This is a phytochemical and zootechnical study on *Physalis peruviana* leaves in streptozotocin induced diabetic rats. This was part of a scientific development program of plant resources used in Congolese traditional medicine for the treatment of diabetes mellitus in which individual and community consequences are well established. Different fractions with hexane, ethyl acetate and the residue were obtained from the hydroalcoholic extract of *P. peruviana* leaves. Phytochemical screening was focused on the usual reactions of characterization based on precipitation and coloration with general reagents. The diabetic conditions were induced in rats by a single administration of streptozotocin (50 mg/kg body weight) intravenously. The positive control group received glibenclamide (6.5 mg/kg body weight) and each test group received 100 mg/kg of body weight. Those groups were compared with a control group which received only a Tween 20 solution (1 ml per 100 g body weight). Zootechnical profiles were evaluated by weight monitoring as well as food and water consumption in rats. Phytochemical screening showed the presence of polyphenols, flavonoids, alkaloids, saponins, tannins, anthocyanins, mucilages, cardiac glycosides, coumarins and betalains in the hydroalcoholic extract and its fractions. A highly significant difference (P < 0.001) of water consumption in opposition to the food intake and weight changes was observed. This study suggested the isolation and characterization of compounds from hydroalcoholic extract from the leaves of *P. peruviana* L. and its fractions for an extensive antidiabetic investigation.

**Key words:** *Physalis peruviana*, phytochemical, antidiabetic activity, streptozotocin, zootechnical parameters.

**INTRODUCTION**

Several pathophysiological processes are involved in the development of diabetes mellitus. These range from autoimmune destruction of the β-cells of the pancreas with a consequent insulin deficiency to abnormalities that result in resistance to insulin action (Armelle et al., 2008; Arika et al., 2016).
Diabetes mellitus is the most prevalent disease in the world, affecting 25% of the population and afflicts 150 million people and is predicted to rise to 300 million by 2025 (WHO, 2002; Babu, 2016). Conventional management of diabetes is expensive and therefore not affordable by many patients, especially in developing nations. More so, conventional drugs are not readily available and have been found to have side effects with long term use (Arika et al., 2015; Deeni and Sadiq, 2002). The distinctive traditional medical opinions and natural medicines have shown a bright future in the therapy of diabetes mellitus and its complications (Ekramul et al., 2002; Arika et al., 2016).

The World Health Organization (WHO, 2002) recommended the use of medicinal plants for the management of DM and further encouraged the expansion of the frontiers of scientific evaluation of hypoglycemic properties of diverse plant species (WHO, 2002; Kwete et al., 2002; Chikezie et al., 2015).

Plants have potential sources of hidden phyto-constituents which can be responsible for solving various potential health problems (Noumi and Yomi, 2001; Kwete et al., 2007). Medicinal plants have curative properties due to the presence of various complex chemical substances of different composition, which are found as secondary plant metabolites in one or more parts of plants (Li et al., 2007; Patil, 2016). They are also associated with reduced risks of cancer, cardiovascular disease, diabetes and lower mortality rates of several human diseases (Momeni et al., 2005; Ozkan et al., 2016).

Physalis peruviana (Solanaceae) is a medicinal plant widely used in folk medicine for treating diseases such as malaria, asthma, hepatitis, dermatitis, cancer, diuretic, rheumatism, antispasmodic, diuretic, antiseptic, sedative, analgesic diseases, and has antidiabetic, antifungal, antibacterial, anti-inflammatory, cataract-cleaning, antidiabetic and anti-parasitic properties (Mariotte et al., 2005; Kasali et al., 2013a; Çakir et al., 2014; Joshi and Joshi, 2015; Lashin et al., 2016; Higaki et al., 2016; Chang et al., 2016).

In a survey conducted in the Eastern part of the Democratic Republic of the Congo (DRC), a number of traditional healers pointed out the use of Physalis peruviana L. leaves for this purpose (Kasali et al., 2013b). The objectif of this study was to analyze the phytochemical composition and evaluate zootechnical parameters of a hydroalcoholic extract and its fractions in diabetic rats.

