MATERIALS AND METHODS

SELECTION OF SPECIES:

Timber yielding tree species Tectona grandis Linn., Acacia nilotica Del. Azadirachta indica A. Juss. and Tamarindus indica L. were selected in the present study. Except Tectona all the species are widely distributed in moist deciduous forest (Dangs forest-waghai), dry deciduous forest (Pavagadh) and scrubland forest (Kutch ie. Bhuj and Nakhatrana). Tectona grandis is not grown in the area of Kutch, where the climatic features like temperature, humidity and annual rainfall are either extreme or minimal.

COLLECTION OF SAMPLES:

Cambial tissues together with the inner phloem and outer xylem was removed from the main trunks of trees of similar age and trunk diameter at a height of about 1.5 meter from the ground using chisel, hammer and grafting knife Each time two trees were sampled to obtain four blocks and no tree being sampled more than once. Samples removed were 60 X 40 mm in length and width respectively and deep enough to include both the current season's growth and the whole of the previous season's annual ring. Monthly samples from each site were taken during first and second week of every month from January 1994 to December 1994. After each collection, the wounds were covered with sealing wax to prevent the entry of pathogens. Simultaneously small pieces of 2-3 years old branches measuring about one cm in diameter were also collected from each

species. Soon after the collection, the samples were fixed in FAA (Berlyn and Miksche 1976) and aspirated shortly afterwards in the laboratory Then samples blocks were cut into pieces approximately 20X20X20 mm with small hacksaw and after a week in FAA, the pieces were transferred to 70% ethanol in a air tight containers for preservation. At the time of each sampling the phenological details of each species were recorded. The meteorological data on maximum and minimum temperature, rainfall and relative humidity for the year 1994 for all the three regions were collected from the Indian Meteorological Centre Ahmedabad

MICROTOMY AND STAINING :

Small block of samples comprising phloem, cambium and xylem of the main stem were microtomed at 15-20 μm thickness in transverse, tangential and radial longitudinal planes from the two different blocks using Leitz sliding microtome. The sections were taken only in transverse plane for the young branches The sections were arranged in series on slide flooded with 70% ethanol and then tied to the slide with fine cotton thread.

The sections were stained in tannic acid-ferric chloride-Lacmoide combination (Cheadle et al. 1953). After staining, the sections were dehydrated through ethanol xylene series and mounted in DPX.

MACERATION

Xylem portion of one mm width bordering the cambial zone was used to macerate fibres and vessel members. Maceration was done with Jeffrey's fluid (Berlyn and Miksche 1976) or combination of hydrogen peroxide and acetic acid (1:1). After thorough washing, the macerated elements were stained with saffranin or toluidine blue.

MEASUREMENTS

Cambial activity was determined by counting the number of undifferentiated layers of cells lying in between the xylem and phloem from transverse sections. Radial enlargement was the basis for distinguishing the cambial zone cells from their immediate derivatives during active period of cambium. The differentiating xylem and phloem cells were determined by their radial enlargement and thin cell walls. The length and width of fusiform cambial cells, tangential diameter of ray cambial cells, ray height, width and the number of rays passing through one cm tangential width of cambium and the dimensions of vessel elements, sieve tube elements and xylem fibres were measured with an occular micrometer scale mounted in Carl Zeiss ampliwal research microscope. The width of xylem growthring was measured directly from the wood blocks with the help of wild Leitz stereo zoom microscope. The number of vessels per 0 5 mm^2 in transverse section of xylem was counted on Metzer projection microscope

For each parameter hundred measurements were selected randomly and they were statistically analysed to determine the mean and standard deviation.

DRAWINGS

The dimensional details and meteorological data were either represented in histograms or plotted on graphs. Schematic diagram was drawn illustrating the seasonal variations in the mean number of cell layers in the cambial zone and differentiating xylem and phloem elements in the main stem.

HISTOCHEMICAL TECHNIQUES

Radial longitudinal sections were used for the histochemical studies Starch was localised by I_2KI (Johansan 1940) and Mercuric bromophenol blue (Mazia et al. 1953) and Coomassie Brilliant Blue R 250 (Eklavya 1979) were employed to localise proteins. For control, tissues were pre-treated with sodium nitrate and acetic acid prior to the staining. For the detection of lipids, sudan black B stain was used (Jensen 1962). For control, tissues were treated with ether ethanol at 60°C and washed in running water prior to the staining. Phenolic contents were indentified with nitroso reaction (Reeve, 1951).

PHOTOGRAPHY

Photographs were taken from stained sections on Zeiss Microscope using ORWO NP 55 Negative film. Black and white positive prints were taken using sterling special grade bromide paper.