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Effect of simultaneous treatment with Ukrain and amitriptyline on selected biochemical serum parameters in rodents

Wpływ jednoczesnego stosowania leku Ukrain i amitryptyliny na wybrane parametry biochemiczne w surowicy krwi gryzoni

INTRODUCTION

Major depressive disorder is a common and serious illness with the potential of becoming the leading cause of disability worldwide. The lifetime prevalence rate is 16.2%, and is expected to increase. In the elderly, prevalence is about 3% in the general population but 15%–25% among nursing home residents [5, 17]. These numbers may be even higher, because it is estimated that clinically significant depression goes untreated in 60% of the elderly [18]. The average age of onset of depression is the mid-20s. The lifetime risk in women is twice the risk in men, and is increased during the reproductive years [5]. Traditional pharmacotherapy includes tricyclic antidepressants (TCAs). These drugs were shown to be comparable or more effective than selective serotonin reuptake inhibitors (SSRIs) and amitriptyline is considered very effective. Unfortunately, TCAs are less well tolerated but efficacy in treatment-resistant depression may be their advantage [1, 9]. TCAs are known for a lot of side effects, the most typical atropinic side effects (dry mouth, blurry vision, constipation, urine retention, tachycardia, sedation and memory impairment), but especially liver dysfunction and interactions with other drugs [3, 14, 21]. TCAs also have a small therapeutic range, so therapeutic drug monitoring is useful. Female gender and higher drug doses increase the risk of side effects [17].

The process of cancer diagnostics and treatment provokes strong stress reaction which may be the cause of depressed mood [2]. The prevalence of psychiatric disorders in cancer patients assessed with a variety of instruments is estimated to be between 25% and 60%. Recent reports suggest that major depressive disorder occurs quite frequently among patients with cancer and the prevalence of depression among these patients is often underestimated, partly because many symptoms of depression (fatigue, weight loss, sleep disruption, loss of appetite,) closely mirror the physiological effects of cancer [16].

Ukrain (i.e. tiophosphoric acid derivative of alkaloids *Chelidonium majus* L.) is a semi-synthetic cytostatic drug [12, 15, 19]. It reduces toxic effects of earlier administered cytostatics as well as prolonging and improving the quality of patients' lives [6]. In addition, drug Ukrain exhibited significant hepatoprotective effect in human and experimental animal hepatitis [10, 13, 20]. These results point to the possibility of its use in the treatment of hepatitis induced as a side effect by different drugs. Because of the lack of literature data on simultaneous treatment with Ukrain and TCAs, our study was aimed to evaluate whether, and in what degree, Ukrain with amitriptyline (AMI) affect the serum biochemical parameters indicating liver function in rodents.

MATERIAL AND METHODS

D r u g s a n d c h e m i c a l s . The following substances were used in the study: Ukrain – i.e. tiophosphoric acid derivative of alkaloids *Chelidonium majus* L. (aqueous high-purity concentrate 1:33, Ukrainian Anti-Cancer Institute, Vienna, Austria), amitriptyline hydrochloride (AMI) from Sigma-Aldrich GmbH (Germany) and also *aqua pro injectione* (Polfa Lublin, Poland). Ready-made diagnostic kits were used to determine: aspartate (AST) and alanine (ALT) aminotransferases activities, total protein level (Cormay Diagnostic S.A., Lublin, Poland) and α-fetoprotein (AFP) level (IBL, Hamburg, Germany).

A n i m a l s . The study was carried out on Albino-Swiss male mice (20–25 g) and male Wistar rats (200–250 g) coming from a licensed breeder. The animals were kept at room temperature (20±1ºC) under a natural day-night cycle in constant environmental conditions (humidity, noise). They had access to food and water *ad libitum*. The experiments were approved by the Local Ethics Committee on Animal Experimentation of the Medical University of Lublin.

