Chula Med J Vol. 53 No. 6 November - December 2008

Sterility and stability of manually diluted 2.5 % phenylephrine hydrochloride for pupillary dilatation*

Juntarut Vaivanijkul**

Kittisak Kulvichit** Wasee Tulvatana**

Vaivanijkul J, Kulvichit K, Tulvatana W. Sterility and stability of manually diluted 2.5 % phenylephrine hydrochloride for pupillary dilatation. Chula Med J 2009 Nov - Dec; 53(6): 489 - 97

Introduction : *Comparing with 10 % phenylephrine hydrochloride, the 2.5 % concentration is potentially safer, and has been proved to be equally effective in pupillary dilatation for posterior eye segment examination, but it is not commercially available in Thailand.*

Objective : *To study the sterility and stability of manually diluted 2.5 % phenylephrine hydrochloride solutions.*

Setting : *Outpatient eye clinic, King Chulalongkorn Memorial Hospital.*

Research design : *Clinical experimental study.*

Methods : *The 10 % phenylephrine hydrochloride was manually diluted with normal saline solution, Tears Naturale II® eye drop, balanced salt solution and Opsil tears® eye drop to make four kinds of 2.5 % phenylephrine hydrochloride solutions. All of the solutions were cultured on day 0, 1, 3, 5, 7, 10, and 14 after dilution to detect microbial contamination. The 2.5 % solutions, in combination with 1 % tropicamide, were then applied to the patients 0, 1, 3, 5, 7, 10, and 14 days after the dilution. The pupillary size before and after application of the solutions was measured under slit lamp biomicroscope.*

* Presented in part as a poster at the 16th scientific meeting of the Royal College of Ophthalmologists of Thailand, Phetchaburi, Thailand, December 2-4, 2005.

** Department of Ophthalmology, Faculty of Medicine, Chulalongkorn University

Results : *All of the microbial culture results were negative. A total of 189 eyes from 101 patients (29 men, 72 women) were recruited. The mean age of the patient was 50.6 ± 17.3 years. The normal saline solution group included 28 patients, the Tear Naturale II® eye drop group included 22 patients, the balanced salt solution group included 25 patients and the Opsil tears® eye drop group included 26 patients. The manually diluted 2.5 % phenylephrine hydrochloride solutions can increase the pupillary size to approximate 7.1 to 8.8 mm.*

Conclusion : *All of the manually diluted 2.5 % phenylephrine hydrochloride solutions in combination with 1% tropicamide has sufficient clinical efficacy for the pupillary dilatation. The sterility and stability of the solutions maintained for at least 2 weeks.*

Keywords : *Pupillary dilatation, Phenylephrine hydrochloride, Stability, Sterility.*

Reprint request : Kulvichit K. Department of Ophthalmology, Faculty of Medicine, Chulalongkorn University, Bangkok 10330, Thailand. E-mail: kulvichit@gmail.com

Received for publication. June 28, 2006.

จันทรัสม์ ไววนิชกุล, กิตติศักดิ์ กุลวิชิต, วสี ตุลวรรธนะ. การปลอดเชื้อและเสถียรภาพของสารละลาย 2.5 % phenylephrine hydrochloride ที่ได้จากการผสมขึ้นเองเพื่อใช้ในการขยายรูม่านตา. จุฬาลงกรณ์เวชสาร 2552 พ.ย. - ธ.ค.; 53(6): 489 - 97

บทนำ : *เมื่อเปรียบเทียบกับ 10 % phenylephrine hydrochloride แล้วพบว่า 2.5 % phenylephrine hydrochloride มีประสิทธิภาพในการขยายรูม่านตาได้ดีเท่า ๆ กัน และยังน่าจะมีความปลอดภัยมากกว่า แต่ไม่มีจำหน่ายในประเทศไทย*

วัตถุประสงค์ : *เพื่อศึกษาถึงการปลอดเชื้อและเสถียรภาพของสารละลาย 2.5 % phenylephrine hydrochloride ที่ได้จากการผสมขึ้นเอง*

สถานที่ทำวิจัย : *แผนกผู้ป่วยนอกทางจักษุวิทยา โรงพยาบาลจุฬาลงกรณ์*

รูปแบบการวิจัย : *การทดลองทางคลินิก*

วิธีการศึกษา : *นำ 10 % phenylephrine hydrochloride มาผสมกับสารละลาย 4 ชนิด ได้แก่ normal saline solution, Tear Naturale II® eye drop, balanced salt solution และ Opsil tears® eye drop เพื่อให้ได้สารละลาย 2.5 % phenylephrine hydrochloride สี่ชนิด สารละลาย ทุกชนิดจะถูกตรวจสอบถึงการปนเปื้อนจากจุลชีพ โดยทำการเพาะเชื้อในวันแรกที่ผสม และวันที่ 1, 3, 5, 7, 10 และ 14 ภายหลังการผสม สารละลายแต่ละชนิดจะถูกหยอดร่วมกับ 1 % tropicamide ให้แก่ผู้ป่วย ตั้งแต่วันแรกที่ผสม และในวันที่ 1, 3, 5, 7, 10 และ14 ภายหลังการผสม โดยใช้ slit lamp biomicroscope วัดขนาดของรูม่านตา*

