IN VITRO COMPARATIVE OF DIFFERENT ACYCLOVIR TABLET FORMULATION IN COMPARISON WITH PURE ACYCLOVIR

Nadheerah Falih Neamah

College of Pharmacy ,University of Basrah,Basrah,Iraq.

(Received 27 May 2012,Accepted 21 June 2012)

Keywords; Veramide**,** Acyclovir**,** tablet formulation**.**

ABSTRACT

The aim of the study was to compare the in vitro performance of different acyclovir tablets of the same strength from different companies under different trade names: Acyclovir actavis 400mg tab ,Acic 400mg tab ,and Veramide 400mg tab., and compare with pure acyclovir.

The comparison include friability, thickness, and hardness as non official tests, and dissolution and weight variation as official tests. Dissolution test is the most important test in which we can determine the real amount of active ingredient (acyclovir) found in each tablet formulation in addition to the rate of drug release from these formulation .

The results reveals that no significant changes observed in thickness, hardness ,and Friability ,all the formulation were found within the limits.

The dissolution rate exerts significant differences after 5min, 51.9% of veramide, 76.4% of acyclovir activs, and acic 58.3% in comparison with 88.5% of acyclovir . After 10 min, only the percentage released from acic.(75.72%) was significantly differ from reference drug acyclovir 94.5% .While the results of this study showed a significant differences of percentage released after 20 min from veramide ,acyclovir actavis ,and acic are (95.7, 97.2,and 99.1 respectively) in comparison with acyclovir(99.5%). Amount of drug that released after40 min ,the percentage of drug released from all test tablets not significantly differ from that released from acyclovir100%..

Conclusion an attempt was made to compare three acyclovir different forms Acyclovi actavis , Acic ,and Veramide and compare the dissolution rate with standard acyclovir .The results showed a significant decrease in dissolution rate after 5 ,and20 min ,only acic exert a significant reduction after 10 min. All tablets released active ingredient in non significant manner after 40 min . further in vivo studies showed be done to reveal how such results affect the pharmacokinetic of mentioned drugs.

INTRODUCTION

Acyclovir, or Zovirax, as it is commonly known, is an antiviral medication used to treat cold sores and genital herpes caused by the herpes virus. It is also prescribed to treat chickenpox and shingles. It is given as a cream or ointment (topical use), oral tablets, or intravenous liquid. The efficacious treatment schedules of oral acyclovir have involved the ingestion of medication five times daily due to low and limited bioavailability(15 to 30%), the requirement for frequent dosing may be inconvenient and result in poor compliance and therapeutic failure(1) ,the absorption is variable and incomplete following oral administration. It is about 20% bound to plasma protein and is widely distributed throughout body tissues. Significant amounts may be found in amniotic fluid, placenta, and breast milk. the main focus of pregnancy studies with acyclovir has been to look for birth defects following first trimester exposure. However, limited information suggests there is no increased risk for other problems such as low birth weight, preterm delivery, or stillbirth(2). Most of acyclovir dose is excreted in the urine as unchanged drug; a small portion is excreted as an oxidized inactive metabolite. The plasma half-life of acyclovir is 3 to 4 hours in patients with normal kidney function and up to 20 hours in patients with renal impairment. Reversible renal dysfunction (azotemia) and neurotoxicity (tremor, seizure, delirium) are dose limiting toxicities of intravenous acyclovir. Adequate hydration and slow drug infusion can minimize the risk of renal toxicity. The efficacy of oral aciclovir is limited as a result of its low bioavailability(3,4)

Acyclovir is converted to its active metabolite via three phosphorylation steps. First, viral thymidinekinase converts acyclovir to acyclovir monophosphate. Next, host cell enzymes convert the monophosphate to the diphosphate and then to the active compound, acyclovir triphosphate, which accumulates only in virus-infected cells. The active metabolite of acyclovir inhibits herpes virus DNA replication in two ways; acts as a competitive inhibitor for the incorporation of deoxyguanosine triphosphate (dGTP) into the viral DNA. In addition, acyclovir that is incorporated into viral DNA acts as a chain terminator because it lacks the 3-hydroxy group necessary for further chain elongation. Viral DNA polymerase becomes irreversibly bound to an acyclovir-terminated DNA chain and is unavailable for further replicative activity. The effect of acyclovir on host cell DNA synthesis is much smaller than its effect on the viral enzyme. Concentrations of acyclovir significantly beyond the therapeutic range are required to inhibit host cell growth.

