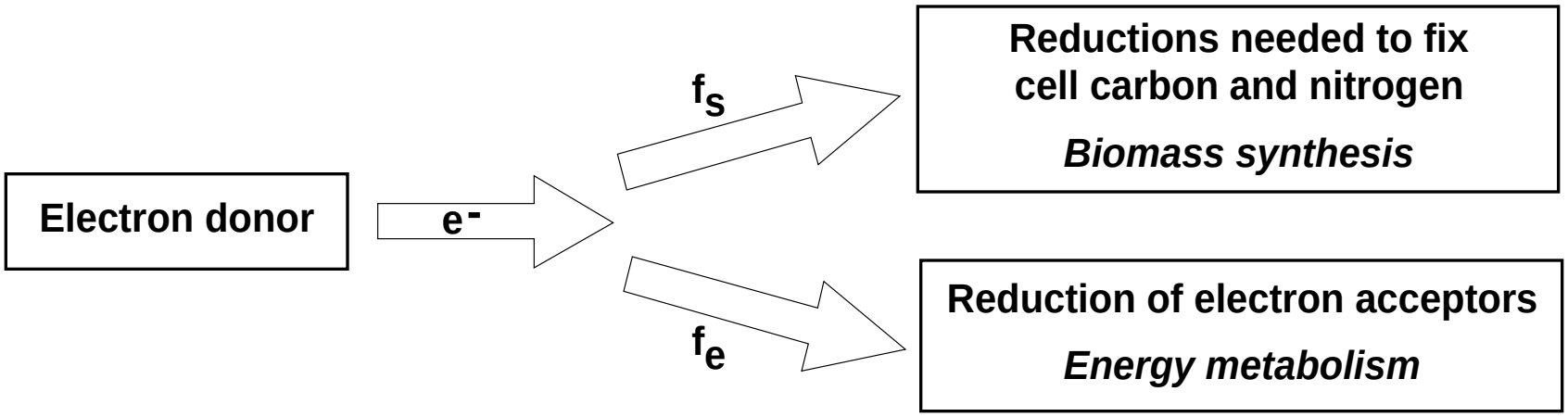


H_2 Threshold Values are Lower in the Presence of the Chlorinated Compound

Culture (enriched on)	H_2 Threshold concentration [ppmv]	
	No Chlorinated compound added	Chlorinated compound added
RC (1,2-D)	343 (±63)	0.25 (±0.17)
KS (1,2-D)	117 (±37)	0.30 (±0.14)
PM (PCE)	320 (±112)	0.21 (±0.16)
PM (*cis*-DCE)	439 (±85)	0.24 (±0.07)
PM (VC)	380 (±117)	0.20 (±0.05)
AuS (PCE)	319 (±86)	0.21 (±0.16)
AuS (*cis*-DCE)	368 (±88)	0.34 (±0.25)
AuS (VC)	253 (±61)	0.19 (±0.13)
RC (PCE)	334 (±25)	0.16 (±0.07)
RC (*cis*-DCE)	413 (±107)	0.21 (±0.08)
RC (VC)	368 (±62)	0.21 (±0.11)

Pathways for Reducing Power Consumption in Biological Systems - f_e and f_s

Electrons derived from electron donor oxidation are diverted to biomass formation (f_s) and energy generation (f_e) (modified after reference 1)