T r e a t m e n t s . Aqueous solutions of AMI (5 mg/kg for mice and 10 mg/kg for rats) and Ukrain (9.5 or 19 mg/kg for mice and 14 or 28 mg/kg for rats) were prepared *ex tempore* and administered intraperitoneally (ip) once daily for 10 days separated or combined in constant volumes 0.1 ml/10 g of mice body weight and 0.5 ml/100 g of rat body weight. Our previous research [7, 8] proved the above-mentioned doses of AMI and Ukrain were effective. The control groups received the same amounts of *aqua pro injectione*.

E x p e r i m e n t a l p r o t o c o l s . The experimental groups consisted of eight animals each. 24 hours after the last injection, the animals were decapitated and the blood was taken and centrifuged for 10 minutes at 3000 rpm. The serum was stored at -20 ºC until biochemical determinations.

S t a t i s t i c a l a n a l y s i s . Results are expressed as mean ± SEM. Statistical significance among groups was determined by Student's *t*-test and *p*-values less than 0.05 were considered significant.

RESULTS AND DISCUSSION

The major depressive disorder occurs quite frequently among patients with cancer [16]. Antidepressant drugs, which have to be used simultaneously with specific oncologic therapy, can induce toxic drug-drug interactions, especially TCAs, though their effectiveness even in treatment-

resistant depression. Post-treatment liver damage may be the consequence of such interaction. The aminotransferases' activities, particularly liver-specific alanine transaminase activity, are basic diagnostic parameters supplying important information on the liver condition. The levels of total protein and AFP in blood serum can also be affected in the case of liver damage.

The results showed the 10-day combined treatment with Ukrain (9.5 or 19 mg/kg for mice and 14 or 28 mg/kg for rats) and amitriptyline (5 mg/kg for mice and 10 mg/kg for rats) caused changes in serum levels of total protein, AFP and activity of aminotransferases compared with those of Ukrain and amitriptyline groups of rodents. The species-depending differences were observed. In mice the activities of ALT and AST were generally decreased (except Ukrain 19 mg/kg with AMI) in serum (Fig. 1, 2). However, in rats the combined administration resulted only in the ALT activity's increase when compared with only AMI group as well as an increase or a decrease (Ukrain 14 or 28 mg/kg with AMI, respectively) when compared with only Ukrain group (Fig. 3) but AST activity remained unaffected. The total protein concentration also differed in mice and rats. In mice pre-treated with Ukrain and AMI this parameter was significantly enhanced (Fig. 4) but decreased in rats (Fig. 5). In addition, this simultaneous treatment caused a very significant increase of AFP concentration only in mice (Fig. 6).

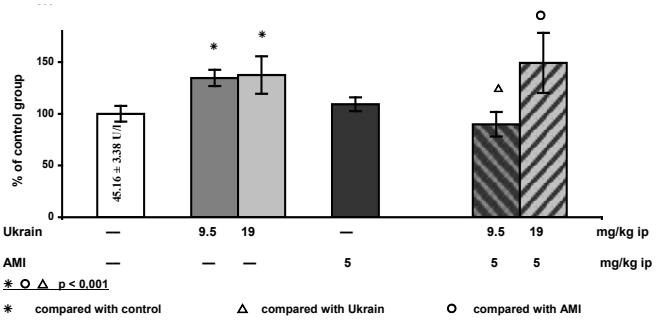

Fig. 1. Effect of 10-day simultaneous treatment with Ukrain and AMI on AST activity in mice serum

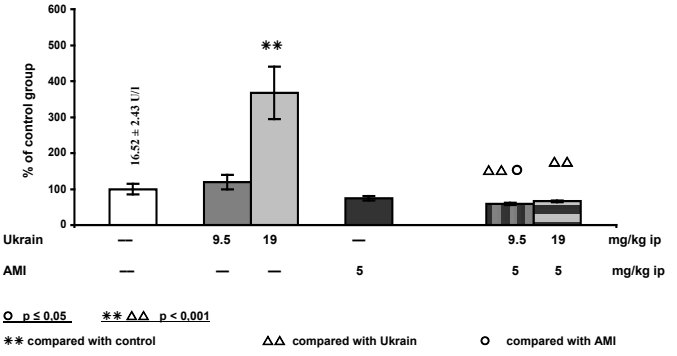

Fig. 2. Effect of 10-day simultaneous treatment with Ukrain and AMI on ALT activity in mice serum