The most common mechanism of resistance to acyclovir involves mutations that result in decreased thymidine kinase activity. Therefore, these viral mutants exhibit cross-resistance to other antiviral agents that require thymidinekinase activation, such as famciclovir, ganciclovir, and valacyclovir . A rare mechanism of acyclovir resistance involves decreased affinity of viral DNA polymerase for the drug. Oral acyclovir is useful in the treatment of Herpes Simplex virus HSV-1 and HSV-2 infections, such as genital herpes, herpes encephalitis, herpes keratitis, herpes labialis, and neonatal herpes. In initial episodes of genital herpes, oral acyclovir has been found to reduce viral shedding, increase the speed of healing of lesions, and decrease the duration of pain and new lesion formation. (5)

Aside from drug hypersensitivity, there are no absolute contraindications to the use of acyclovir, A potentially fatal disorder, thrombotic thrombocytopenic purpura hemolytic uremic syndrome, has been reported in immunocompromised individuals. Probenecid has been shown to inhibit the renal clearance of acyclovir.(6).

MATERIAL AND METHODS

Evaluation of Tablets

Hardness

The force required to break a tablet in a diametric compression i.e. Hardness or tablet crushing strength was measured using Monsanto tablet hardness tester. The hardness was measured in terms of kg/cm2.

Thickness

Thickness of tablet was measured by using vernier calipers. Three tablets were selected at random from each batch and average thickness in mm was reported.

Weight variation

Twenty tablets from each batch were selected at a random and average weight was determined. Then individual tablets were weighed and was compared with average weight.

Friability

Friability of the tablet determined using Roche friabilator. It subjects the tablet to the combined effect of abrasion and shock in a plastic chamber revolving at 25rpm and dropping a tablet at height of 6 inches in each revolution. reweighed sample of tablets was placed in the friabilator and were subjected to the 100 revolutions. Tablets were de-dusted using a soft muslin cloth and reweighed.

The % friability (F) is given by the formula.

$$F = \frac{W_{intial} - W_{final}}{W_{intial}} * 100$$

Where, $W_{initial}$ is total weight of tablets before subjecting to the test .W_{final} is total weight of tablets after subjecting to the test (7,8). It must be less than or equal to1%

In vitro drug release

The dissolution test that made on tablets done by using dissolution apparatus 2 (paddle apparatus) set at 50 rpm for 40min in 0.1N HCl as gastric fluid simulator in 37°c .Aliquots of 10 ml was collected at predetermined time intervals and replenished with an equivalent volume of fresh medium. The samples were filtered through a filter paper and diluted suitably with 0.1 N HCl and were analyzed using UV/visible spectrophotometer at 252 nm. Percent cumulative drug release was plotted against time in minutes to obtain dissolution profile. The UV absorbance of

the resultant solutions was used to determine the concentration of that solutions by using the equation of standard curve (calibration curve) of acyclovir .(9)

RESULTS

Hardness, Thickness ,and . Friability

Thickness and hardness of all the formulations ranged in between 3.62mm to 4.16mm and lower hardness 3.5± 0.129099 kg/cm2(of acic) to (6.1± 0.05) kg/cm2(Veramide), respectively. Friability of all the tablets were found to be less than 1% which was in accordance to the IP specifications for friability and which confirms the mechanical stability of tablets.% drug content of all the formulations was found to be in the range of 99.24%(aciclovir actavis) to 100.9% (acic) which was acceptable. Also weight variation of all the formulation batches was found to be in the permissible limits of ± 5%.(Appendix contain tables of results)

The dissolution rate

In the present study the percentage of drug that released after 5 min ,exert significant differences . 51.97402% of veramide, 76.44932% of acyclovir activs, and acic 58.3328% in comparison with 88.55 of acyclovir. The last amount that released was 207.8mg from veramide tab.,as in fig :1

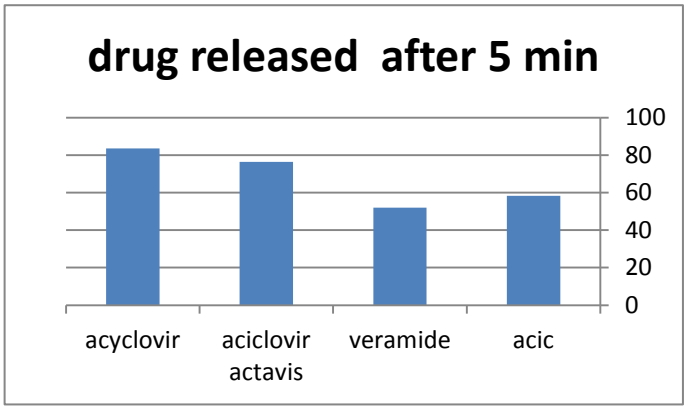

Fig 1:The amount of drug released after 5 min, significant differences in percentage of the amount released from each tab., in comparison with acyclovir