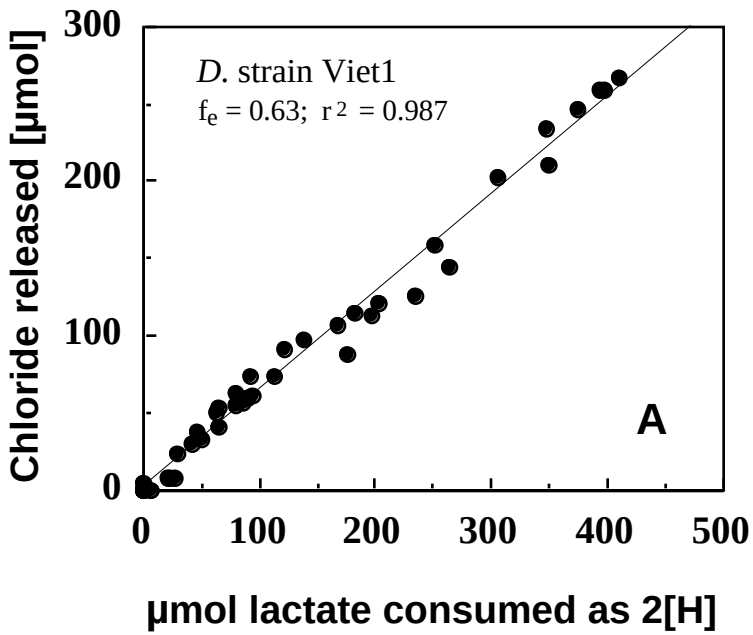
The f_e value indicates the fraction of electrons released in electron donor oxidation that is directed towards the reduction of the terminal electron acceptor.

The fraction of electrons derived from the electron donor that is incorporated into biomass is known as the synthesis fraction, f_s.

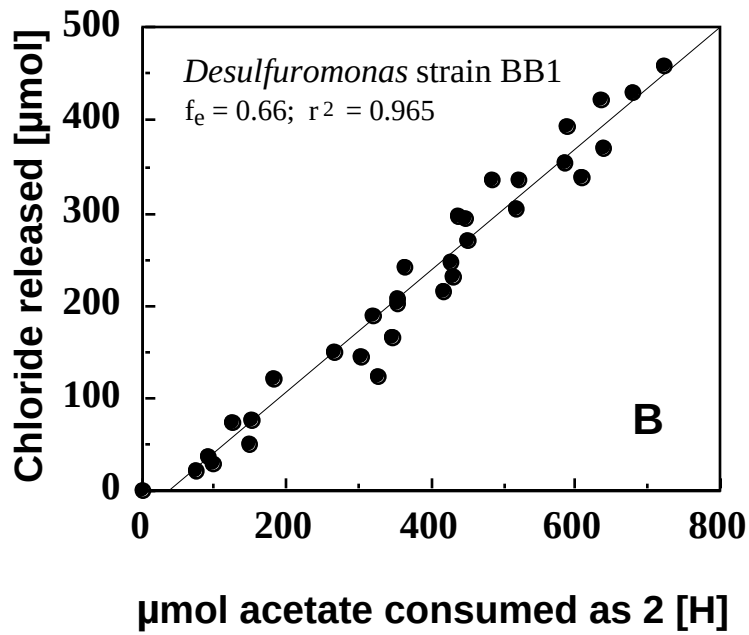
Graphical Determination of f_e

Values for f_e can be estimated from the slope of the regression line obtained by plotting the reducing equivalents (as 2[H]) generated in the oxidation of the electron donor against the amount of electron acceptor dechlorinated.

