DETERMINATION OF XENOPUS INDEX AND HAEMOLYTIC INDEX IN FRUITS OF *SAPINDUS MUKOROSSI* GAERTN. (SAPINDACEAE) AND SEEDS OF *ENTADA SCANDENS* BENTH. (MIMOSACEAE)

BY MARTHE BLYBERG

From the Department of Pharmacognosy, Institute of Pharmacy, University of Oslo, Norway

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Two drugs, known from the Burmese and Indian folk medicines, the fruits of *Sapindus mukorossi* and the seeds from *Entada scandens*, were examined for their effect on Xenopus larvae. As a comparative test the haemolytic index of the drugs was also determined.

The investigations showed that parts of both drugs contained substances which were toxic for Xenopus larvae and which haemolysed blood. The fruit wall from *Sapindus mukorossi* and the seed kernel from *Entada scandens* had a Xenopus index of approximately 7,000 and 400 respectively. The corresponding haemolytical indexes were approximately 7,000 and 150.

THE fruits of *Sapindus mukorossi*, Indian soap nuts, and the seeds of *Entada scandens* have for a long time been used in Burma and India as saponin drugs and fish poisons. It has also been claimed that the entada seeds possess anthelmintic properties.

According to the available literature both drugs contain saponins[1,2], and as East-Asiatic drugs in recent years have attained increasing importance in Europe, it was felt to be of interest to determine the content of saponin-like substances in the two drugs.

The drugs were examined according to the Xenopus method of Karma and von Schantz[3–5]. The haemolytical index of both drugs was also determined.

EXPERIMENTAL

Determination of Xenopus Index

The investigations were aimed at finding the drug concentration at which the average time of death of the Xenopus larvae is exactly 1 hour. This concentration is termed the hour-concentration. The Xenopus index is then defined as the amount of water, expressed in g., with which 1 g. of crude drug or of pure saponin must be diluted to obtain the hour-concentration.

For the decoctions, coarse powder of the drugs (Pharm. Fennic. VII, screen No. 4) and for the tests, chlorine-free water was used. The decoctions were made according to the original papers[3,4], but the extraction was in a boiling water bath.

For the experiments a larva strain was used for which the hour-concentration of a decoction of the standard drug "Cortex Qvillajae XV" was determined as 0·038 per cent. The corresponding hour-concentration of the normal strain is 0·025 per cent[4]. Accordingly the correction factor is 0·65.

The Fruit of Sapindus mukorossi

The fruit was divided into the fruit wall, the seed coat and the seed kernel.

The fruit wall. Preliminary tests made with solutions of different strength prepared from a 5 per cent decoction showed that the hour-concentration lay between 0·1 and 0·01 per cent. A subsequent experiment with dilutions of a 0·1 per cent decoction (pH 6·3) showed that the

FIG. 1. The fruits of *Sapindus mukorossi* Gaertn. (Sapindaceae).

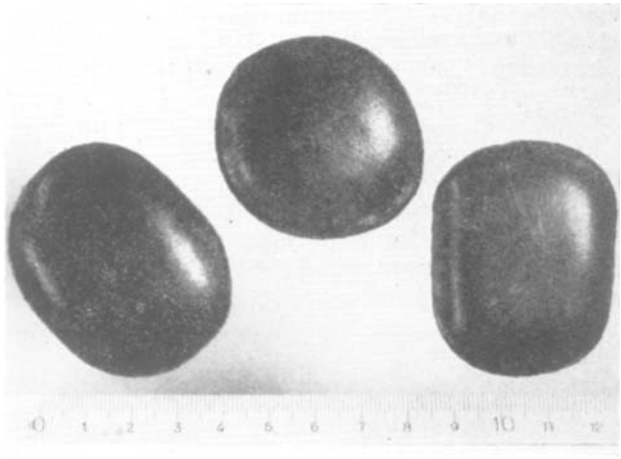

FIG. 2. The seeds of *Entada scandens* Benth. (Mimosaceae).

hour-concentration lay between 0·03 and 0·02 per cent. By reducing the concentration intervals gradually (Table I) the hour-concentration was determined as 0·022, corresponding to 0·014 when adjusted to the normal strain which gives a Xenopus index of approximately 7,000.

The seed coat. Preliminary tests showed that the larvae survived 1 to 2 hours in 5 per cent decoction, which indicates that saponins cannot be detected by this means.

The seed kernel. Preliminary tests showed that the larvae were not injured in a 5 per cent decoction. Accordingly no saponins could be demonstrated by this means.

The Seed of Entada scandens

The seeds were divided into seed coat and seed kernel.

The seed coat. Preliminary tests with solutions of different strength prepared from a 5 per cent decoction showed that the time of death of the

TABLE I

DEATH TIMES OF LARVAE TO WHICH DECREASING CONCENTRATIONS OF THE DECOCTION OF THE FRUIT WALL OF *S. mukorossi* WERE ADDED

Concentration of the decoction in per cent	Number of larvae	Mean value of death time in min.
0·0300	9	29
0·0275	6	40
0·0250	17	45
0·0225	13	55
0·0200	13	75

larvae varied greatly and that there was no clear connection between the concentration of the decoctions and the time of death of the larvae. Accordingly there is no clear evidence that saponins are present in the seed coat.

The seed kernel. Preliminary tests made with dilutions of different strength prepared from a 5 per cent decoction showed that the hour-concentration lay between 1 and 0·1 per cent.

TABLE II

DEATH TIMES OF LARVAE TO WHICH DECREASING CONCENTRATIONS OF DECOCTION OF THE SEED KERNEL OF *E. scandens* WERE ADDED

Concentration of the decoction in per cent	Number of larvae	Mean value of death time in min.
0·400	4	43
0·375	5	48
0·350	5	69
0·325	5	70

A subsequent experiment with dilutions of a 2 per cent decoction (pH 5·6) showed that the hour-concentration lay between 0·4 per cent and 0·3 per cent. By reducing the concentration intervals gradually (Table II) the hour-concentration was determined as 0·36, corresponding to 0·24 when adjusted to the normal strain which gives a Xenopus index of approximately 400.

Determination of the Haemolytic Index

The experiments were made according to a modification of Büchi, Hippenmeyer and Dolder[6]. The sodium citrate solution and phosphate buffer were prepared according to Sandberg[7].

The decoctions were prepared with buffer solution. Otherwise the method described under determination of Xenopus index was followed. Sheep blood was used.

Under the same conditions the haemolytical index of pure saponin Merck was determined: 0·08 mg. pure saponin produced full haemolysis in 2 ml. of 1 per cent blood-suspension, which gives a haemolytic index of approximately 25,000.

The Fruit of Sapindus mukorossi

The fruit wall. 0·3 mg. of drug produced full haemolysis in 2 ml. of 1 per cent blood-suspension. HI = 7,000 (approx.).

The seed coat and the seed kernel. 10 per cent decoctions of the seed coat and the seed kernel gave no haemolysis, and thus no saponins were demonstrated.

The Seed of Entada scandens

The seed coat. A 10 per cent decoction gave no haemolysis.

The seed kernel. 14 mg. of drug produced full haemolysis in 2 ml. of 1 per cent blood-suspension. HI = 150 (approx.).

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