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A MODIFIED SOXHLET EXTRACTOR FOR USE IN TOXICOLOGICAL ANALYSIS

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A soxhlet extractor which can be used at room temperature and under reduced pressure has been described. It is used in toxicological analysis and for the extraction of plant materials.

Key words: Soxhlet, Continuous, Reduced pressure.

INTRODUCTION

The continuous soxhlet extractor has long been in use. However, no attempt to use the extractor under reduced pressure has been recorded. Curry and Phang [1] devised a continuous tissue extractor. This is used at ordinary temperature and under reduced pressure, and a large quantity of tissue can be efficiently extracted. In laboratories where this extractor is not available an alternative can be found in the modified continuous soxhlet extractor described below.

The ordinary soxhlet extractor does not function under reduced pressure. Under such a condition it has been found that the soxhlet will not siphon. This difficulty is however overcome by a little modification to the ordinary soxhlet extractor.

MATERIALS AND METHODS

Analar and analytical grade of the glycosides were purchased from BDH Chemical Ltd., Dorset, England and Sigma Chemical Company, U.S.A.

The modification to the ordinary soxhlet extract consists of a polythene tubing connections on the lower end of the ordinary soxhlet. The lower end of this tubing dips into the solvent in the evaporation flask (Fig. 1).

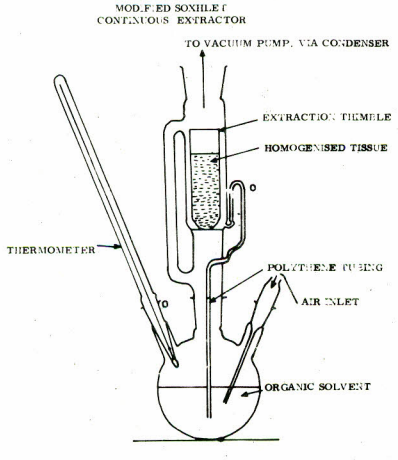

Fig. 1.

Application of the modified soxhlet extractor in extracting glycosides. 2 mg of digitoxin was added to 25 gm of liver and then homogenised in alcohol to obtain a reasonably thick slurry. This was then mixed with 1 gm anhydrous sodium sulphate, put in a thimble, covered with cotton wool and introduced into the soxhlet extractor. This was then connected to vacuum pump. (Edwards High vacuum ISP 30C, Edwards High vacuum Crawley, England). Alcohol was poured into the three necked evaporation flask which was placed in a water bath maintained at 50^{o}. A Thermometer was attached to one neck of the evaporation flask while an air inlet was connected to the third. The extraction was carried out for eight hours under reduced pressure 13.3 pa (0.1 mm Hg). The alcoholic extract was evaporated to dryness in vacuo and the residue taken in 20 ml warm absolute alcohol. Precipitate so formed was filtered off and the filtrate again evaporated to dryness in vacuo. The residue was taken in 10 ml 0.001 N H_2SO_4 shaken vigorously for 15 min and filtered. The filtrate was extracted with 20 ml light petroleum (60^{o} - 80^{o}) followed by 20 ml solvent ether. Both organic phases were rejected. The aqueous phase was adjusted to pH 6 with dilute ammonium hydroxide. It was then extracted with 20 ml chloroform. The chloroform layer was removed, concentrated, purified by paper chromatography eluted. The concentration of digitoxin in the eluate was determined by the method of Jelliffe [2]. Curry-Phang continuous extractor.

2 mg of digitoxin was added to 25 gm of liver homogenised and extracted for 8 hours using the Curry-Phang extractor. The extract so obtained was treated as above.

Both methods of extraction were used for the extraction of glycosides namely; aesculin, amygdalin, digitoxin, sennoside B, thevetin etc. obtained from homogenised liver. The percentage recovery by each method was determined.

RESULTS AND DISCUSSION

The percentage recovery of each glycoside is as shown in Table 1.

Table 1

Glycoside	Curry-Phang recovery %	Modified soxhlet extractor recovery %
Digitoxin	60	50
Thevetin	55	50
Digoxin	20	20
Amygdalin	72	70
Singrin	72	71
Solanin	40	38
Anthraquinone	20	20
Aesculin	20	20
Phloridzin	40	41
Sennoside B	20	18

The values represent an average of three determinations for each of the glycosides.

From the result it can be seen that the modified soxhlet extractor gave comparative results as the Curry-Phang extractor. The advantages of the modified soxhlet apparatus, are that the extraction is at room temperature and under reduced pressure. The working conditions are so mild that glycosides and other labile compounds are not destroyed. It is also simple and easy to handle. In addition, it requires comparatively smaller quantity of organic solvent. It can also be used for the extraction of plant materials.

The method were applied to recover glycosides from liver homogenate. In these two methods the extraction of different glycosides (individually added) differ from glycoside to glycoside. The percentage recovery was found to vary from as high as 72% for singrin through 60% for digitoxin to as low as 20% for aesculin, digoxin, sennoside B and anthraquinone. Glycosides which gave low recovery, gave similar results with direct extraction method also.

The low recoveries were therefore not due to inefficiency of the Curry-Phang extractor or the modified soxhlet extractor. The 60% recovery for digitoxin compares well with the value obtained by other workers [1,3,4]. Besides it requires comparatively smaller quantity of organic solvent and without loss of alcohol by air under vacuum.

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