After 10 min the percentage of amount released were not significant differences of a viramide tab. 80.52%, and aciclovire actavis 90.6% in comparsion with control drug. However the precentage released from acic.(75.72%),which was significantly differ from reference drug acyclovir 94.5% as in fig 2

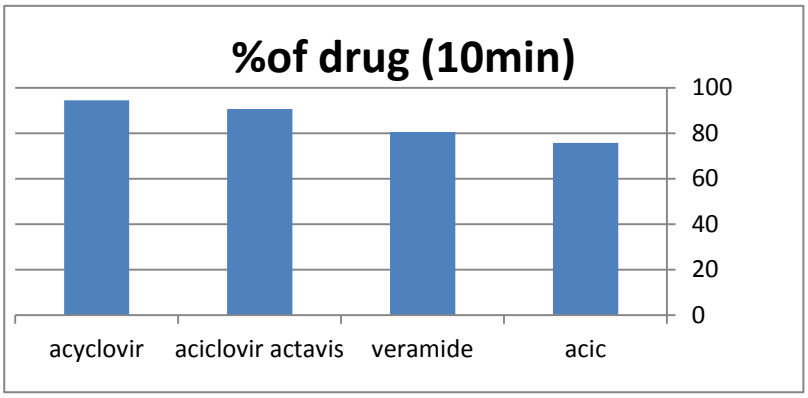

Fig 2: significant amount that released from aciclovire actavis ,and viramide ,and acic, in comparison with acyclovir

Amount of drug that released after 20min ,the results of this study showed a significant differences of percentage released after 20 min from veramide ,&acyclovir actavis ,and acic are (95.7,& 97.2,and 99.1 respectively) in comparison with acyclovir(99.5%)(fig3)

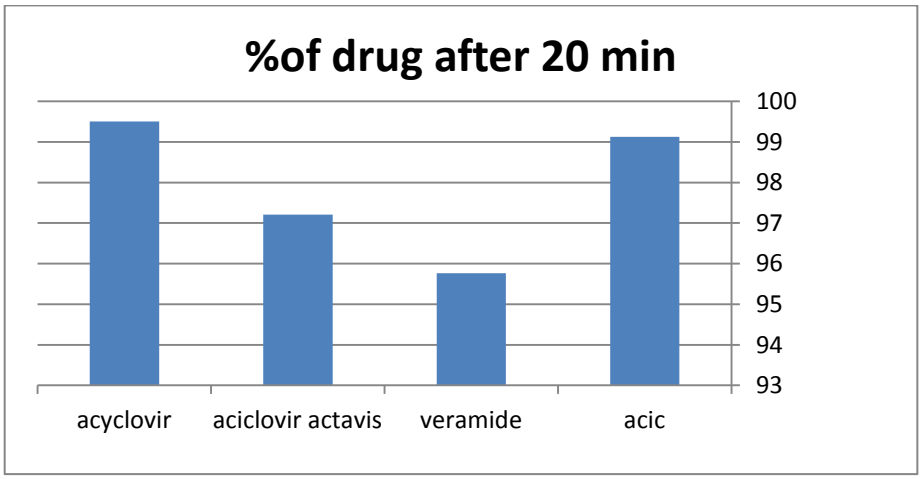

Fig3: shows a significant differences in percentage of the drug released from test tablets in comparison with reference drug acyclovir

Amount of drug that released after40 min ,the percentage of drug released from acic ,veramide, and acyclovir activis (100.9,99.7,and99.2% respectively) not significantly differ from that released from acyclovir.(fig,4) .All results were shown in fig5

