A Clinical Trial on the Safety and Efficacy of Oil of Bergamot 30% and Sun Exposure combination versus Sun Exposure alone in the Management of Residual Hypomelanosis of Pityriasis Versicolor

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Abstract

This study compared the safety and efficacy of Oil of Bergamot with sun exposure, and that of sun exposure alone in the management of the resulting hypomelanosis of Pityriasis versicolor. There was indeed a difference in the melanin and erythema indices between the two treatments with results favoring the former in terms of improvement of the condition of interest.

Introduction: Pityriasis versicolor is a skin problem in a tropical country like the Philippines. It is characterized by pinkish to hypopigmented patches usually affecting trunk and extremities. The residual hypomelanosis of this condition is a common concern for most patients. There is no known treatment for it except to wait for its spontaneous resolution for a period of six months or more, a solution which is unacceptable for many. Oil of Bergamot is an extract from the plant Citrus bergamia containing the active ingredients of 5-methoxypsoralen and 8-genoxypsoralen, members of psoralen compound which is currently being used along with UVA (PUVA) for several dermatologic conditions such as Vitiligo and Progressive Macular Hypomelanosis. It is a photosensitizing and phototoxic agent which aids in the turning back of melanocytes in conditions characterized by reduced pigment cells.

Methods: This was a single-blind study among patients aged 11 to 65 years old diagnosed with Pityriasis versicolor clinically and through a confirmatory KOH examination. There were 28 patients that were included in the study with no dropouts. Patients were instructed to split their backs into halves, right and left, one side subjected to application of oil followed by 30-minute sun exposure while the other was solely subjected to sun exposure, with the assignment of treatment sides decided by a third party. The duration of treatment was four weeks with weekly determination of melanin and erythema indices via a mexameter.

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Results: ANOVA of repeated measures was used to determine if there was a difference between the two treatment groups for both the dependent variables melanin and erythema. With a p value of <0.0001, the melanin and erythema significantly improved after the treatment of oil of bergamot and sun exposure.

Conclusion: The combination of Oil of bergamot and 30-minute sun exposure significantly increased the melanin and erythema levels of the hypomelanotic lesions brought about by Pityriasis versicolor as compared to sun exposure alone**.** Adverse reactions were limited and were of mild intensity demonstrating that a short contact with the oil is sufficient enough to produce the needed erythema but inadequate in yielding strong hypersensitivity reactions.

Keywords: Hypomelanosis; Pityriasis versicolor; Oil of bergamot; Mexameter; melanin index; erythema index

INTRODUCTION

Skin color has always been an important issue since time immemorial. Many individuals, particularly during olden times, would look primarily on one's complexion, unfortunately deducing judgments solely from it. Though the latter seems no longer taking place these days, the habit of giving so much value on a person's skin appearance still holds true up to this modern time. This is where the role of us, dermatologists, comes in. We are entrusted to take well care of the largest organ system in a human's body.

Pigmentation whether in the form of hyper- or hypomelanosis is a taxing dermatologic problem to both the clinician and patient. It is a condition which on the part of the former, is extremely difficult to manage while for the latter, there is exhaustion on the waiting process for its resolution. While there are several products and procedures available in the market to address skin darkening, there is barely anything known to public for its opposite. Several dermatoses bring about alteration in skin color. For hyperpigmentation, causes vary from sun damage to hypersensitivity reactions to contact dermatitis. As for hypopigmentation, sources include inflammatory dermatoses such as psoriasis and seborrheic dermatitis, dermatophytoses, mycosis fungoides, vitiligo and many others.

In the Philippines, being a tropical country, Saprophyte infections are not infrequent, with Pityriasis versicolor being one of the most commonly seen cases. It is characterized by slightly scaling macules that can be hypopigmented, pink or salmon-colored or hyperpigmented. This superficial mycosis which is common among young adults is said to be caused by the yeast Malasezzia sp., of which M. globosa and M. sympodialis most frequently have been identified in scales of lesions.[1] Current management for this infection includes several topical antifungal preparations such as Selenium sulfide, 2.5%, Ketoconazole, Ciclopirox etc. However, these treatments while they can manage to relieve the redness and pruritus, the residual hypopigmentation remains to be unsolved.

Oil of Bergamot, an essential oil from Citrus bergamia (Bergamot orange) which is being used in various expensive fragrances, is proven to be an effective treatment for Vitiligo. Bergaptene, also known as 5-methoxypsoralen and Bergamotin, also called as 5-Geranoxypsoralen, are the major furocoumarin contents of this oil, and regarded as highly photoreactive psoralens.[2] Therefore, products containing Bergamot oil are said to be photosensitizing and highly phototoxic causing erythema and eventual repigmentation to occur in vitiligo patients.

