REFERENCES

[1.] Baker, S., and Price, K. 2006. Ultraviolet Radation [Online]. Available from: http://www.biospherical.com/hsf/student/page3.html [2006, August 21]

[2.] Canadian Centre for Occupational Health and Safety. 2006. What is ultraviolet radiation [Online]. Available from: http://www.ccohs.ca/oshanswers/phys_agents/ultravioletradiation.html [2006, August 21]

[3.] Herring, D. 2005. Health concerns and protection [Online] Available from: http:// en.wikipedia.org/wiki/Ultraviolet [2006, August 21]

[4.] LeVee, G.J.; Oberhelman, L.; Anderson, T.; Koren, H. and Cooper, K.D. UVA II exposure of human skin results in decreased immunization capacity, increased induction of tolerance and a unique pattern of epidermal antigen-presenting cell alteration. *Photochem Photobiol.* **65** (1997): 662-629.

[5.] Lowe, N.J. and Shaath, N.A.; Pathak, M. A. "Sunscreen development, evaluation and regulatory aspects" 2nd ed, New york:Mercel Dekker Inc, 1990, 216-233.

[6.] Dransfield, G.; Guest, P.J.; Lyth, P.L.; Mcgyrvey, D.J. and Truscott, T.G. Photoactivity test of TiO_2-based inorganic sunscreens part I: Non-aqueous dispersions. *J. Photochem.Photobiol.* **59** (2000): 147-151.

[7.] Hancock-chen, T. and Scaiano, J.C. Enzyme inactivation by TiO_2 photosensitization. *J. Photochem.Photobiol.* **57** (2000): 193-196.

[8.] Wamer, W.G.; Yin, J.J. and Wet, R.R. Oxidative damage to nucleic acids photosentized by titanium dioxide. *Free Rad. Biol. med.* **23** (1997): 851-858.

[9.] Umbach, W. "Cosmetic and Toiletries Development, Production and Use" New York: Ellis Horwood Limited, 1991, 96.

[10 U.S. Food and Drug Administration. "Sunscreen drug products for over-the-counter human use" Washington D.C.: FDA Press, 1999, 27666-27693.

[11.] Leweke, U.H. and Lippold, B.C. Absorption of sunscreens and other compounds through human skin In vivo deriviation of a method to predict maximum fluxe. *Pharmaceut. Res.* **12** (1995): 1354-1360.

[12.] Heyden, C.G.J.; Roberts, M.S. and Benson, H.A.E. Systemic Absorption of Sunscreen After Topical Application. *The Lancet.* **350** (1997): 863-864.

[13.] Gupta, V.K.; Zatz, J.L. and Rerek, M. Percutaneous absorption of sunscreens through micro-yucatan pig skin in vitro. *Pharm. Res.* **16** (1999): 1602-1607.

[14.] Potard, G.; Laugel, C.; Baillet, A.; Schaefer, H. and Marty, J.P. Quantitative HPLC Analysis of Sunscreens and Caffeine During In Vitro Percutaneous Penetration Studies. *Int .J. Pharm.* **189** (1999): 249-260.

[15.] Sarveiya, V.; Risk, S. and Benson, H.A.E. Liquid chromatographic assay for common sunscreen agents:application to in vivo assessment of sin penetration and systemic absorption in human volunteers. *J. of Chromatogr. B.* **803** (2004): 225-231.

[16.] Chatelain, E.; Gabard, B. and Surber, C. Skin penetration and sun protection factor of five UV filters: Effect of the vehicle. *Skin Pharmacol. And Appl. Skin Physiol.* **16** (2003): 28-35.

[17.] InstitutionRecherche, L'Oreal. Percutaneous absorption of Mexoryl sx in human volunteers:comparison with in vitro data. *Pharmacol. Appl. Skin Physiol.* **16** (2003): 343-355.

[18.] Janjua, N.R.; Mogensen, B.; Andersson, A.M.; Petersen, J.H.; Henriksen, M.; Skakkebaek, N.E. and Wulf, H.C. Systemic absorption of the sunscreens benzophenone-3, octyl-methoxycinnamate and 3-(4-methyl-benzylidene) camphor after whole-body topical application and reproductive hormone levels in humans. *J. Invest. Dermatol.* **123** (2004): 57-61.

[19.] Kertesz, Z.S.; Szikszai , Z.; Gontier, E.; Moretto, P.; Surlève-Bazeille, J.E.; Kiss, B.; Juhász, I.; Hunyadi, J.; and Kiss, Á.Z. Nuclear microprobe study of TiO_2-penetration in the epidermis of human skin xenografts. *Nucl. Instr. And Meth. In Phys. Res. B.* **231** (2005): 280-285.

[20.] Menzel, F.; Reinert, T.; Vogt, J. and Butz, T. Investigations of percutaneous uptake of ultrafine TiO_2 particles at the high energy ion nanoprobe LIPSION. *Nucl. Instr. And Meth. In Phys. Res. B.* **82** (2004): 219-220.

[21.] Godwin, D.A.; Kim Nae-Hwa and Linda, A.F. Influence of Transcutol® CG on the skin accumulation and transdermal permeation of ultraviolet absorbers. *Eur.J. Pharm. Biopharm.* **53** (2002): 23-27.

[22.] Yener, G.; Incegul, T. and Yener, N. Importance of Using Solid Lipid Microspheres as Carriers for UV filters on The Example Octylmethoxycinnamate. *Int.J. Pharmaceutics.* **258** (2003): 203-207.

[23.] Jimenez, M.M.; Pelletier, J.; Bobin, M.F. and Martini, M.C. Influence of encapsulation on the in vitro percutaneous absorption of octyl methoxycinnamate. *IntJ. Pharmaceutics.* **272** (2004): 45-55.

[24.] Simeoni, S.; Scalia, S. and Benson, H.A.E. Influence of cyclodextrins on in vitro human skin absorption of the sunscreen, butyl-methoxydibenzoylmethane. *IntJ. Pharmaceutics.* **280** (2004): 163-171.

[25.] The saftyline institute. 2006. layer and structures of the skin [Online] . Available from: www.safetyline.wa.gov.au/.../images/l129_06.jpg. [2006, August 22]

[26.] Williams, A.C. and Barry B.W. "Skin absorption enhencer Critical Reviews in the Therapeutic Drug Carrier Systems." **9** (1992): 305-353.

[27.] Barry, B.W. "Structure, function, diseases, and topical treatment of human skin. In:Dermatological Formulations, Percutaneous absorption" New york: Mercel Dekker Inc, 1983, 1-48.

[28.] Barry, B.W. Mode of action of penetration enhancers in human skin. *J.Control.Rel.* **6** (1987): 85-97.

[29.] Wiechers J.W. The barrier function of the skin in relation to percutaneous absorption of drugs. *Pharm. Weekbl. Sci. Ed.* **6** (1989): 185-198.

[30.] Guy, R.H. and Hadgraft, J. Transdermal drug delivery. *Pharmacy International.* **43** (1986): 112-116.

[31.] Barry, B.W. "Penetration enhancers, mode of action in human skin, pharmacology and the skin" New york: Mercel Dekker Inc, 1987, 121-137.

[32.] Smith, K.L. "Penetrant characteristics influencing skin absorption, methods for skin absorption" Florida: CRC Press, 1990, 23-34.

[33.] Frantz S.W. "Instrumentation and methodology for in vitro skin diffusion cells in methodology for skin absorption, methods for skin absorption, Florida: CRC Press, 1990, 35-39.

[34.] Tojo, K. "Design and calibration of in vitro permeation apparatus, transdermal controlled systemic medications" New York: Marcel Dekker Inc, 1987, 127-158.

[35.] Barry B.W. "Methods for studying percutaneues absorption, percutaneous absorption" New york: Mercel Dekker Inc, 1983, 234-295.

[36.] Friend D.R. In vitro permeation techniques. *J.Control. Rel.* **18** (1992): 235-248.

[37.] Skelly J.P.; Shah V.P.; Maibach H.I.; Guy R.H.; Wester, W.C.; Flynn, G. and Yacobi A. FDA and AAPS report of the worshop on principles and practices of in vitro percutaneous penetration studies:Relevance to bioavailability and bioequivalence. *Pharm. Res.* **3** (1987): 265-267.

[38.] Behl, C.R.; Kumar. S.; Malick, A.W.; Patel, S.B.; Char, H. and Pimontese. "Choice of membranes for in vitro skin uptake studies and general experimental techniques, methods for skin absorption" Florida: CRC Press, 1990, 1-21.

[39.] Moore, L. and Chien Y.W. Transdermal drug delivery:A review of its pharmacokinetics, and pharmacodynamics. *Therapeutic Drug Carrier Systems.* **4** (1988): 285-349.

[40.] Smisterova, J.; Ensing, K. and de Zeeuw R.A. Methodological aspect of quantitative receptor assay. *J.Pharm. Biomed. Anal.* **6** (1994): 723-745.

[41.] Kiistala, U. Suction blister device for separating viable epidermis from dermis in human skin. *J.Invest.Dermatol.* **50** (1968): 129-137.

[42.] Ihlberg, L.; Haukipuro, K.; Risteli, L.; Oikarinen, A.; Kairaluoma M.I. and Risteli J. Collagen synthesis in intact skin is suppressed during wound healing. *Ann. Surg.* **217** (1993): 397-405.

[43.] Vermeer, B.J.; Reman, F.C. and van Gent C.M. The determination of lipids and proteins in suction blister fluid. *J. Invest Dermatol.* **73** (1979): 303-305.

[44.] Marja, J.; Peter M.E. and Frederklk A. Determination of 8-methoxyphsoraler in suction blister fluid and serum by liquid chromatography. *Clin.Chem.* **26** (1980): 1825-1828.

[45.] Surber, C.; Wilhelm, KP.; Bermann, D. and Nagel, R. *In vitro* skin penetration of acitretin in volunteers using three sampling techniques. *Pharm Res.* **10** (1993): 1291-1294.

[46.] Ronald, G.; Janice, K.; Phillip, T.; Robert C.M.; Bryon, F.L. and Willium B.M. Sress-related changes in proinflammatory cytokine production in wounds. *Arch. Gen. Psychiatry.* **56** (1999): 450-456.

[47.] Tapani, L.; Riitta, K. and Pekka, A. Suction blisters for the investigation of radiation reactions in human skin. *Acta Oncologica.* **41** (2002): 313-314.

[48.] Nanda, S.; Relhan, V.; Grover, C.M. and Reddyl, B.S.N. Suction blister epidermal grafting for management of eyelid vitiligo:special conciderations. *Dermatologic surgery*. **32** (2006): 387-392.

[49.] Brain, K.R.; Walters, K.A.; Green, D.M.; Brain, S.; Loretz, L.J.; Sharma, R.K. and Dressler, W.E. Percutaneous penetration of diethanolamine through human skin *in vitro*: Application from cosmetic vehicles. *Food and Chemical Toxicology* **43** (2005):681-690.

[50.] EnchantedLearning.com. 2006. Skin Anatomy [Online]. Available from: http://www.enchantedlearning.com/subjects/anatomy/skin/.[2006, August 30]

APPENDICES

APPENDIX A

Calculation of penetration of each sunscreens.

Percent penetration of each sunscreen as compared to OMC. Percent penetration was calculated as follow:

$$\% \text{ penetration} = \frac{\text{amount of sunscreens determined in receptor fluid}}{\text{amount of applied sunscreens}} \times 100$$

Determination of sunscreens in receptor fluid was done through the calibration curves using UV/VIS spectrophotometry. Percent penetration of each sunscreens by Franz diffusion cell are shown in Figure A.1-A.14.UV-Vis spectra of the withdrawn receptor fluids are shown in Figure A.15 –A .18.

