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ORAL GLUCOSE TOLERANCE TEST (OGTT) WITH A COMBINATION OF *COLOCASIA ESCULENTA* STEMS AND *EICHHORNIA CRASSIPES* AERIAL PARTS

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ABSTRACT

Background: In previous experimental studies, it was observed that various parts of *Colocasia esculenta* (L.) Schott possess antihyperglycemic potential. The objective of the present study was to determine the antihyperglycemic effect of combined methanol extracts of *Colocasia esculenta* stems and *Eichhornia crassipes* (Mart.) Solms aerial parts (known for antioxidant properties) in glucose-challenged mice. **Methods**: Antihyperglycemic activity was determined through oral glucose tolerance test (OGTT) in mice. **Results:** Individual administration of methanol extract of *Colocasia esculenta* stems (MECE) and *Eichhornia crassipes* aerial parts (MEEC) at a dose of 400 mg each per kg body weight to glucose-loaded mice reduced blood glucose levels by 41.1 and 21.5%, respectively, compared to control (untreated) mice. When given in combination, the combined extracts reduced blood glucose level by 42.4% in glucose-challenged mice. By comparison, a standard antihyperglycemic drug, glibenclamide, when administered at a dose of 10 mg per kg body weight, reduced blood glucose level by 46.1%. **Conclusion**: The combination of MECE and MEEC was not more effective than MECE alone despite reported antioxidative potential of MEEC.

KEYWORDS: Antihyperglycemic, *Colocasia esculenta*, *Eichhornia crassipes*, glibenclamide, OGTT.

BACKGROUND

Colocasia esculenta (L.) Schott. is an Araceae family plant, which is known as 'cocoyam' in English and 'panikochu' in Bengali. The plant is cultivated for its edible corms and stems, but also can be found in the wild in marshy regions. The leaves and the corms (sometimes called tubers) attain a large size. *Eichhornia crassipes* (Mart.) Solms belongs to the Pontederiaceae family, and is known in English as 'water hyacinth' and in Bengali as 'kachuripana'. This aquatic plant can be found in profusion in practically all water bodies of Bangladesh, and apart from limited use as organic fertilizer, is considered a weed throughout the country.

The antihyperglycemic and antinociceptive potentials of various parts of *Colocasia esculenta* by themselves or in combination with plant parts of other plant species has previously been reported by us.[1-4] Diabetes is a disorder where the body cannot metabolize glucose normally, leading to elevated blood glucose levels. This glucose intolerance, in turn can lead to other severe and more complicated disorders like diabetic retinopathy (eye disorders), diabetic nephropathy (kidney disorders), cardiovascular disorders, foot problems, and diabetic neuropathy (nerve disorders). Increase in oxidative stress due to diabetes can be the cause for these disorders.[5] The antioxidant potential of *Eichhornia crassipes* has variously been reported,[6-8] which may prove useful in diabetes. We had been screening local plants for their blood glucose lowering ability for a number of years,[9-37] because there are no existing medications for curing diabetes, and allopathic antidiabetic drug(s) are not always affordable or available to rural people. As such, the objective of the present study was to evaluate the antihyperglycemic activity of a combination of methanolic extract of *Colocasia esculenta* stems and *Eichhornia crassipes* aerial parts through oral glucose tolerance test (OGTT).

METHODS

Plant material collection and extraction

Stems of *Colocasia esculenta* and aerial parts of *Eichhornia crassipes* were collected from Mohammadpur area in Dhaka city during September 2016. Plant specimens were taxonomically identified by a trained botanist at the University of Development Alternative. The sliced air-dried stems of *Colocasia esculenta* and aerial parts of *Eichhornia crassipes* were grounded separately into a fine powder and the powders extracted separately with methanol (1:5, w/v) for 48 hours. The extracts (MECE for *Colocasia esculenta* and MEEC for *Eichhornia crassipes*) was evaporated to dryness at 40^{o}C and stored at -20^{o}C till use.

Chemicals and Drugs

Glibenclamide and glucose were obtained from Square Pharmaceuticals Ltd., Bangladesh. All other chemicals were of analytical grade.

