Phytolacca americana (Phytolaccaceae)
(syn.: P. decandra, P. vulgaris)

English: Pocan, poke, pokeweed, Virginian poke, American nightshade, garged weed, pigeon berry, red ink plant, Virginian poke
French: Laque, raisin d`Amerique, phytolaque Ital.: Amaranto, erba cremosina, fitolacca, lacca sanguinella, tinta, uva di Spagna, uva turca
Chinese: Hao-lu, Tang-lu
German: Kermesbeere, Scharlachbeere, Schminkbeere

African vernacular names:
Chagga: Ibesa, mbesa, ingorosoi, ivesa, ingorism Masai: Oladaba
Shambala: Hoko Zulu: Umaheneni, ingub-ivumile

The plant
The plant genus Phytolacca encloses 35 species with a close relativity and similar characteristics. These different species are used in same applications, therefore.
P. americana is native to the temperate regions of Northern America, now propagated or cultivated over the whole world. In Europe it is running wild. Shoots are used as legume.
P. icosandra, a shrub native to tropical America from Mexico to Peru and on the Caribbean islands, is now naturalized in the hilly and mountainous regions of Central and East Java. In Australia, New Zealand and South Africa it is a noxious weed. In East Africa P.dodecandra is used in schistosomiasis control.

Phytolacca americana plants are shrubs or trees, annual, likewise perennial, the stems are erect up to 1-3 m heighth. The leaves are pinnate, opposite or appearing whorled, ovate, 10-15 cm long, 4-12 cm broad, margins entire, often undulated. Flowers: Inflorescences stand axillary, in racemes, radially symmetric and hypogynic; the sepals (5) are rounded often persistent, originally white or green, later on purple together with the peduncles of the leaves. The fruits are red berries, finally becoming black, seeds with perisperm, gleaming black,10 mm in diameter.

Plant parts used
The roots, the berries, the leaves

Constituents
In an early work from 1975 following aglycones of P.dodecandra berries were found: Oleanolic acid 66.2 %, bayogenin 14.9 %, hederogenin 8.9 %, 2-hydroxyoleanolic acid 6.5 %, which all are 28-carboxyoleanenes. The saponins in the berries of the following species like P.americana, P.dioica, P.octandra, P.rivinoides, and P.esculenta are said to be composed of 28,30-dicarboxy- and/or carbomethoxy oleanenes (9).
In the whole plant triterpene saponines and Phytolacca mitogenes, in the leaves the flavanoles astragalin and isoquercetrin, and in the epidermis cells oxalate crystals (2) are

found. In newer works no references about the “alkaloid” phytolaccin” could be invented.

Constituents of the ripe berries

Pharm. definition: Phytolaccae americanae fructus, poke berries

Betacyanes: Phytolaccin, the red coloured sap of the fruits contains mainly (95 %) of betanin.

Triterpensaponines: The hydrolysis resulted in the aglyca esculentic acid, jaligonic acid, phytolaccagenic acid, pokeberrygenine, and a little bit of acinosolic acid. In the seeds 3-acetyloleanolic acid and 3-acetylaleuritolic acid could be found. In the seeds lot of triterpene alcohols like amyrin, cycloartenol, lanosterol, lupeol could be found together with starch, proteins (10 %),PAP-S, a specific protein and further fatty oils (11-13 %).

Lignanes: From the seeds the neolignanes Americanine A, B, D and americanol A and isoamericanol A could be isolated.(2).

Saponines: The dried berries of P. dodecandra, known as “endod” or “soapberry” contain up to 25 % (w/w) saponines. Monodesmosidic oleanolic acid acts as a molluscicidal saponine, while bidesmosidic compounds of oleanolic acid are inactive, like hederogenin and bayogenin. The isolated saponines are white amorphous powders, soluble in methanol. Mass-spectrometric values were in the range 499 (A-H) and 661 (M-H). Sugars were glucopyranose and rhamnose (13).

For quantitative determination a HPLC method exists. Monodesmosidic oleanon saponines of the berries are determinable at 254 nm as their 4-bromophenacyl derivatives, whereas non-derivatized bidesmosidic saponins, lacking a free carboxyl group, are determined at 206 nm.

Cold water extraction brings predominantly molluscicidal monodesmosidic saponines, hot water extraction gives mainly non-molluscicidal bidesmosidic saponins (11).

The content of the saponins in the berries varies with the season. The highest content of the berries of P. dodecandra is during the dry season and just before the onset of the rain (7).

Constituents of the root

Pharm. definition: Phytolaccae Americanae radix, poke root.

Triterpensaponines: Phytolaccosides A, B (= phytolacca saponine G), D, D_2, E (= phytolacca-saponine E) and Phytolacca saponine B.

After acid hydrolysis the aglyca were: Esculentic acid, jaligonic acid, jaligonic acid-30-methyl ester (=phytolaccagenin, esculentic acid-30-methylester (phytolaccagenic acid). The saponines are monodesmosidic mostly with a short unbranched sugar chain from which glucose and xylose could be derived.

