STUDY PROTOCOL

TITLE

"Efficacy and safety of topical administration of timolol maleate 0.5% solution in the treatment of Infantile Hemangioma in Early Proliferative Stage" Randomized Control Trial

Protocol version	Version: 4. Date: 09/23/2014
Research product	Timolol maleate 0.5%
Code:	IIBSP-TIM-2013-156
Indication	Infantile Hemangioma
Promoter	Institut de Recerca de l'Hospital de la Santa Creu i Sant Pau – IIB Sant Pau c/ Sant Antoni Maria Claret, 167 08025 Barcelona Phone: 93 553 78 69 Fax: 93 553 78 12
Research Coordinator	Dra. Eulalia Baselga Dermatology Service Hospital de la Santa Creu i Sant Pau c/ Sant Antoni Maria Claret, 167 08025 Barcelona Phone: 93553 70 02 E-mail: ebaselga@santpau.cat dra.baselga@gmail.com
Research Monitor	Pablo Bros/ Nàdia Llavero Institut de Recerca de l'Hospital de la Santa Creu i Sant Pau – IIB Sant Pau c/ Sant Antoni Maria Claret, 167 08025 Barcelona Phone: 93 553 76 35 Fax: 93 553 78 12

GOOD CLINICAL PRACTICE COMPLIANCE

This trial is designed to comply with Section ICH E6 Guideline for Good Clinical Practice, as it has been implemented in Europe on January 17, 1997 (CPMP-CPMP/ICH /135/95 Guideline) and in the USA dated May 9, 1997 (Guidance for the Industry E6 Note for Guidance on Good Clinical Practice: Consolidated Guidance, April 1996); local regulatory requirements and the directive on clinical trials 2001/20/EC of the European Parliament of April 4, 2001.

Signature sheet

Title: **"Efficacy and safety of topical administration of timolol maleate 0.5% solution in the treatment of Superficial Infantile Hemangioma in Early Proliferative Stage"**
Randomized Control Trial

Code: IIBSP-TIM-2013-156

Protocol version 4. Date: 09/23/2014

Promoter: Institut de Recerca de l'Hospital de la Santa Creu i Sant Pau – IIB Sant Pau

The undersigned agrees to carry out the study as specified in the protocol and in accordance with Good Clinical Practices.

Principal Investigator

Eulalia Baselga _____ _____
Signature Date

Coordinator of the Central Clinical Research Unit:

Claudia E. Delgado _____ _____
Signature Date

Research Monitor:

Pablo Bros _____ _____
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Nàdia Llavero Signature Date

Quality Control Monitor

Jordi Virgili _____ _____
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Central Clinical Research Unit Administrator:
Rosa M. Antonijoan _____Signature Date

CLINICAL TRIAL PROTOCOL INDEX

1. SYNOPSIS……

1.1 Trial Identification	EUDRACT number: 2013-005199-17 Promoter protocol code: IIBSP-TIM-2013-156 Version: 4. Date: 09/23/2014
1.2 Clinical Trial Title	*"Efficacy and safety of topical administration of timolol maleate 0.5% solution in the treatment of Infantile Hemangioma in Early Proliferative Stage" Randomized Controlled Trial*
1.3 Promoter Identification	Institut de Recerca de l'Hospital de la Santa Creu i Sant Pau – IIB Sant Pau c/ Sant Antoni Maria Claret, 167 08025 Barcelona Phone: 93 553 78 69 Fax: 93 553 78 12
1.3.1 Research Monitor	Pablo Bros/ Nàdia Llavero Institut de Recerca de l'Hospital de la Santa Creu i Sant Pau – IIB Sant Pau c/ Sant Antoni Maria Claret, 167 08025 Barcelona Phone: 93 553 76 35 Fax: 93 553 78 12
1.4 Financial Source	Dermatology Service Funds
1.5 Research Study Coordinator	Dr. Eulalia Baselga Dermatology Service Hospital de la Santa Creu i Sant Pau c/ Sant Antoni Maria Claret, 167 08025 Barcelona Phone: 93553 70 02 E-mail: ebaselga@santpau.cat dra.baselga@gmail.com
1.6 Ethics Committee Reference	Ethics in Clinical Research Committee of the Hospital de la Santa Creu i Sant Pau.
1.7 Centers where the trial will be conducted	Dermatology Service of the Hospital de la Santa Creu i Sant Pau
1.8 Justification and relevance of the study	Infantile hemangioma is the most common benign vascular tumor in childhood. Most of the lesions are not present at birth. They manifest in the first days of life with a precursor lesion that establishes the extension of the lesion; after that, it has a rapid proliferative stage during the 5th to 7th week of