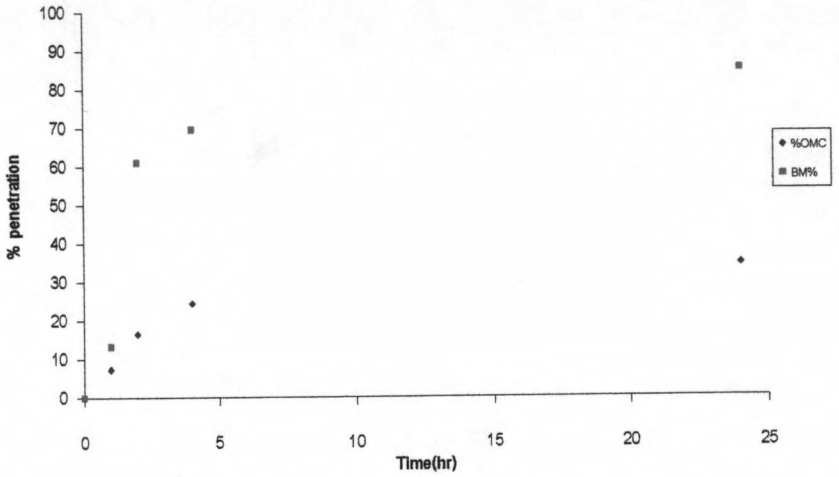

Figure A.1 % penetration of butyl methoxy dibenzoylmethane (■) and OMC (◆) by Franz-diffusion cell.

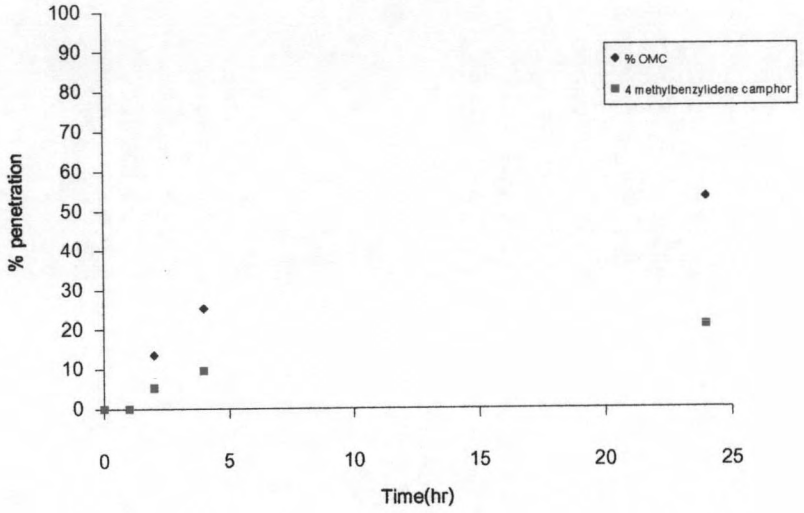

Figure A.2 % penetration of 4-methylbenzylidene camphor (■) And OMC (◆) by Franz-diffusion cell.

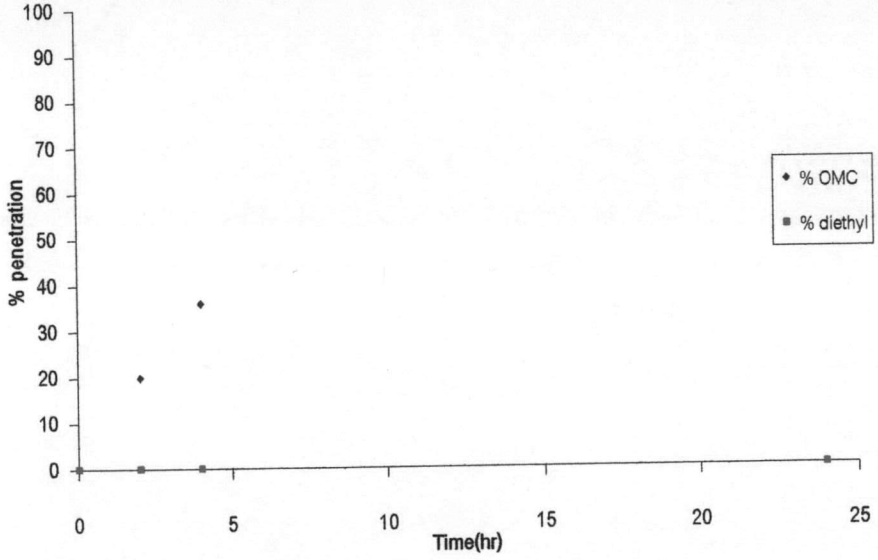

Figure A.3 % penetration of diethyl-2,4,5-trimethoxybenzalmalonate (■) and OMC (◆) by Franz-diffusion cell.

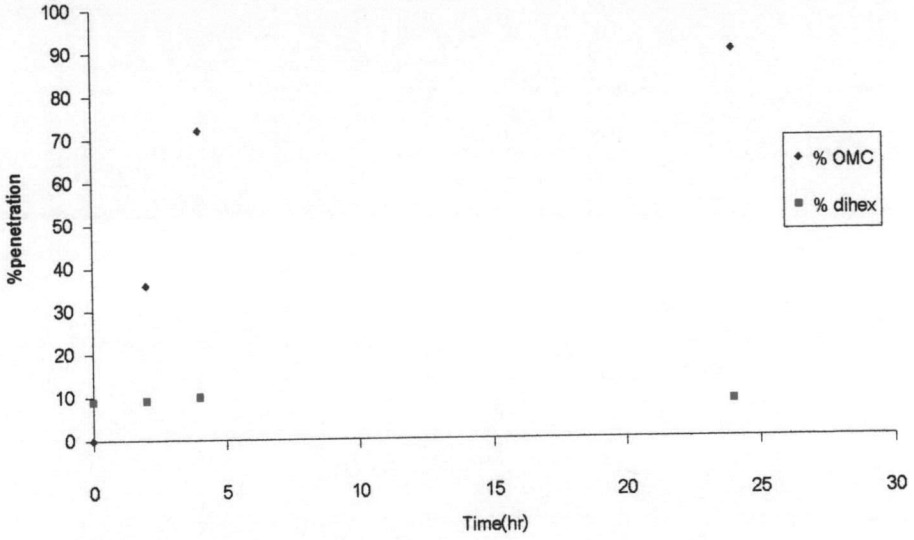

Figure A.4 % penetration of dihexyl-2,4,5-trimethoxybenzalmalonate (■) and OMC (◆) by Franz-diffusion cell.

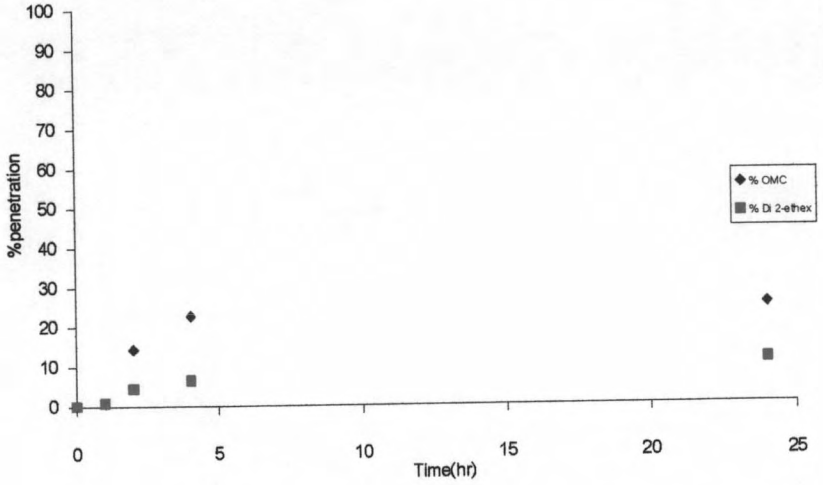

Figure A.5 % penetration of di(2-ethylhexyl)-2,4,5-trimethoxybenzalmalonate (■) OMC (◆) by Franz-diffusion cell.

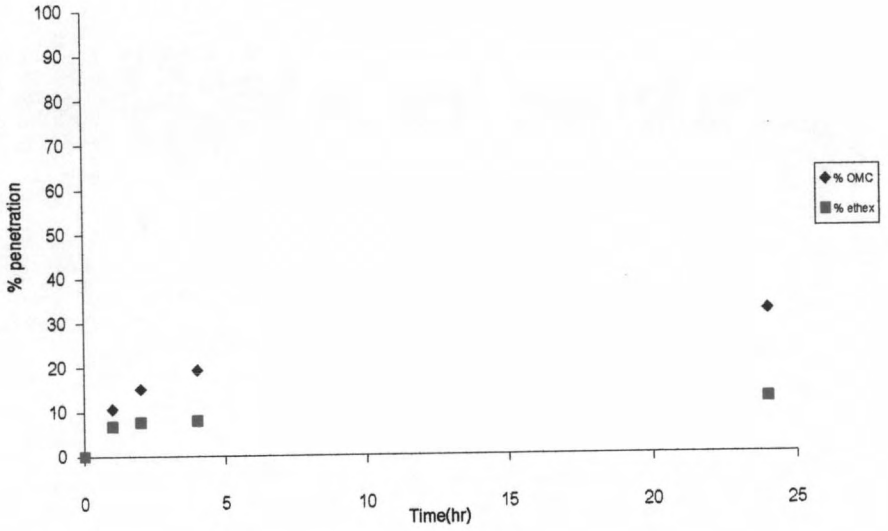

Figure A.6 % penetration of ethylhexyl-2,4,5-trimethoxycinnamate (■) and OMC (◆) by Franz-diffusion cell.

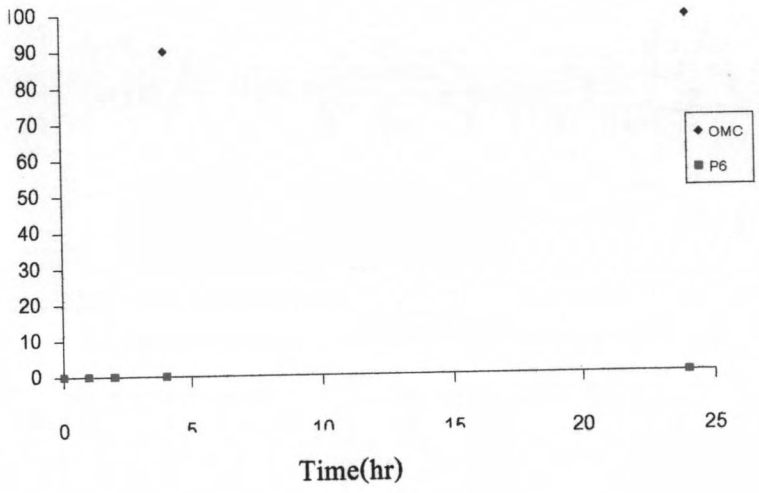

Figure A.7 % penetration of poly-(6-hydroxy-hexyloxy) cinnamic acid (■) and OMC (◆) by Franz-diffusion cell.

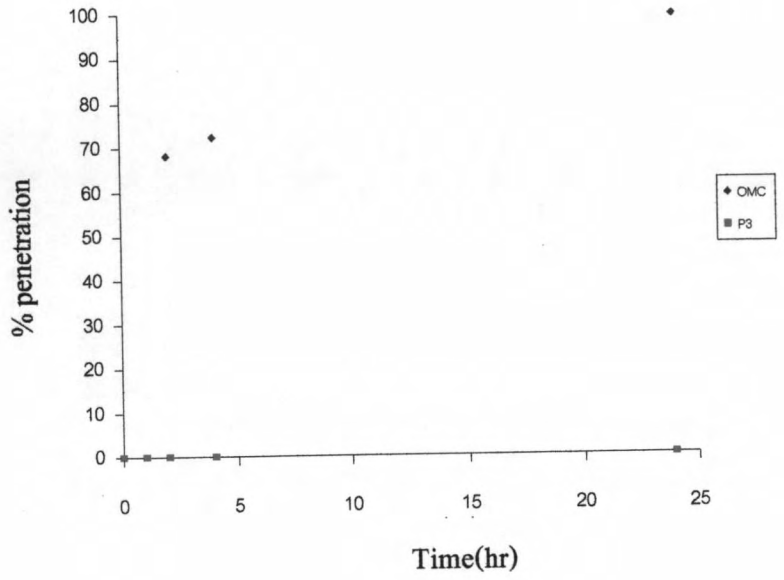

Figure A.8 % penetration of poly-(3-hydroxy-propoxy)cinnamic acid (■) and OMC (◆) by Franz-diffusion cell.