The main saponine is phytolaccoside E.

Proteins: Out of the fresh root five glycoproteins (Pa-1 to Pa-5) could be won (PWM = Poke Weed Mitogen). They are mitogenic active, with 19 % cysteine and 4-5 % carbohydrate, and PAP-R (Poke weed antiviral protein from the root) an antiviral compound.

Further compounds are: 0.13 %-0.16 % histamine, gamma-aminobuttyric acid, alpha spinasterol, stigmasterol, starch, saccharose, and potassium salts like KNO_3 (2).

Traditional uses

Use of the berries:

In North America Red Indians drank tea made from the berries against rheumatism. The early colonists used the sap of the berries against skin lesions accompanied with cancer.

In the Appalachian Mountains dried fruits in pap were applied on wounds. Wine growers took the red fruits for colouring (and adulterating) wines.

Use of the root:

Red Indians used the pulverised root for the preparation of ointments against cancer. In Spain a pomade made from roots is applied on skin eruptions and ringworm disease.
In America the dried roots were employed against rheumatism, catarrh, dysmenorrhoea, dyspepsia, tonsillitis, laryngitis, conjunctivitis, mumps, ring worm disease, syphilis, itch, scabies, and as an emetic and against constipation, finally (2).
In the European Homoeopathy the fresh root is used against rheumatism of the joints, against flu and angina.

Results of experimental studies

Molluscicidal activity

In Purwodadi, Java, the methanol and water extract of P. icosandra berries , showed molluscicidal activity at 200 µg/ml, and 25 µg/ml, against the snail Biomphalaria glabrata. Monodesmosidic saponines were active against snails with a minimum concentration of 3.1 µg/ml. Bidesmosidic saponines were inactive.
But in water bidesmodic saponines are transformed into monodesmosidic ones. Authors from South Africa argue that an enzyme, located in the seeds is necessary for this transformation (10).
In South East Asia P. icosandra is recommended for the local control of schistosomiasis.
In Zimbabwe the berries of P. dodecandra were found to be 50 % lethal against the snail Bulinus globosus. The most toxic berries were collected from the drier and hotter places of the country. The toxicity depends from the geographical feature of the place (8).

Determination of Phytolacca saponines

For the determination of Phytolacca saponins in water during molluscidic actions two methods are possible under field conditions:
Thin layer chromatography and a haemolytic method .
The test subject was the snail Biomphalaria with “endod”, the saponine of P.dodecandra. The concentration of the saponins was between 2 mg/l and 6 mg/L.
The active LC 50 and LC 90 values were 2.57 and 2.92 mg/l, respectively. The controlling TLC method gave a correlation coefficient of 0.99 for saponines above 1.5 µg .According these results the haemolytic test can be used under field conditions (6).
After a short contact (30 min) of the berry extract from P.dodecandra with miracidia of Schistsoma mansoni the LC 50 value was 8.2 ppm.
Schistosoma haematobium, exposed to the (sublethal) dose of 3 ppm for 30 min or overnight in open air ponds reduced the infection rate up to 3.5-5.6 fold, compared with controls. The application of the toxicant in low doses can reduce the infection of the snails at such foci, where transmission is likely to occur (4).
In Ethiopia for controlling schistosomiasis the effects of aqueous extracts from P.dodecandra (“endod”) were investigated on the larval stadium, cercaria, and miracidia. Aqueous extracts of the berries prevented snails from being infected by miracidia at a concentration of 4 ppm. Pre-treatment of the cercaria with 12 ppm of the extract completely inhibited infection of mice by cercaria and significantly reduced egg deposition and worm establishment in the mice ($P < 0.05$). The extract of the berries can be a good tool against schistosomiasis in fresh water (1).
The biogradability of the saponins in aqueous extracts from berries of P.dodecandra was evaluated under OECD standardized conditions with HPLC measurement and hemolytic assays. Measurements of saponines with HPLC and with a haemolytic assay showed no significant differences ($p<0.05$). The saponine concentrations in water were stable for two days. The concentration rapidly decreased during the third or fourth day.

Lethal concentrations for the snails (LC50) were 9.6 mg/ml and 6.8 mg/ml, respectively. Under aerobic conditions saponines are biodegraded within 10 days.
According to the authors the use of these extracts is environmentally acceptable (5).

Toxicity

Toxicity tests according to the OECD Guidelines for Premarket Chemicals were conducted with a standard extract from the unripe (!) berries of P. dodecandra. All tests on mammalians were classified non-toxic or slightly toxic.

But the eye irritation toxicity test indicated severe activity

Eye protection is recommended urgently during the berry crushing and the handling of the dry powder. Further exotoxicity tests resulted that these extracts are not more toxic than synthetic molluscicides (3).