	life, reaching 80% of its size at 3-5 months followed by a stage of involution that can last from 3 to 5 years. Most infants are evaluated by primary care physicians/pediatricians during the first days of life, where a precursor lesion can already be seen. These explain its benignity and natural history to involution. Most of these lesions are benign and do not need treatment, or one can be expectant; however, in a group of patients, the lesions grow exponentially, causing stress in the parents who seek the help of a specialist, which in most cases occurs after 5 months of age on average, where even if we start treatment, a large percentage will show skin sequelae such as changes in color, anetodermic skin, telangiectasias, fibro-fatty tissue, etc. A certain percentage of hemangiomas have special characteristics depending on the location, extension, and involvement of natural or organic orifices, which can put the patient's life at risk. There are multiple therapeutic alternatives reported to date, with systemic corticosteroids being considered the first choice of treatment, even with all the side effects that this entails. In 2008, the usefulness of β-adrenergic blockers was reported; in this case, propranolol hydrochloride in the treatment of complicated hemangiomas, revolutionizing the current treatment of hemangiomas, since they slow progression and promote involution quickly; however, side effects such as hypotension, hypoglycemia, and bronchial hyperresponsiveness have been reported, which can limit its use. In 2010, the usefulness of timolol maleate 0.5% in solution was reported in the treatment of a superficial infantile hemangioma on the eyelid, showing significant involution of the lesion. As of this date, several case reports, case-control series, and only one randomized clinical study have been published, where they have demonstrated the usefulness of this drug, as well as its safety in infants, establishing it as an off-label treatment for not complicated superficial hemangiomas. These articles highlight the function of timolol, which exerts an inhibitory effect on the growth and slow regression of the lesion with minimal side effects, both in the early proliferative phase and in the stabilization phase of the hemangioma. In both cases, with both propranolol and timolol used in our practice compassionately, we have realized that when using these drugs in the early stages of the lesion, we find a higher success rate and less risk of sequelae; this is also supported

	by previous publications of the drugs. This work aims to provide information to date unknown on the efficacy of timolol maleate in newborns with infantile hemangiomas to decrease progression and growth during the early proliferative phase, and we will also evaluate the safety of the drug in this age group. The use of timolol maleate 5 mg/ml solution has been approved in Spain since 1980 for the treatment of hypertensive conditions of the eye (open-angle glaucoma) in both adults and children. It currently has the marketing number 55.236 with the last authorization renewal date in October 2009. However, it is not approved for use in the treatment of infantile hemangiomas.
1.9 Study design. Study Phase	Placebo-controlled double-blind randomized clinical trial 2-group parallel design Phase IIa Study Single-center
1.10 Main Objective	<u>Main objective:</u> To analyze the efficacy of timolol maleate 0.5% in solution in the involution of infantile hemangioma in the early proliferative phase.
1.11 Experimental and control drugs	Experimental drug: Timolol maleate 0.5% in solution, in topical administration. We will use a dose of 2 drops every 12 hours (equivalent to 0.5 mg/timolol) giving a gentle massage on the lesion. The duration of treatment will be 24 weeks. Control drug: Placebo based on saline solution, in topical administration. We will use a dose of 2 drops every 12 hours giving a gentle massage on the lesion. The duration of treatment will be 24 weeks. To evaluate the compliance with treatment in both groups, at the end of each of the eye drops, the empty containers will be collected by the HSCSP Pharmacy.
1.12 Main variable for evaluation	**Main variable:** Complete/almost complete resolution of infantile