On the other hand, Narrowband UVB light therapy (NB-UVB) is a treatment which uses the optimal part of the UVB light spectrum to cause repigmentation of one's natural skin coloring.[3] This mechanism of action is currently taken into advantage as treatment for hypopigmented dermatoses such as vitiligo and progressive macular hypomelanosis.

Therefore, it is the aim of this study to work around the strong points of these two treatments as a means of finding an effective and safe treatment to the challenging repercussions of Pityriasis versicolor which is decreased skin pigmentation. However, with the highly demanding requirement in the frequency of hospital visits as well as the cost of Narrowband UVB treatment, in this experimental research, sun exposure will serve as its alternative.

REVIEW OF RELATED LITERATURE

Pityriasis versicolor (also known as Tinea versicolor) is a common skin complaint characterized by various pigmentary changes of the skin, usually affecting the chest, back, neck and arms. The patches may be pink, coppery brown or paler than surrounding skin, which can often be associated with pruritus. The hyperpigmentation or hypopigmentation of skin depends on the outcome of interactions between Malassezia yeasts and the skin, such as lipoperoxidation process, stimulus of inflammatory cell to melanocytes, and increased thickness of keratin layer.[4]

Confirmation of diagnosis is made in two ways: The first one is via 10 % to 20% Potassium Hydroxide (KOH) microscopy wherein hyphae and yeast cells are seen resembling the so-called 'spaghetti and meat-balls'. The second one is by means of Wood's light (long wave Ultraviolet A) which shows a yellow-green fluorescence over the affected areas among one out of three patients.[5]

Pityriasis versicolor is more common in hot, humid climates or in those who sweat heavily, so it may recur each summer. It does not appear to predispose affected areas to sunburn even when it causes pale white marks.[6] It is here where the cosmetic concern comes in, the consequent hypopigmented patches over the affected areas.

The oil of Bergamot, an essential oil extracted from Citrus bergamia or Bergamot orange which is a fragrant fruit the size of an orange, with a yellow color similar to a lemon, is widely being used in the perfume industry. The oil has the ability to combine with an array of scents to form a bouquet of aromas which complement each other. Approximately one third of all men's and about half of women's perfumes contain bergamot essential oil.[7]

Having 5-methoxypsoralen and 5-geranoxypsoralen as its components, Bergamot oil is both photosensitizing and phototoxic. Its photosensitizing properties aid in turning back on melanocytes in conditions with reduced amount of the said pigment cells given the proper stimulation via ultraviolet light.[8] Taking lead of this property the oil is being used as one of the treatment for vitiligo, a skin condition characterized by depigmented patches due to destruction of melanocytes as well as of other dermatologic conditions characterized by hypopigmentation. In a study done by Kinley et.al they made use of hairless mice to assess the epidermal melanogenesis induction of topical furocoumarins (active ingredient of Bergamot oil) along with UVA radiation. Each subject was subjected into a 12-day application of topical furocoumarin followed by exposure to subphototoxic doses of UVA.

The results of which have shown evidences of melanogenesis such as increased density and size of melanocytes.[9] In another experimental study of Romano et.al the in vitro activity of Bergamot oil against clinical isolates of dermatophytes was determined. The outcome of which suggested its potential use against common species of dermatophytes.[10] In a similar study done by the same group of researchers, they measured the Minimum Inhibitory Concentration (MIC) of bergamot oil against clinical isolates of Candida species from which the group have found a probable usage of Citrus bergamia as a topical treatment against Candidal infections.[11] These studies therefore make the Oil of Bergamot more appealing in the management of Pityriasis versicolor not only in terms of its value in the restoration of skin color but as well as the possibility of decreasing the growth and thus recurrence, of Malasezzia infection.

Interestingly, similar hypomelanotic conditions such as vitiligo and progressive macular hypomelanosis can both be managed with either Narrowband UVB or topical/oral Psoralen and UVA (PUVA). In a study done by Wu CS et. al., they have proposed that PUVA is best used in the active stages of vitiligo as it slows down the destruction of melanocytes (MC) thereby creating a favourable environment for MCs to survive. On the other hand, NBUVB irradiation will best suit stable vitiligo with its ability to stimulate both proliferation and migration of MCs.[12] Conversely, Progressive Macular Hypomelanosis (PMH), a condition characterized by ill-defined, nummular, hypopigmented spots on the skin due to decreased pigmentation of epidermis has been shown to be effectively and safely treated using NBUVB. In a small uncontrolled and non-blinded clinical trial done by Kim MB et.al, 9 out of 16 patients (56.2%) experienced more than 90% repigmentation while 13 of 16 patients (81.3%) had at least 50% repigmentation after several weekly sessions of NBUVB.[13] While in 2010, Duarte et al. reviewed patients seen in a phototherapy clinic between 1997 and 2008 to evaluate the therapeutic response to PUVA photochemotherapy or Narrowbandband UVB. No significant difference between these two treatments after 16 sessions or three months of treatment has been found with majority of the patients showing more than 50% of skin repigmentation, with 65% classified as cured or much improved.[14]