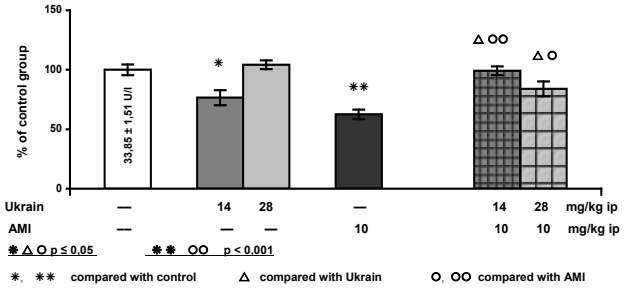

Fig. 3. Effect of 10-day simultaneous treatment with Ukrain and AMI on ALT activity in rat serum

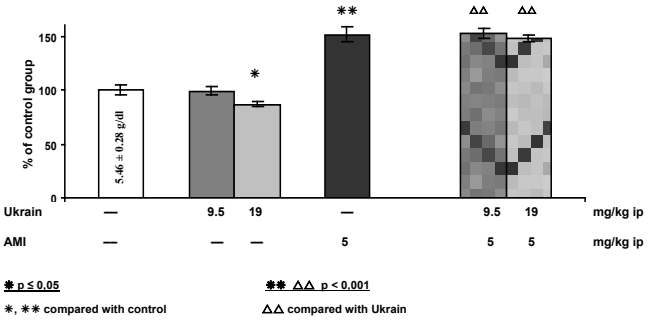

Fig. 4. Effect of 10-day simultaneous treatment with Ukrain and AMI on total protein level in mice serum

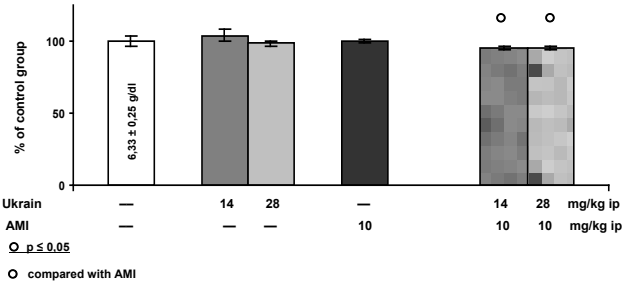

Fig. 5. Effect of 10-day simultaneous treatment with Ukrain and AMI on total protein level in rat serum

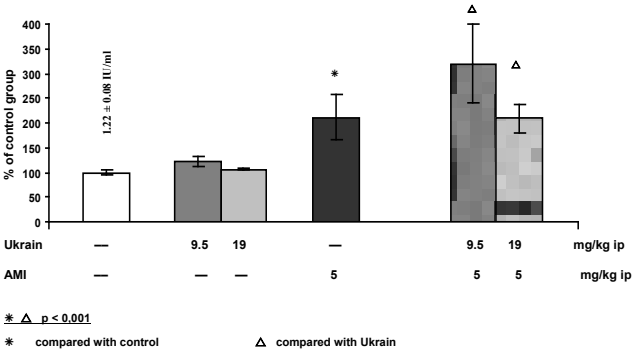

Fig. 6. Effect of 10-day simultaneous treatment with Ukrain and AMI on AFP level in mice serum

Our research indicated that amitriptyline administered at 5 mg/kg to mice and 10 mg/kg to rats for 10 days led to the decrease of ALT activity in rat serum (Fig. 3) but did not affect the activities of both transaminases in mice serum. The concentration of total protein after AMI treatment was significantly increased only in mice sera (Fig. 4) such as AFP level in these samples (Fig. 6). The 10-day treatment with Ukrain 9.5 mg/kg and 19 mg/kg increased AST and ALT (only 19 mg/kg) activities in mice blood serum (Fig. 1, 2). On the contrary, Ukrain administered at 14 mg/kg to rats decreased ALT activity in rat blood serum (Fig. 3), which is in agreement with previous studies [7, 11].

