

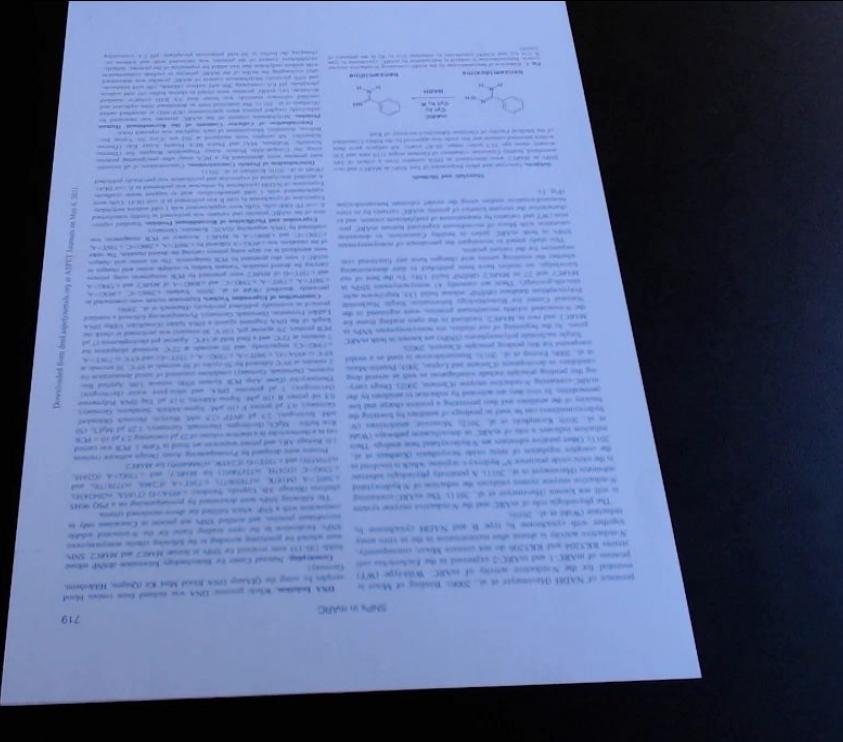
# Challenge Accepted

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## How to Escape the Quicksand While Engineering a Computer Vision Application