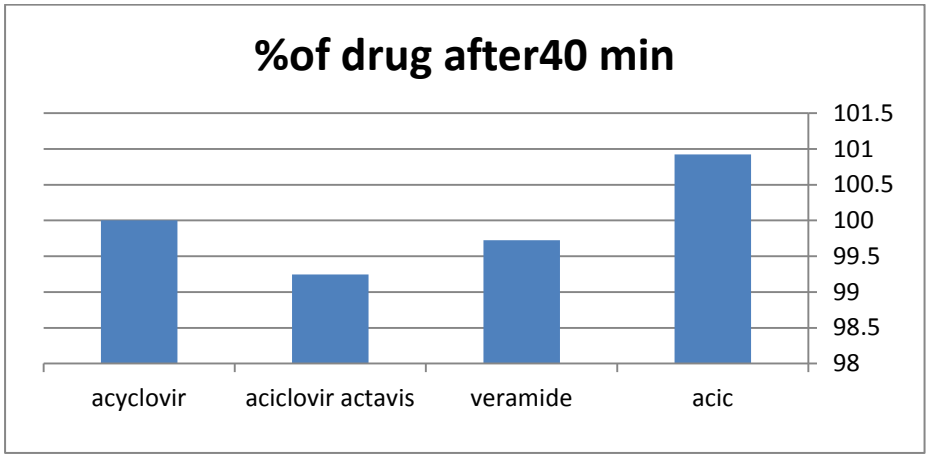

fig4: reveals that no significant differences among test drugs in comparison with reference drug

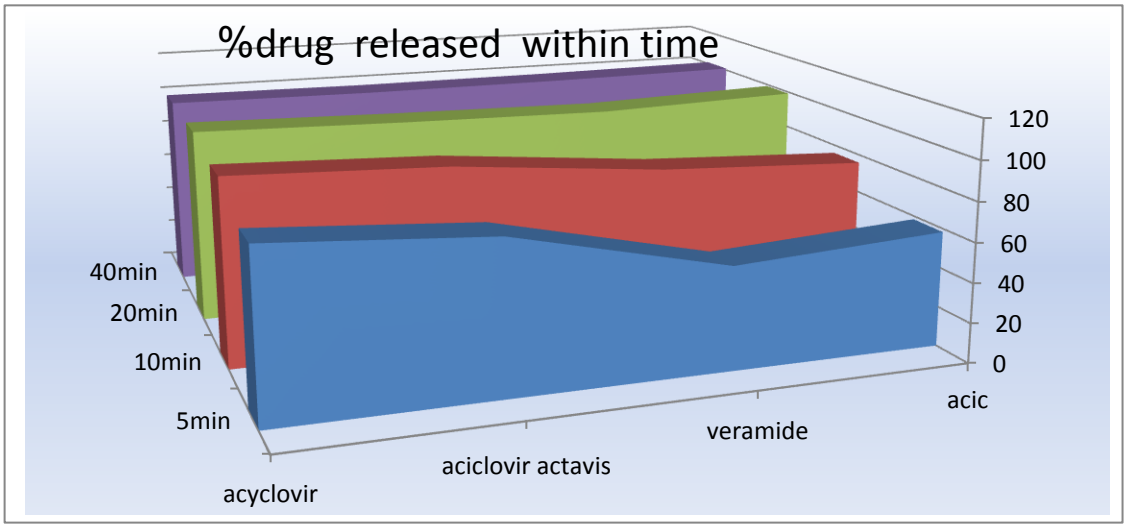

Fig 5: exerts the percentage released from test drugs in all time of estimation in comparison with control drug

Appendix

Friability test

Veramide	Acyclovir actavis	Acic
W1= 10.2gm	W1= 9.8gm	W1= 10.6gm
W2=10.18gm	W2= 9.784gm	W2= 10.565gm
Loss%=0.2%	Loss%=0.16%	Loss%= 0.33%

Weight Variation (uniformity of weight) of tablets .Weight of tablet 324 mg or more then %error = ±5%

Veramide	Acyclovir actavis	Acic (mg)	Tab No.
510.8	490	530.2	1
510	490	529.4	2
510.2	490.1	530	3
510.3	490	530	4
510.1	489.8	529.8	5
510.5	489.9	530	6
510	490.2	530.6	7
499.2	490	530.1	8
499.7	490.3	530.2	9
510	490	530	10
499.7	489.7	530.2	11
510.3	490.55	529.8	12
499.8	490.1	529.9	13
510	49.45	529.7	14
510	490	529.8	15
510.2	489.9	530.3	16
499.5	490.4	530	17
510	490	530	18
499.8	489.6	530.1	19
510	490	529.9	20
510	490	530	Mean
25.5	24.5	26.5	5%
535.5	514.5	556.5	Upper
474.5	465.5	503.5	Lower

