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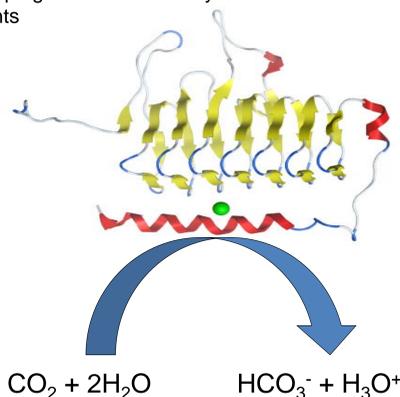
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Summary

Current CO₂ capture solvents for coal-fired power plants (e.g. MEA) give rise to unacceptably high energy losses due to the high energy needed to regenerate these solvents. Solvents that can be regenerated with much less energy (e.g. MDEA) are limited by slow capture rates, giving rise to prohibitively large absorption tower requirements. We aim to decrease this parasitic energy demand and decrease the impact on cost of electricity (COE) to <35% by developing low-cost biocatalysts for the acceleration of more desirable solvents



Codexis is applying state of the art biotechnology in an effort to develop low-cost catalysts for efficient carbon capture. Enzymes from nature catalyze CO₂ hydration in virtually all living systems, however these enzymes are not evolved to be stable or active in carbon capture solvents. Codexis' Directed Evolution technology has allowed development of low-cost industrial biocatalysts for the chemical manufacturing industry. Our recent work has now shown promise in applying this technology to carbon capture. Codexis is using ARPA funds in a program designed to create biocatalysts that are highly active and long-lived in desirable carbon capture solvents.

Approach

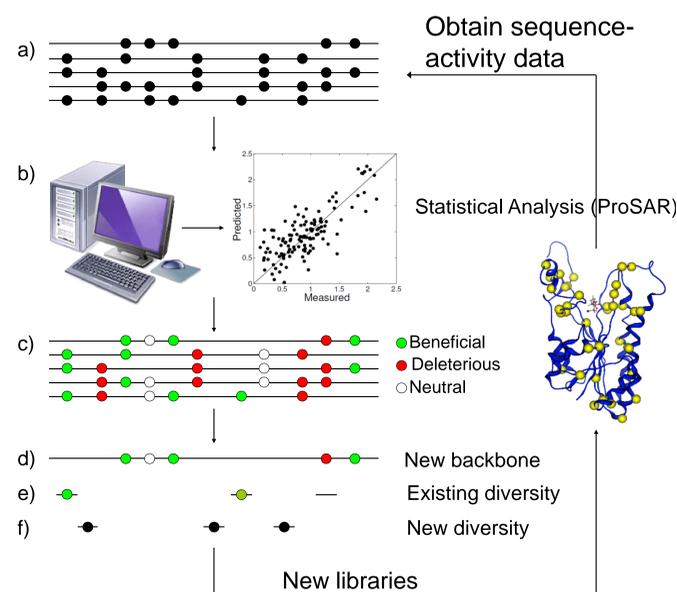
Acceleration of CO₂ hydration in tertiary amines such as N-methyl-diethanolamine (MDEA) is advantageous because these solvents provide a substantially lower heat of regeneration and use only one equivalent of amine (vs. 2 for MEA) to absorb CO₂. Thus tertiary amines such as MDEA can dramatically reduce the parasitic energy demand and increase the capture capacity. However, without a catalyst, absorption of gaseous CO₂ into aqueous tertiary and hindered amines is slow, leading to a need for large solvent volumes and prohibitively tall absorption units. In addition, MDEA is less corrosive, of higher capacity, more inert, and less volatile than MEA, so lower cost materials of construction and less solvent make-up may be required and allow for smaller absorber and stripper units.

The ability of a CA to accelerate the absorption of CO₂ into aqueous amines has been demonstrated for amines such as MEA, MDEA and AMP.¹ However, presently available CAs are woefully inadequate in terms of their cost, stability and activity in amine and carbonate capture solvents.

Solvent Characteristics	MEA	MDEA
Theoretical capacity (mol CO ₂ /mol amine)	0.5	1
Volumetric capacity (mol of CO ₂ /L solvent)	0.098	0.151
Heat of regeneration (kJ/mol CO ₂)*	167	100
Theoretical column height *(m)	15	600
Absorption rate (k ₂ at 25°C, M ⁻¹ s ⁻¹)	6000	5

* Heat of regeneration and column height from Aspen Plus® modeling.

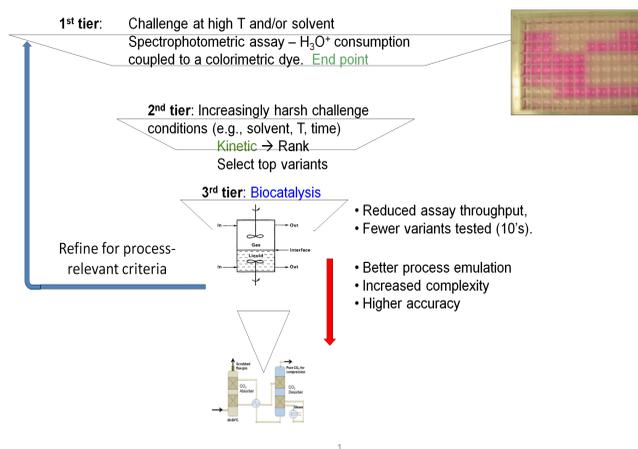
Carbonic anhydrases are some of the fastest known enzymes; each enzyme can catalyze the hydration of up to 1 million CO₂ molecules per second² and so, under capture conditions, CO₂ capture could be as much as three orders of magnitude faster than MEA. In principle, CA may be used with any basic aqueous solution (to neutralize H⁺) to accelerate the capture of CO₂ as HCO₃⁻. Thus, our approach is to use state of the art enzyme evolution technology to create CA's that are highly active and stable under carbon capture conditions. Codexis has applied these methods to the creation of enzymes for the chemical manufacturing industry. Codexis has successfully used its advanced directed evolution technology to increase the activity of other enzymes by more than 6 orders of magnitude in mixed aqueous-organic media,³ enabling several commercial manufacturing processes. We are now applying this innovative approach to CA activity and stability.