Bettina Heinlein

# The Idea



# Strategies

Test Early

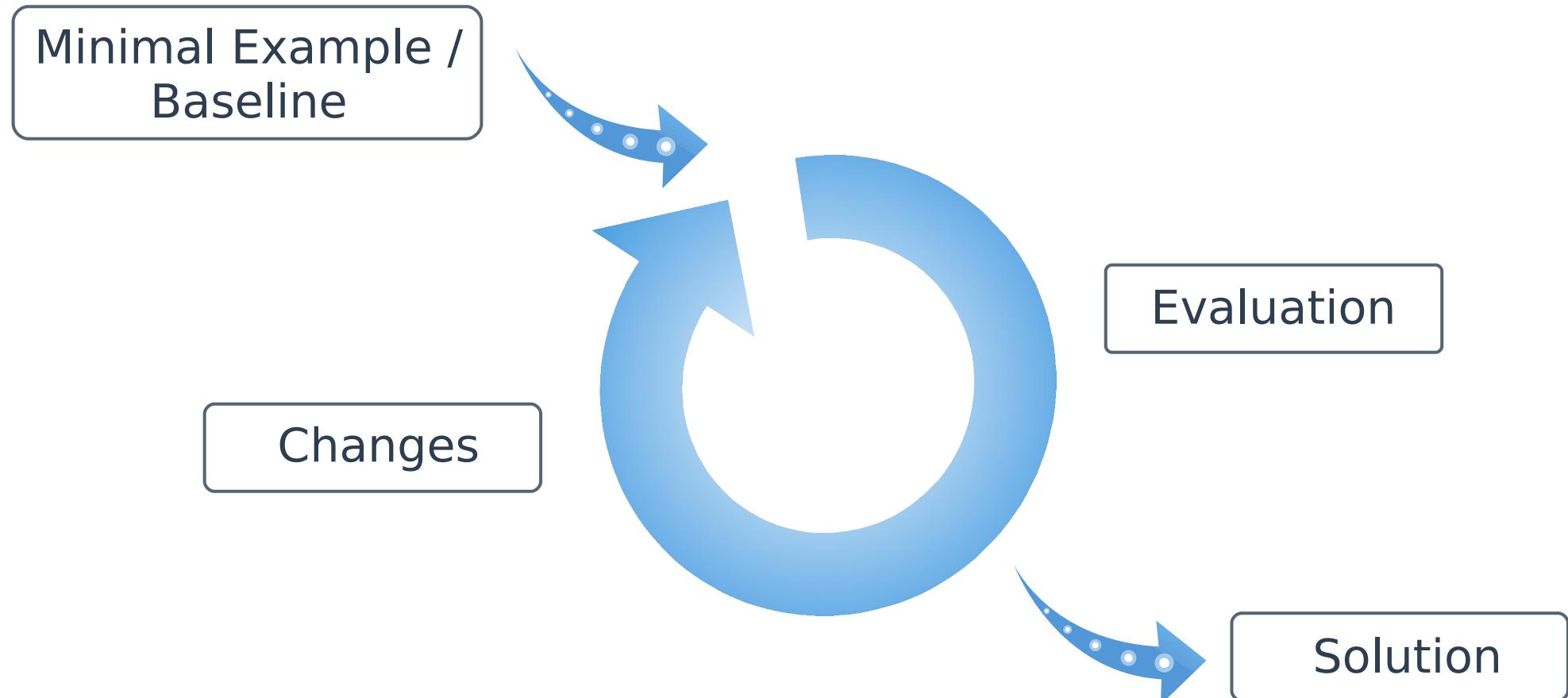
Update Mental Model

Fast-feedback Loops

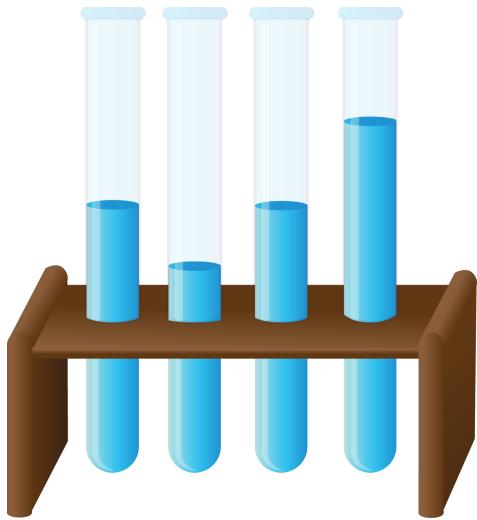
Gather Feedback

Evaluate Trade-Offs