Amount of drug released after 5 min

p value	% of DR	Amount of RD mg	SD	Mean con of drug released(RD)	drug
	83.55	334.2	8.784	0.003114	acyclovir
0.000495011	58.3328	233.3312	0.0566	0.002593	acic
0.000495011	51.97402	207.8961	0.05886	0.00231	veramide
0.002617833	76.44932	305.7973	0.06138	0.003398	aciclovir actavis

Amount of drug released after 10 min

p value	% of DR	Amount of RD mg	SD	Mean con of drug released(RD)	drug
	94.5	378.1	0.0005766	0.004395	acyclovir
0.037274961	75.72946	302.9178	3.48305	0.003366	acic
0.066080724	80.52854	322.1142	1.74152	0.003579	veramide
0.291485303	90.6066	362.4264	3.01641	0.004027	aciclovir actavis

Amount of drug released after 20 min

p value	% of DR	Amount of RD mg	SD	Mean con of drug released(RD)	drug
	99.5	398.1111	9.68379	0.004762	acyclovir
0.004117615	99.12497	396.4999	2.04212	0.004406	acic
0.000544823	95.76561	383.0625	4.39714	0.004256	veramide
0.002568564	97.20534	388.8213	1.06646	0.00432	aciclovir actavis

Amount of drug released after 20 min

p value	% of DR	Amount of RD mg	SD	Mean con of drug released(RD)	drug
	100	400	5.17347	0.004457	acyclovir
0.358898032	100.9246	403.6985	1.23144	0.004486	acic
0.414429413	99.72485	398.8994	1.74152	0.004432	veramide
0.229423661	99.24494	396.9798	4.60764	0.004411	aciclovir actavis

DISSCUSION

Tablets which can rapidly dissolve or disintegrate in the oral cavity have attracted a great deal of attention. Rapidly dissolving or disintegrating tablets are not only indicated for people who have swallowing difficulties, but also are ideal for active people.(7) Failure to respond to acyclovir therapy may arise from an inadequate dose

(frequency of dose or total daily dose); patient noncompliance; malabsorption in the intestine; or, resistant viral strains. The need for readily absorbed oral antiviral agents has been identified as imperative for treatment of viral diseases to both patient populations(10)

Few direct comparisons of antiviral medications have been done, and we are not aware of any other studies(11) .Acyclovir is a white, crystalline powder with the molecular formula $C_8H_{11}N_5O_3$ and a molecular weight of 225. The maximum solubility in water at 37°C is 2.5 mg/mL.

Acyclovir is commonly used as the free acid form in solid dosage forms, whereas the sodium salt is used in parental dosage forms. The solubility was vary slightly with pH and lowest

solubility of 2.3 mg/mL at pH 5.8 at 37°C. Absorption of acyclovir from the gastrointestinal tract is variable and incomplete; 10–30% of an oral dose may be absorbed . This poor systemic bioavailability is considered to be a result of the characteristics of the drug itself and not its delivery vehicle . Because of its high hydrophilic nature, absorption of acyclovir occurs mainly by passive diffusion mechanism and it is slow, variable and incomplete manner.

In vitro release profiles of all three tablets were comparatively evaluated ,

Hardness can affect the disintegration. So if the tablet is too hard, it may not disintegrate in the required period of time. And if the tablet is too soft, it will not withstand the handling during subsequent processing such as coating or packaging. In general, if the tablet hardness is too high, first tablets were check for their disintegration before rejecting the patch. And if the disintegration is within limit, the patch will be accepted. In the present study the all the formulation batches was found to be in the permissible limits of ± 5%.

An instrument called friabilator is used to evaluate the ability of the tablet to withstand abrasion in packaging, handling, and shipping. The tendency of tablets to powder, chip, or fragment is called Friability; and this can affect the elegance appearance, consumer acceptance of the tablet, and also add to tablet's weight variation or content uniformity problems. It must be less than or equal to1% but if more we do not reject the tablets as this test is non-official. The mechanical stability of tablets in the present study was within normal limits(<1%).

The increase in dissolution rate of the drug may be due to increase in wettability, hydrophilic nature of the carrier and due to reduction in drug crystallinity.(12)

Dissolution profile of the veramide batches, acic and aciclovir actavis were studied and compared with the dissolution profile of pure acyclovir (Fig. 1)

The dissolution rate exerts significant differences after 5min in comparison with control, all active ingredient released from test tablets were less than control. Aciclovir actavis was the higher percentage of drug released from other test tablets . After 10 min , the amount released from acic.(75.72%),which was significantly differ from reference drug acyclovir 94.5%. While reduction of percentage released from veramide,&acyclovir actavis(95.7,97.2 respectively) are not significantly important .

