ON FASTING
BLOOD SUGAR LEVEL
The pharmacological investigations were carried out on albino rats of uniform strain originally obtained from Hindustan Antibiotics, Pimpri and then bred and reared in our laboratories. The weight of rats ranged between 140 - 160 G and only male rats were included in this study. The rats were given a uniform diet mixture throughout the period of experimentation. They were fasted for at least 18 hours and water was withdrawn during the experiment. They were kept in separate cages and coloured with dye during administration of drugs with different concentrations.

Rats were immobilized with anaesthetic ether if necessary and were put into bell shape glass jar in order to withdraw the blood. Blood was drawn by cutting the tip of the tail by means of a sharp scissors and blood collected directly into a micro-pipette so as to measure exactly 0.05 ml. Blood sugar was determined colorimetrically by King's method (1956). In each case, fasting blood sugar (F.B.S.) was determined and re-checked after a week. Further experimentation was carried out only in those rats where the F.B.S. was generally constant.
Hypoglycaemic response on fasting Blood sugar level in albino rats.

Compounds were given at three doses level which was arrived at by previous experimentation regarding their suitability for minimum and maximum responses. Compounds' solutions were made in 1% acacia and given orally by means of a blunt hypodermic curved needle. For testing one compound at one dose level three groups of rats were taken, each group containing 5 rats and F.B.S. was determined in each rat. After administration of compounds blood sugar level was determined at 1.5, 3, 5, 7, 24, 48 and 60 hours and mean percentage fall in blood sugar level was determined. Each compound was tested in a dose of 10 mg/kg, 50 mg/kg and 100 mg/kg. Blood sugar level was seen upto 60 hours as in most compounds blood sugar level returned to normal. Results with these compounds are tabulated in Tables 2, 3, 4, 5, 6, 7 & 8.

Colorimetric estimation of blood sugar -

Principle: With this method (Asatoor and King, 1954) glucose is estimated in pure solution and in blood. The results obtained are identical with those found with the previously described titration method (King et al, 1956). The proteins are precipitated by sodium tungstate and copper sulphate (Somogyi, 1931) and the filtrate is treated with a modified Harding and Downs sugar reagent, from which the iodate is omitted. The cuprous oxide formed is
estimated by the blue colour produced with an arseno-molybdic acid solution (Nelson, 1944), which yield almost identical results.

**Methods:**

**Test:** 0.05 ml. of whole blood is pipetted into 3.9 ml. of isotonic sodium sulphate-copper sulphate solution in a conical centrifuge tube. 0.05 ml. of sodium tungstate is added and the mixture is well shaken. The precipitated proteins and copper tungstate are spun down in the centrifuge. 2 ml. of the superant fluid ( = 0.025 ml. of blood) are mixed with 2 ml. of modified Harding B reagent in a 1/3 in. diameter test tube.

**Standard:** 2 ml. of the standard glucose solution is treated in the same way as the blood filtrate.

**Blank:** 2 ml. of isotonic sodium sulphate-copper sulphate mixed with 2 ml. of modified Harding B solution.

The tubes, stoppered with cotton wool, are placed in a boiling water-bath for exactly 10 minutes. After immediate cooling 6 ml. of the arseno-molybdic acid reagent is added and 5 ml. of distilled water (10 or 15 for very densely coloured tests). A wave length of 700 mp used in measuring the colours.