	hemangioma at week 24 of treatment, where complete resolution is defined as a complete improvement of the lesion, and almost complete resolution is defined as the existence of a minimal degree of telangiectasia, erythema, skin thickening, soft tissue swelling and/or distortion of anatomical landmarks. Assessment of this variable will be carried out by comparing the blind centralized independent qualitative evaluations of the photographs of the IH from W24 with the baseline period. Treatment success will be defined as a centralized "complete/near-complete resolution" assessment of the IH at W24 compared to baseline. This qualitative assessment is the one that has been used in all childhood hemangioma treatment studies since changes in thickness are difficult to measure in other ways. In a previous study of hemangiomas and imiquimod, carried out in our department, an attempt was made to measure color using a colorimeter and although the colorimeter was capable of measuring discoloration changes, the differences in scoring had no clinical significance. On the other hand, the objective measurement of the thickness of the hemangioma is often not possible due to the existence of a deep component. For these reasons, a qualitative measurement has been chosen on a scale of 1-4 of the different variables (complete resolution, improvement, stabilization, and worsening)-where "almost complete resolution" is defined as a minimum degree of telangiectasia, erythema, skin thickening, soft tissue swelling and/or distortion of anatomical landmarks as recorded in the CRF.
1.13 Study population and number of patients	Patients between 10 and 60 days old will be included, who must have at least: - Focal or segmental hemangioma, both superficial and mixed between 0,3 and 5 cm in size, at any location on the body surface; or - Precursor of hemangioma defined as pinkish macules with a whitish halo in the periphery, clinically characteristic of the precursors of hemangiomas in childhood; or - "abortive" or minimally proliferating hemangioma, defined as a telangiectatic angioma showing proliferation in <5% of the hemangioma surface.

	This is a proof of concept study. There are no data on the evolution at 24 weeks of localized infantile hemangiomas. We postulate complete or near-complete resolution in 10% of patients in the placebo group and expect 40% success in the timolol group. Based on these hypotheses, a sample size of 70 patients will suffice, distributed equally between both groups (with a risk of αlpha of 5% (0.05%) and a β error of 20% (0.80) and a power of 90%. A loss percentage of no more than 10% is considered. Stratification by age is not foreseen since the inclusion age is only 10-60 days. Stratification by growth phase will also not be done, since at 10-60 days the hemangiomas are all in the proliferation phase. Finally, the size of the sample does not allow stratification by location or type of hemangioma. The characteristic of these patients will be vascular tumors, newborns, which do not have any previous treatment, without associated diseases, derived from primary care consultations and pediatricians in the area, as well as those who arrive at the HSCSP service.
1.14 Statistical analysis	In this study, we will carry out two types of analysis: one of intention to treat, which will include all patients who have been randomized, even if they have only received a single dose, and the other, analysis by protocol: where patients who have completed the study will be included. A statistician blinded to the study will carry out the statistical analysis. Continuous Baseline and demographic characteristics will be summarized by sample size, mean, standard deviation (SD), minimum, median, and maximum. Categorical Baseline data will be summarized by number and proportions in each category. For data that are not normally distributed, medians and interquartile ranges will be used. The $X2$ analysis will be used to determine response predictors. The response to treatment will be assessed using multiple regression analysis adjusted for repeated measurements and other predictor variables such as age at initiation of treatment, sex, dose, etc. An analysis of the full set of analyzes will be performed. A value of 5% will be considered significant. Main efficacy analysis: Success rates (complete/near-complete resolution at W24) will be compared between timolol maleate in solution and

