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Effects of Aqueous Leaf Extract of Pride of Barbados (*Caesalpina pulcherrima*) on the Activities of Some Liver Function Enzymes and Blood Glucose Concentration in Normal Rabbits

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Abstract: Some of the effects of the aqueous leaf extract of Pride of Barbados (*Caesalpina pulcherrima*) on the activities of plasma glutamic pyruvate transaminase (GPT), glutamic oxaloacetate transaminase (GOT), acid phosphatase (AP), alkaline phosphatase (ALP) and blood glucose concentration in normal rabbits were carried out. All the rabbits used which were of New Zealand strain weighed between 1.00 to 1.60 kg. About 100ml aqueous leaf extract of this plant was administered daily to the test rabbits orally through clean drinking troughs for a period of 15 days. Analyses were conducted using the blood collected intravenously from the larger veins at the back of the ears of rabbits. Aqueous leaf extract of Pride of Barbados did not affect plasma GOT, GPT, AP and ALP activities (P > 0.05). However, plasma glucose concentration increased significantly (P < 0.05) on the 9th, 11th, 13th and 15th days of administration of the aqueous leaf extract to the rabbits. It might be unsafe to abuse the use of this extract as liver tonic as it may create serious health problems for the diabetics due to the likely escalation of complications arising from hyperglycemia. Non – diabetics however, may be placed on a routine check of their fasting blood sugar as they use the aqueous leaf extract of Pride of Barbados.

Key words: Aqueous leaf extract, pride of Barbados, liver function enzymes, glucose

Introduction

Pride of Barbados is a genus of tropical evergreen shrub, which belongs to the fabacae of bean family. Its other common names are dwarf poinciana, and Bird of paradise (Russel et al., 1997). It is a leguminous plant belonging to the leguminosae family, which is the second largest family among the dicotyledonous plants (Prohp et al., 2006a,b). The immature seeds of Pride of Barbados are edible while the mature seeds are toxic. The toxicity is due to the presence of tannins which when ingested could cause symptoms such as nausea, vomiting and diarrhea (Russel et at., 1997). Pride of Barbados is a distinctive plant of great medicinal value. Methyl alcohol extracts of the dried bark have been shown to have in vitro activities against staphylococcus aureus. Root extract with methanol is also known to have in vitro activity against staphylococcus aureus and Escherichia coli (Khan et al., 1980). The aqueous extracts of the fresh leaves have a strong in vitro anti fungal activities against usilage maydis (Singh and Pattak, 1994). In Suriname's traditional medicine, the leaves are purgatives and could be used against kidney stone, malaria, fever and bronchitis. They are also used for abortion due to its contents of garlic acids, tannins and hydrocyanic acids. A lot of works have been documented on some nutrient potentialities of the extra - cotyledonous deposits of this legume (Prohp and Alaiya, 2003, Prohp and Maduemezia, 2004, Prohp et al.,

2004, Prohp et al., 2006a, Prohp et al., 2006b). The stem and roots are however, cytotoxic. Traditionally the aqueous fresh leaf extract of Pride of Barbados are administered as liver tonic (crescentbloom.com.1998). This study is therefore designed to understand if there might be any possible side effects of this extract on some plasma enzymes vis-a -vis the liver and other organs of those using it as a type of tonic. Glucose assays were also conducted on rabbits exposed to the aqueous extract with the view of assessing its possible anti – diabetic properties.

Materials and Methods

Pride of barbados: Fresh leaves of this plant were obtained from the main campus of Ambrose Alli University, Ekpoma, Edo State, Nigeria.

Experimental animals: Adult male and female rabbits of the same strain (New Zealand) weighing between 1.00-1.600 kg were obtained from Benin City, Edo State, Nigeria. The rabbits were kept in standard cages, and fed on growers' mash and water for about 2 weeks of acclimatization to the new environment. Subsequently they were maintained on growers' mash, water (for control) and aqueous extract of fresh leaves of Pride of Barbados (for test) throughout the duration of the experiment (ad libitum).

Reagents: Glucose oxidase, Glutamic pyruvate transaminase, Glutamic oxaloacetate transaminase, Acid phosphatase, Alkaline phosphatase, Creatine kinase and α - Amylase kits were procured from Randox Laboratories Ltd., United Kingdom. All other reagents were of analytical grades.

