The presence of an oxalacetate carboxylase in parsley root has been re-
ported previously from this laboratory. The reaction catalyzed by this
system has been found to be reversible, since it fixes C\textsubscript{14}O\textsubscript{2} into oxalacetate
during the decarboxylation of the latter in the presence of NaHCO\textsubscript{3}.

The enzyme was prepared essentially as previously reported and com-
pletely freed from pyruvic carboxylase by prolonged dialysis. The prep-
aration was lyophilized and made up to the desired strength by dissolving
in water. The decarboxylation reaction, measured in 0.1 M acetate buffer,
pH 5.0, in the presence of 0.01 M MnCl\textsubscript{2}, follows first order kinetics. The
difference between the first order rate constants, \( k = (2.303/t) \log (C_0/C) \), observed with active and with heat-inactivated enzyme, divided
by mg. of enzyme preparation, may be used as a measure of activity. For
the enzyme employed in the experiments reported, \( k = 0.075 \text{ min}^{-1} \) per
mg.

The exchange experiments with oxalacetate were conducted by the pro-
cedures recently described. The initial reaction mixture contained 1.2
\( \times 10^{-2} \) M phosphate buffer, pH 6.0, 10\textsuperscript{-3} M MnCl\textsubscript{2}, 4.25 \( \times 10^{-2} \) M oxalacetate, 3.6 \( \times 10^{-2} \) M NaHCO\textsubscript{3} containing C\textsubscript{14}, and 180 mg. of enzyme in a
total volume of 10 ml. The reaction was incubated at 30\textdegree for 8 minutes,
at which time approximately half of the oxalacetate had been decarbox-
ylated. The activity of the \( \beta \)-carboxyl carbon liberated by aniline citrate
was at this time found to be 0.49 \( \pm 0.02 \) per cent of the activity of the
NaHCO\textsubscript{3} of the medium at the end of the incubation period. The control
run with the heat-inactivated enzyme showed no detectable fixation after
30 minutes incubation.

The enzyme preparation used in these experiments contains a malic
dehydrogenase active with triphosphopyridine nucleotide. This activity
can be demonstrated by measuring the reduction of 2,6-dichlorophenol
indophenol. It can also be detected by measuring the reduction of cyto-
chrome c in the presence of cytochrome reductase and TPN. In a typical
experiment of the latter type, the reaction mixture initially contained 3.7
\( \times 10^{-3} \) M glycylglycine, pH 7.4, 3.7 \( \times 10^{-4} \) M MnCl\textsubscript{2}, 1.65 \( \times 10^{-5} \) M TPN,

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1 Vennesland, B., and Felsher, R. Z., Arch. Biochem., 11, 279 (1946).
500 \gamma of cytochrome reductase, \(4.59 \times 10^{-5} \text{M cytochrome c, } 5.5 \times 10^{-4} \text{M L-malic acid, and 1.5 mg. of enzyme in a total volume of 1.35 ml. At 26^\circ, the specific reaction rate constant for the first order reduction (per mg. of enzyme used) was 0.038 \text{min}^{-1} \text{corrected for the control without malic acid.}

In accordance with expectations, the parsley root preparation was found to be capable of fixing CO\(_2\) in the malate fraction when pyruvate and malate were incubated with C\(^{14}\)O\(_2\). There is no oxidation-reduction between pyruvate and malate, since the preparation contains no lactic dehydrogenase. Fumarase is likewise absent.

These results suggest that the synthesis of the plant dicarboxylic acids may occur by way of an initial Wood-Werkman reaction, as has been found to be the case with bacteria and animal tissues.\(^3\)

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