Whereas, the exact mechanism of hypopigmentation in pityriasis versicolor is not known with recent discoveries pointing towards a role of tryptophan-derived metabolites of Malassezia furfur, similarly it can take advantage of phototherapy relying on its ability to induce maturation of existent melanocytes thus accelerating the repigmentation process.[15]

Both UVA and UVB comprised a certain portion of sun's rays thereby making direct exposure to sunlight, a good source of UV radiation. In particular, UVB are the main rays which are said to cause sunburn. However, excessive exposure to sunlight can increase an individual's predisposition to skin cancers such as melanoma and squamous cell carcinoma. In as much as UV radiation is warranted amongst patients with dermatologic conditions which can benefit from exposure to phototherapy but with limited ability to obtain such in a hospital facility due to lack of time and/or for financial reasons making direct sunlight exposure a good alternative, it is of importance still to advise avoidance of sun from 10 am to 4 pm when UVR intensity is at its peak in so doing, eluding the harmful consequences of too much sun exposure.[16]

Accordingly, proving the efficacy of these different modes of treatment for pigmentary disorders will be difficult if done in a subjective way. Therefore, to minimize this point of error, the researcher will be using a device used to measure skin color that would provide a reproducible means and sensitive means of quantifying small skin color differences. Park es et al. in 2006 have concluded that the Mexameter is an objective pigment-measuring device which can be used to achieve a more accurate diagnosis of hypopigmentary disorders, and that the relative melanin index which represents the relative pigment levels, might be a more effective parameter than the melanin index (MI) itself for comparing pigmentation differences.[17]

In conclusion, the researcher took advantage of the strong points of these literatures with some modifications in order to allow its feasibility in the current setting of the trial with much consideration to the study patients.

GENERAL OBJECTIVE

To compare the safety and efficacy of Oil of Bergamot 30% and sun exposure combination versus sun exposure alone in the restoration of the normal skin color in post-hypopigmented lesions of Pityriasis-versicolor.

SPECIFIC OBJECTIVES:

1. To measure the pigmentation level of the skin by means of Mexameter at weeks 0,1,2,3, and 4 of treatment.
2. To discern the relationship between Melanin Index and the erythema measurement.
3. To determine the adverse effects associated with the use of both treatments via patient's self--assessment forms.

METHODOLOGY

STUDY DESIGN

A single-blinded clinical trial on the safety and efficacy of oil of Bergamot 30% and sun exposure combination versus sun exposure in the management of the accompanying hypomelanosis of Pityriasis versicolor among normal healthy patients with Pityriasis versicolor, who were diagnosed at the Dermatology Out-Patient Department of Jose R. Reyes memorial Medical Center.

STUDY POPULATION

Normal healthy male and female patients aged 11 to 65 years old with Pityriasis versicolor over back area. Diagnosis was made by means of clinical history and examination and confirmed by means of Potassium hydroxide (KOH) examination. Duration and history of prior treatment was not considered. A total of 28 patients were enrolled in this study.

Exclusion criteria include known hypersensitivity reaction to any form of fragrances (known to contain Bergamot oil), and concomitant medical conditions known to be exacerbated by Ultraviolet exposure such as Lupus Erythematosus. Pregnant and breast-feeding mothers were also excluded from the study.

MATERIALS

The oil of Bergamot 30% was purchased by the researcher from a local supplier packaged in a plastic opaque jar ensuring its stability. Sunblock gel from another local company was also provided by the researcher to warrant the skin protection of non-involved skin areas of all study patients.

RANDOMIZATION, TREATMENT ALLOCATION, AND BLINDING

The randomization of back areas of the subjects was done by a third party via computer-generated method. The oil was handed to the subject by the said party who re-instructed the patient as to its application and as to which side should it be placed. In this experiment, the researcher as well as the physician performed the mexameter readings were blinded.

STUDY INTERVENTION AND ASSESSMENT

Diagnosed patients were treated primarily with topical and/or oral antifungal opted by the primary clinician. A repeat KOH was performed to check for clearance of dermatophyte infection.