ผลการศึกษา : *ผลการเพาะเชื้อจุลชีพของทุกตัวอย่างไม่พบเชื้อ ทำการศึกษาใน 189 ตาจากผู้ป่วย 101 ราย เป็นเพศชาย 29 ราย หญิง 72 ราย อายุเฉลี่ย 50.6 ± 17.3 ปี มีผู้ป่วยในกลุ่มที่ได้รับ normal saline solution, Tear Naturale II® eye drop, balanced salt solution และ Opsil tears® eye drop ทั้งสิ้น 28, 22, 25 และ 26 รายตามลำดับ สารละลาย 2.5 % phenylephrine hydrochloride ที่ได้จากการผสมขึ้นเองสามารถขยายม่านตาได้เฉลี่ย 7.1-8.8 มม.*

สรุป : *สารละลาย 2.5 % phenylephrine hydrochloride ที่ได้จากการผสมขึ้นเองทุกชนิด เมื่อหยอดร่วมกับ 1 % tropicamide มีประสิทธิภาพทางคลินิกเพียงพอในการขยายม่านตา ซึ่งสารละลายดังกล่าวทุกชนิดมีการปลอดเชื้อและเสถียรภาพอย่างน้อย 2 สัปดาห์*

คำสำคัญ : *การขยายรูม่านตา, การปลอดเชื้อ, เสถียรภาพ, ฟีนีลเอฟรีน ไฮโดรคลอไรด์*

Pharmacologic pupillary dilatation is necessary for a thorough posterior eye segment examination. Mydriasis can be achieved by synergistic effect of sympathomimetic drugs and parasympatholytic drugs, usually by instillation of 10 % phenylephrine hydrochloride and 1 % tropicamide eye drops. The former drug has a potent sympathomimetic effect. In one drop, there is 3-4 times the maximum safe dose for intravenous administration of the drug, even to a young healthy adult.[1] In the manufacturer package insert, the company recommends to instil the drug not for more than 1 drop per eye per hour. There have been reports of serious adverse events of 10 % phenylephrine hydrochloride eye drop such as severe hypertension,[1,2] acute myocardial infarction,[1] acute subarachnoid haemorrhage,[2] ventricular arrhythmia,[3] and death.[1] The unfavourable cardiovascular consequences are more pronounced in diabetic patients, and patients receiving reserpine or guanethidine.[4]

Although a previous study has shown that the cardiovascular effects are not different in 2.5 % and 10 % phenylephrine groups during ophthalmic surgeries,[5] a larger controlled trial has shown that 2.5 % concentration of phenylephrine had lower effect on blood pressure and pulse rate when compared with its 5 % counterpart.[6] The rise in blood pressure and heart rate was seen significantly in 5 % group immediately after anaesthesia.

When comparing the effectiveness of 2.5 % phenylephrine with 10 % phenylephrine, the pupillary dilatation power has been clinically proven to be similar in light-coloured irides,[7,8] diabetic patients with various iris colours,[9] and dark-coloured irides.[10,11] A much lower concentration (0.5 %) of phenylephrine may not be as effective as 2.5 % phenylephrine in dark-coloured irides, as seen in a study of Chinese population which found that 0.5 % phenylephrine in combination with 0.5 % tropicamide has a lower efficacy than 2.5 % phenylephrine in combination with 1 % tropicamide.[12]

Due to possible serious adverse events, the 2.5 % solution is recommended to be used cautiously in low birth weight infants, elderly, and patients who take drugs that might enhance the cardiovascular side effects such as atropine, monoamine oxidase inhibitors, and tricyclic antidepressants.[13]

The 2.5 % phenylephrine hydrochloride has clinical effectiveness in pupillary dilatation as 10 % phenylephrine but with potentially fewer side effects, and therefore should replace 10 % phenylephrine in general use. Unfortunately, 2.5 % phenylephrine hydrochloride eye drop is not commercially available in Thailand. Therefore, we conducted a study to evaluate the sterility and stability of the manually diluted 2.5 % phenylephrine by various available solvents to find a suitable formula for a self-prepared solution used for pupillary dilatation.