The results of this study showed a significant differences of percentage released after 20 min from veramide ,&acyclovir actavis ,and acic are (95.7,& 97.2,and 99.1

respectively) in comparison with acyclovir(99.5%) after 20 min, while the drug released were not significantly differ from control drug after 40 min.

The results in the present study is agreed with other study ,which showed that the physical properties like tablet hardness, weight values obtained and acyclovir content uniformity in both test and reference product were consistent and within the acceptance range(13), The disintegration
test results demonstrated that acyclovir tablets held different release kinetics compared to reference product as anticipated.(14,15). The dissolution behaviors of all commercially available acyclovir tablet products dissolved quicker in 0.1N HCL, medium, which was more than 85% of drug substance is released within 30 minutes(16) .

Conclusion an attempt was made to compare three acyclovir different forms Acyclovir actavis , Acic ,and Veramide and compare the dissolution rate with standard acyclovir .the results showed a significant decrease in dissolution rate after 5 ,and20 min ,only acic exert a significant reduction after 10 min. All tablets released active ingredient in non significant manner . further in vivo studies showed be done to reveal how such results affect the pharmacokinetic of mentioned drugs.

دراسة للمقارنة خارج الجسم بين انواع مختلفة من حبوب الاسايكلو فير بالمقارنة مع الاسايكلوفير النقي

نظيرة فالح نعمة

فرع الادوية والعلوم السريرية , كلية الصيدلة, جامعة البصرة ,البصرة,العراق.

الخلاصة

اقيمت هذه الدراسة للمقارنة خارج الجسم لحبوب ايكلو فير بنفس القوة من مختلف الشركات وباسماء تجارية مختلفة :ايكلوفير اكتفيس 400 ملغم ،اساك 400ملغم،وفيرامايد 400ملغم

المقارنة تتضمن قابلية التحمل ،السمك، والصلابة كاختبارات غير تقنية ،و اختلافات التحلل والوزن كاختبارات تقنية

اختبار التحلل اهم الاختبارات التي يمكن تحديد الكمية الحقيقة للمادة الفعالة الموجودة في تركيب الحبوب بالاضافة الى معدل اطلاق الدواء من تركيبة الدواء.امتصاص اشعة فوق البنفسجية للمحلول الناتج المستخدم لتحديد تركيز المحاليل بواسطة معادلة المنحنى القياسي للاسايكلو فير .اظهرت النتائج ليس هناك اي تغيير معتد احصائيا في السمك والصلابة وقابلية التحمل.جميع التركيبات ضمن الحدود

معدل التحلل ظهر هناك فرق معتد احصائيا بعد 5 دقائق 51ز97%للفيراميد,76ز44%للاسايكلوفير اكتيفيس ،و58.33%للاساك بالمقارنة مع 88.55%للاسايكلوفير..بعد 10 دقائق النسبة المئوية التي تتحرر من الاسايك 75.72% ,التي تختلف بشكل معتد احصائيا عن دواء السيطرة 94.5% . بينما النتائج في هذه الدراسة اظهرت اختلافات معتدة احصائيا بعد 20دقيقة من الافيرامايد ,الاسايكلو فير,و

الاساك(95.7,97.2,99.1%بالترتيب). اما كمية الدواء التي اطلقت من بعد40دقيقة من جميع الحبوب المختبرة لاتختلف بشكل معتد احصائيا عن التي اطلقت من شؤغؤمخرهق100%.

كنتيجة محاولة لجعل مقارنة بين ثلاثة من مختلف حبوب الاسايكلوفير اسايكلوفير اكتيفس والاسايك والافيراميد,ومقارنة معدل التحرر مع الاسايكلوفير القياسي .النتائج اظهرت هناك انخفاض في معدل التحلل التي تؤثر بعد 5,ووبعد20 دقيقة,وفقط الاساك اظهرانخفاض بشكل معتد احصائيا بعد 10 دقائق.جميع الحبوب اطلقت المادة الفعالة بشكل غير معتد احصائيا بعد40دقيقة .دراسات داخل الجسم يجب ان تجرى لتوضيح مدى تاثير هذه النتائج على الحركة الدوائية للاوية المذكورة

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