Our approach to the directed evolution starts with a diverse set of genes that encode for mutational variations (random, natural and computationally designed from enzyme structure) of a target biocatalyst. We recombine (shuffle) these mutations to produce new biocatalyst variants (see Figure above). By screening a wide range of genetic variants under process relevant conditions in a tiered fashion (see Figure below), we are able to make very large changes in enzyme function and stability by accumulating beneficial genetic diversity over several rounds of evolution. From each round of evolution, the genes coding the best biocatalyst variants are put back into the next round of evolution and recombined combinatorially to accumulate the beneficial mutations until biocatalysts are created that meet or exceed performance targets. Codexis has made large investments in the development and purchase of robotics and high HTP instrumentation and sample handling allowing the testing of 1000's of genetic variants and to enable ProSAR, our proprietary bioinformatics technology.

Recombination of multiple mutations by DNA shuffling, coupled with statistical analysis of protein sequence-activity relationships (ProSAR) allows for very large modifications in biocatalyst structure and function. Such major changes are needed in cases where the process reaction conditions are a drastic departure from the natural environment in which the wild-type (WT) biocatalyst(s) evolved; in this case to obtain a highly stable CA in high concentrations of organic amine and elevated temperatures.

CA screening: tiered approach

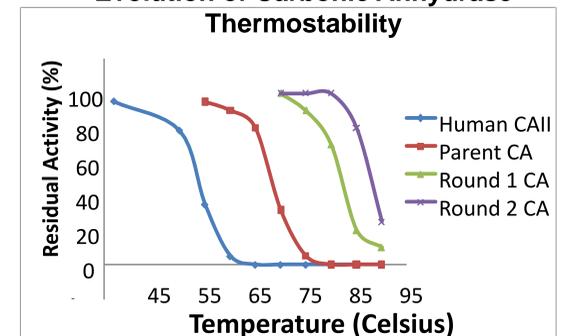


The three elements of biocatalyst cost are: 1. activity (loading), 2. stability (lifetime) and 3. fermentation titer (manufacturing cost). We will use our directed evolution technology to increase the activity and stability of the enzyme and in parallel, we will integrate our acceleration catalyst into existing solvent based carbon capture process designs so that simple retrofitting of existing coal-fired power plants will be made possible. We will use our proprietary strain engineering technology and industrial enzyme production strain to develop a high titer fermentation process which will allow us to produce the biocatalyst at large scale and low cost.

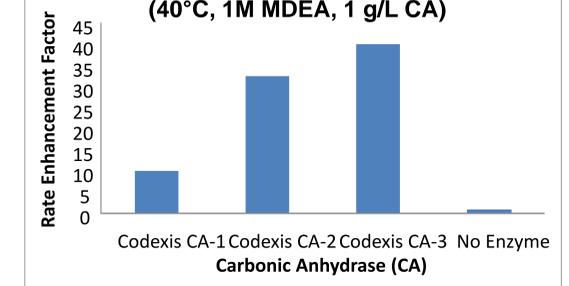
Results of our proof of concept studies to increase thermostability, activity and stability under carbon capture conditions are shown in the following 3 figures.

Proof of Concept Results

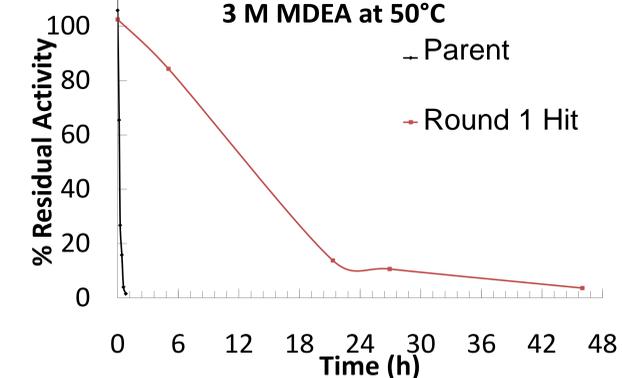
Evolution of Carbonic Anhydrase



CO₂ Absorption Acceleration (40°C, 1M MDEA, 1 g/L CA)



Stability of Top CA Evolvent (3 M MDEA at 50°C)



Key Milestones and Deliverables

Year 1	<ul style="list-style-type: none"> Projected COE < 50% based on catalyst performance Develop biocatalyst with > 1 week stability under target capture conditions
Year 2	<ul style="list-style-type: none"> Develop biocatalyst with > 3 months stability Produce 100 kg enzyme for pilot plant runs Projected COE < 35% based on catalyst performance

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1. Voyer, N.; Fradette, S. In 7th CCS Conf, Pittsburgh, PA, May, 2008; Exchange Monitor Publications: Pittsburgh, PA, 2008.
 2. Khalifah, R.; Silverman, D. N., Carbonic Anhydrase Kinetics and Molecular Function, The Carbonic Anhydrase In Plenum Press: New York, 1991; pp 49-64.
 3. Luetz, S.; Giver, L.; Lalonde, J., Engineered enzymes for chemical production. *Biotechnology and Bioengineering* 2008, 101, (4), 647-653.
 Acronyms COE: Cost of electricity, MDEA: monomethyl,-diethanolamine, MEA: monoethanolamine, ProSAR: Protein structure activity relationship.

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