Preparation of leaf extract: The leaves were removed from the stalk, weighed and washed thoroughly with distilled water. The clean leaves were then boiled in distilled water (1g/10ml) for three hours on the 1st day, two hours on the 2nd day and one hour on the 3rd day. After cooling to room temperature, filtration was carried out using Buchnner funnel to eliminate debris. Clear extracts were stored at -21°C until needed. (Onoagbe *et al.*, 1999)

Administration of leaf extract: The aqueous extract from the leaves of Pride of Barbados was administered orally through clean drinking troughs to a set of 3 test rabbits ad libitum. Each rabbit was force fed with at least 100ml of the extract (after an overnight fast) on the first day of the experiment after collecting blood samples at zero day. Subsequently, the extract was fed to the test rabbits for a minimum of 15 days. Drinking troughs depleted of extract were immediately replenished with known volume of extract. The volume of extract consumed was the difference between initial volume given and the volume left after 24 hours (Onoagbe et al., 1999).

Collection of blood: The blood was collected intravenously through the large veins at the back of the rabbit's ear. Prior to blood collection the ear was sterilized by dabbing with cotton wool soaked in methylated spirit. Petroleum jelly was then used to make the hair lie flat on the surface to make the vein conspicuous. Blood collection was carried out using sterile needles and syringes. Lithium heparinized (for plasma enzyme assays) and fluoride oxalate (for glucose assays) sample tubes were used in blood collection. The blood sample was rocked slightly and centrifuged at 3000 r.pm for 5 minutes. The supernatant was then stored in the freezer at -21°C until analyzed (Onoagbe et al., 1999).

Assays: Blood glucose and enzyme assays were carried out according to the procedures described by Randox Laboratories Ltd, United Kingdom.

Statistics: Data were analyzed using the student t- test.

Results

The results have been presented in Tables 1, 2, 3, 4 and 5. Table 1 shows the summary of mean plasma values of glucose concentration (mg/100ml). Tables 2 and 3 show the plasma glutamic pyruvate transaminase and

glutamic oxaloacetate transaminase activities (U/I) respectively. Tables 4 and 5 are the result for plasma acid phosphatase and alkaline phosphatase activities (U/I). Statistical analyses using the student t-test showed that the aqueous extract of Pride of Barbados has no significant effect (P > 0.05) on the activities of some of the liver function enzymes. However, plasma glucose concentration increased significantly (P < 0.05) on the 9^{th} , 11^{th} , 13^{th} and 15^{th} days of administration of the aqueous leaf extract to the normal rabbits.

Table 1: Summary of mean plasma glucose concentration (mg/100ml) of normal rabbits administered aqueous leaf extract of Pride of Barbados for 15 days

Days	Control	Test
0	80.29 ± 11.84	77.27±6.93
1	79.15 ± 12.02	44.43±3.68
3	50.46 ± 4.86	59.25±8.84
5	60.22 ± 3.06	76.37±3.69
7	49.99 ± 2.42	68.04±7.34
9	67.09 ± 6.64	*141.66±30.05
11	59.71 ± 5.55	*140.23±12.37
13	74.97 ± 4.79	*156.83±3.64
15	59.24 ± 3.48	*157.00±4.81

Values are mean \pm S.E.M. of three separate determinations from six rabbits. *Significant increases (P < 0.05) from the control.

Table 2: Summary of mean plasma glutamic pyruvate transaminase activities (U/I) of normal rabbits administered aqueous leaf extract of Pride of Barbados for 15 days

Days	Control	Test
0	55.07 ± 2.54	47.60 ± 10.23
1	52.27 ± 3.87	58.27 ± 3.22
3	45.33 ± 5.08	39.47 ± 6.99
5	43.87 ± 8.49	67.33 ± 13.69
7	61.87 ± 13.87	46.40 ± 6.06
9	57.60 ± 12.83	81.20 ± 6.71
11	54.00 ± 15.83	54.67 ± 7.63
13	63.27 ± 19.71	56.67 ± 9.43
15	65.11 ± 14.47	58.66 ± 8.61

Values are mean \pm S.E.M. of three separate determinations from six rabbits. Values not significantly different (P > 0.05).

Discussion

Plants generally have varied compositions depending upon species. Some plants are economically important while others are of medicinal value. Those that are of medicinal value are often used as herbal remedy for the restoration and maintenance of good health. Some herbs have been considered as drugs and therefore generally safe and effective (Treasure, 2000). Herbs have been associated with broad actions on a number physiological systems in concert unlike pharmaceutical drugs usually designed to elicit a specific effect. Some are of the opinion that some herbal plants are usually oriented in the same general therapeutic direction and are complementary or synergistic, often non - specific but very rarely adverse (Ibiblio.org., 2000; Treasure, 2000).

Table 3: Summary of mean plasma glutamic oxaloacetate transaminase activities (U/I) of normal rabbits administered aqueous leaf extract of Pride of Barbados for 15 days.