# Fast-feedback Loops



# Early Testing with (Realistic) Data



# Question and Update Your Mental Model



**What am I  
missing?**



# Gather Feedback Early

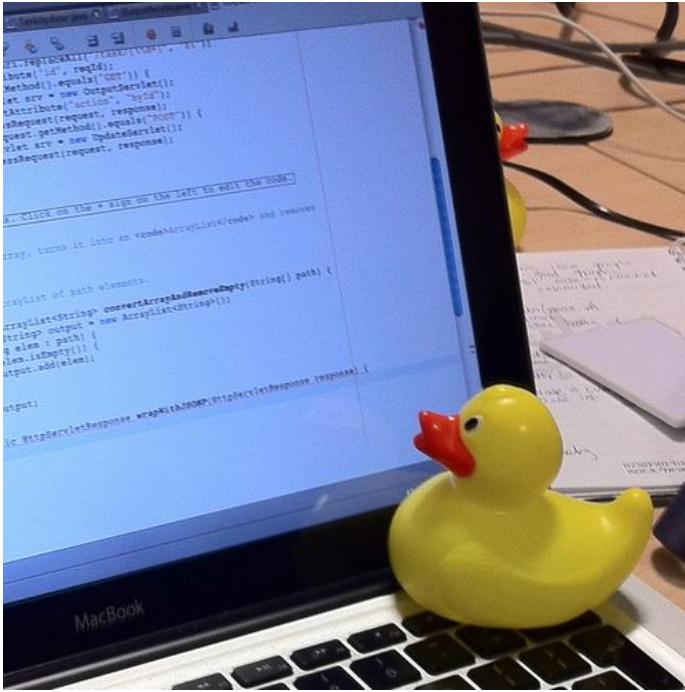
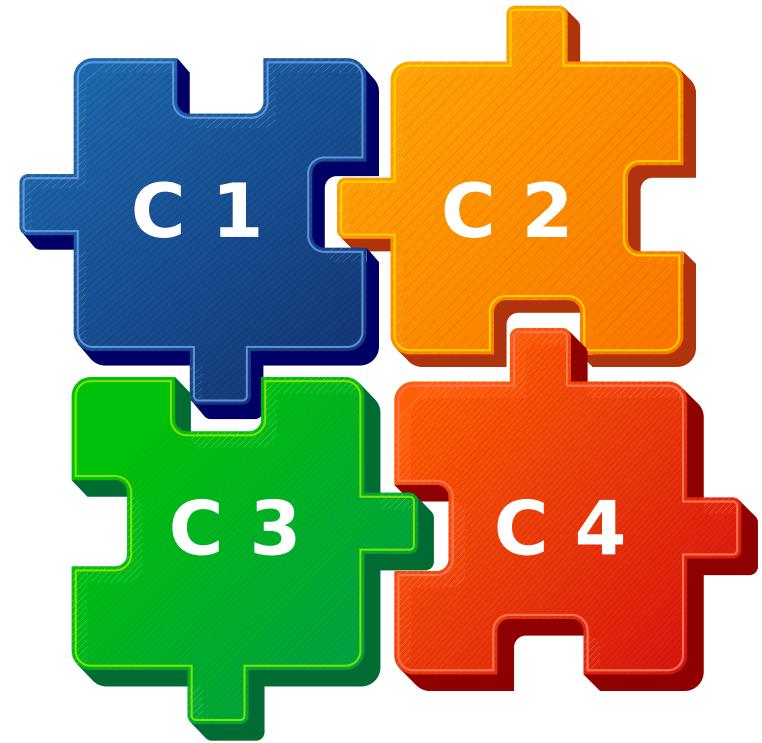
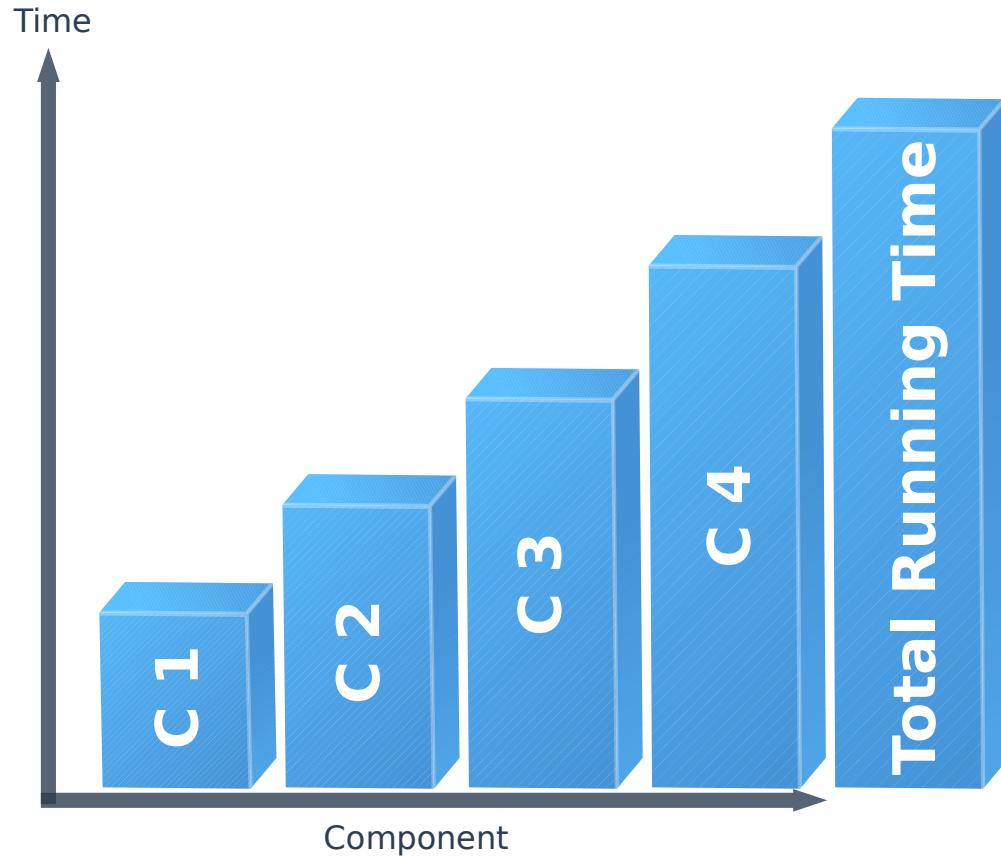