Methods

The study protocol was approved by the Ethics Committee of the faculty. The manually diluted 2.5 % phenylephrine hydrochloride was prepared by the hospital's pharmaceutical unit under a sterile technique. The solvents used were normal saline solution (Thai Otsuka Pharmaceutical, Samutsakorn, Thailand), Tears Naturale II® eye drop (Alcon-Couvreur, Puurs, Belgium), balanced salt solution (Baxter Corporation, Canada) and Opsil tears® eye drop

(Silom Medical, Bangkok, Thailand). One mL of 10 % phenylephrine Hydrochloride (Mono drop® 10 % Neo-Synephrine®, Winthrop Laboratories, New York, USA) was diluted with 3 mL of the solvents to make four different kinds of 2.5 % phenylephrine hydrochloride solutions. After dilution, all of the solutions were stored at 4 degree Celsius. Cultures for aerobic bacteria, anaerobic bacteria, and fungus were done at day 0, 1, 3, 5, 7, 10, and 14 after dilution.

Each of 2.5 % phenylephrine hydrochloride solutions was applied to consecutive outpatients of ophthalmologic department who required pupillary dilatation on day 0, 1, 3, 5, 7, 10, and 14 after dilution. The exclusion criteria were patients with previously diagnosed hypertension, diabetes mellitus, any heart diseases, iris abnormalities, any lesion of optic nerve or oculomotor nerve, or other ophthalmic conditions that might interfere with the pupil size measurement. Those patients with a history of intraocular surgery, using mydriatic drug or long-term use of miotics were also excluded.

One drop of 2.5 % phenylephrine hydrochloride solution was instilled into the patient's lower fornix, followed by one drop of 1 % Tropicamide at 5 minutes later, for three times in 10 minutes apart. The pupillary size was measured under slit lamp biomicroscope just before the first drop and at 30 minutes after the last instillation. Approximately 3-5 patients were recruited for the dilutions kept for each period of time.

Results

The culture results of all kinds of 2.5 % phenylephrine solution on day 0, 1, 3, 5, 7, 10, and 14 were negative.

A total of 189 eyes from 101 patients (29 men and 72 women) were included in the study. The mean age of the patient was 50.6 ± 17.3 years. The normal saline solution group, the Tear Naturale II® eye drop group, the balanced salt solution group, and the Opsil tears® eye drop group included 28, 22, 25 and 26 patients, respectively (Table 1).

Table 1. Demographic data of the patients (total n = 101).

Patient data	Type of 2.5 % phenylephrine hydrochloride solution			
	NSS	TN II	BSS	Opsil
Age (years)				
Mean	53.4	50.2	48.6	49.7
Range	(21 - 74)	(17 - 84)	(17 - 83)	(6 - 80)
Gender (n)				
Male	8	8	7	6
Female	20	14	18	20

NSS = Normal saline solution, TN II = Tear Naturale II® eye drop,
BSS = balanced salt solution, Opsil = Opsil tears® eye drop

Mean pupil size before dilatation with each solution on day 0, 1, 3, 5, 7, 10, and 14 after mixture are shown in Table 2. Mean pupil size at 30 minutes after dilatation with each solution on day 0, 1, 3, 5, 7, 10, and 14 after mixture are shown in Table 3. All of the 2.5 % phenylephrine solutions along with 1 % tropicamide can dilate the pupil up to 7.1-8.8 mm. Mean difference of pupil size before and after dilatation with each solution on day 0, 1, 3, 5, 7, 10, and 14 after mixture are shown in Table 4. The solutions can increase the pupil size for at least 4 mm from baseline.

Table 2. Mean (range) of the pupil size at baseline in millimetres.

Day after mixture	NSS	TN II	BSS	Opsil
0	2.6 (2.1 - 3.0)	1.9 (1.9 - 2.3)	2.8 (2.4 - 3.0)	2.6 (2.0 - 3.0)
1	2.4 (2.0 - 3.0)	2.4 (2.2 - 2.5)	3.2 (2.5 - 5.0)	2.2 (1.8 - 2.5)
3	2.2 (2.0 - 2.3)	2.3 (2.1 - 2.5)	2.3 (2.0 - 2.5)	2.2 (2.0 - 2.4)
5	2.1 (1.8 - 2.5)	2.6 (2.3 - 3.0)	2.4 (2.0 - 3.0)	2.4 (2.0 - 3.0)
7	2.4 (1.8 - 3.0)	2.5 (2.0 - 2.8)	2.3 (2.0 - 2.7)	2.0 (1.8 - 2.2)
10	2.3 (2.1 - 2.7)	2.4 (2.2 - 2.8)	2.2 (2.0 - 2.5)	2.7 (2.3 - 3.0)
14	2.5 (2.2 - 3.4)	2.3 (2.1 - 2.4)	2.2 (2.0 - 2.5)	2.1 (2.0 - 2.2)

NSS = Normal saline solution, TN II = Tear Naturale II® eye drop,

BSS = balanced salt solution, Opsil = Opsil tears® eye drop

Table 3. Mean (range) of the pupil size at 30 minutes in millimetres.