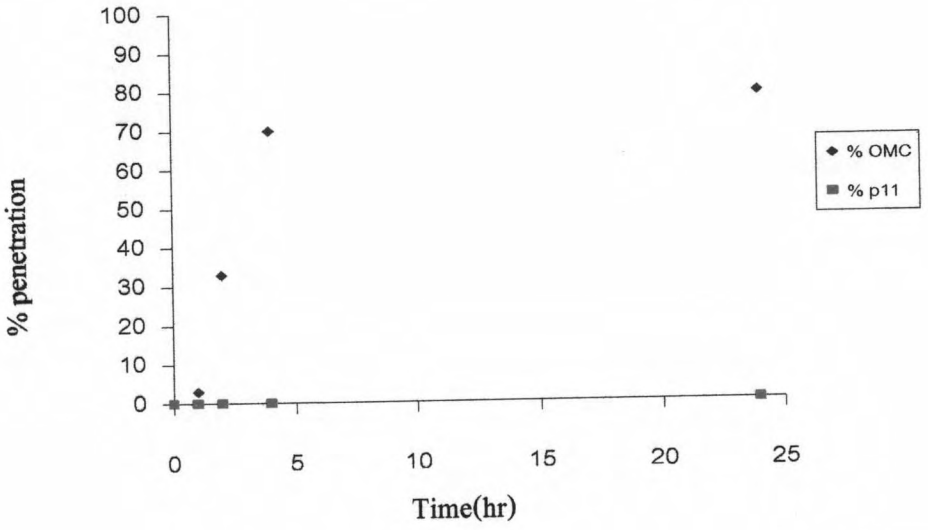

Figure A.9 % penetration of poly-(11-hydroxy-undecyloxy)cinnamic acid (■) and OMC (◆) by Franz-diffusion cell.

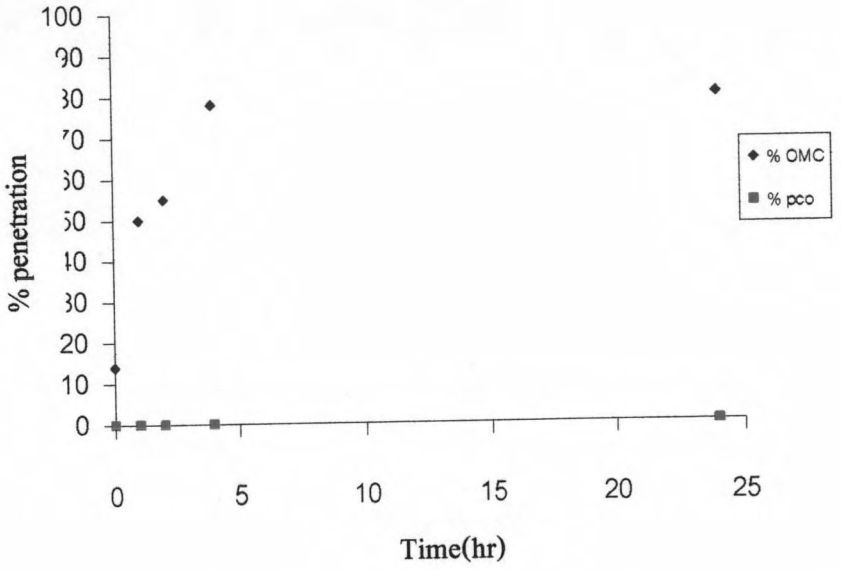

Figure A.10 % penetration of poly(p-propoxycinnamate)-co-(p-undecyloxy cinnamate) (■) and OMC (◆) by Franz-diffusion cell.

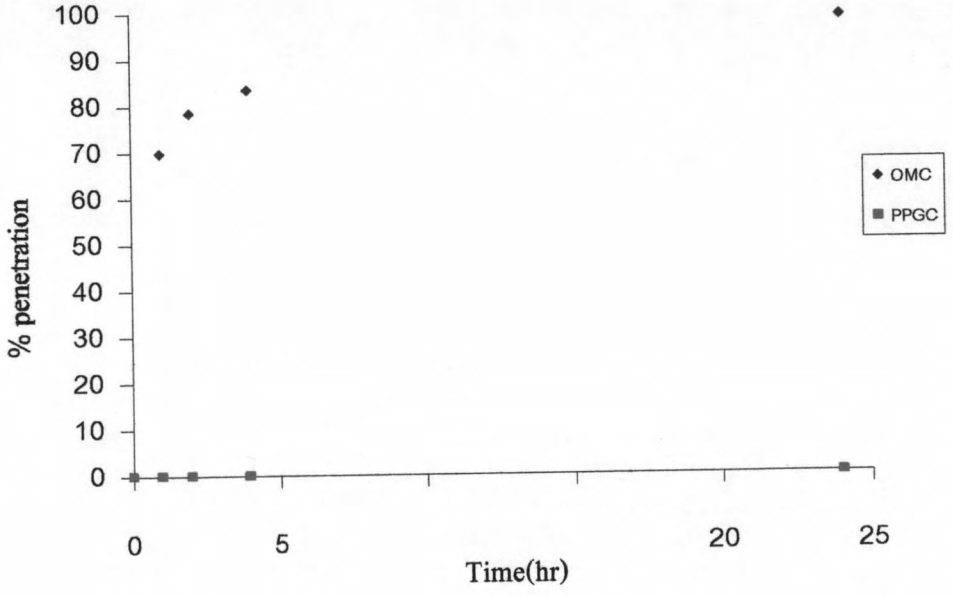

Figure A.11 % penetration of poly penta ethylene glycol cinnamate (■) and OMC (◆) by Franz-diffusion cell.

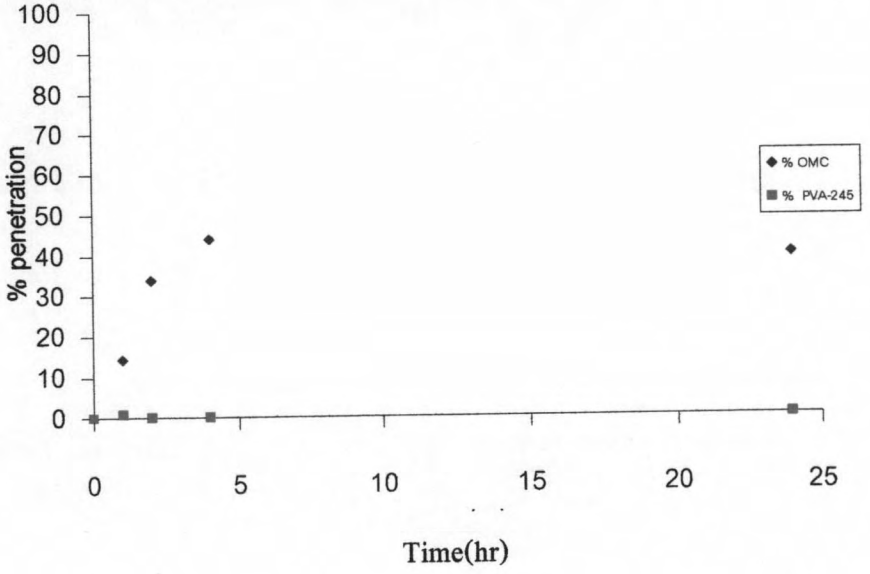

Figure A.12 % penetration of poly[vinyl 2,4,5 trimethoxycinnamate(vinyl alcohol)]copolymer (■) and OMC (◆) by Franz-diffusion cell.

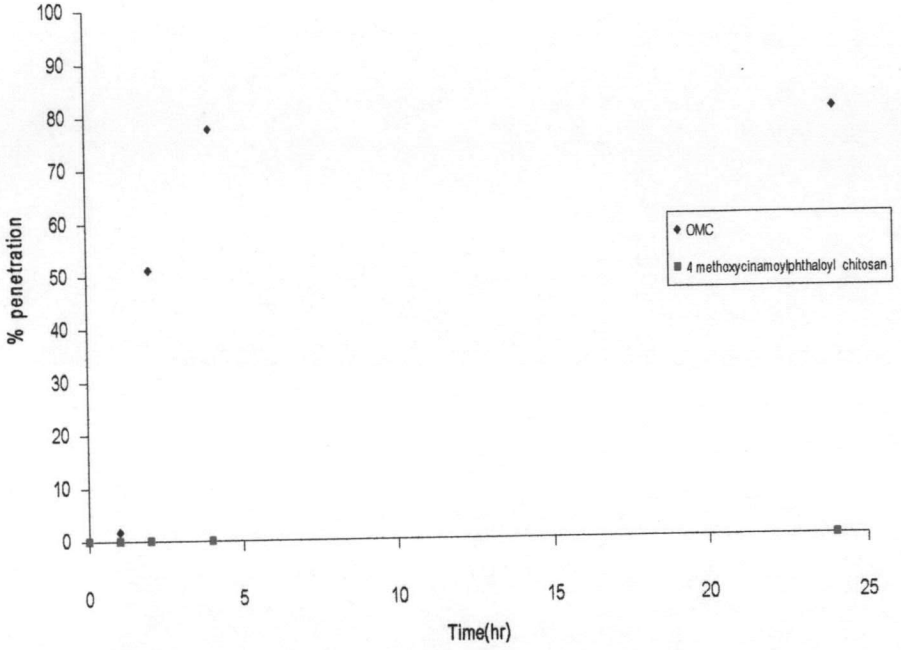

Figure A.13 % penetration of 4-methoxycinamoylphthaloylchitosan (■) and OMC (◆) by Franz-diffusion cell.

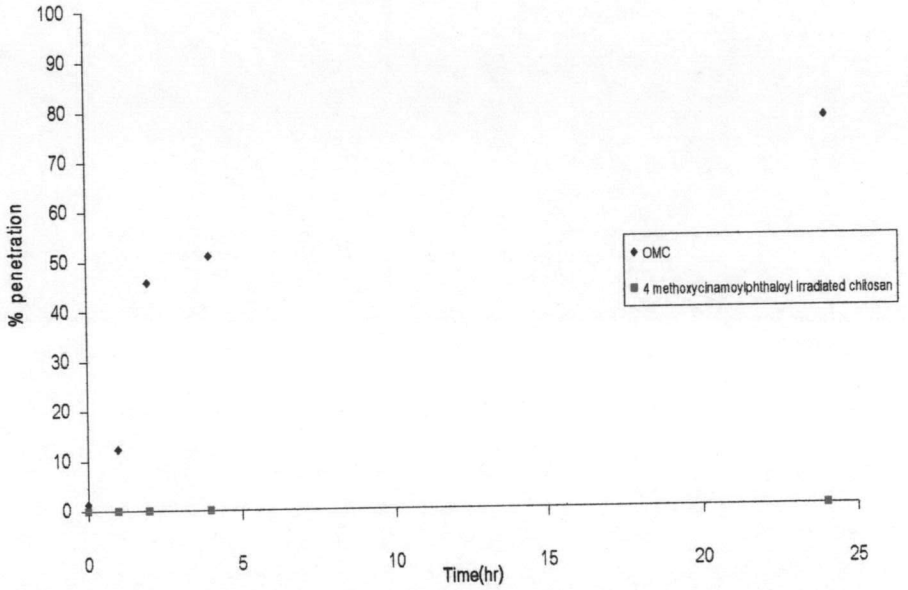

Figure A.14 % penetration of 4-methoxycinamoylphthaloylirradiated chitosan (■) and OMC (◆) by Franz-diffusion cell.

