The *Clostridium* species are gram-positive, spore-forming rods, either strictly anaerobic or microaerophilic. Bacteria in the genus differ in their fermentations. *Clostridium butyricum*, synonymous with *C. saccharobutyricum*, typically produces the following from 100 moles fermented glucose: 233 moles hydrogen, 196 moles carbon dioxide, 43 moles acetic, and 75 moles butyric acid. Other species of the genus producing large quantities of butyric acid are: *Clostridium amylobacter*, *Clostridium pasteurianum*, and *Clostridium lactoacetophilum*, which, as the name implies, is active toward lactate in the presence of acetate.

Closely related to butyric is the butylic fermentation. Butylic clostridia—*C. butylicum*, *Clostridium acetobutylicum*, and others—can continue butyric fermentation to a more advanced stage, leading to production of butanol and other volatile solvents.

Butyric Acid Fermentation.—In the formation of butyric acid, C_3 compounds may be utilized (glycerol by *C. acetobutylicum*, pyruvate by *C. butylicum*, and lactate by *C. lactoacetophilum*). Consequently it appears that butyric acid formation is not a direct result of the splitting of a hexose into C_4 and C_2 constituents.

In the light of the demonstrated importance of coenzyme A in the oxidation of butyric acid, Barker[19] formulated the following series of reactions:

$$2\,CH_3\cdot CO\cdot COO^- + 2CoA \underset{+2CO_2}{\overset{-2CO_2}{\rightleftharpoons}} 2\,\underset{\text{acetyl-CoA}}{CH_3\cdot CO\sim CoA} \underset{+H_2O}{\overset{-H_2O}{\rightleftharpoons}} \underset{\text{acetoacetyl-CoA}}{CH_3\cdot CO\cdot CH_2\cdot CO\sim CoA} \quad \text{(R55)}$$

$$CH_3\cdot CO\cdot CH_2\cdot CO\sim CoA \underset{-2H}{\overset{+2H}{\rightleftharpoons}} \underset{\text{vinylacetyl-CoA}}{CH_2{=}CH\cdot CH_2\cdot CO\sim CoA} \text{ or } \underset{\text{crotonyl-CoA}}{CH_3\cdot CH{=}CH\cdot CO\sim CoA} + H_2O \quad \text{(R56)}$$

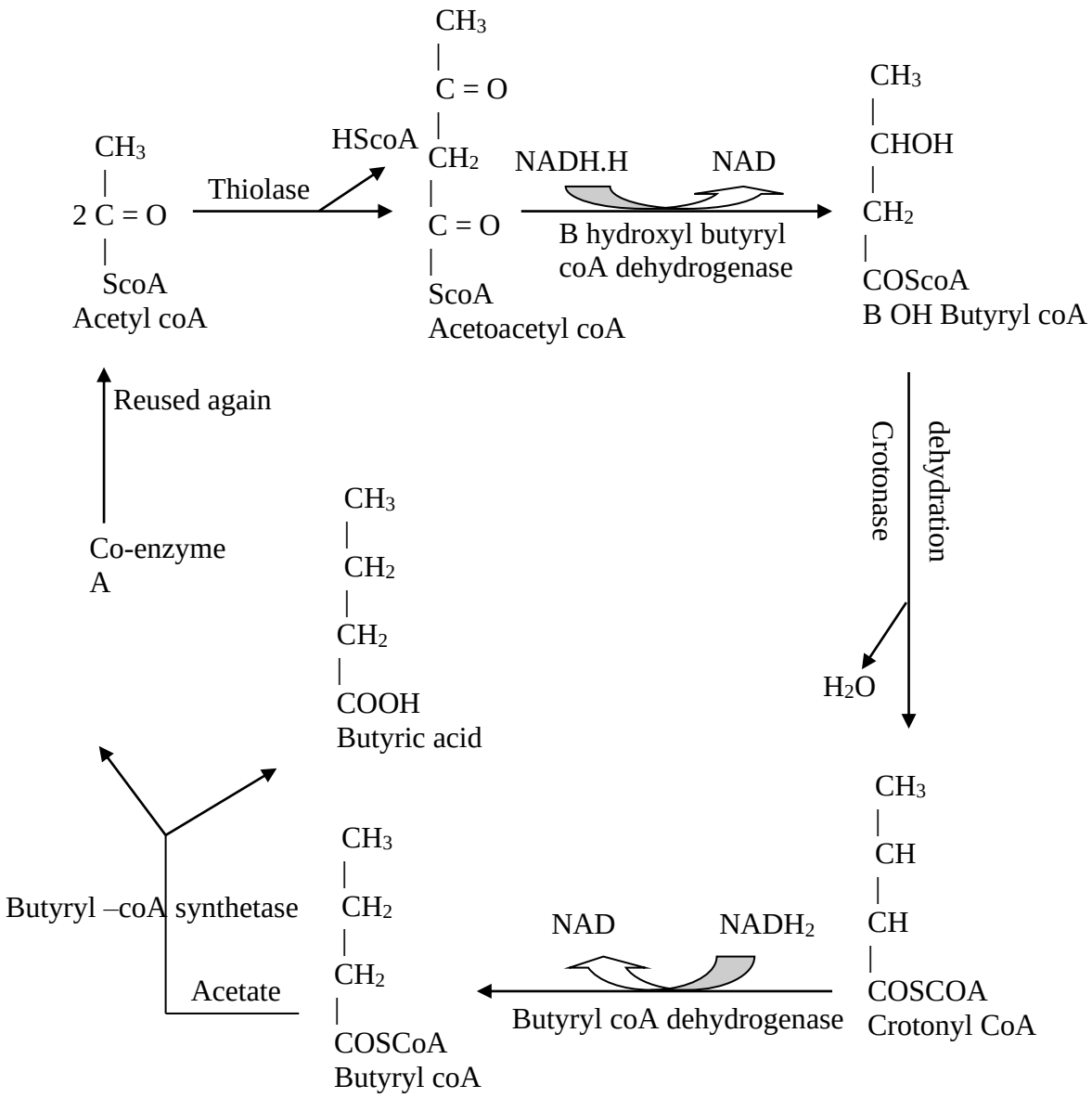
$$\text{vinylacetyl-CoA} \underset{-2H}{\overset{+2H}{\rightleftharpoons}} \underset{\text{butyryl-CoA}}{CH_3\cdot CH_2\cdot CH_2\cdot CO\sim CoA} \quad \text{(R57)}$$

$$\text{butyryl-CoA} \rightleftharpoons \text{butyrate} + \text{CoA} \quad \text{(R57a)}$$

Attempts to demonstrate the conversion of pyruvate to butyrate met with difficulties. *C. butylicum* converts pyruvate into acetate, CO_2 and H_2 at an optimum pH of 5.0. Cell-free extracts, in the presence of phosphate, form acetyl phosphate, CO_2, and H_2.

The mechanism postulated for conversion of acetate to butyrate also explains conversion of lactate to butyrate by *C. lactoacetophilum*. Acetate is required and the condensation again involves CoA, and is mediated by an independent condensing apoenzyme. One mole of added acetate is consumed per mole of butyric acid formed.

In the formation of butyric acid from lactic acid by *Butyribacterium*



Butyric acid fermentation:

In the light of the demonstrated importance of coenzyme A in the oxidation of butyric acid, Barker formulated the following series of reactions:

$$2CH_3.CO.COO + 2CoA \xrightarrow[+2CO_2]{-2CO_2} 2CH_3.CO.CoA \quad (Acetyl\text{-}coA)$$