MATERIALS AND METHODS

Study sites

The present study was undertaken at the laboratory of Pharmacognosy, Faculty of Medicine and Pharmacy, Official University of Bukavu/Republic Democratic of Congo and chemical study of medicinal plants, bacteria, fungi and endophytes were done at Faculty of Sciences, University of Yaounde 1/Cameroon; Phytochemical laboratory of Higher Teachers’ Training College (Faculty of Sciences, University of Yaounde 1) and laboratory of Toxicological and Pharmacological Studies (Faculty of Medicine and Biomedical Sciences/ University of Yaounde 1). This study was conducted between September 2015 and April 2016.

Plant material

The leaves of P. peruviana L. (Solanaceae) were collected at Lwiro (Center for Research in Natural Sciences, Democratic Republic of Congo) situated at 50 km from Bukavu (South Kivu, Democratic Republic of Congo). They were identified a by Mr. Gentil IRAGI of Botany Department of this center and compared with voucher specimen No.2044. The leaves were air-dried and powdered for analysis.

Preparation of hydroalcoholic extract and its fractions

800 g of the powdered leaves of P. peruviana were macerated with 6 L of 70% EtOH (Jothi et al., 2015) for 48 h and the combined filtrate (using the Whatman filter paper No. 1) was evaporated under reduced pressure using a rotary evaporator. A dried extract with a yield of 28.95% was obtained. One part of the filtered hydroalcoholic extract was stored in a refrigerator at 4°C. Another part of this extract was soaked in hexane and decanted into a funnel. The hexane fraction was concentrated in a rotary evaporator (BÜCHI 461 water Bath). This operation was repeated several times until total exhaustion (the solution has become colorless). The same operations were carried out with ethyl acetate. The residue from this fraction was concentrated under reduced pressure using a rotary evaporator. The following yields were obtained: 3.19 and 25.06%, respectively, for hexane and ethyl acetate.

Animals

Healthy male albino Wistar rats (body weight 175 ± 10.6 g) aged 2-3 months, were used in the study. The rats were maintained under standard laboratory conditions at 27.75 ± 1°C, and normal photo period (12 h dark/12 h light) was used for the experiment. The rats were acclimatized to the laboratory conditions a week prior to the experiment.

The experimental protocol and the maintenance of the experimental animals was done in accordance with the regulations of the Organization for Economic Co-operation and Development (OECD) guide since in Cameroon, the ethics committee focuses only clinical studies. The animal experiment protocols were carried out in accordance with the guidelines of the ICH on preclinical pharmaceutical testing in mouse (OECD, 2001; Tsague et al., 2016).

Animal ethical regulatory consideration

Healthy male albino Wistar rats (body weight 150 to 250 g) were ethically required for use for the experiment according to
the ICH guidelines.

The experimental protocol and the maintenance of the experimental animals was done in accordance with the regulations of the OECD guide, the EU parliament directives on the protection of animals used for scientific purposes, since in Cameroon, the ethics committee focuses only on clinical studies. The animal experiment protocols was carried out in accordance with the guidelines of the ICH on preclinical pharmaceutical testing in mouse (OECD, 2001; Akbarzadeh et al., 2007).

Phytochemical screening

Qualitative phytochemical tests of R. heudelotti methanolic extract were carried out according to Odebiyi and Sofowora (1978) methods to identify some components such as alkaloids, saponins, tannins, flavonoids, polyphenols and anthraquinones.

Test for alkaloids: 0.5 g of the sample was stirred with 5 ml of 1% aqueous HCl on a steam bath and then filtered. 1 ml of the filtrate was treated with a few drops of Mayer’s reagent and a second 1 ml portion was treated similarly with Dragendorff reagent. Turbidity or precipitation with either of these reagents was taken as evidence for the presence of alkaloids in the extract.