	placebo using a logistic regression model. The odds ratio and the corresponding 95% confidence interval will be provided as a measure of treatment effect. Complementary analysis: The main analysis of the protocol repeated on the set is composed of all FAS patients without any significant protocol deviation or other source of bias for the main criterion analysis and characterized by a minimum treatment exposure of 28 days (4 weeks) and with the main criterion of efficacy available. Sensitivity analysis: The proportion of successes in the main criterion will be analyzed using a logistic regression model. Secondary criteria analysis: Description of its evolution by treatment group. Comparison of IH evolutions between the treatment groups using a logistic regression model. Criteria for the persistence of efficacy: Qualitative evaluations performed by the investigator (4-point scale: complete resolution, improvement, stabilization, worsening) of the evolution of the hemangioma will be compared between the treatment groups at the week 36 visit, compared with week 24, using a logistic regression model.
1.15 Ethical Considerations	The study will be carried out in strict accordance with international ethical recommendations for research and clinical trials in humans. Likewise, the standards set out in the Declaration of Helsinki will be guaranteed and will be developed in accordance with the protocol and standard operation procedures (SOPs) that ensure compliance with the standards of Good Clinical Practice (GCP). The researcher must explain to the parents or the authorized legal representative of the patient, the nature of the study, its purpose, procedures, estimated duration, the potential risks and benefits related to participation in the study, as well as any inconvenience that this may entail. Parents or guardians must be advised that participation in the study is voluntary and that the study can be abandoned at any time, without affecting subsequent treatment, or the relationship with the professionals treating the patient.

	For this purpose, an information/consent form has been designed for parents or the authorized legal representative, which is attached. No other medical risks will be added as a result of this study. The possible discomfort that could be noticed is the close follow-up in the first month of treatment and the application of the treatment; this for the parents does not pose any risk to the infant, neither pain nor additional exploration. Patients may not benefit from participating in this study; however, the results of the study will allow us to establish whether timolol is effective in the involution of early proliferative phase infantile hemangioma. Due to the presence of this tumor exclusively in early childhood, all patients who are recruited will be infants. This is an independent promoter study for academic purposes. Both the principal investigator and the collaborators will not receive any type of sponsorship or honorarium for conducting this study. The diagnosis of infantile hemangioma is clinical; it is not necessary to carry out any additional exploration such as a biopsy or other invasive study, or others that carry a risk for the patient. In some cases, a radiological study can be carried out, such as magnetic resonance, and this is to rule out involvement in hemangiomas associated with syndromes, which is an exclusion criterion in our study. Direct access to the original documents obtained from this study will be provided to the study sponsor, who will carry out monitoring, audits, and regulatory inspections related to the trial, always ensuring the identity of the patient and solely for supervision purposes. As a control drug, we will use an isotonic saline-based eye drop in topical administration, which will not cause any extra risk to the patient. Since hemangioma is a benign tumor in which management with an expectant attitude towards involution is accepted, we consider it permissible, from a methodological and ethical point of view, the use of a placebo-type control drug to evaluate the net effect of the experimental treatment. To date, the experimental drug is not approved for the study indication.

1.16 Treatment Duration	Study period per patient: 24 weeks of treatment and 12 weeks of post-treatment follow-up. Planned patient recruitment dates: Start: July 2014 End: December 2014 Scheduled visits: baseline visit (S0), Visit S2, S4, S8, S12, S24, and S36.
1.17 Safety Evaluation	The safety endpoints will be based on the following assessments: - Recording of adverse events at all visits - Recording of heart rate, blood pressure, pulmonary auscultation, liver palpation, and global physical examination at all visits. - Local tolerability at week 2, 4, 8, 12, and 24: A 4-point scale will be used (very good tolerability, good tolerability, poor or very poor tolerability). o Very good tolerability: the absence of subjective signs or physical signs of local side effects. o Good tolerability: transitory subjective signs, without physical signs, nor need to modify the frequency of application of the product. o Poor tolerability: persistence of subjective signs or physical signs of local side effects, which make it necessary to modify the frequency of application of the product but not to suspend treatment. o Very poor tolerability: subjective and/or physical signs that make it necessary to suspend treatment.
1.18 Study Calendar Study Duration	The first patient to include: July/01 /2014 Inclusion of the last patient: December 31, 2014 Completion of the last patient in the study: September 31, 2015 Study completion: September 31, 2015 The total duration of the study: 15 months