Desulfuromonas sp. BB1
PCE ⟶ *cis*-DCE

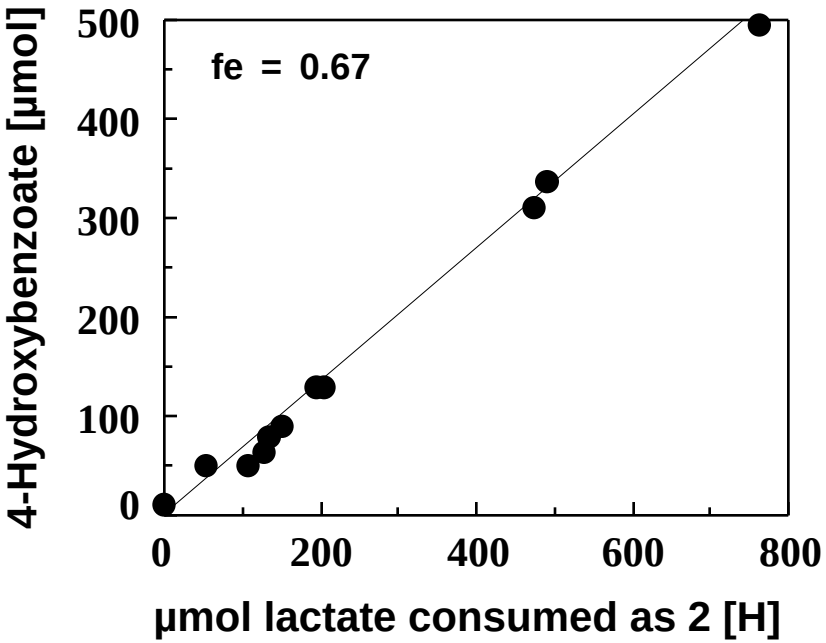


Desulfitobacterium sp. Viet1
PCE ⟶ TCE

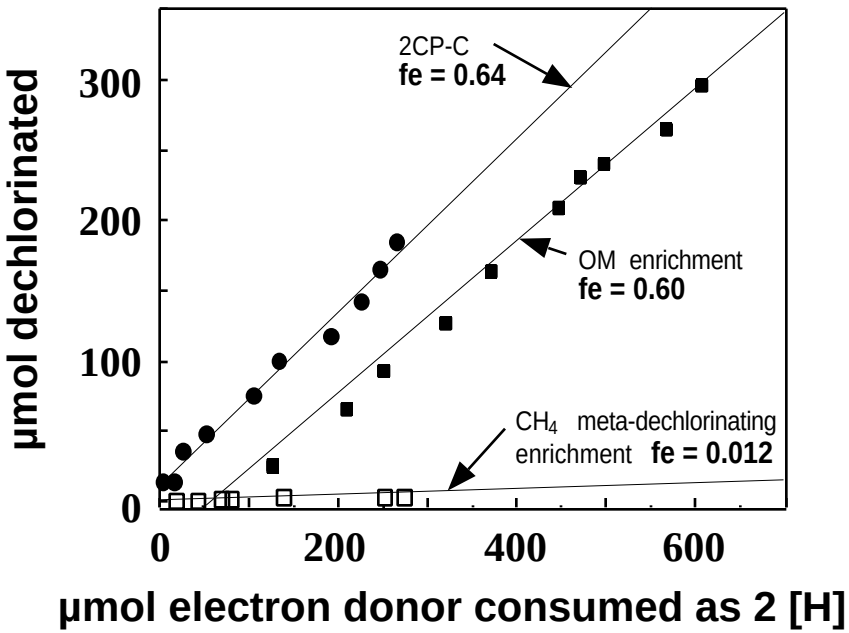


Desulfitobacterium chlororespirans

3Cl-4-HBA ⟶ 4-HBA



Strain 2CP-C	2-CP	⟶	Phenol
OM enrichment	2,5diCP	⟶	Phenol
CH_4 enrichment	3-CP	⟶	Phenol



Values for f_e were determined for four different cultures of halorespiring organisms and ranged from 0.63 to 0.67. In contrast, f_e values were below 0.05 in mixed methanogenic cultures that co-metabolized the chlorinated compound.

Conclusions

- **Hydrogen threshold measurements can indicate halo-respiration (chloridogenesis) as the dominant TEAP**
- **Halorespiring organisms reduce the hydrogen concentration below 0.4 ppmv in the presence of the chlorinated electron acceptor**
- **Hydrogen threshold measurements confirm that all chloroethenes, including VC, can be used as terminal electron acceptors**
- **Halorespiring organisms exhibit f_e values in the range of 0.6 to 0.7**

References

1. **Criddle, C. S., L. M. Alvarez, and P. L. McCarty.** 1991. Microbial processes in porous media, p. 639-691. In J. Bear and M. Y. Corapcioglu (ed.), Transport processes in porous media. Kluwer Academic Publishers, Dordrecht, The Netherlands.

2. **Löffler, F. E., J. M. Tiedje, and R. A. Sanford.** 1998. The fraction of electrons consumed in electron acceptor reduction (f_e) and hydrogen thresholds as indicators of halorespiratory physiology. Submitted to Appl. Environ. Microbiol.

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