Days	Control	Test
0	66.80 ± 22.54	81.20 ± 12.13
1	50.00 ± 10.81	72.00 ± 2.40
3	47.60 ± 7.63	40.10 ± 7.57
5	47.60 ± 7.63	40.00 ± 7.63
7	34.40 ± 10.23	28.8 ± 6.36
9	52.00 ± 26.53	92.40 ± 13.93
11	30.80 ± 6.30	54.80 ± 12.83
13	24.80 ± 8.15	54.60 ± 25.32
15	25.56 ± 5.32	55.00 ± 12.38

Values are mean ± S.E.M. of three separate determinations from six rabbits. Values not significantly different (P > 0.05).

Table 4: Summary of mean plasma acid phosphatase activities (U/I) of normal rabbits administered aqueous leaf extract of Pride of Barbados for 15 days

Days	Control	Test
0	6.00 ± 1.15	6.67 ± 1.76
1	8.00 ± 2.00	9.33 ± 3.33
3	8.00 ± 3.06	16.00 ± 2.31
5	6.67 ± 0.67	13.33 ± 2.40
7	14.67 ± 1.76	17.33 ± 2.67
9	9.33 ± 0.67	13.33 ± 2.40
11	12.67 ± 2.40	9.33 ± 3.53
13	11.33 ± 1.76	12.67 ± 2.40
15	12.48 ± 0.98	11.97 ± 2.21

Values are mean \pm S.E.M. of three separate determinations from six rabbits. Values not significantly different (P > 0.05).

Table 5: Summary of mean plasma alkaline phosphatase activities (U/I) of normal rabbits administered aqueous leaf extract of Pride of Barbados for 15 days.

Days	Control	Test
0	9.33 ± 1.33	18.67 ± 4.67
1	9.33 ± 1.33	14.00 ± 8.08
3	14.00 ± 8.08	14.00 ± 8.08
5	23.33 ± 4.67	14.00 ± 8.08
7	14.00 ± 0.00	9.33 ± 1.67
9	9.33 ± 1.69	23.33 ± 4.81
11	18.67 ± 4.14	18.67 ± 4.14
13	4.67 ± 1.02	18.67 ± 4.67
15	7.89 ± 2.11	17.93 ± 4.18

Values are mean \pm S.E.M. of three separate determinations from six rabbits. Values not significantly different (P > 0.05).

Herbs used as herbal remedies are administered in various ways. Some can be drunk while others are applied to the skin as ointment (Bradford, 1997). Most of the standard pharmaceutical drugs are derivatives of plants (Joyce and Christopher, 1992; Chevallier, 1996). In some of these plants the active ingredients are in the leaves. Good examples are cocaine, from the leaves of Tulsi (Oscimum sanctum) widely used to cure malaria, liver disorder, cold and cough and aloe (Vern gheekwar) from thick and fleshy leaves of aloe contain several glucosides and used as purgatives after extraction. Others are Ballandona, from dried leaves of Atropa belladonna, used externally as pain reliever to check excessive perspiration while neem, obtained from

Azadirachta indica is used against jaundice and in various skin diseases (Burton, 1993; Foster, 1993; Bilgrami et al., 1997). The leaves of Pride of Barbados are used against malaria fever, bronchitis and kidney stones. Literature sources show that the aqueous leaf extract of this legume can also be used as liver tonic and for hepatotoxic protection (crescent bloom.com.1998). The extract from the leaves of Pride of Barbados and Aloe, Vern Gheekwar, are used as purgatives (Bilgrami et al., 1997). According to Khan et al., 1980, the roots are cytotoxic and have been used against bacterial infection such as staphylococcus spp and Escherichia coli.

Analyses of the activities of some basic liver function enzymes in the plasma can be used to indirectly access the integrity of tissues after being exposed to certain pharmacological agent(s). The enzymes studied are liver markers whose plasma concentrations above the homeostatic limits could be associated with cirrhosis, lesions or other damages to the liver by agents considered unsafe.

This investigation shows that the use of the aqueous leaf extract of Pride of Barbados as liver tonic in some part of the world may not interfere with the integrity of the liver and perhaps other organs of the body. This is against the backdrop of non-significant effect (P > 0.05) of the aqueous leaf extract of this plant on the enzymes studied. However, significant increases (P<0.05) in glucose concentration were obtained on the 9th, 11th, 13th and 15th days of administration of extract to normal rabbits. It is also very likely that the use of this aqueous extract consistently as a liver tonic might pose a serious health problem if abused.

In conclusion, therefore, it might be necessary to suggest that non-diabetics using this herbal extract as liver tonic be placed on a routine check of their blood glucose level. For diabetics the use of this extract may aggravate hyperglycemic complications on the long term.

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