Test for saponins: The ability of saponins to produce frothing in aqueous solution and to haemolysed red blood cells was used for the screening test. 0.5 g of plant extract was shaken with water in a test tube. Frothing which persisted on warming was taken as evidence for the presence of saponins.

Test for tannins: 0.5 g of dried extract was stirred with 5.0 ml of distilled water. This was filtered and ferric chloride reagent was added to the filtrate. A blue-black precipitate was taken as evidence for the presence of tannins.

Test for phenol and polyphenols: 0.5 g of plant extract was heated for 30 min in a water bath. 3 ml of 5% FeCl₃ was added to the mixture, then followed by the addition of 1 ml of 1.00% potassium ferrocyanide. The mixture was filtered and green (phenol) and blue (polyphenol) colours were observed.

Test for anthraquinones: 0.5 g of plant extract was shaken with 5 ml of benzene, filtered and 2 ml of 10% ammonia solution was added to the filtrate. The mixture was shaken and the presence of a pink or violet colour in the ammoniacal (lower) phase indicated the presence of free hydroxy anthraquinones.

Test for flavonoids: 0.5 g of plant extract was dissolved in 5 ml of NaOH at 1 N. The change of the yellow colour obtained after adding HCl 1 N indicated the presence of flavonoids.

Zootecchnical study in Streptozotocin-induced diabetic rats

Induction of diabetes

Diabetes was induced in fasted rats injecting 50 mg/kg streptozotocin (Sigma, France) in the tail vein. STZ was dissolved in 0.1 M citrate buffer (pH 4.5) (Tadjeddine et al., 2013). Streptozotocin induces diabetes within 3 days by destroying the beta cells (Ziane et al., 2015). After 48 h of STZ administration, blood glucose level of each rat was determined (Anthikat et al., 2016). Before induction, all rats were fasted 12 h (Ngueguim et al., 2016). Rats with serum glucose level above 300 mg/dl were considered as diabetic (Khathi et al., 2013).

Experimental protocol

The rats were divided into six groups of five rats in each group.

Group 1: Untreated rats (control), received vehicle alone (1% tween 20, 1 ml per orally); Group 2: Rats treated with 6.5 mg/kg of glibenclamide, positive control; Group 3: Rats treated with 100 mg/kg of hydroalcoholic extract of P. peruviana; Group 4: Rats treated with 100 mg/kg of the hexane fraction of P. peruviana; Group 5: Rats treated with 100 mg/kg of the ethyl acetate fraction of P. peruviana; Group 6: Rats treated with 100 mg/kg of the residue fraction of P. peruviana.

All rats were administered single dose of drug (orally) daily for 28 days. Daily administration was through a gastric gavage by inducing a gastric tube (Galiterrez et al., 2014). The day of administration of first dose was considered the zero day of treatment.

At the end of the experimental period, all animals were deprived of food overnight and then sacrificed by cervical decapitation after anesthetizing by either inhalation (Saini and Sharma, 2013). One touch electronic Glucometer (One Touch Ultra®) was used for glucose measurement.

Water consumption and food intake

The body weight of each rat was measured once each week and the total amount of food consumed was recorded 3 times per week (Gutierrez et al., 2005).

Body weight monitoring

Body weights of all animals in each group were monitored using a top loader weighing balance throughout the experimental period (Ofusori et al., 2012).

Statistical analysis

All results were expressed as mean ± standard error (SE) for each sample. Statistical analysis was performed using GraphPad Prism 5.02 statistical package (GraphPad Software, USA). The data were analyzed by one way analysis of variance (ANOVA) followed by Turkey’s multiple comparison post test. Differences between groups were considered to be significant at P < 0.05.

RESULTS

Phytochemical analysis

The results in the Table 1 represent the phytochemical analysis of some fractions from the hydroalcoholic extract of P. peruviana leaves. According to these results, the phytochemical analysis showed the presence of tannins and saponins in all fractions except in the hexane fraction. However, the method used did not show resins and oxalates. In addition, the highest percentage of positive tests were obtained from the hydroalcoholic extract (37.5%) respectively, followed by fractions with ethyl acetate residue (25%) and ethyl acetate (25 %), and finally the hexane fraction (12.5%).
Table 1. Phytochemical screening of *Physalis peruviana* leaves.