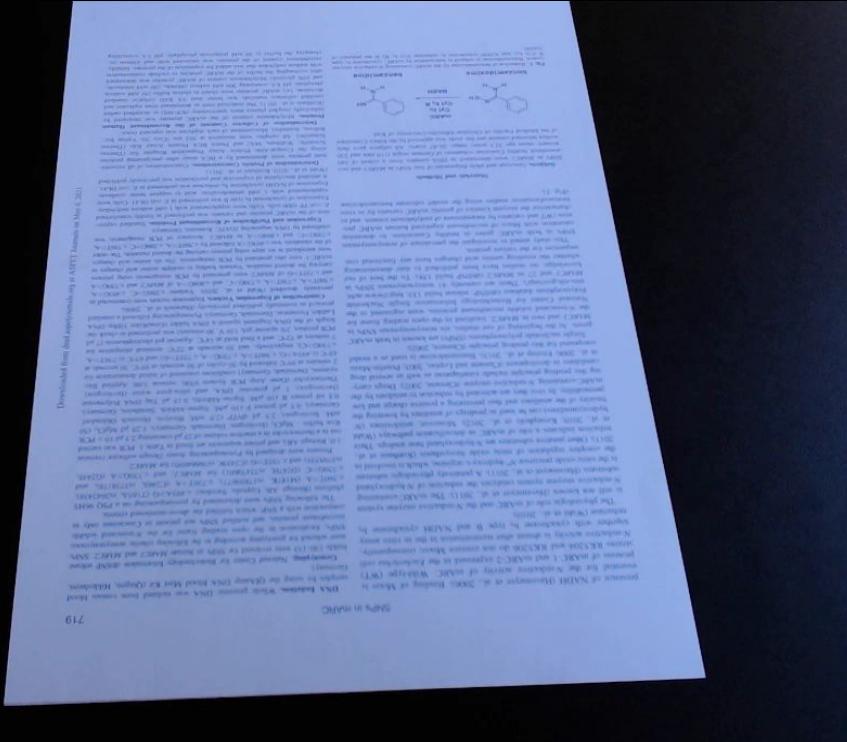
Image: CC BY-SA 3.0 License



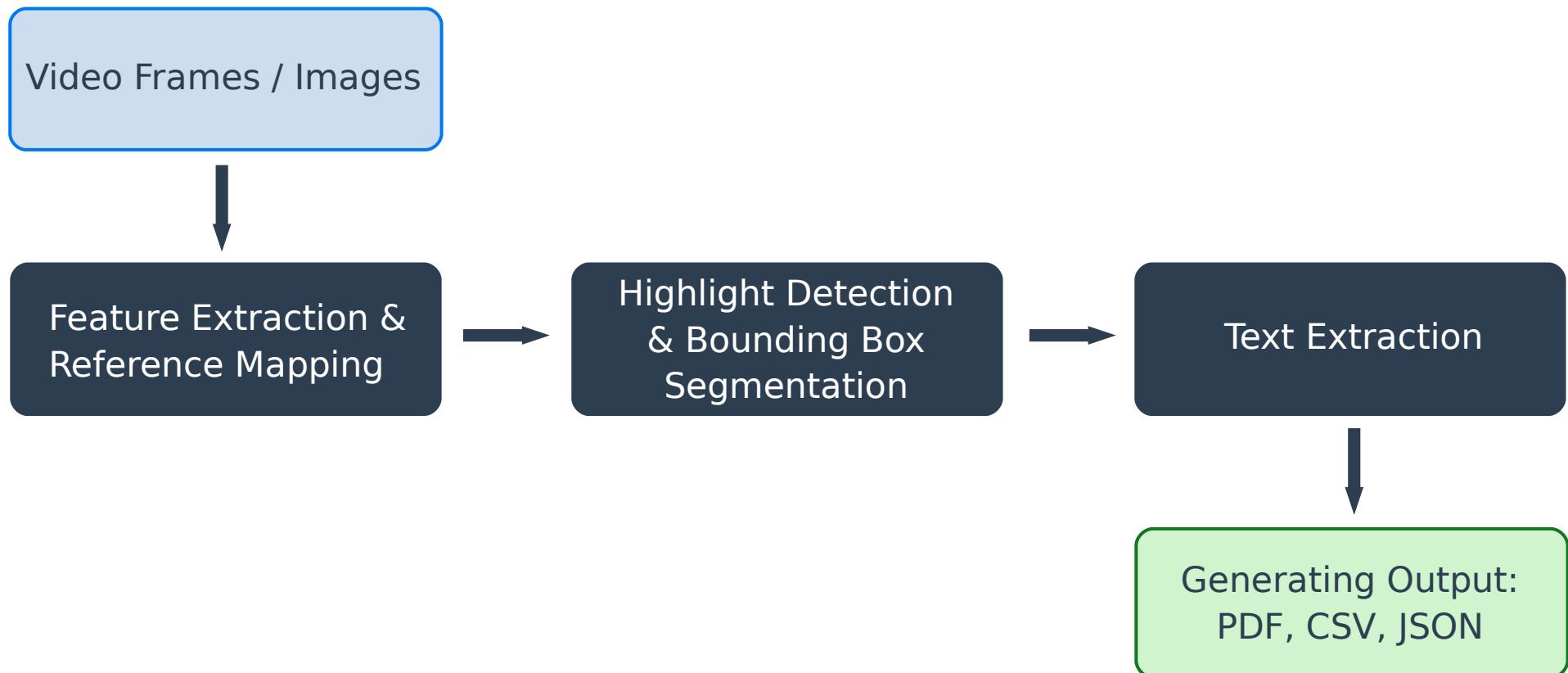
# Evaluate Trade-Offs (Mind the Priorities)



# The Idea



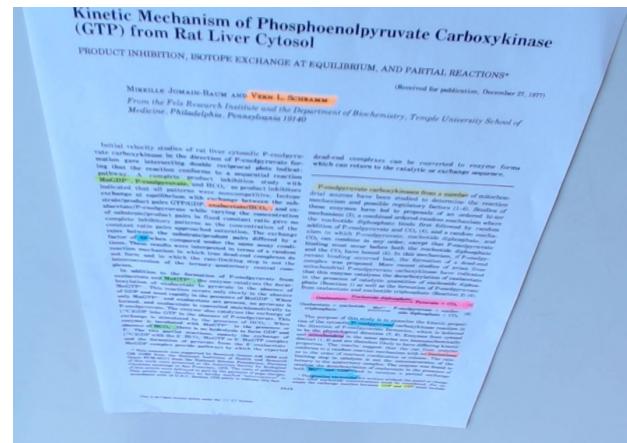
# Pipeline



# Mapping of Image to Reference Image

## Challenges

- Fast feature detection and extraction (given the camera placement)
- Identifying the reference image



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## Kinetic Mechanism of Phosphoenolpyruvate Carboxykinase (GTP) from Rat Liver Cytosol

PRODUCT INHIBITION. ISOTOPE EXCHANGE AT EQUILIBRIUM. AND PARTIAL REACTIONS\*

(Received for publication, December 27, 1977)

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Initial velocity studies of rat liver cytosolic P-enolpyruvate carboxykinase in the direction of P-enolpyruvate formation gave intersecting double reciprocal plots indicating that the reaction conforms to a sequential reaction pathway. A complete product inhibition study with MnGDP, P-enolpyruvate, and HCO<sub>3</sub><sup>-</sup>, as product inhibitors indicated all patterns were noncompetitive. Isotope exchange studies with oxalacetate and MnGDP, and with substrate/product pairs (GTP/GDP, malate/oxaloacetate/HCO<sub>3</sub><sup>-</sup>) and with MnGDP, P-enolpyruvate, while varying the concentration of substrate/product pairs in fixed constant ratio gave no complete inhibitory patterns as the concentration of the constant ratio pairs approached saturation. The exchange rates between the substrate/product pairs differed with dead-end complexes, can be converted to enzyme forms which can return to the catalytic or exchange sequence.