On follow-up, the residual hypomelanosis was managed. Each patient had the affected body area divided into two areas. One area was subjected to the application of Bergamot oil 30% 30 minutes prior to sun exposure while the other side was directly exposed to the sun without any other intervention. Both sides had exposure between 6 am to 9 am to avoid the intense burning heat past the given time. In addition, patients were instructed to apply sunblock gel on normal skin, 30 minutes prior to going out while the involved areas with oil were washed off with water after heat exposure prior to the application of sunblock gel.

To address the possible result-altering external factors, every patient was instructed to have avoidance of sun exposure after the given time and to protect their skin upon stepping outside via use of umbrella and wearing of proper shirt at all times.

On doing the mexameter reading, two representative lesions were chosen from each side by the researcher and by the third party who handled the said device. The representative lesions that were chosen were based on which portion is the whitest or most hypopigmented of all. Photos were taken on every reading to mark the representative lesions.

CLINICAL ASSESSMENT

Baseline photos and mexameter readings were taken prior to the initiation of therapy. Patients were asked to follow-up on a weekly basis for a period of 4 weeks wherein a questionnaire regarding the adverse effects was requested to be answered. Repeat photography and Mexameter readings were done on every follow-up.

Primary endpoint was the restoration of normal skin color as per evidence of clinical photographs and the increase in the level of pigmentation via the mexameter. The grading were as follows: Grade 1 for < 50% of repigmentation or interpreted as slightly improved; Grade 2 for 50% to 79% of repigmentation or moderately improved; Grade 3 for 80% to 99% or much improved; Grade 4 for 100% or cured. Secondary endpoint was the number of weeks needed to achieve at least 50% of re-pigmentation.

STOPPING GUIDELINES

A subject would have been withdrawn from the study upon any signs of severe irritation such as blistering, intense erythema and pruritus. Patients who used other topical preparations or those who would were unable to comply with the proper sun exposure as well as the wearing of protective gears would have been taken out of the study.

SAMPLE SIZE

Based on the 2013 census of the JRRMMC Department of Dermatology, Out Patient Department, 237 cases of Pityriasis versicolor were seen out of the total of 30,328 patients. The researcher set the maximum permissible error at 5%, α at 5% and confidence level at 95%. Open Epi results indicated that a minimum of 28 patients was needed to achieve a 95%

level of confidence, precision of 5% given an estimated with outcome to be at 80%. The researcher needed minimum of 28 patients to achieve a 95% level of confidence, precision of 5% considering a 20% drop outs.

RESULTS

DISCUSSION

There were 28 patients enrolled in the study. Majority of which were males comprising 78.57% of the study population. The mean age of patients was 23.3 years old. In the study, each subject's back area was divided into two. One half of the back was subjected to application of Oil of Bergamot followed by 30-minute sun exposure while the other half was solely exposed to sun for a similar duration as the former. The mean percentage of melanin was 14.46% and erythema was 11.98% on baseline (see table 1).

ANOVA of repeated measures was used to determine if there was a difference between the two treatment groups for both the dependent variables melanin and erythema. With a p value of <0.0001, it can be concluded that the melanin and erythema significantly improved after the treatment of oil of bergamot and sun exposure (see table 2). A post hoc comparison (tukey) test was used to determine at which week showed marked improvement. With p-values of < 0.0001 for all the treatment weeks, it showed that using the combination of oil of bergamot and sun exposure significantly increased the percentage of erythema and melanin.

Fundamentally, the increase in the melanin and erythema values is projected amongst the areas subjected into the combination of the oil and sun exposure. As mentioned in the previous section, the aim of this study is directed on taking advantage of the oil of bergamot's photosensitizing and phototoxic properties. With sufficient stimulation of ultraviolet radiation, the melanocytes are expected to be stimulated following the application of the oil on a hypopigmented spot and thus, break open into a phototoxic reaction priming the development of erythema over the area and eventually leading to its repigmentation. As for the areas which were merely exposed to the sun, the increase in their melanin and erythema indices can be accounted by the induction of maturation of the existing melanocytes by the adequate amount of ultraviolet radiation received.

Conversely, adverse reactions were noted in a number of subjects in areas applied with the Oil of Bergamot. As mentioned, it is both photosensitizing and phototoxic prompting the development of some unwarranted symptoms among patients, particularly those with strong personal and/or family history of atopy. Reportedly, 5 out of 28 patients or 17.8% developed mild pruritus upon contact with the oil which eventually resolved with rinsing of the area. There were 3 out of 28 or 10.7% who experienced sting or a burning sensation upon application of the oil which once more disappeared after washing it off. There were however, only 2 out of 28 patients or 7.1% who had few papules over the area involved both of which having resolution following the application of an emollient cream for a few days. Fortunately, none of the 28 subjects had experienced blister formation. All of the symptoms mentioned were signs of a hypersensitivity reaction to the Oil of bergamot and sun exposure combination. On the other hand, sun exposure alone showed no capacity of inducing such reactions with none of the areas subjected to it had the development of the said symptoms.