<table>
<thead>
<tr>
<th>Metabolite</th>
<th>Reagent methods</th>
<th>HydAE</th>
<th>HexF</th>
<th>EthAF</th>
<th>ResEAF</th>
</tr>
</thead>
<tbody>
<tr>
<td>Polyphenols</td>
<td>Ferric chloride</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>Lead acetate</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Flavonoids</td>
<td>Iso amyl alcohol/Mg + hydrochloric acid</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>Hodger</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Alkaloids</td>
<td>Wagner</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>Mayer</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
</tr>
<tr>
<td>Cardiac glycosides</td>
<td>Glacial acetic acid/ FeCl₃ + Sulfuric acid</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Saponosides</td>
<td>Frothing test</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Tannins</td>
<td>Ferric chloride</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Anthocyanins</td>
<td>Sulfuric acid/Ammonia</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Quinones</td>
<td>Sodium hydroxyde</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Mucilages</td>
<td>Ethanol 95°</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>Resins</td>
<td>Glacial acetic acid/ H₂SO₄</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Betalains</td>
<td>Sodium hydroxyde</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Terpenoids and steroids</td>
<td>Liebermann-Burchard</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Coumarins</td>
<td>Ferric chloride/HNO₃</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Oxalates</td>
<td>Glacial acetic acid</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>


Zootechnical evaluations

Water consumption and food intake

The water consumption showed a highly significant increase (** = P > 0.001) in all animals treated against healthy animals (control), however intake food variation showed a highly significant decrease (P > 0.001) in all animals treated as compared to the healthy animals (Figure 1).

Body weight monitoring

There was a highly significant (p< 0.001) body weight changes in all treated rat groups as compared to the control the group (Figure 2). Between groups treated (with glibenclamide and plant extract/fractions), there was no significant difference (P<0.05). There was a significant difference (p< 0.05) between the control group and those treated with the aqueous alcoholic extract.

Assessment of weight variation of organs

From the study as shown in Table 2, there was no significant difference (p< 0.05) in the change of organ weight in all animals treated for heart, liver, brain, spleen and the two kidneys. However, a significant difference (p< 0.05) in weight variation of pancreas, liver, brain, lungs and testicles was noticed (Table 2).

DISCUSSION

There was an uneven distribution in this study of secondary metabolites in the hydroalcoholic extract and its fractions. Saponins and tannins tests were positive in the extract and in two of its fractions (ethyl acetate and its residue fractions), and having 25% of positive tests. Polyphenols compounds, flavonoids, anthocyanins, mucilage, cardiac glycosides and coumarin represented 9.4% in each category. This category is followed by grouping betalains (6.2%). Alkaloids represented 15.6% of positive tests for three different reagents used, which gave an average of 5.2% per reagent. Steroids and terpenes represented 3.1%. Quinones, resins, and oxalates are not found (0%).

Many preceding studies reported the presence of some secondary metabolites in the fruit or the leaves of *P. peruviana*. Some studies indicated that cardiac glycoside, alkaloid, saponins, tannins, steroids and terpenoids and flavonoids were present while anthraquinones were absent.
in the leaves (Moabe et al., 2013; Magambu et al., 2014). A phytochemical screening in regeneration plant, callus from seed, leaf and fruits from mother plants of *P. peruviana*, showed the presence of alkaloids, glycosides, cardiac glycosides, saponins, phenol, sterol, tannins, flavonoids and diterpene (Lashin and Elhaw, 2016). A phytochemical investigation of the crude ethanolic extract of *P. peruviana* L. revealed the presence of phenols, flavonoids, phytosterols, glycosides, sterols, saponins, tannins and alkaloids (Ahmed, 2014). Previous phytochemical studies have isolated a number of compounds from *P. peruviana*, such as ticloidine, withanolides, phenolics and phytosterols (Gautam et al., 2015).
Table 2. Weight variation of organs.