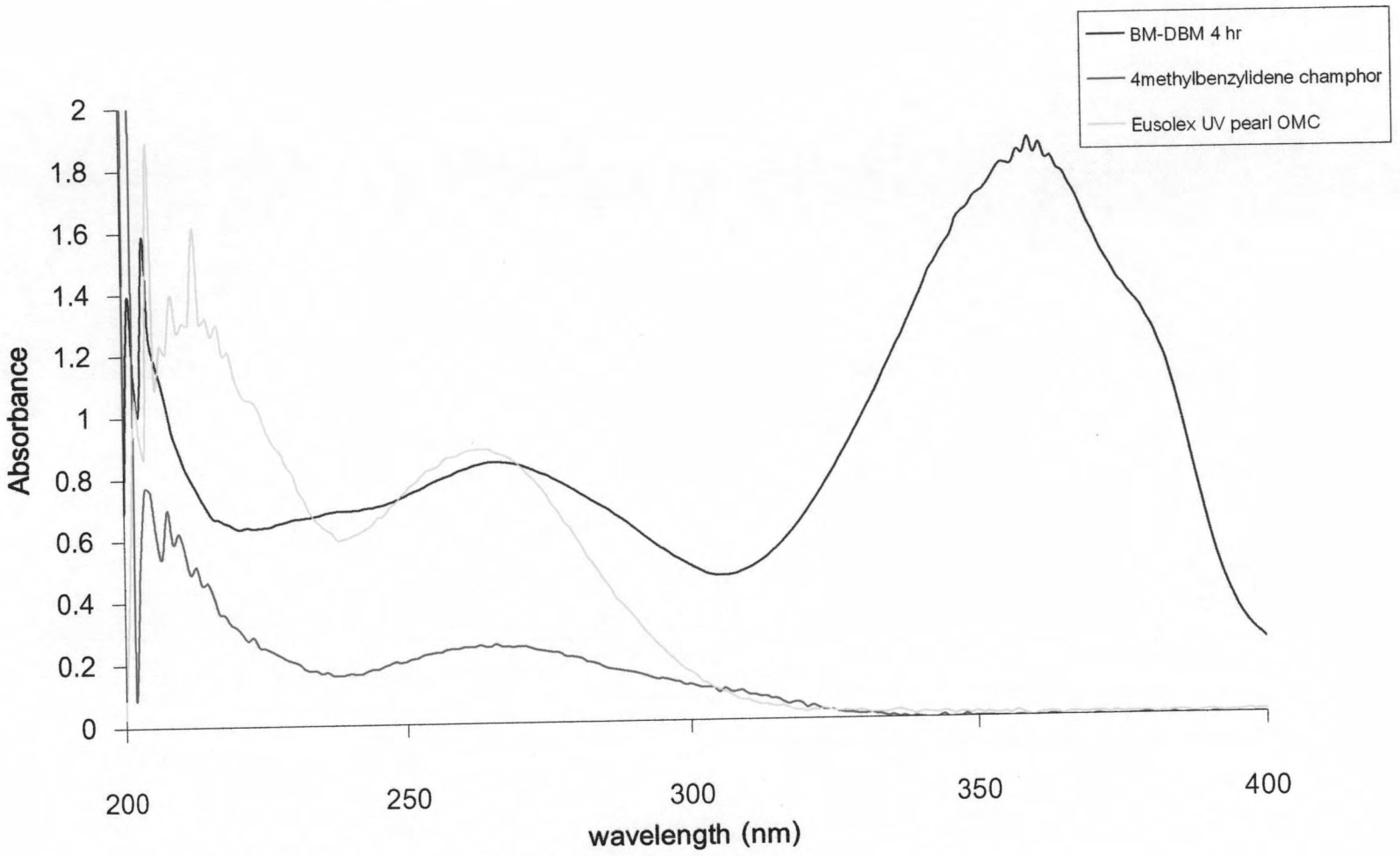

Figure A.15 UV absorbance spectra of commercial organic UV filters after 24 h in diffusion cell.

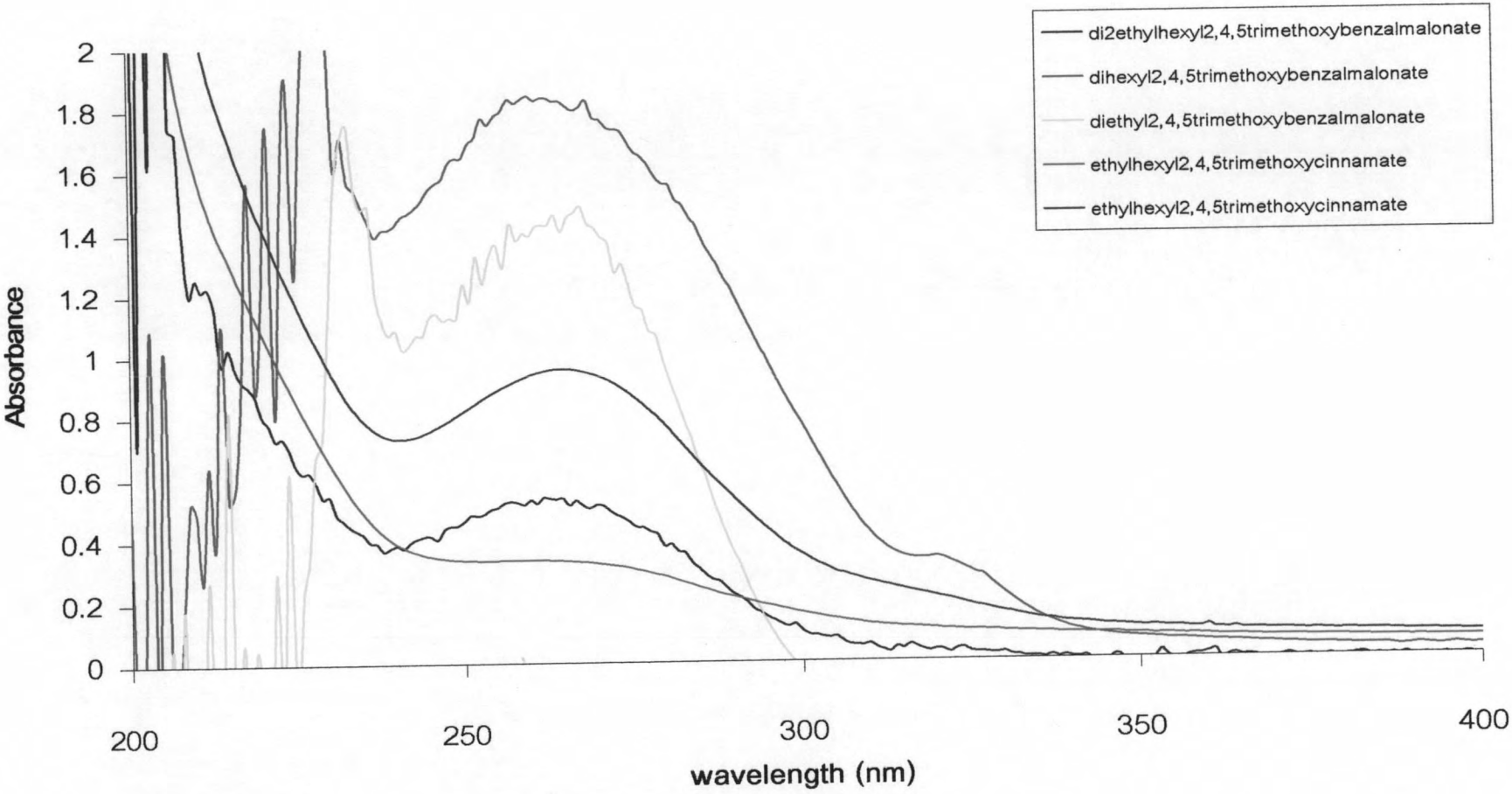

Figure A.16 UV absorbance spectra of newly developed small organic UV filters after 24 h in diffusion cell.

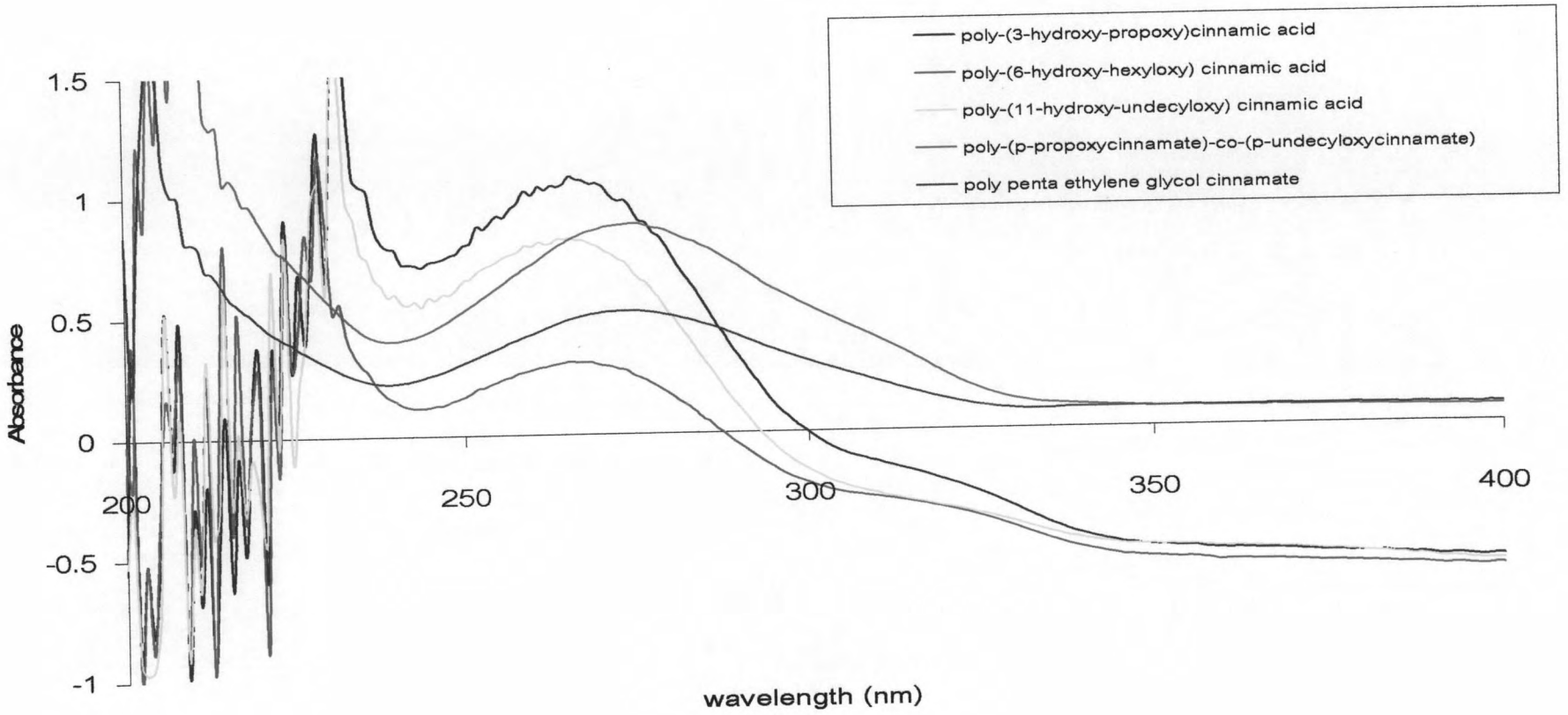

Figure A.17 UV absorbance spectra of polymeric UV filters after 24 h in diffusion cell.