P-enolpyruvate carboxykinase from a number of mitochondrial sources have been studied to determine the reaction mechanism and possible regulatory factors (1-6). Studies of these enzymes have had to rely primarily on the substrate/product pairs (GTP/GDP, malate/oxaloacetate/HCO<sub>3</sub><sup>-</sup>) and ex-

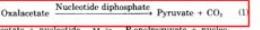
change of equimolar amounts of P-enolpyruvate and oxaloacetate/oxaloacetate while varying the concentration of substrate/product pairs in fixed constant ratio gave no complete inhibitory patterns as the concentration of the constant ratio pairs approached saturation. The exchange rates between the substrate/product pairs differed with dead-end complexes, can be converted to enzyme forms which can return to the catalytic or exchange sequence.

In addition to the formation of P-enolpyruvate from oxaloacetate and MnGTP, the enzyme catalyzes the decarboxylation of oxaloacetate to pyruvate in the absence of MnGTP. This reaction occurs only slowly in the absence of GDP and most rapidly in the presence of MnGDP. When only MnGTP<sup>-</sup> and oxaloacetate are present, no pyruvate is formed, and oxaloacetate is converted stoichiometrically to P-enolpyruvate. This enzyme also catalyzes the exchange of [<sup>14</sup>C]GDP for GTP in the absence of P-enolpyruvate. This exchange is stimulated by the presence of HCO<sub>3</sub><sup>-</sup>. When enzyme is incubated with MnGTP<sup>-</sup> in the presence or absence of HCO<sub>3</sub><sup>-</sup>, there is no hydrolysis to form GDP and P<sub>i</sub>. The two partial reactions, namely, the exchange of [<sup>14</sup>C]GDP with the E-HCO<sub>3</sub>-MnGTP or E-MnGTP complex and the formation of pyruvate from the E-oxaloacetate-MnGDP complex provide pathways by which the expected

\* To whom reprint requests should be addressed. Research Grants AG-16899 and GM-20888 from the National Institutes of Health and a Research Career Development Grant PCM-8017 from the National Science Foundation. Portions of this work were presented at the American Society for Biological Chemistry meetings in San Francisco, 1976. The costs of publication of this article were defrayed in part by the payment of page charges. This article must therefore be hereby marked "advertisement" in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.

dead-end complexes can be converted to enzyme forms which can return to the catalytic or exchange sequence.

P-enolpyruvate carboxykinases from a number of mitochondrial sources have been studied to determine the reaction mechanism and possible regulatory factors (1-6). Studies of these enzymes have led to proposals of an ordered bi-ter mechanism (3), a combined ordered-random mechanism where the substrates enter the active site one at a time, and a random mechanism (4) in which P-enolpyruvate, nucleotide, and MnGDP bind in a random fashion. These studies have been based on the assumption that the substrate enters before both the nucleotide and MnGDP. This may not be the case, however, since the nucleotide and the CO<sub>2</sub> can combine in any order, except that P-enolpyruvate binding must occur before both the nucleotide diphosphate and the CO<sub>2</sub> have bound (5). In this mechanism, if P-enolpyruvate binding occurred first, the formation of a dead-end complex would be avoided. MnGDP studies avoid liver cytosolic P-enolpyruvate carboxykinase have indicated that this enzyme catalyzes the decarboxylation of oxaloacetate in the presence of catalytic quantities of nucleoside diphosphate or oxaloacetate and nucleotide triphosphate (Reaction 2) (6).



The purpose of this article is to examine the kinetic properties of the cytosolic P-enolpyruvate carboxykinase reaction in the direction of P-enolpyruvate formation, which is believed to be the physiological direction (7, 8). Enzymes from cytosol and mitochondria in the same species are immunologically distinct (1, 2) and are therefore likely to have differing kinetic properties. The results suggest that the rat liver enzyme conform to a random reaction mechanism with no limitations to the rate of reaction. The enzymatic reaction is limited by the rate of P-enolpyruvate formation, which is the rate limiting step in catalysis is not the interconversion of the ternary to the quaternary complex. The enzyme was found to catalyze the decarboxylation of oxaloacetate in the presence of both Mn<sup>2+</sup> and GDP<sup>-</sup> and to catalyze a partial exchange of the carboxylic acid group by the phosphate group.