With regards to the grading of improvement which has been set for this clinical trial, chi-square was used to determine the difference between the two groups and with a p value of 0.136, there was not much of a difference visually between the two. However, in terms of patient's satisfaction, majority of them verbally expressed their satisfaction with the usage of Bergamot oil along with the 30-min sun exposure pushing them to continue with the management even after the conclusion of their participation in this trial.

In conclusion, the combination of Oil of bergamot and 30-minute sun exposure significantly increased the melanin and erythema levels of the hypomelanotic lesions brought about by Pityriasis versicolor as compared to sun exposure alone. Adverse reactions were limited and were of mild intensity demonstrating that a short contact with the oil is sufficient enough to produce the needed erythema but inadequate in yielding strong hypersensitivity reactions.

As for the author's recommendation, it is recommended that subsequent trials will include a larger population with a longer duration of study. None of the patients have been noted to develop a 100% repigmentation of their lesions which is likely due to the very limited course of the clinical trial. It is also advisable that two arms will be assigned, one with the similar Oil of bergamot with sun exposure versus the other with no intervention at all. This is to see the difference in the repigmentation time between an interventional group and the one which will rely solely on spontaneous resolution.

TABLES AND FIGURES

Table 1. Demographic characteristics of patients on baseline

Subjects N=28	Mean
Age	23.3
Gender	22:6
Melanin	14.26%
Erythema	11.98%

Table 2. Melanin Index (Oil of Bergamot 30% + Sun Exposure)

	WEEK					
Patient	**0**	**1**	**2**	**3**	**4**	**Improvement**
1	200	234	296	297	312	2
2	360	427	457	494	506	1
3	80	87	96	99	99	1
4	374	417	450	499	520	1
5	101	143	183	187	198	3
6	325	390	471.2	481	507	2
7	170.33	209	255	258	281	2
8	215.3	280	308	318	340	2
9	213	240	289	315	343	2
10	257	290	328	332	360	1
11	300	330	401	425	465	2
12	197	224.2	294	313	325.33	2
13	220	256	357	372	401	3
14	336	361	408	420	474.67	1
15	223	234	314		355	2
16	198.2	217	284.33	301	330.33	2
17	115	137	165	201	214	3
18	320	347.67	398	427	484	2
19	234	269	322	334	360	2
20	280	317	356	371	426	2
21	219	245	290	315	336	2
22	303	338	389.3	429.3	478	2
23	180	200	264	286.67	329	3
24	236	292	329	336	384	2
25	254	299	326.33	353	396	2
26	165	194	256	270	304.67	3
27	301	386	469	475	494	2
28	312	357	423	434	499	2

Table 3. Melanin Index in Percentage Increase (Oil of Bergamot 30% + Sun Exposure)

	WEEK					
Patient	**0**	**1**	**2**	**3**	**4**	**Improvement**
1	200	17	48	48.5	56	2
2	360	18.6	26.9	37.2	40.5	1
3	80	8.8	20	23.8	23.8	1
4	374	11.5	20.3	33.4	39	1
5	101	41.5	81.18	85	96	3
6	325	20	45	48	56	2
7	170.33	23	50	51.3	65	2
8	215.3	30	43	48	58	2
9	213	12.7	35.6	47.8	61	2
10	257	12.8	27.6	29.2	40	1
11	300	10	33.7	41.7	55	2
12	197	13.8	49	58.9	65.1	2
13	220	16.3	62.2	69	82.3	3
14	336	7.4	21.4	25	41.2	1
15	223	4.9	40.8	45.7	59.1	2
16	198.2	9.5	43.5	51.8	66.6	2
17	115	19.1	43.5	74.8	86	3
18	320	8.6	24.3	33.4	51.2	2
19	234	15	37.6	42.7	53.8	2
20	280	13.2	27.1	32.5	52.1	2
21	219	11.9	32.4	43.8	53.4	2
22	303	11.6	28.5	41.7	57.8	2
23	180	11.1	46.7	59.3	82.8	3
24	236	23.7	39.4	42.4	62.7	2
25	254	17.7	28.5	39	55.9	2
26	165	17.6	55.1	63.6	84.6	3
27	301	28.2	55.8	57.8	64.1	2
28	312	14.4	35.6	39.1	60	2