<table>
<thead>
<tr>
<th>Organs</th>
<th>Group 1</th>
<th>Reference</th>
<th>Exp. I</th>
<th>Exp. II</th>
<th>Exp. III</th>
<th>Exp. IV</th>
</tr>
</thead>
<tbody>
<tr>
<td>Heart</td>
<td>0.66±0.09</td>
<td>0.73±0.04</td>
<td>0.57±0.10</td>
<td>0.59±0.06</td>
<td>0.48±0.06</td>
<td>0.55±0.14</td>
</tr>
<tr>
<td>Lung</td>
<td>2.20±0.27</td>
<td>2.57±0.08</td>
<td>1.06±0.19**</td>
<td>1.78±0.58</td>
<td>1.05±0.1**</td>
<td>1.90±0.14</td>
</tr>
<tr>
<td>Liver</td>
<td>5.33±0.5</td>
<td>7.40±0.56**</td>
<td>5.72±0.58</td>
<td>5.68±0.62</td>
<td>5.58±0.39</td>
<td>5.69±0.64</td>
</tr>
<tr>
<td>Brain</td>
<td>1.62±0.05</td>
<td>1.52±0.07</td>
<td>1.52±0.12</td>
<td>1.42±0.13*</td>
<td>1.55±0.08</td>
<td>1.55±0.08</td>
</tr>
<tr>
<td>Pancreas</td>
<td>0.77±0.09</td>
<td>0.48±0.04**</td>
<td>0.57±0.05*</td>
<td>0.45±0.1**</td>
<td>0.55±0.01**</td>
<td>0.52±0.1*</td>
</tr>
<tr>
<td>Spleen</td>
<td>0.76±0.14</td>
<td>0.71±0.15</td>
<td>0.69±0.15</td>
<td>0.65±0.09</td>
<td>0.50±0.10</td>
<td>0.96±0.07</td>
</tr>
<tr>
<td>Right kidney</td>
<td>0.67±0.06</td>
<td>0.85±0.01</td>
<td>0.79±0.11</td>
<td>0.67±0.11</td>
<td>0.62±0.06</td>
<td>0.63±0.02</td>
</tr>
<tr>
<td>Left kidney</td>
<td>0.73±0.05</td>
<td>0.84±0.03</td>
<td>0.77±0.11</td>
<td>0.69±0.14</td>
<td>0.62±0.09</td>
<td>0.63±0.05</td>
</tr>
<tr>
<td>Right testicle</td>
<td>1.52±0.41</td>
<td>1.39±0.03</td>
<td>1.03±0.46</td>
<td>0.72±0.2**</td>
<td>0.62±0.03**</td>
<td>0.62±0.01*</td>
</tr>
<tr>
<td>Left testicle</td>
<td>1.54±0.33</td>
<td>1.42±0.01</td>
<td>1.08±0.41*</td>
<td>0.71±0.1**</td>
<td>0.56±0.01**</td>
<td>0.61±0.02*</td>
</tr>
</tbody>
</table>

The values are expressed as mean ± SEM of the respective groups (n=5). The weight values of groups are compared with normal control animals, value *p< 0.05 and **p< 0.01.

Exp. I: Group treated with 100 mg/kg of hydroalcoholic extract of the plant; Exp. II: Group treated with 100 mg/kg of the hexane fraction of plant; Exp. III: Group treated with 100 mg/kg of the ethyl acetate fraction of plant, Exp. IV: Group treated with 100 mg/kg of the residue fraction of the plant.

**P. peruviana** contain the pseudo-steroids (physalines) and glycosides which show the anticancer activity. From the aerial parts of *P. peruviana*, various withanolide glycosides have been isolated. From the whole plant material, there is isolation of two withanolides (Sharma et al., 2015). Three new physalin steroids, physalin III, physalin IV, 3-O-methylphysalin X, together with five known physalins were isolated from the 80% EtOH extract of calyces of *P. alkekengi* var. francheii (Yu et al., 2013).

