1.1 INTRODUCTION

By common usage, wall-less plant cells with its usual contents intact is called a protoplast. Due to the recent advances in the technique of removing plant cell wall by enzymatic hydrolysis, it is now possible to obtain viable plant protoplasts from a variety of tissues. The protoplasts like intact cells are found to be totipotent and can regenerate the cell wall, divide and redivide mitotically to form a callus which on differentiation may give rise to an intact plant. Since the protoplasts are naked cell systems, a variety of foreign substances ranging from macromolecules to cell organelles and intact microorganisms can be effectively inserted in to the protoplasts (vide Reinert and Bajaj, 1977). Further, the protoplasts can be induced to undergo fusion which is a physiochemical process and is mostly independent of the type of protoplast taking part.

In addition to the intact protoplasts, various types of incomplete or subprotoplasts like vacuoplasts, cytoplasts and miniprotoplasts can also be produced experimentally (vide Korn, 1974, Lorz et al., 1976, Wallin et al., 1978). These miniprotoplasts, cytoplasts and vacuoplasts are of immense help in selectively transferring the genetic factor without the cytoplasm or vice versa into a protoplast through fusion.

Besides, such subprotoplasts obtained from fruits have also been used for fusion experiments (Binding and Kollmann, 1976). All these structures have a common feature in that they are membrane-bound and sensitive to osmotic pressure of the medium. Their potentiality in cellular engineering are also recognized.

Comparable to the protoplasts and subprotoplasts is a group of subcellular structures which are formed naturally in the ripening fruits. These fruit protoplasts are extremely diverse in their origin, content, nature and dimension.

In tomato such fruit protoplasts arise from the parenchymatous interplacental tissue after pollination (Smith, 1935). The cell multiplication ceases early and as the fruit develops the most pronounced effect is the softening of this tissue during ripening finally resulting in the breaking of the cells releasing their contents in to the locular cavity. The locular sap of a ripening tomato fruit thus contains numerous free cells, protoplasts, subprotoplasts, cytoplasts and many spherical membrane bound units of diverse nature. The occurrence of similar membrane-bound protoplasmic units have also been observed in case of other solanaceous berries (Raj and Herr, 1973) and in the ripening fruits of grapes, plum, strawberry, cherry and asparagus (Cocking and Gregory 1963). The origin of these membrane-bound units are not fully understood. The locular sap of the ripe tomato fruit therefore can be considered as a reservoir of naturally released protoplasts and various types of protoplasmic units, in contrast to the enzymatically isolated protoplasts.

The role of various cell wall hydrolytic enzymes have been attributed for the softening and the degeneration of the interplacental tissue of tomato. The work of Foda (1957), Hobson (1963, 1964), Raj and Herr (l.c) showed that pectic enzymes like pectin esterase and polygalacturonase play a significant role in softening of the fruits during ripening. Although the occurrence of cellulase in tomato fruits has been detected by Hall as early as 1963, its role in fruit softening still remained controvertial. Hobson (1968) concluded that in tomato fruits cellulase plays a minor role in softening, where as Sobotka and Watada (1970) emphasized the role of cellulase in fruit softening.

The present investigation, was carried out to study the various aspects of the interplacental tissue, including

i) the ultrastructure,

ii) detection and histochemical location of endogenous cellulase activity,

and iii) original properties of the protoplasmic units.