Table 4. Melanin Index (Sun Exposure Alone)

	WEEK					
Patient	**0**	**1**	**2**	**3**	**4**	**Improvement**
1	169.67	200	227	242	274	2
2	295	381	399	435	483	2
3	146	166	169	172	178	1
4	382	417.33	496.33	521	545	1
5	194	212	248	257.67	280	1
6	293.33	324	371	396	410.67	1
7	150	170.33	184	198	205	1
8	264	304	363	385	401	2
9	260	295	338.33	347	360	1
10	227	259	298	316	347.33	2
11	313	369	398	439	457	1
12	164.67	174.	193	198	229	1
13	228	269	288	303	355.67	2
14	314.33	347	385	427	472.33	2
15	223	258	292	317	340.67	2
16	238	256	277	302.6	333	1
17	108	115	137	149	165	2
18	347	384	458	495	517	1
19	223	244.67	265	287	323.33	1
20	290	317	352	395	431	1
21	187	201.2	240	244	275	1
22	329	378	409	454	490	1
23	205	249	267	294	313	2
24	224.67	253	270.33	291	331	1
25	276	294	326	388	422	2
26	195	227.2	259	270	289	1
27	276	311	349	376	408	1
28	315	347	389	430.67	472	1

Table 5. Melanin Index in Percentage Increase (Sun Exposure Alone)

	WEEK					
Patient	**0**	**1**	**2**	**3**	**4**	**Improvement**
1	169.67	17.8	33.7	42.6	61.4	2
2	295	29.2	35.2	47.5	63.7	2
3	146	13.6	15.8	17.8	21.9	1
4	382	9.2	29.9	36.4	42.7	1
5	194	9.3	27.8	32.8	44.32	1
6	293.33	10.5	26.5	35	40	1
7	150	13.5	22.7	32	36.7	1
8	264	15.1	37.5	45.8	51.9	2
9	260	13.5	30.1	33.5	38.5	1
10	227	14	31.3	39.2	53	2
11	313	17.9	27.2	40.3	46	1
12	164.67	6	28.3	20.2	39	1
13	228	18	26.3	32.9	56	2
14	314.33	10.4	22.5	35.8	50.3	2
15	223	15.7	30.9	42.1	52.8	2
16	238	7.6	16.4	27.1	39.9	1
17	108	6.5	26.9	38	52.8	2
18	347	10.7	32	42.7	49	1
19	223	9.7	18.8	28.7	45	1
20	290	9.3	21.4	36.2	48.6	1
21	187	7.6	28.3	30.5	47	1
22	329	14.9	24.3	38	48.9	1
23	205	21.5	30.2	43.4	52.7	2
24	224.67	12.6	20.3	29.5	47.3	1
25	276	6.5	18.1	40.6	52.8	2
26	195	16.5	32.8	38.5	48.2	1
27	276	12.7	26.4	36.2	47.8	1
28	315	10.2	23.5	36.7	49.8	1

Table 6. Erythema Index (Oil of Bergamot 30% + Sun Exposure)

	WEEK				
Patient	**0**	**1**	**2**	**3**	**4**
1	265.33	286	331	348	395
2	399	433	427	455	479
3	362	384	408	447	470
4	385	429.67	467	488	475
5	245	288.33	288	292.33	315
6	354	400.67	447	460	484
7	227.33	274	290	311	347
8	247	267	296	327	365
9	226.67	288	344	361	387
10	292	314	374.67	412	438
11	321	379	401	458.33	489
12	200.67	215.33	236	250	294.67
13	241.2	268	285	323	361.67
14	367	402	424	496	562
15	269	296	337	354	395
16	238	256.33	289	307.67	345
17	139	155	179.67	197	219
18	358.2	395	458	501.33	540.67
19	283.33	297	321	375	424.67
20	303	328	371.33	414	455
21	256	277	293	336	384
22	311	338	343.33	366	429
23	215	264	301.67	316	355
24	267	294	345	380	407.33
25	298	347	356.33	382	422
26	202	236.67	251	274	314
27	319.67	344	379	428	479.67
28	344	381	417	475.33	524

Table 7. Erythema Index in Percentage Increase (Oil of Bergamot 30% + Sun Exposure)