Withanolides are natural steroidal lactones produced mainly by plants in the Solanaceae that often have many health benefits such as anti-inflammatory activity (Ahmed, 2014). *Physalis* (L.) species contain various carbohydrates, lipids, minerals, vitamins and phytosteroles (Sharma et al., 2015). This genus contains calystegins, alkaloids from nortropane, and steroidal glycoalcaloids from spirosolane (Jouzier et al., 2005; Xu et al., 2013).

A number of phytochemical are known, some of which include: alkaloids, saponins, flavonoids, tannins, glycosides, anthaquinones, steroids and terpenoids. They do not only protect the plants but have enormous physiological activities in humans and animals. These include cancer prevention, antibacterial, antifungal, antioxidative, hormonal action, enzyme stimulation and many more. Phytochemicals are responsible for the medicinal activity of plants and they have protected human from various diseases (Savithramma et al., 2011). Many classes of plants secondary metabolites, such as alkaloids, terpenoids, polyphenols, flavonoids and many others show promising antidiabetic potentials. These natural constituents may act as a promising source of delivering oral hypoglycemic effect with minimal side effects (Singab et al., 2014).

According to the results, the administration of a single dose of Streptozotocin (50 mg/kg weight body) increased water consumption. Other studies have shown that diabetes mellitus is characterized by classical symptoms such as polyphagia, and polydipsia which are exhibited in HFD-STZ diabetic rats and this may be attributed to the impaired glucose homeostasis as a result of insulin inefficiency (Akbarzadeh et al., 2007). Water consumption was inversely related to food intake, this was an indication that the decrease in food intake in diabetic animals was linked to the significant amount of sugars in the blood that had an impact on the index of satiety. High levels of sugars were associated with decreased appetite and short-term food intake as has been reported also (Anderson and Woodend, 2003). Oral treatment with the fruit extract of *P. peruviana* to diabetic group of rats decreased food and fluid consumptions which could be due to improved glycemic status (Sathyadevi et al., 2014).

This study showed a significant decrease in final weight, weight gain at p< 0.001 and also food intake at p< 0.05 as compared to control group. Both *Physalis* powder and juice treated groups showed a significant decrease in final weight (p< 0.01 and 0.05, respectively), weight gain and FER.
at \( p < 0.05 \) as compared to the control group. Aqua *Physalis* extract and methanol *Physalis* extract treated groups showed non-significant difference in these parameters as compared to the control group. *Physalis* powder, juice, aqua *Physalis* and methanol extract treated groups showed a significant increase in final weight, weight gain, food intake as compared to reference group (Hafez et al., 2011). This study also showed differences in some changes in organ weight and body weight. Diabetic condition has been known to be associated with weight loss as reported by Anderson and Woodend (2003). The weight loss recorded in untreated diabetic animals could be a symptom of ill health, which may have been caused by the release of free radicals (Abdelmoaty et al., 2010).

The streptozotocin-induced diabetes rats showed significant loss of body weight with respect to the extract treated and controlled groups. Kumar et al. (2011) reported that antidiabetic and antihyperlipidemic effects is best induced in rat models using streptozotocin-induced rat models for better comparison with test plants, and give a better result profile of the test battery. With respect to the reference group, the inability of the plant to improve the animals' weight, at the end of treatment was observed, although a stabilization of weight was recorded at the end of treatment.

**Conclusion**

The result of this present study showed that the hydroalcoholic extract of *P. peruviana* and its fractions contains many secondary metabolites which will be used against diabetes. It is interesting to isolate and characterize some compounds of this plant and to extend their antidiabetic potential investigations.

**CONFLICT OF INTERESTS**

The author(s) have not declared any conflict of interests.

**ACKNOWLEDGEMENTS**

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**REFERENCES**


Genève. OMS: P 65.