Day after mixture	NSS	TN II	BSS	Opsil
0	8.0 (7.2 - 9.0)	8.4 (7.8 - 8.8)	8.3 (7.0 - 9.0)	8.2 (7.7 - 9.3)
1	7.7 (7.0 - 8.4)	7.5 (6.8 - 8.0)	8.8 (8.5 - 9.0)	7.7 (7.4 - 8.0)
3	7.6 (7.0 - 8.2)	7.1 (5.5 - 8.2)	8.2 (7.5 - 8.5)	8.1 (7.4 - 9.0)
5	7.7 (7.1 - 8.1)	7.9 (7.5 - 8.5)	7.8 (7.5 - 8.0)	7.3 (6.9 - 7.5)
7	8.0 (6.8 - 9.2)	7.8 (7.5 - 8.0)	8.4 (8.0 - 8.8)	7.2 (6.5 - 7.9)
10	7.8 (7.5 - 8.2)	7.6 (6.5 - 8.9)	7.8 (7.1 - 8.4)	8.8 (8.3 - 9.3)
14	7.8 (6.9 - 9.0)	7.7 (7.2 - 8.3)	8.3 (7.5 - 9.2)	7.8 (7.5 - 8.0)

NSS = Normal saline solution, TN II = Tear Naturale II® eye drop, BSS = balanced salt solution,

Opsil = Opsil tears® eye drop

Table 4. Increase in pupil size after dilatation with each solution. Values are in mean (range) in millimetres.

Day after mixture	NSS	TN II	BSS	Opsil
0	5.4 (4.8 - 6)	6.2 (5.9 - 6.6)	5.4 (4.0 - 6.6)	5.9 (4.7 - 6.8)
1	5.3 (4.5 - 5.8)	5.2 (4.2 - 5.8)	5.6 (4.0 - 6.5)	5.5 (4.9 - 6.2)
3	5.4 (4.7 - 6.0)	5.1 (3.0 - 5.8)	5.8 (5.2 - 6.4)	5.9 (5.1 - 6.6)
5	5.6 (5.1 - 6.0)	5.3 (5.2 - 5.5)	5.3 (4.8 - 5.8)	4.9 (4.2 - 5.5)
7	5.6 (3.9 - 6.7)	5.4 (5.0 - 6.0)	6.1 (6.0 - 6.2)	5.2 (4.5 - 5.9)
10	5.5 (5.2 - 5.8)	5.2 (4.3 - 6.2)	5.6 (4.8 - 6.4)	6.1 (5.9 - 6.4)
14	5.4 (4.7 - 5.7)	5.5 (5.0 - 6.0)	5.9 (5.2 - 6.8)	5.7 (5.3 - 6.0)

NSS = Normal saline solution, TN II = Tear Naturale II® eye drop,

BSS = balanced salt solution, Opsil = Opsil tears® eye drop

Discussion

Although there were some reports of adverse drug reactions possibly associated with ocular use of 2.5 % phenylephrine hydrochloride, the 2.5 % phenylephrine hydrochloride is potentially safer than the 10 % concentration, and has been proved by many studies to be equally effective for pupillary dilatation.[7-11]

We have demonstrated that the manually diluted solutions with four different kinds of commercially available solutions are clinically effective. The pupil size in all solutions at different time after mixture was sufficient for a complete posterior eye segment examination. The average pupil size after instillation with 2.5 % phenylephrine hydrochloride in combination with 1 % tropicamide was 7.1-8.8 mm and the average increase in pupil size was 4.9-6.2 mm. Our results are compatible with the previous studies. In a study using 1 % cyclopentolate in combination with phenylephrine eye drops, the mean pupil size in the 2.5 % group and 10 % group were 8.0 mm and 8.2 mm, respectively.[8] Another study in diabetic population found that in combination with 1 % tropicamide, the mean (SD) increase in pupil size in the 2.5 % group and 10 % group were 3.56 (1.26) mm and 3.66 (1.30) mm, respectively.[9] A study in Chinese subjects using 1% tropicamide and 2.5 % phenylephrine found a mean (SD) pupil diameter after dilatation of 7.00 (1.06) mm and a mean increase of pupil size of 3.70 (0.97) mm.[12]

A question may be raised if this clinical effectiveness is from tropicamide eye drop alone or by the combinations of drugs. From our previous study, we found that with tropicamide alone the mean (SD) pupillary size at 30 minutes after the instillation is approximately 6.2 (0.76) mm,[14] comparing with another study where phenylephrine was used in combination with 1 % tropicamide, the mean (SD) pupil size was 7.35 (0.90) mm with 2.5 % phenylephrine and 7.58 (0.77) mm with 10 % phenylephrine eye drop.[10] These results support the synergistic effects of both drugs in pupillary dilatation.