Animals

Swiss albino mice, which weighed between 14-18g were used in the present study. The animals were obtained from International Centre for Diarrhoeal Disease Research, Bangladesh (ICDDR, B). The animals were acclimatized for three days prior to actual experiments. During this period, they were kept in a temperature controlled room (25^{o}C) and given standard mice chow and water *ad libitum*. The study was conducted following approval by the Institutional Animal Ethical Committee of University of Development Alternative, Dhaka, Bangladesh.

Oral glucose tolerance tests (OGTT) for evaluation of antihyperglycemic activity

Oral glucose tolerance tests were carried out as per the procedure previously described by Joy and Kuttan (1999)[37] with minor modifications. Briefly, fasted mice were grouped into five groups of five mice each. The various groups received different treatments like Group 1 received vehicle and served as control, Group 2 received standard drug (glibenclamide, 10 mg/kg body weight). Group 3 received 400 mg MECE per kg, Group 4 received 400 mg MEEC per kg, and Group 5 received a combination of 400 mg MECE plus 400 mg MEEC per kg. All substances were orally administered. Following a period of one hour, all mice were orally administered 2g glucose/kg of body weight. Blood samples were collected 120 minutes after the glucose administration through puncturing heart. Blood glucose levels were measured with a glucometer. The percent lowering of blood glucose levels were calculated according to the formula described below.

Percent lowering of blood glucose level = $(1 – W_e/W_c) X 100$,

where W_e and W_c represents the blood glucose concentration in glibenclamide or various extracts administered mice (Groups 2-5), and control mice (Group 1), respectively.

Statistical analysis

Experimental values are expressed as mean ± SEM. Independent Sample t-test was carried out for statistical comparison. Statistical significance was considered to be indicated by a p value < 0.05 in all cases.[25]

RESULTS

Oral glucose tolerance test (OGTT) results

Administration of 400 mg per kg MECE alone led to 41.1% reduction in blood glucose level in glucose-challenged mice, compared to control (untreated) mice. At the same dose, administration of MEEC alone led to 21.5% reduction in blood glucose level in glucose-challenged mice, compared to control (untreated) mice. A combination of 400 mg MECE plus 400 mg MEEC administered together led to 42.4% reduction in blood glucose level. By comparison, a standard antihyperglycemic drug, glibenclamide, when administered at a dose of 10 mg per kg body weight, reduced blood glucose level by 46.1%. Thus at the dose tested (400 mg per kg), MECE as well as (MECE + MEEC) demonstrated nearly comparable ability to glibenclamide in its antihyperglycemic activity. However, there was very little synergistic effect of MECE and MEEC when both extracts were administered in combination.

Table. 1: Effect of MECE, MEEC, and (MECE + MEEC) on blood glucose level in hyperglycemic mice following 120 minutes of glucose loading.

Treatment	Dose (mg/kg body weight)	Blood glucose level (mmol/l)	% lowering of blood glucose level
Control	10 ml	5.94 ± 0.10	-
Glibenclamide	10 mg	3.20 ± 0.09	46.1*
(MECE)	400 mg	3.50 ± 0.11	41.1*
(MEEC)	400 mg	4.66 ± 0.12	21.5*
(MECE + MEEC)	400 mg each	3.42 ± 0.15	42.4*

All administrations were made orally. Values represented as mean ± SEM, (n=5); $^{*}P < 0.05$; significant compared to hyperglycemic control animals.

DISCUSSION

Although no synergistic antihyperglycemic effect was seen with a combination of MECE and MEEC, the combination may still prove useful. While the main function of MECE can be reduction of elevated blood glucose level, the function of MEEC can be to prevent development of other complications arising from diabetes through its antioxidant capacity.[6] Moreover, both plants are readily available in Bangladesh and can be obtained at practically no cost throughout the year. As such, further studies can prove beneficial in obtaining antidiabetic drugs from both plant species.

CONCLUSION

The results suggest that methanolic extract of stems of *Colocasia esculenta* (MECE) and methanolic extract of aerial parts of *Eichhornia crassipes* (MEEC) possess antihyperglycemic

effects as demonstrated through OGTT. However, very little synergistic action was observed when the extracts were administered together.

CONFLICTS OF INTEREST

The author(s) declare that they have no competing interests.

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