1.19 Flow Diagram of the Study

Procedure	Treatment Period						Follow-up
	S0	S2	S4	S8	S12	S24	S36
Informed consent	X						
Inclusion/Exclusion Criteria	X						
Review of demographic data	X						
Review of medical history/concomitant diseases	X						
Randomization	X						
Global physical examination	X	X	X	X	X	X	X
Height, weight, pulmonary auscultation, liver palpation, vital signs	X	X	X	X	X	X	X
IH photographs	X	X	X	X	X	X	X
Qualitative evaluations of the IH carried out in the center	X	X	X	X	X	X	X
Qualitative evaluations of the photographs	X	X	X	X	X	X	X
Qualitative evaluations of efficacy carried out by the researcher at the center		X	X	X	X	X	X
Qualitative evaluations of efficacy carried out by the parents at the center		X	X	X	X	X	X
Review of adverse events		X	X	X	X	X	X
Local tolerability		X	X	X	X	X	
Study drug delivery to the parents as needed	X		X	X	X		
Review of previous/concomitant drugs	X	X	X	X	X	X	X
Pick up study drug and compliance assessment			X	X	X	X	

2. Glossary

AA	Adverse Events
SAE	Severe Adverse Event
AE AT	Adverse event appearing during treatment
GCP	Good Clinical Practice
CEIC	Ethics Committee
OC	Observed cases
COS	Classification by organ and systems
CRF	Case report form
INN	International Nonproprietary Name
Fas	Full set analysis
HR	Heart rate
ES	End of Study
ET	End of treatment
RR	Respiratory rate
IH	Infantile Hemangioma
LOCF	Last observation carried forward
bpm	beats per minute
mm	Millimeter
WHO	World Health Organization
OR	Odds Ratio
RP	Research product
NWP	Normalized Work Procedure
AP	According to protocol
AR	Adverse Reaction
UAR	Unexpected Adverse Reaction
SUAR	Severe Unexpected Adverse Reaction
NMR	Nuclear magnetic resonance
W	Week
PT	Preferred term

3. General Information

3.1 Clinical Trial Title

"Efficacy and safety of topical administration of timolol maleate 0.5% solution in the treatment of Superficial Infantile Hemangioma in Early Proliferative Stage" Randomized Controlled Trial

Protocol Code: IIBSP-TIM-2013-156

Protocol version 4. Date: 09/23/2014

3.2 Description of Study Products

3.2.1 International Nonproprietary Name (INN)

Experimental drug
Timolol maleate 0.5% eye drop solution

Control drug
Saline eye drop solution

3.2.2 Chemical Denomination

Experimental drug
®CUSIMOLOL 0,5%

Control drug
Saline eye drop solution

3.2.3. Molecular form

Experimental drug
$C_{13}H_{24}N_4O_3S \cdot C_4H_4O_4$
Molecular weight: 432.50

Control drug
NaCl

3.2.4. Qualitative and Quantitative Composition

Experimental drug

Every ml of CUSIMOLOL 0.5% contains 5 mg of timolol maleate.
Excipients: monosodium phosphate, disodium phosphate, sodium chloride, benzalconium chloride and purified water.

Control drug

Contains 9 grams of NaCl or 154 mEq of Cl and 154 mEq de Na+ in 1 liter of H2O, with an osmolarity of 308 mOsm/L.

3.2.5. Pharmaceutical form

Experimental drug

Eye drop solution
Transparent, colorless or clear yellow solution

Control drug

Eye drop solution
Transparent, colorless or clear yellow solution

3.2.6. Dose and route of administration

Dose	Route of Administration	Frequency
2 drops	Topical	Every 12 hrs

3.2.7. Drug Supplier

Pharmacy Department Hospital de la Santa Creu i Sant Pau

3.3 Data of the Promoter

Institut de Recerca de l'Hospital de la Santa Creu i Sant Pau – IIB Sant Pau
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3.4 Research Monitor Data