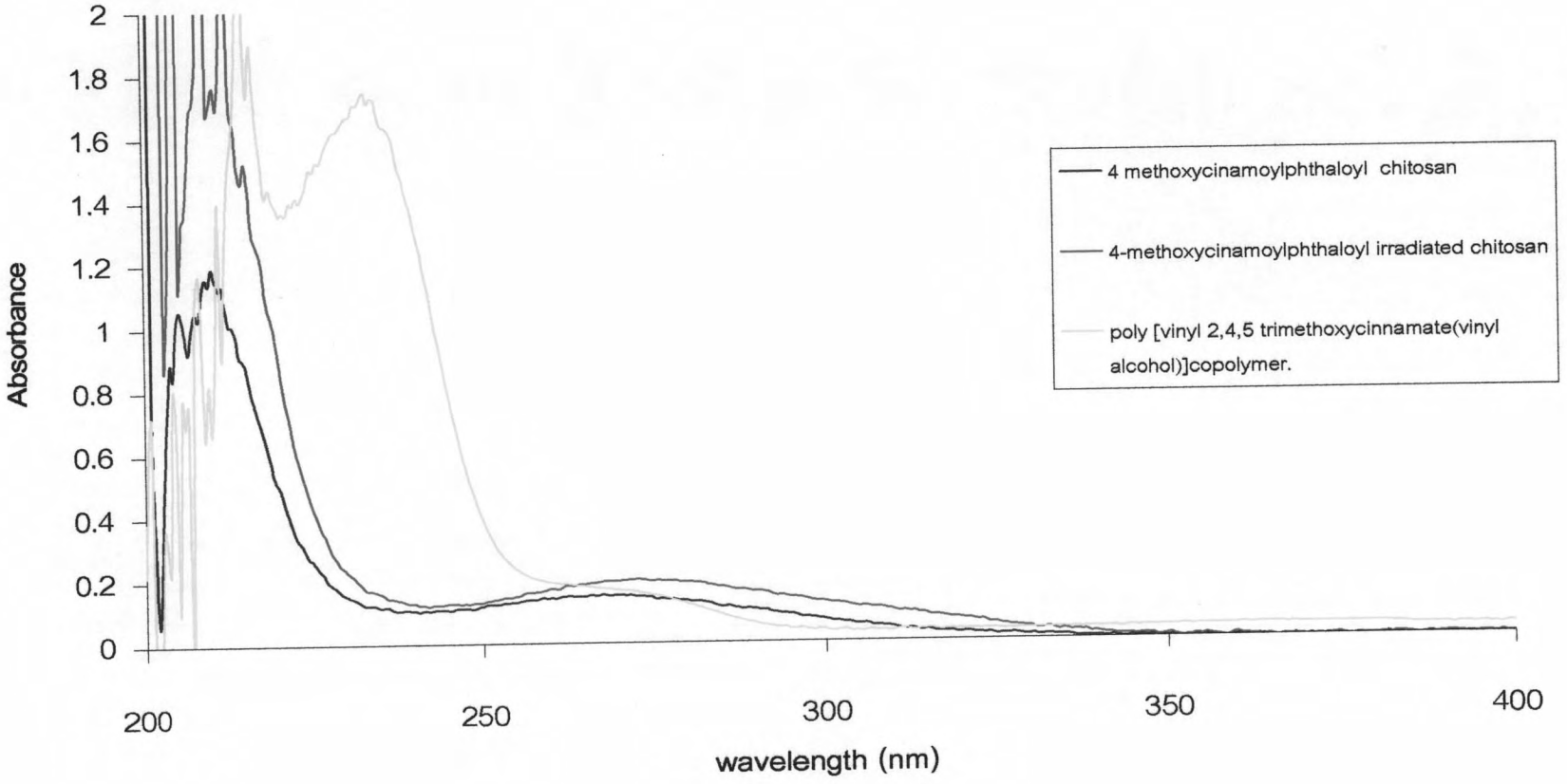

Figure A.18 UV absorbance spectra of polymeric UV filters after 24 h in diffusion cell.

APPENDIX B

UV –Vis absorbance spectra of SBF withdrawn from the volunteers are shown in Figure B.1-B.5.

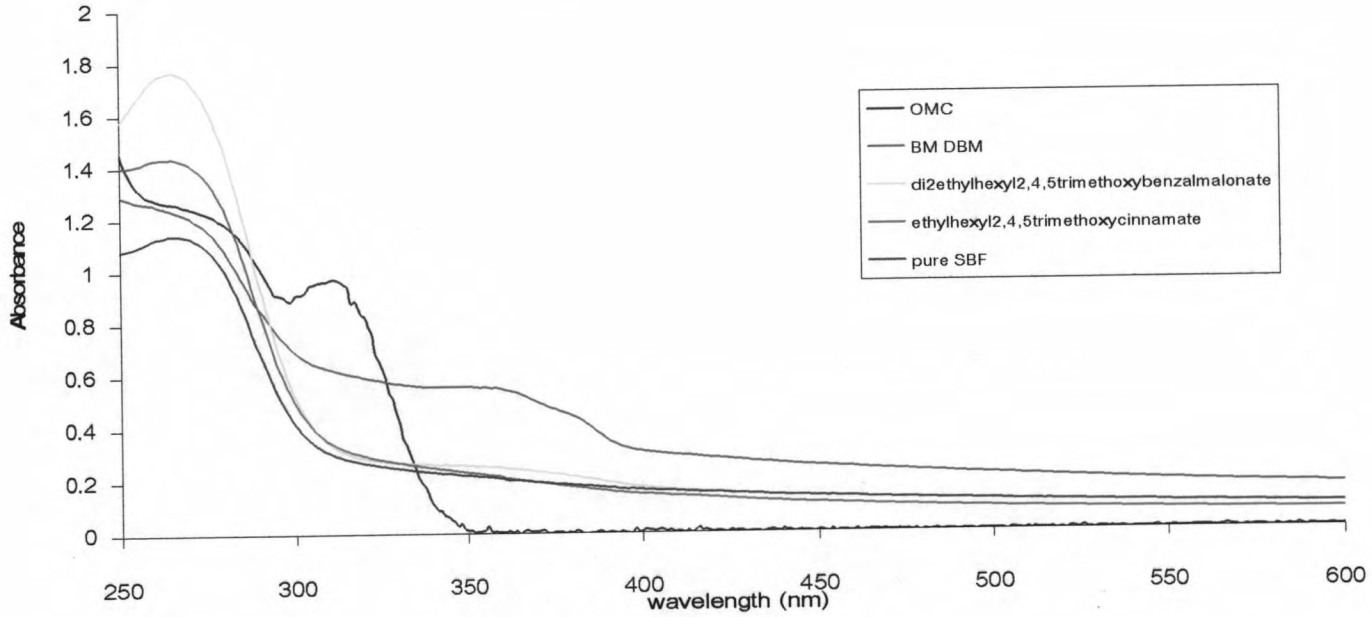

Figure B.1 UV-Vis absorbance spectra of SBF withdrawn after its roof skin had been applied with sunscreen spiked lotion for 3 hr; first volunteer.

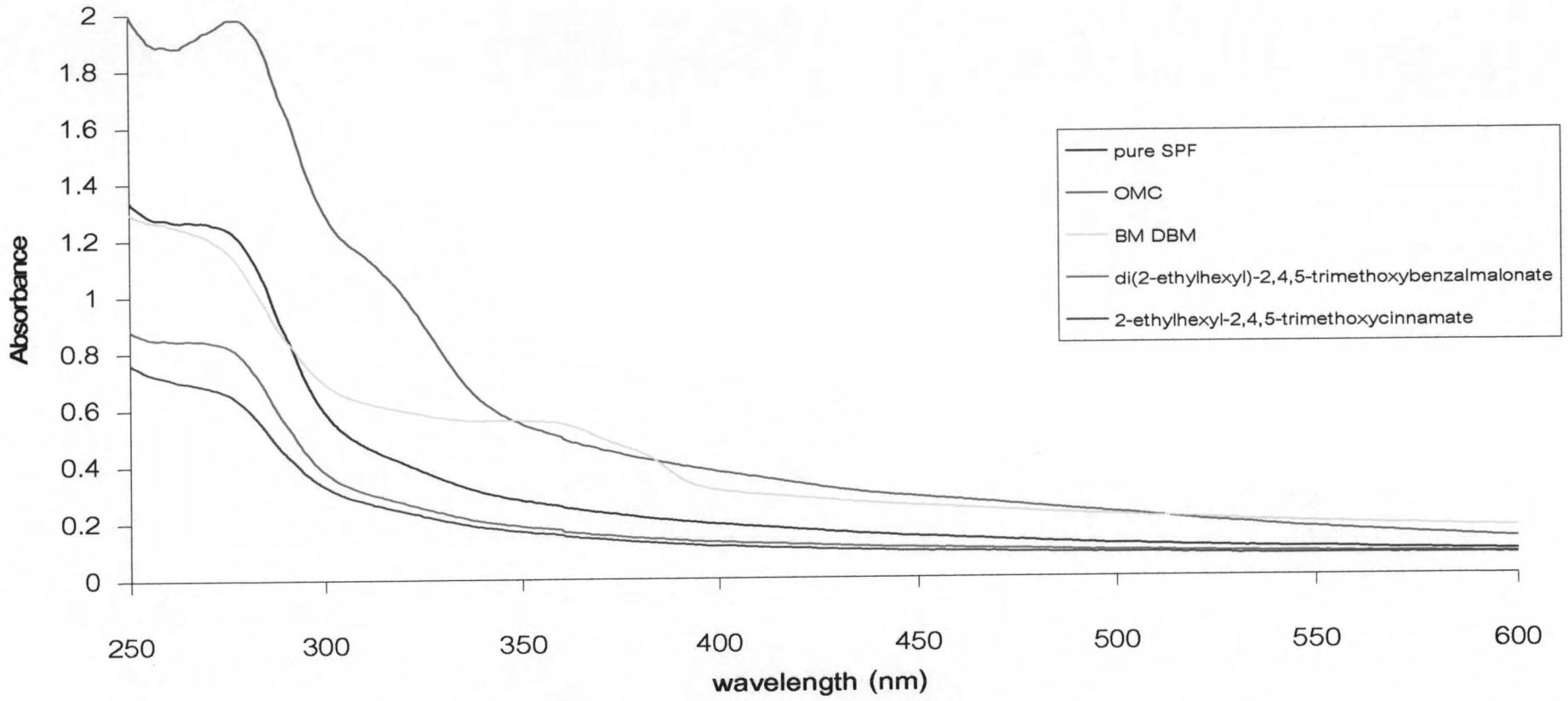

Figure B.2 UV-Vis absorbance spectra of SBF withdrawn after its roof skin had been applied with sunscreen spiked lotion for 3 hr; second volunteer.