	WEEK				
Patient	**0**	**1**	**2**	**3**	**4**
1	265.33	7.79	24.8	31.2	50.8
2	399	8.5	7.01	14	20
3	362	6.07	12.7	23.5	29.8
4	385	11.6	21.3	26.7	23.4
5	245	17.7	17.6	19.3	28.6
6	354	13.2	26.3	30	36.7
7	227.33	20.5	27.6	36.8	52.6
8	247	8.1	19.8	32.4	47.7
9	226.67	27	51.8	59.3	70.7
10	292	7.53	28.3	41.1	50
11	321	18.1	37.3	42.8	52.3
12	200.67	7.3	17.6	24.6	47.1
13	241.2	11.1	18.2	3.9	50
14	367	9.5	15.5	35.1	53.1
15	269	10	25.3	31.6	46.8
16	238	7.7	21.4	29.2	45
17	139	11.5	29.3	41.7	57.6
18	358.2	10.3	27.9	40	51
19	283.33	4.8	13.3	32.4	49.9
20	303	8.25	22.6	36.6	50.2
21	256	8.2	14.5	31.3	50
22	311	8.7	10.4	17.6	37.9
23	215	22.8	40.3	47	65
24	267	10.1	29.2	42.3	52.5
25	298	16.4	19.6	28.2	41.6
26	202	17.2	24.3	35.6	55.4
27	319.67	7.61	18.6	33.9	50
28	344	10.8	21.2	38.2	52.3

Table 8. Erythema Index (Sun Exposure Alone)

	WEEK				
Patient	**0**	**1**	**2**	**3**	**4**
1	269	322	327	323	371.6
2	363	418	410	457	482
3	248	266	256	260.33	316
4	405.67	425	447	471	508
5	232	265.67	297	274	306
6	312	348	395	370	399
7	184	211.67	235	248	276
8	298	339	378	363	389.33
9	283.67	341.67	357	368	374
10	254	297	315	328	352
11	346.33	388	397.67	411	453
12	213.2	246.33	255	278	299
13	265.33	294	310.67	328	352
14	346	371	399.33	416	440
15	272	301	315	347	368
16	266	285	298.33	312.67	330
17	159	189	219	237	231.67
18	380	412.33	437	459	464
19	245.67	268	295.67	302	339
20	319	348	369	381	410
21	208	246.67	263.67	272	319
22	355.33	386	401.33	417	454
23	237	252	274.33	290	307.67
24	242	268	294	318	339
25	290	319.67	337	342	384
26	234	250.67	282	305	317
27	305	347.33	384	372.67	385
28	338	360	395	415	425

Table 9. Erythema Management in Percentage Increase (Sun Exposure Alone)

	WEEK				
Patient	**0**	**1**	**2**	**3**	**4**
1	269	19.7	21.6	20	38.1
2	363	15.1	12.9	25.9	32.8
3	248	7.25	3.22	4.7	27.4
4	405.67	4.8	10.2	11.2	25.2
5	232	14.5	28	18.1	31.9
6	312	11.1	26.6	18.6	27.9
7	184	15	27.7	34.8	50
8	298	13.8	26.8	21.8	30.6
9	283.67	20.8	25.9	29.7	31.8
10	254	29	24	30.3	38.9
11	346.33	12	14.8	18.7	30.8
12	213.2	15.5	19.6	30.4	40.2
13	265.33	10.8	17.1	23.6	32.7
14	346	8.1	15.4	20.2	27.1
15	272	10.7	15.8	27.6	35.3
16	266	7.1	12.2	17.5	24
17	159	18.9	37.7	49.1	45.7
18	380	8.5	15	20.8	22.1
19	245.67	9.1	20.4	22.9	38
20	319	9.1	15.7	19.4	28.5
21	208	18.6	26.8	30.8	53.3
22	355.33	8.6	13	17.4	27.8
23	237	6.3	15.8	22.4	29.8
24	242	10.7	21.5	31.4	40.1
25	290	10.2	16.2	17.9	32.4
26	234	7.12	20.5	30.3	35.5
27	305	13.9	25.9	22.2	26.2
28	338	6.51	16.9	22.8	25.7

Table 10. ANOVA Table

Subjects N=28	Oil of bergamot and sun exposure				sun exposure alone				p value
weeks	**1**	**2**	**3**	**4**	**1**	**2**	**3**	**4**	
Melanin	16.07	39.38	46.94	59.61	12.85	26.61	35.71	47.43	<0.0001
Erythema	11.73	22.99	32.37	43.8	12.24	19.54	23.58	33.21	<0.0001