The major limitation in our study is that we did not systematically evaluate the reported adverse drug effect of 2.5 % phenylephrine hydrochloride solution such as severe hypertension, syncope, myocardial infarction, tachycardia, arrhythmia, and fatal subarachnoid haemorrhage. However, we did not find any serious side effects during the entire course of the study.

In conclusion, all of the manually diluted solutions of 2.5 % phenylephrine hydrochloride have clinically sufficient potency for the pupillary dilatation. Their sterility and stability maintained for at least 2 weeks.

Acknowledgement

The study was financially supported by the Ratchadapiseksompotch Fund, Faculty of Medicine, Chulalongkorn University, Bangkok, Thailand.

Conflicts of interest

The authors declare no conflicts of interest in the study.

References

1. Fraunfelder FT, Scafidi AF. Possible adverse effects from topical ocular 10 % phenylephrine. Am J Ophthalmol 1978 Apr; 85(4): 447-53
2. McReynolds WU, Havener WH, Henderson JW. Hazards of the use of sympathomimetic drugs in ophthalmology. AMA Arch Ophthalmol 1956 Aug; 56(2): 176-9
3. Vaughan RW. Ventricular arrhythmias after topical vasoconstrictors. Anesth Analg 1973 Mar; 52(2): 161-5
4. Kim JM, Stevenson CE, Mathewson HS. Hypertensive reactions to phenylephrine eyedrops in patients with sympathetic denervation. Am J Ophthalmol 1978 Jun; 85(6): 862-8
5. Malhotra R, Banerjee G, Brampton W, Price NC. Comparison of the cardiovascular effects of 2.5 % phenylephrine and 10 % phenylephrine during ophthalmic surgery. Eye 1998; 12 (Pt 6): 973-5
6. Hakim OJ, Orton RB, Cadera W. Topical 2.5 % and 5 % phenylephrine: comparison of effects on heart rate and blood pressure. Can J Ophthalmol 1990 Dec; 25(7): 336-9
7. Neuhaus RW, Hepler RS. Mydriatic effect of phenylephrine 10 %(aq) vs phenylephrine 2.5 % (aq). Ann Ophthalmol 1980 Oct;12(10): 1159-60
8. Tanner V, Casswell AG. A comparative study of the efficacy of 2.5 % phenylephrine and 10 % phenylephrine in pre-operative mydriasis for routine cataract surgery. Eye 1996; 10(Pt 1): 95-8
9. Weiss JS, Weiss JN, Greenfield DS. The effects of phenylephrine 2.5 % versus phenylephrine 10 % on pupillary dilation in patients with diabetes. Retina 1995; 15 (2): 130-133
10. Kulvichit K. Clinical equivalence between a combination of 1 % tropicamide/2.5 % phenylephrine hydrochloride and 1 % tropicamide/10 % phenylephrine in pupillary dilatation efficacy in dark irides: a double blind randomized controlled trial [dissertation]. Bangkok: Chulalongkorn University; 2001.
11. Yospaiboon Y, Luanratanakorn P, Noppawinyoowong

C. Randomized Double-blind Study of Phenylephrine 2.5 % vs 10 % on Pupillary Dilation. J Med Assoc Thai 2004 Nov; 87(11): 1380-4

12. Lam PTH, Poon BTM, Wu WK, Chi SCC, Lam DSC. Randomized clinical trial of the efficacy and safety of tropicamide and phenylephrine in preoperative mydriasis for phacoemulsification. Clin Experiment Ophthalmol 2003 Feb; 31(1): 52-6

13. Fraunfelder FT, Meyer SM. Possible cardiovascular effects secondary to topical ophthalmic 2.5 % phenylephrine. Am J Ophthalmol 1985 Mar 15; 99(3): 362-3

14. Pukrushpan P, Tulvatana W, Kulvichit K. Intraocular pressure change following application of 1 % tropicamide for diagnostic mydriasis. Acta Ophthalmol Scand 2006 Apr; 84(2): 268-70