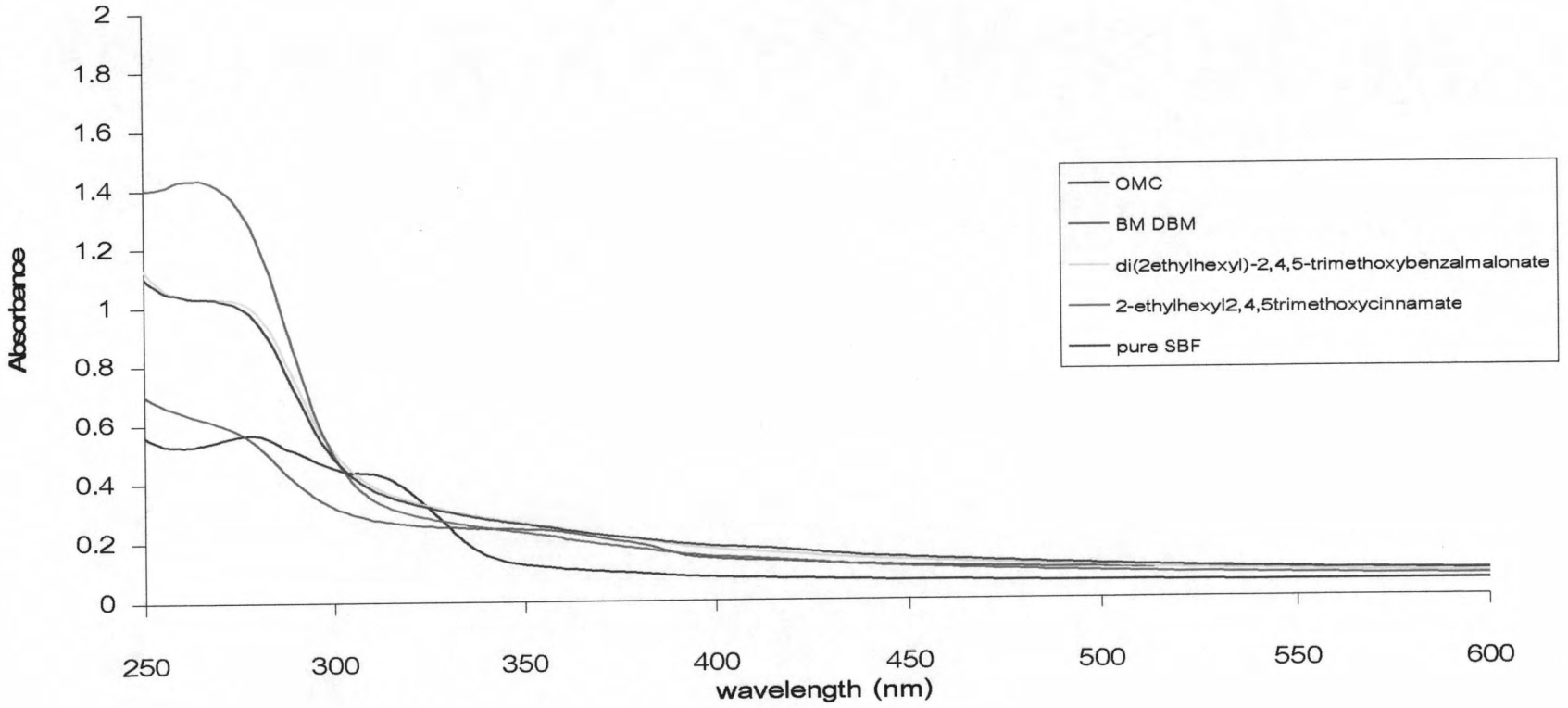

Figure B.3 UV-Vis absorbance spectra of SBF withdrawn after its roof skin had been applied with sunscreen spiked lotion for 3 hr; third volunteer.

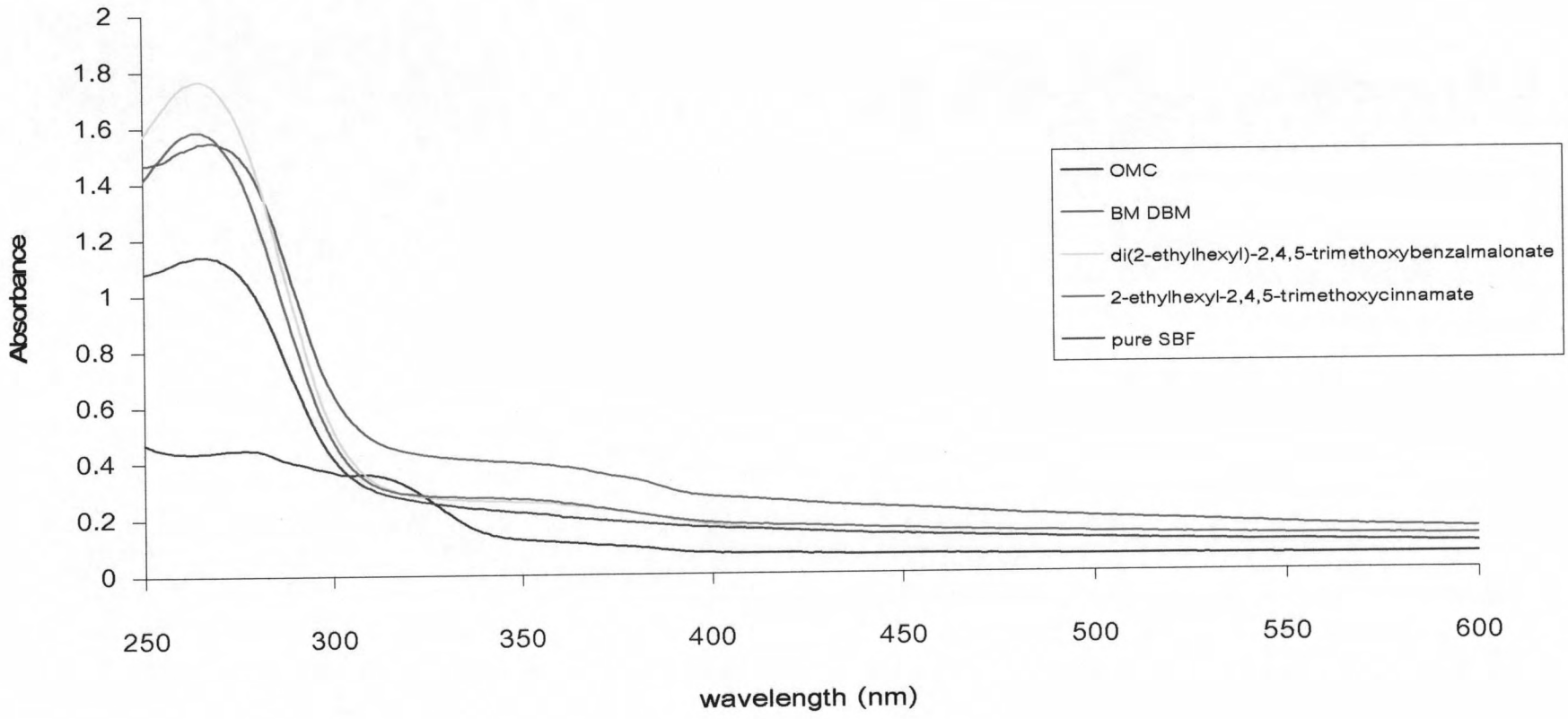

Figure B.4 UV-Vis absorbance spectra of SBF withdrawn after its roof skin had been applied with sunscreen spiked lotion for 3 hr; forth volunteer.

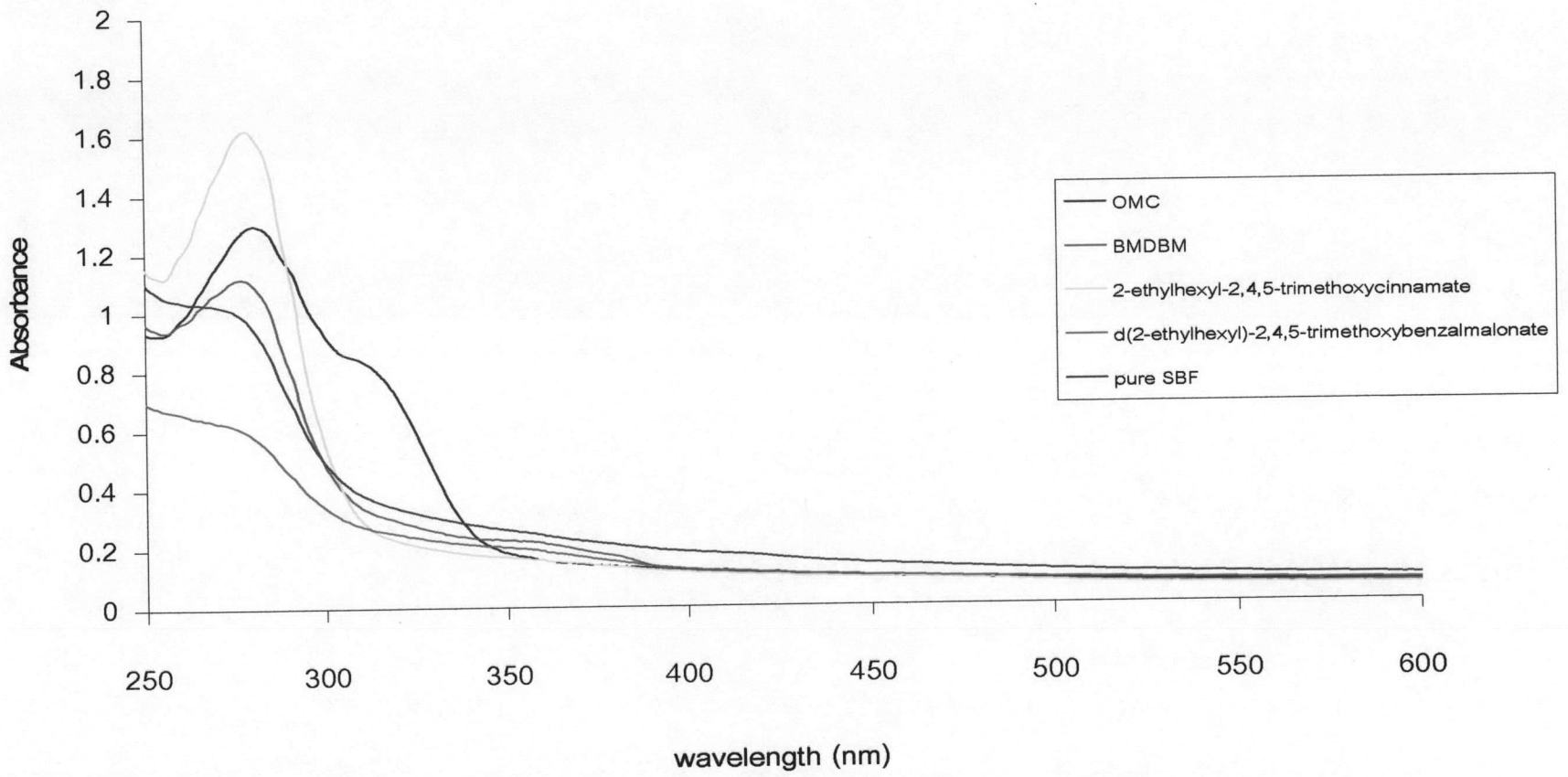

Figure B.5 UV-Vis absorbance spectra of SBF withdrawn after its roof skin had been applied with sunscreen spiked lotion for 3 hr; fifth volunteer.

APPENDIX C

Penetration of sunscreens through human epidermis. Figure C.1 is shown percent penetration of OMC at various times. Percent penetration was obtained from SBF withdrawn at time after the suction blister roof was applied with sunscreen. Percent penetration was calculated as follow : % penetration $= \frac{\text{amount of UV filters obtained in SBF}}{\text{amount of applied UV filters}} \times 100$

Amount of UV filter obtained in SBF was obtained from the calibration curves constructed using SBF from five volunteers. Figure C.2-C.6 show % penetration of each UV filters found in SBF (after 3 hr) of each volunteer.