Amitriptyline, the most widely used tricyclic antidepressant, has been associated with very rare but severe incidences of hepatotoxicity in patients. While the mechanism of idiosyncratic hepatotoxicity remains unknown, it is proposed that metabolic activation of amitriptyline and subsequent covalently binding of reactive metabolites to cellular proteins play a causative role [21]. Literature data confirmed the damaging effect of AMI on liver cells characterized by enhanced activity of ALT [14]. Ukrain does not show hepatotoxic effect [4, 12, 19]. It can even decrease the number of side effects and improve the effectiveness of the human hepatitis treatment [20]. The hepatoprotective effect of Ukrain in experimental hepatitis in rodents was also observed [10, 13].

The present study indicates that the observed changes in examined biochemical parameters in rodents treated with Ukrain and amitriptyline together pointed out to significant differences between both species. Our research also showed that Ukrain did not act as a protection against the adverse effect of amitriptyline on the liver functional state. In addition, the results may suggest the possibility of hepatic disorder when Ukrain and amitriptyline are administered together for a long time. We suppose the results can be used to avoid or limit this adverse effect.

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SUMMARY

The effects of 10-day treatment of rodents with Ukrain and amitriptyline (AMI) on selected biochemical serum parameters indicating liver function have been examined. In the samples of rodents' serum AST and ALT activities, total protein and α-fetoprotein (AFP) levels were determined. The results indicated 10-day simultaneous administration of Ukrain and AMI significantly decreased ALT and AST activities in mice (except Ukrain 19 mg/kg with AMI). However, in rats the combined treatment led to ALT activity's increase when compared with only AMI group as well as an increase or a decrease (Ukrain 14 or 28 mg/kg with AMI, respectively) when compared with only Ukrain group but did not affect AST activity. In mice serum the investigated proteins' concentrations (total protein and AFP) were significantly enhanced in comparison with proper control groups. However, in rat serum only total protein level was decreased. Therefore, adverse changes in the examined biochemical parameters indicating liver function disorder were observed after combined administration of Ukrain and AMI to rodents. Our results may be helpful in clinical practice to avoid this effect.

Keywords: Ukrain, amitriptyline, hepatotoxic effect, rodents

STRESZCZENIE

W pracy oceniano wpływ jednoczesnego 10-dniowego stosowania leku Ukrain oraz amitryptyliny (AMI) na wybrane parametry biochemiczne świadczące o funkcji wątroby gryzoni. W surowicy krwi zwierząt oznaczono aktywność AST i ALT oraz stężenie α-fetoproteiny (AFP) i białka całkowitego. Na podstawie uzyskanych wyników stwierdzono, że 10-dniowe stosowanie u gryzoni leku Ukrain w kombinacji z AMI zmniejszało aktywność ALT i AST (z wyjątkiem Ukrainu 19 mg/kg i AMI) w surowicy myszy. Jednakże u szczurów obserwowano zwiększenie aktywności ALT w porównaniu z AMI jak również zwiększenie jej bądź zmniejszenie (odpowiednio Ukrain 14 lub 28 mg/kg łącznie z AMI) w porównaniu z lekiem Ukrain oraz brak wpływu na aktywność AST. Z kolei stężenia AFP oraz białka całkowitego w surowicy krwi myszy poddanych łącznemu działaniu leku Ukrain i AMI były istotnie większe w porównaniu z odpowiednimi grupami kontrolnymi. Natomiast u szczurów obserwowano tylko istotne zmniejszenie stężenia białka całkowitego. Nieprawidłowości w badanych parametrach biochemicznych w surowicy krwi zwierząt wskazują na możliwość wystąpienia zaburzenia czynności wątroby po łącznym 10-dniowym podaniu leku Ukrain z amitryptyliną, co może okazać się istotne w praktyce klinicznej.

Słowa kluczowe: Ukrain, amitryptylina, hepatotoksyczność, gryzonie