Figure 1. Graphic Comparison of Melanin Index

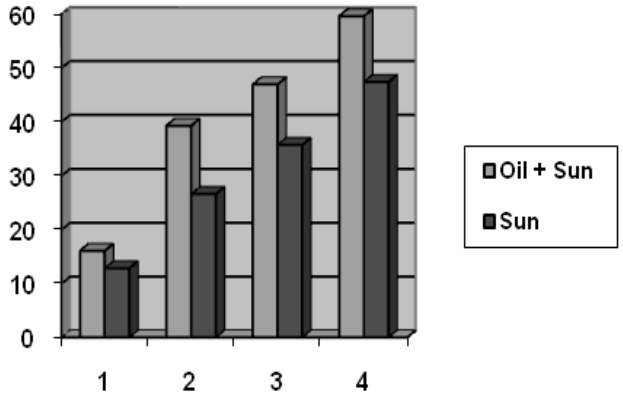
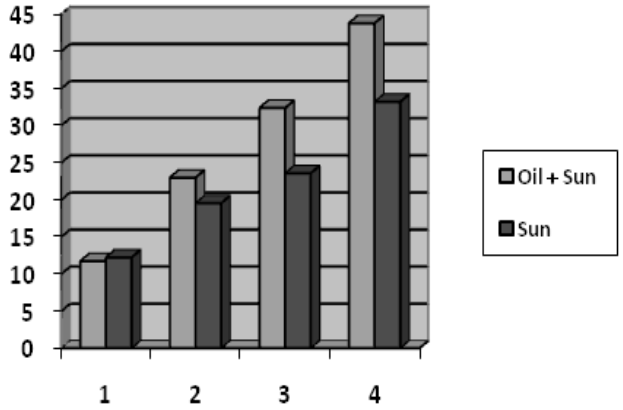


Figure 2. Graphic Comparison of Erythema Index



REFERENCES

1. MorishitaN., Sei Y. Microreview of Pityriasis versicolor and malazessia species. Mycopathologia. 2006 Dec; 162(6):373-6
2. Kejijova K., Jirova D., Bendova H., Kandarova H., Weidenhoffer Z., Kolarova H., Liebsch M. Phototoxicity of bergamot oil by in vitro techniques in combination with human patch tests. Toxicology In Vitro. 2007; 21(7):1298-303
3. Handog EB, Dayrit JF. Mycology in the Philippines, revisited. Nippon Ishinkin Gakkai Zasshi. 2005; 46 (2):71-6
4. Gupta AK, Bluhm R, Summerbell. Pityriasis versicolor. Journal of the European Academy of Dermatology & Venearology 2002; 16:19
5. Oakley A., Review: Pityriasis versicolor. Journal of the European Academy of Dermatology and Venearology 2005 March; 19(2):147
6. Hyun JP, Yang WL, Yong BC, Kyu JA. Skin characteristics in patients with Pityriasis versicolor using non-invasive method, MPA 5. Annals of Dermato logy. 2012 Nov; 24(4):444-52
7. Brannt WT, Schaedler, Karl A. Practical treatise on animal and vegetable fats and oils
8. Placzek M, Fromel W., Eberlein B., Gilbertz KP et al., Evaluation of phototoxic properties of fragrances. ACTA Derm Venearol. 2007; 87(4):312-6
9. Sapam R., Agrawal S., Dhali TK. Systemic PUVA vs narrowband UVB in the treatment of vitiligo: A Randomized controlled trial study; Dept of derm & veneer. Int J Dermatol 2012 Sep; 51(9):1107-15
10. Kim MB, Kim GW, Cho HH et al. Narrowband UVB treatment of Progressive macular hypomelanosis. Journal of American Academy of Dermatology. 2012: 66:598-605

11. Clarys P., Alewaeters K., Lambrecht, Barel AO, Clarys P. Skin color measurements: comparison between three instruments: the chromameter, the derma spectrometer and the mexameter. Skin Research and Technology. 2000 Nov; 6(4): 230-238

12. WU CS., Lan CC., Wang LF., Chen GS., Wu CS.,Yu HS. Effects of psoralen plus ultraviolet A irradiation on cultured epidermal cells in vitro and patients with vitiligo in vivo. Br J Dermatol. 2007 Jan;156(1):122-9

13. Kim MB, Kim GW, Cho HH et al. Narrowband UVB treatment of progressive macular hypomelanosis. J Am Acad Dermatol. 2012 Apr;66(4):598-605.

14. Thoma W[1], Krämer HJ, Mayser P. Progressive macular hypomelanosis: an epidemiological study and therapeutic response to phototherapy. An Bras Dermatol. 2010 Sep-Oct;85(5):621-4. 15

15. Jennifer Smith. A guide to healthy sun exposure. Danielplan.com/healthyhabits

16. Park ES, Na JI, Kim SO, Huh CH, Youn SW, Park KC. Application of a pigment measuring device--Mexameter--for the differential diagnosis of vitiligo and nevus depigmentosus. Skin Res Technol. 2006 Nov;12(4):298-302.