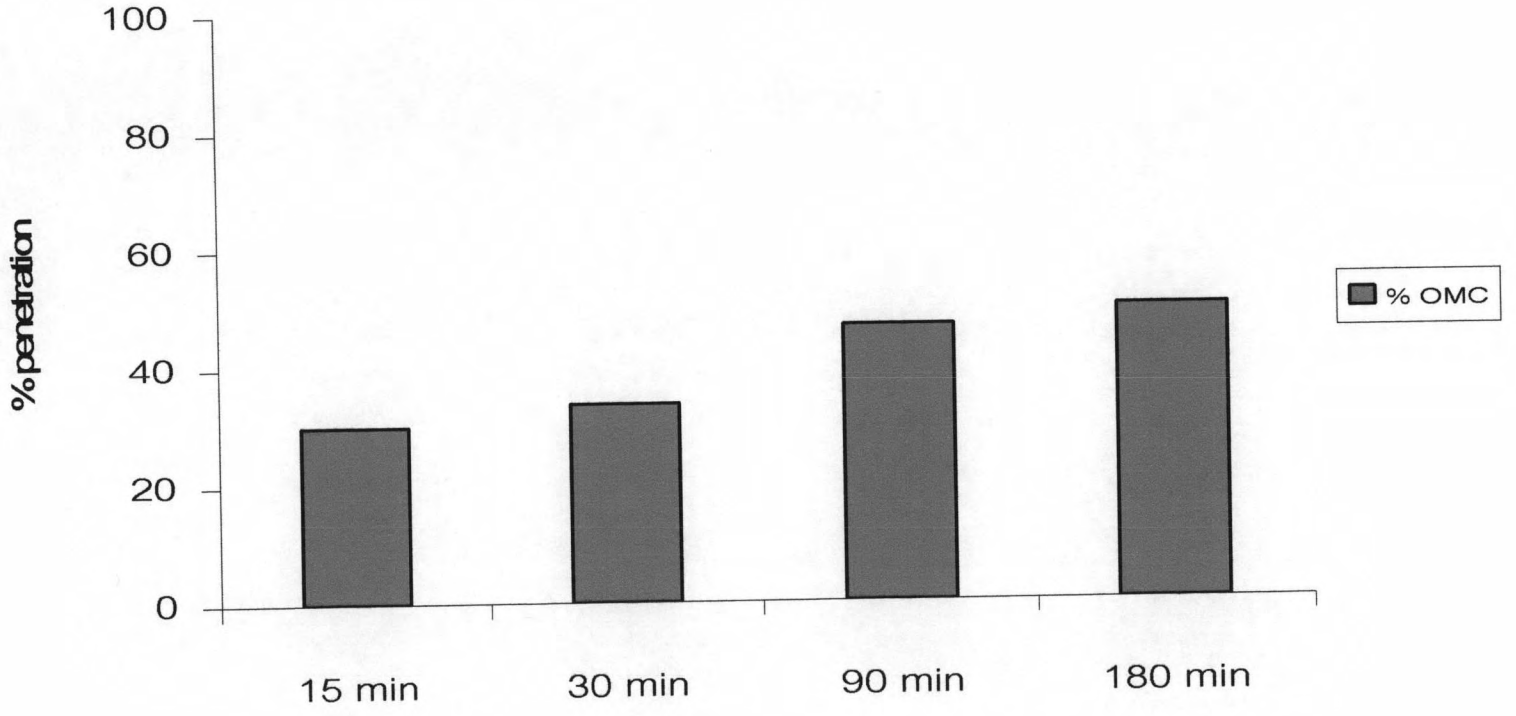

Figure C.1 % penetration of OMC at various times during the suction blister experiment.

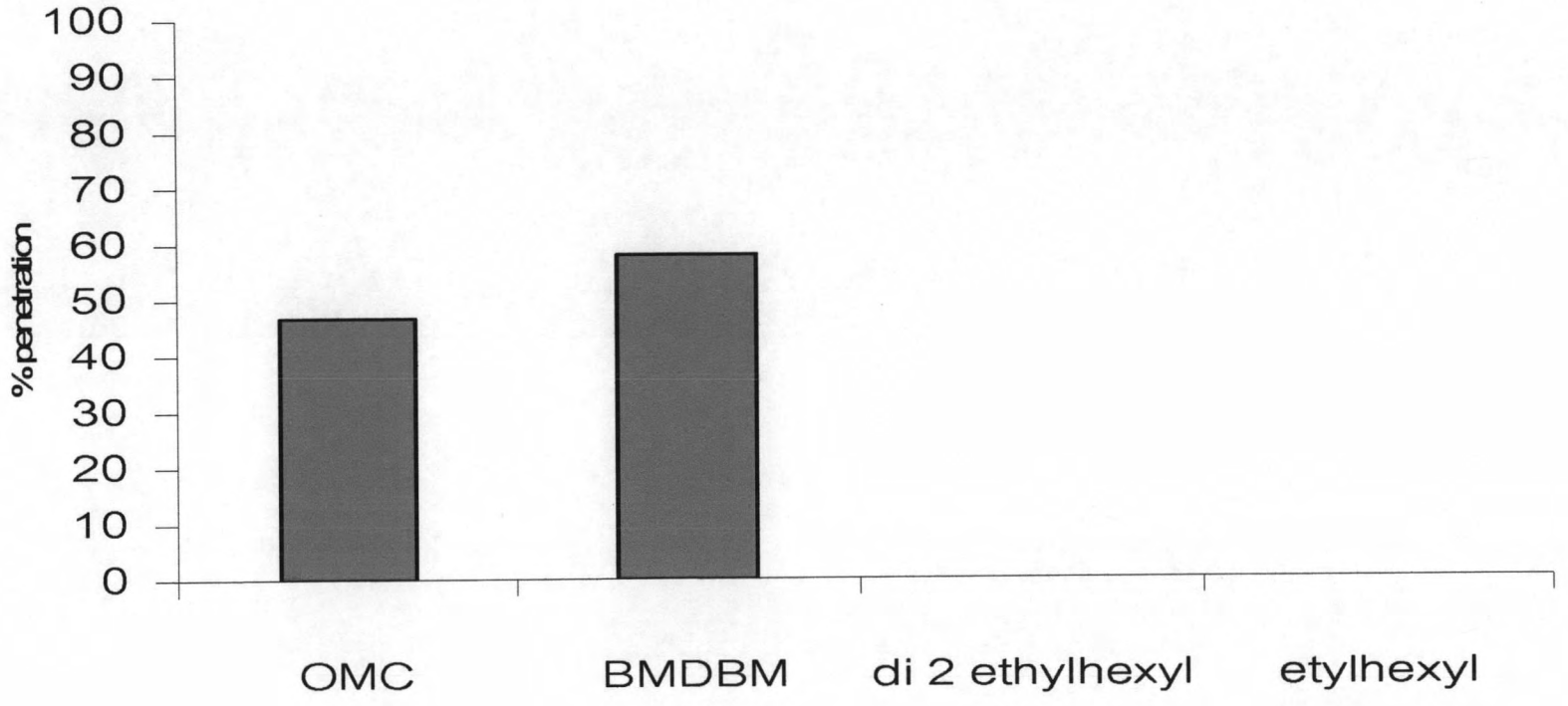

Figure C.2 % penetration of OMC, BMDBM, di(2-ethylhexyl)-2,4,5-trimethoxybenzalmalonate and 2-ethylhexyl-2,4,5 trimethoxycinnamate at 3 h in first human volunteer

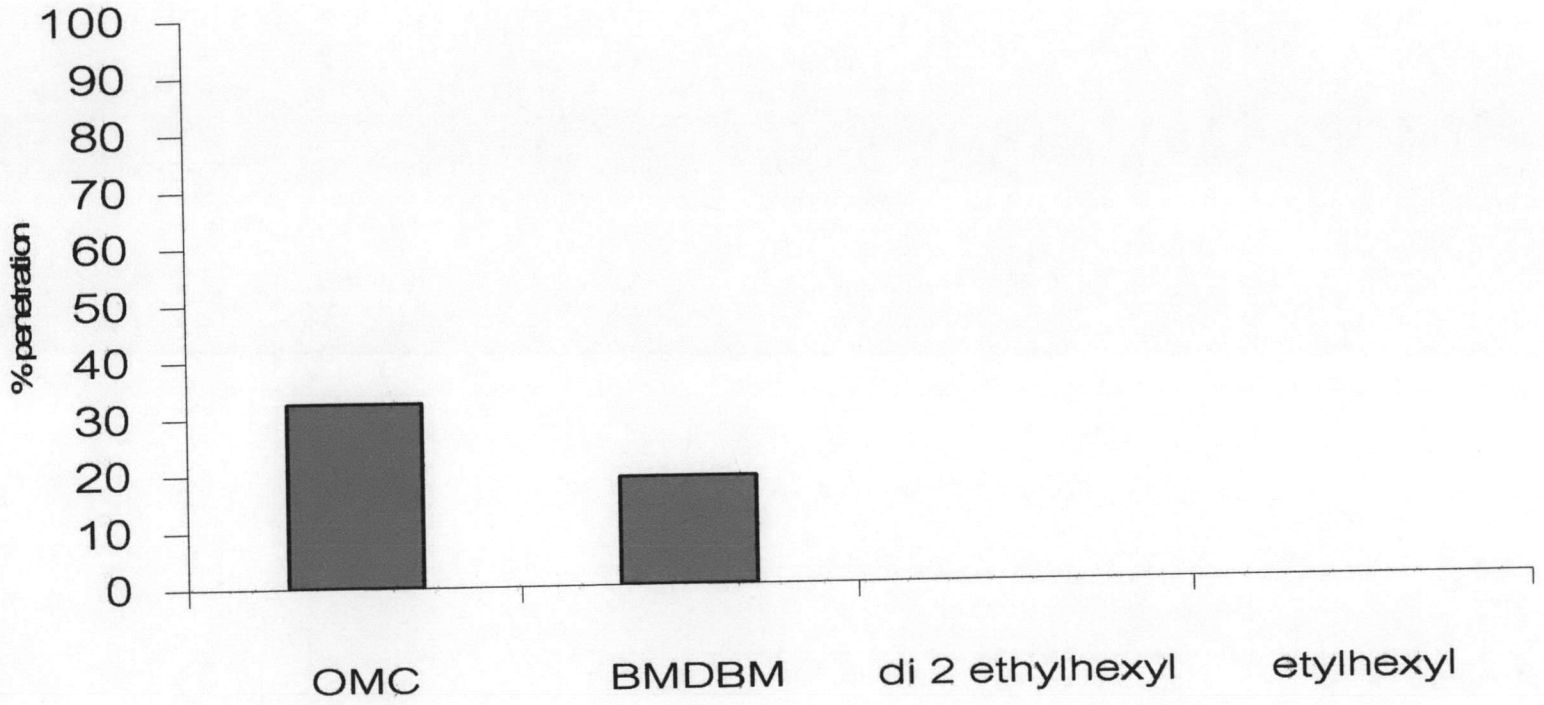

Figure C.3 % penetration of OMC, BMDBM, di(2-ethylhexyl)-2,4,5-trimethoxybenzalmalonate and 2-ethylhexyl-2,4,5 trimethoxycinnamate at 3 h in second human volunteer