Test of the butanol extract of P. dodecandra

In a study on nontargeted animals the toxicity of butanol extracts from P. dodecandra was investigated:

- Groups of 10 mosquito fish (Gambusia affinis) and 8 bluegills (Lepomis macrochirus) were exposed to the butanol extract in 300 ml water from 0.3 - 2.0 ppm.
- Groups of 10 tropical snails (Biomphalaria glabrata) and of 10 pond snails (Physa ssp) were exposed to the extract in 50 ml water in a range 1.0-5 ppm.

After 24 h exposure, the test subjects were transferred to extract-free water and observed for additional 24 h.
The butanol extract was lethal for 50 % of the fish and snails at the relatively low concentrations of less than 3.0 ppm. The fish were 2 – 4 times more sensitive (12).

Haemolytic and spermatocidal activity

For bovine erythrocytes the hemolytic activity of single saponins was between 1.9 - 60 µg/ml. The spermatocidal activity was between 250 µg/ml till 2000 µg/ml (MIC). These concentrations cause 100 % inhibition of the motility of human spermatozoids after 3 minutes (13).

Results of clinical studies

No data were available

Evaluation

In the plant family Phytolaccaceae, and especially in the plant species Phytolacca saponines are the predominant chemical substances. Such saponins are effective on membranes, beginning with those of the cells till such ones of whole organs.
Saponins penetrate into the cells themselves or facilitate the entry of accompanying substances. The efficacy is dependent from their concentration. In low concentrations the saponines can give support against ailments. So they are good for wounds and the treatment of superficial mycotic infections. In higher concentrations they are toxic or lethal, as the experiments with fish and snail show. So extracts of berries are applied in ponds or fresh water against snails, too.
For the application with humans recommendations can be given only for outer use, like ointments with low concentrations and applications for short terms.

The internal use with men must be argued against.

In field campaigns the extracts of berries are good tools for control of schistosomiasis.

Phytolacca americana and other related species

Controlling of schistosomiasis	*** * ***
Against skin ailments	*** ***
For internal application with men	**- - -**

References Phytolacca

1. Birrie H, Balcha F, Erko B et. al. (1998) Investigation in the cercaricidal and miracidiacidal properties of Endod (Phytolacca dodecandra) berries (type 44) East Afr Med J 75,5:311-14 Pub Med 9747 oo6
2. Hänsel R, Keller K, Rimpler H ,Schneider G, Hrsg. (1993) Hagers Handbuch der Pharm. Praxis 5th Edition Vol.5,Springer Berlin, Heidelberg New York p.360 ff
3. Lambert JD, Temmink JH, Marquis J et al. (1991) Endod: Safety evaluation of a plant molluscicide Regul Toxicol Pharmacol 14, 2:189-201 Pub Med 179 2353
4. Madhina D, Shiff C (1996) Prevention of snail miracidia interactions using Phytolacca dodecandra (endod) as a miracidiacid: an alternative approach to the focal control of schistosomiasis Trop Med Int Health 1, 2: 221-6 Pub Med 8665 388
5. Moolgard P,Chihaka A,Lemmich E et al. (2000) Biodegradability of the molluscicidal saponins of Phytolacca dodecandra Regul Toxicol Pharmacol 32, 3: 248-55 Pub Med 1116 2718
6. Monkiedje A, Wall JH, Englande AJ, Anderson AC (1990) A new method for determing concentrations of Endod-S (Phytolacca dodecandra) in water during mollusciciding Environ Sci Health B 25, 6: 777-86 Pub Med 2280 058
7. Ndamba J, Lemmich E, Moolgard P (1994) Investigation of the diurnal,ontogenetic and seasonal variation in the molluscicidal saponine content of Phytolacca dodecandra aqueous berry extracts Phytochem 35,1: 95-9
8. Ndamba J, Chandiwana SK (1988) The geographical variation in the molluscicidal potency of Phytolacca dodecandra (Endod) berries in Zimbabwe Prop Geogr Med 40, 1: 34-8
9. Parkhurst RM (1975) The chemotaxonomy of Phytolacca species Indian J Chem 13, 7: 757-8 Pub Med 1225 8562
10. Parkhurst RM, Mtupha BM, Liang YS et al. (1989): The molluscicidal activity of Phytolacca dodecandra I: Location of the activating esterase Biochem Biophys Res Commun 158, 2: 436-9
11. Slacanin I, Marston A, Hostettmann K (1988) High-performance liquid chromato-graphic determination of molluscicidal saponines from Phytolacca dodecandra (Phytolaccaceae) J Chromatogr 448, 2: 265-74
12. Stobaeus JK, Heath GE, Parkhurst RM et al. (1990) A laboratory study of the toxicity of the butanol extract of endod (Phytolacca dodecandra) on two species of freshwater fish and two species of aquatic snails Vet Hum Toxicol 32,3: 212-6 PubMed 2353429
13. Treyvaud V, Marston A, Dyatmiko W, Hostettmann K (2000) Molluscicidal saponines from Phytolacca icosandra Phytochem 55: 603-9