The kinase nucleotides are written without the metal or charge when total nucleotide concentrations must be considered. For example the exchange reaction between GDP and GTP must include

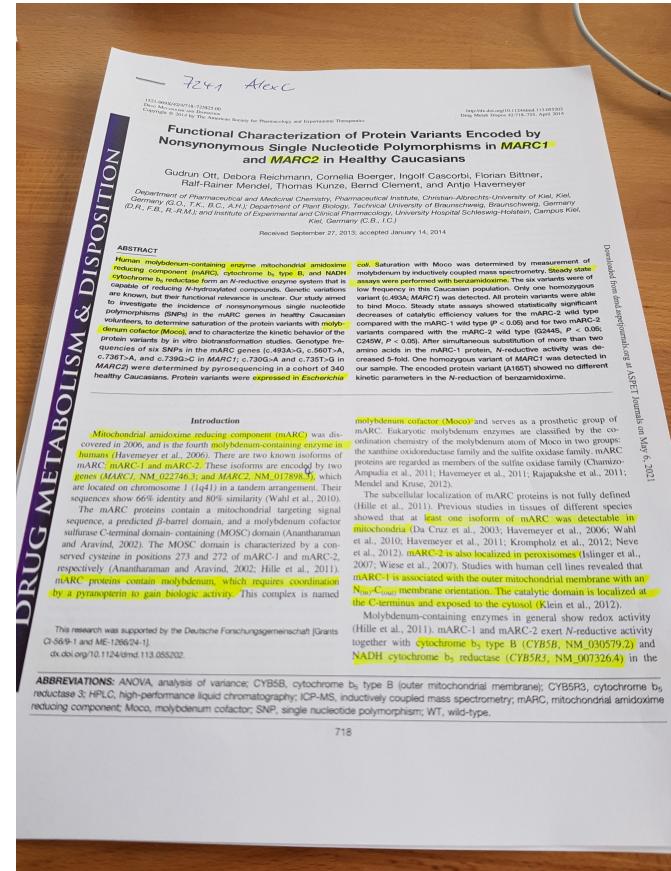
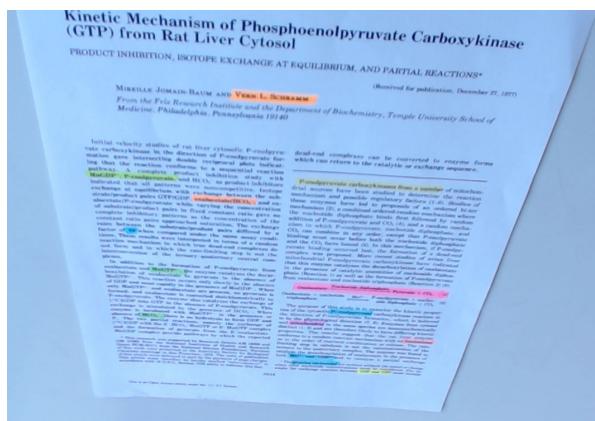
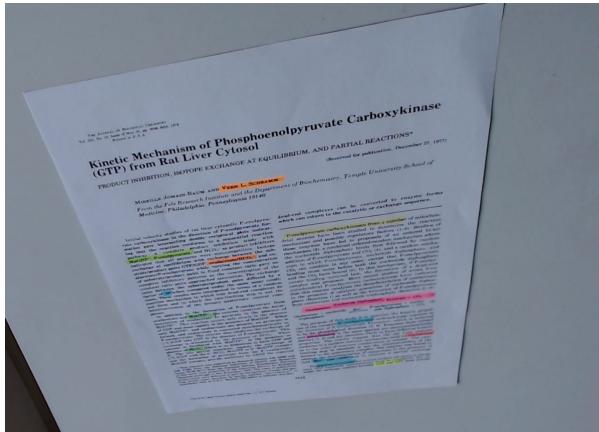
3648

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# Robust Highlight Detection

## Challenges

- Illumination
- Highlighter colors, work environment



# Text Extraction

## Challenges

- Extracting subscripted and superscripted characters

## Ground truth

oxalacetate/ $\text{HCO}_3^-$

## PDF Parsing

oxalacetate/ $\text{HCO}_{,,-}$

## OCR with Tesseract

oxalacetate/ $\text{HCO}_{,}$

# Strategies

Test Early

Update Mental Model

Fast-feedback Loops

Gather Feedback

Evaluate Trade-Offs

# Stay curious!

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