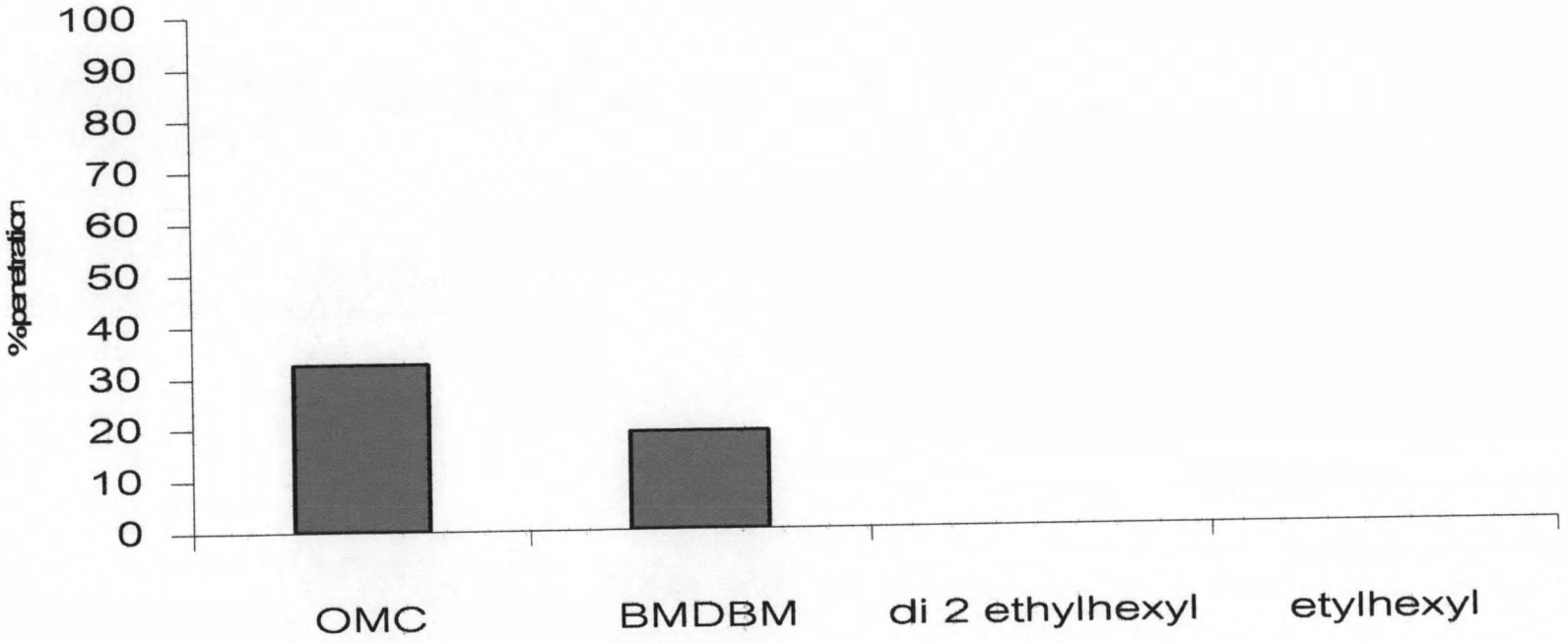

Figure C.4 % penetration of OMC, BMDBM, di(2-ethylhexyl)-2,4,5- trimethoxybenzalmalonate and 2-ethylhexyl-2,4,5 trimethoxycinnamate at 3 h in human third volunteer

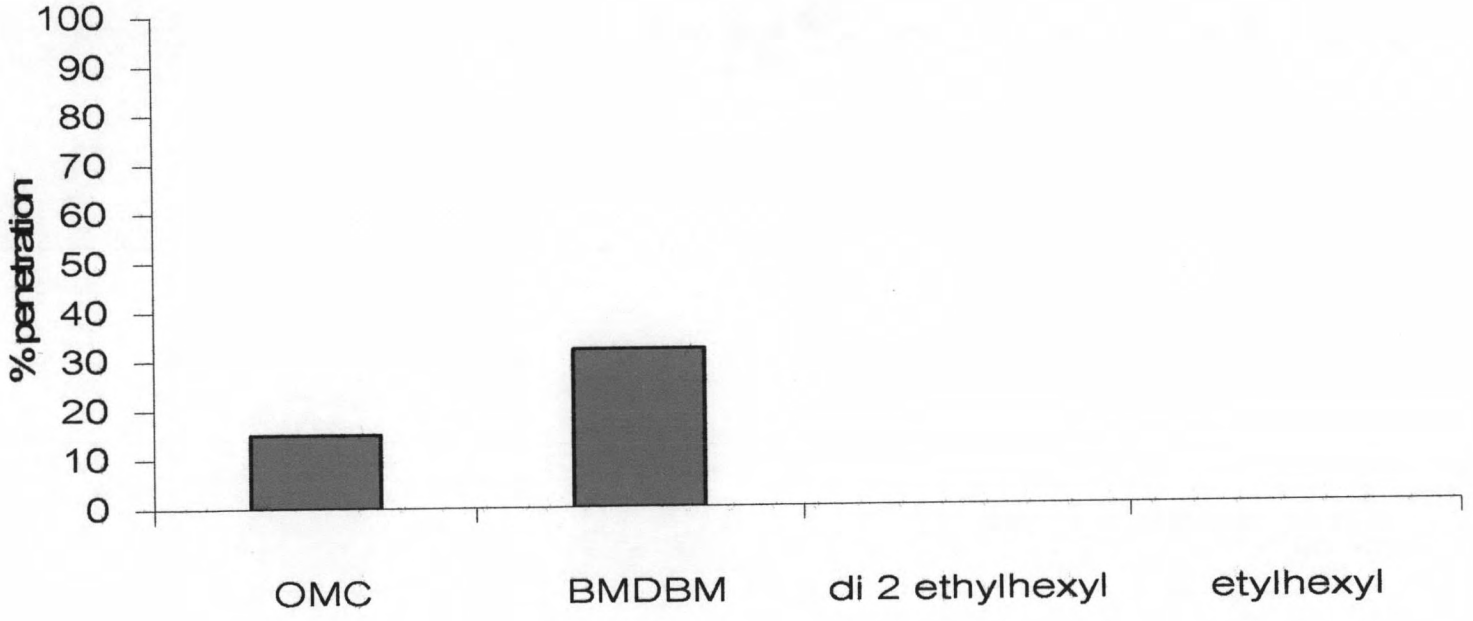

Figure C.5 % penetration of OMC, BMDBM, di(2-ethylhexyl)-2,4,5-trimethoxybenzalmalonate and 2-ethylhexyl-2,4,5 trimethoxycinnamate at 3 h in forth human volunteer

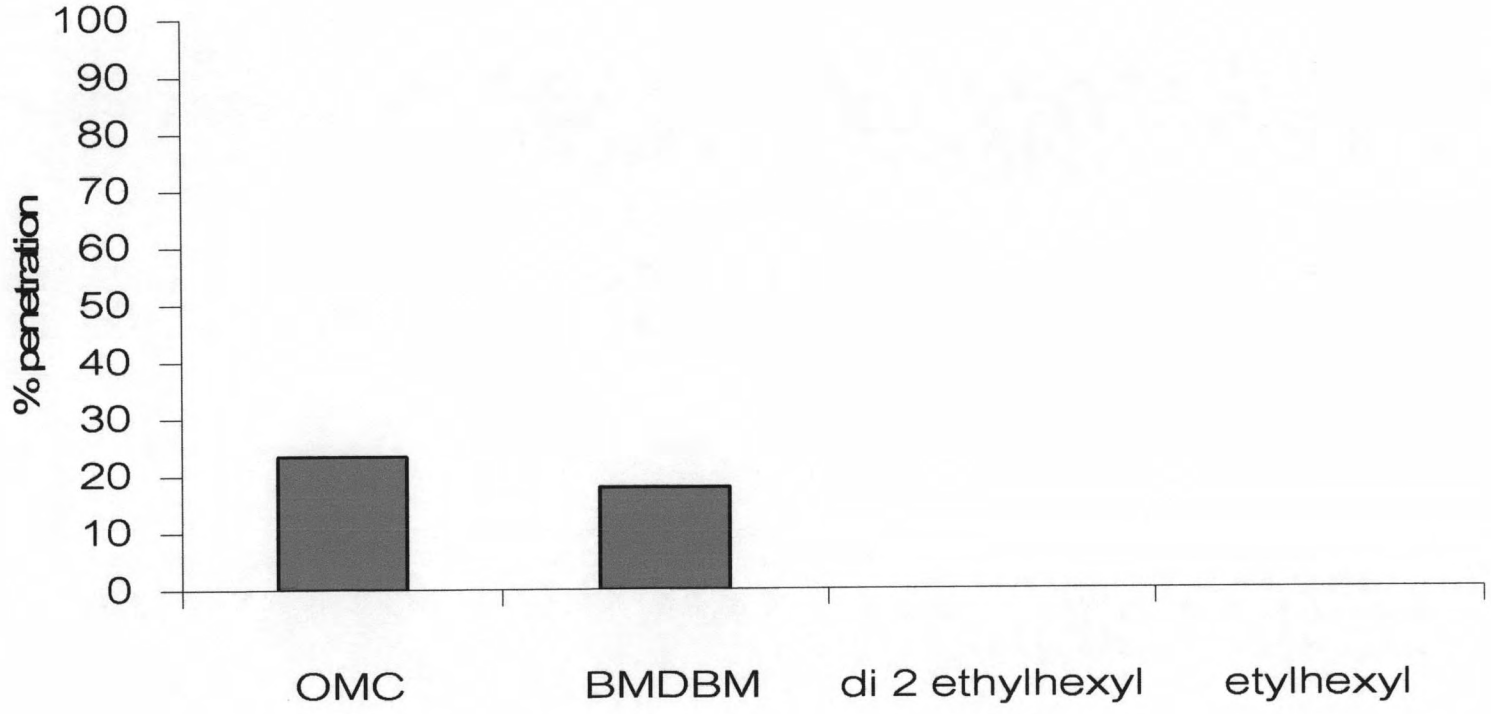

Figure C.6 % penetration of OMC, BMDBM, di(2-ethylhexyl)-2,4,5-trimethoxybenzalmalonate and 2-ethylhexyl-2,4,5-trimethoxycinnamate at 3 h in fifth human volunteer

APPENDIX D

T –test and Pearson's test statistic

Example % penetration of OMC

repeat	1	2	3	4	5
Suction blister	46.75	74.25	32.5	15	23.4
Diffusion cell	40.34	37.44	45.42	36.23	42.8

X	Y	X^2	Y^2	XY
46.75	40.34	2185.56	1627.33	1885.89
74.25	37.44	5513.06	1401.70	2779.92
32.50	45.42	1056.25	2062.90	1476.15
15.00	42.80	547.56	1831.84	1001.52

$\Sigma X = 191.9$ $\Sigma Y = 202.9$ $\Sigma X^2 = 9527.43$ $\Sigma Y^2 = 8236.42$ $\Sigma XY = 7686.93$

$$r = \frac{\Sigma XY}{\sqrt{(\Sigma X^2)(\Sigma Y^2)}}$$

$$r = \frac{7686.93}{\sqrt{(9527.43)(8236.42)}}$$

$$r = 0.8677$$

$r > 0$ indicates a positive linear relationship between the two variables.

$r < 0$ indicates a negative linear relationship between X and Y.

High correlation $r \geq 0.80$ or $r \leq -0.80$

Medium correlation $0.50 < r < 0.80$ or $-0.80 < r < -0.50$

Low correlation $-0.50 \leq r \leq 0.50$

Hypothesis Testing

$H_0: \rho = 0$

$H_1: \rho \neq 0$

The test statistic is $t = \frac{r}{\sqrt{(1-r^2)/(n-2)}}$ $df = N-2$

$$t = \frac{0.8677}{\sqrt{(1-0.8677^2)/(5-2)}}$$

$$t = 3.275$$

t_{table}; $t_{.05}$ df 3 = 3.182

$t_{calculate} > t_{table}$ ∴ Reject H_0: $\rho = 0$ We conclude that have signifiant correlation

ρ = the population Pearson correlation coefficient

r = The sample correlation coefficient

t table

d.f.	α	
	0.050	0.010
1	12.706	63.657
2	4.303	9.925
3	3.182	5.841
4	2.776	4.604
5	2.571	4.032
6	2.447	3.707
7	2.365	3.499
8	2.306	3.355
9	2.262	3.250
10	2.228	3.169

The thickness of baby mice skin

Baby mice skin	Thickness (mm)	Thickness (μm)
1	2.005	2005
2	1.803	1803
3	1.901	1901
4	1.701	1701
5	1.601	1601
6	1.701	1701
7	1.501	1501
8	1.801	1801
9	2.001	2001
10	1.601	1601
Average	1.716	1716
Standard deviation	0.172	163.7

VITA

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