SUMMARY

The accumulation of citric acid in the mature lemon has been attributed to a block in the TCA cycle at the aconitate level. As respiration continues at this stage, this raises a question regarding its mechanism. The possibility suggests itslef that as the fruit matures protein may be broken down into aminoacids which are utilized for respiratory purposes. If this is the case we should expect changes in the contents of protein and aminonitrogen and in the activities of enzymes concerned with carbohydrate and aminoacid metabolism. The present studies were designed to test this hypothesis. Fruit tissues of Citrus acida were investigated at two stages of development for (a) enzymes and intermediates of glycolytic cycle (b) protein, aminonitrogen and free aminoacids and (c) activities of enzymes concerned with glutamate metabolism.

Data obtained on these studies show that the increase in citric acid content in the mature fruit is associated with decreases in total sugar, free sugar, glucose, sugar phosphates and lactic acid. Glucose-1-phosphate could not be detected in mature fruit tissue.

Data on the specific activities of the glycolytic enzymes as well as pyruvate dehydrogenase and citrate synthetase in young and mature fruits show that although hexokinase activity

increases in the mature fruit the rate of glycolysis is decreased. Both pyruvate dehydrogenase and citrate synthase contents increased. This pattern is consistent with the operation of the glycolytic cycle and the formation of citric acid in the fruit.

In a separate study in this department it is found that enzymes involved in the formation of ascorbic acid from glucose via glucose-1-phosphate and D-glucuronolactone are very active in young fruit tissues. It is possible that the glycolytic activity in young fruit tissues helps in generating glucose-1-phosphate required for ascorbic acid formation.

Since aconitase is absent in the mature fruit the question arises regarding the mechanism of respiration in the mature fruit tissues. It is found that during this stage protein nitrogen decreases and aminonitrogen, glutamic acid and aspargine increase. It is also found that glutamic acid can be used for respiration by the mature fruit tissue. While the addition of glutamate has no effect on oxygen uptake in the young fruits it increases the same by 50 per cent in mature fruit. Addition of glutamic acid increases the formation of ammonia but when aspartate is also added this increase is reversed accompanied by an increase in asparagine content. The specific activities of the enzymes concerned with the breakdown of protein and the utilization of glutamate are high in the mature fruit.

The results of these studies suggest that the mode of respiration of the mature fruit is by the conversion of glutamate to 2-oxo-glutarate which gets further oxidised by the Krebs cycle. The formation of asparagine from aspartic acid and ammonia may be the mechanism of detoxification by which the ammonia formed by this reaction is removed.

Glutamate dehydrogenase, glutamate decarboxylase, alanine and aspartate aminotransferase were partially purified. Citrate, which accumulates in mature fruit, does not seem to have any effect on these enzymes.

Glutamate dehydrogenase seems to be a sulphydryl enzyme. It resembles the enzyme from other plant sources regarding Km and optimum pH and that from animal sources regarding heavy metal inhibition.

Glutamate decarboxylase resembles the enzyme isolated from other plant sources like squash regarding optimum pH. PCMB has no effect on this enzyme suggesting that it is not a sulphydryl enzyme.

While alanine aminotransferase resembles in general the enzymes isolated from plant and animal sources aspartate aminotransferase resembles that from hog in its optimum pH. These enzymes when heated for 2 minutes at 60^{o} and then dialysed lose their activities which can be restored on addition of pyridoxal phosphate. Both are found to be sulphydryl enzymes.