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3.5 Researchers Data

3.5.1 Research Coordinator and Principal Investigator

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3.5.3 Biometry and Data Administrator

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3.6 Predicted Trial Duration

Total estimated duration of the study would be 15 months

a) Planned patient recruitment dates:

Recruitment start date: July 2014
Recruitment end date: December 2014

b) Study period:

24 weeks of treatment and 12 weeks of post-treatment follow up per patient.

c) Programmed patient visits:

1) Selection (Visit 0)
2) Week 2
3) Week 4 (+/- 3 days)
4) Week 8 (+/-3 days)
5) Week 12 (+/-3 days)
6) Week 24 (+/- 7 days) End of treatment
7) Week 36 (+/-7 days) End follow up period without treatment

4. Background, justification and Objectives

4.1. Background and Justification of the study

Background

Infantile Hemangioma (IH) is the most common vascular tumor in childhood, with an approximate incidence of 10% in the pediatric population. In general, they are benign tumors, with a wide spectrum of clinical presentation, which varies from small superficial lesions to others that leave significant sequelae and can put the patient's life at risk.

Infantile Hemangiomas have a unique and peculiar natural history, where most are not present at birth, they have a rapid proliferation stage, followed by a slow involution stage. Recently it has been reported that the early proliferative stage is present in the first 5.5 - 7.5 weeks of life, the moment of maximum growth, completing 80% of its size between 3 - 5 months of life. The involution stage is reached between 3 - 5 years of life, where we find the final sequelae of the hemangioma, which varies from no cutaneous changes to those that leave anetoderma, telangiectasias, changes in coloration, and redundant fibrofatty tissue.

A certain percentage of hemangiomas have special characteristics depending on the location, extension and involvement of natural or organic orifices, which can put the patient's life at

risk. There are multiple therapeutic alternatives reported to date, considering systemic corticosteroids the treatment of the first choice, even with all the side effects that this entails, such as hypertension, hyperglycemia, general malaise, exogenous Cushing syndrome, and an increase in the number of infections.

Since 2008, the efficacy of propranolol hydrochloride in the treatment of hemangiomas was discovered by chance. Currently, treatment with this non-selective β-blocker is recommended in complicated, ulcerated hemangiomas that are life-threatening due to organic compromise or those that leave significant disfigurement. Other new recommendations are segmental distribution, psychosocial stress, or failure to other treatments. However, the potential secondary risks such as hypotension, hypoglycemia, bronchial hyperresponsiveness make it difficult to justify its use in patients with smaller or less severe lesions.

In 2010, the efficacy of timolol maleate 0.5% in solution was reported for the treatment of a periorbital hemangioma, another β-blocker, an inhibitor of β adrenergic receptors in a non-selective manner, used in the treatment of glaucoma for more than 30 years. This topical therapeutic alternative is currently used in the treatment of superficial infantile hemangiomas, both localized and segmental, an indication supported by case reports and case series. There is only one randomized controlled study where safety, effectiveness, and tolerance are reported in patients with superficial infantile hemangiomas.

None of the 2 drugs have been approved to date, but numerous treated patients, as well as multiple new studies, overwhelm us with the favorable results obtained both in terms of efficacy and safety, since side effects are present, but have been reported as minimal in frequency, previously noted.

After compassionate use with timolol in some of our patients, we have the impression that if treatment is started in the very early stages of the appearance of the hemangioma and when there is only a superficial component, we can avoid the progression of the hemangioma and the need for systemic treatment with propranolol.

Justification

We know that propranolol and timolol are effective both in the early proliferative phase and in the late proliferative phase, although the specific mechanisms of action by which they inhibit proliferation and promote involution of these lesions are unknown. To date, there are no studies that demonstrate that the initiation of early treatment is better than the one that is established after the early proliferation stage, with which we establish the hypothesis that when starting topical treatment in the detection or the first 4- 5 weeks inhibits the growth of the lesion and thus reduce the need to start systemic treatment with the secondary risks that it entails. This will allow us to assess the efficacy and potential impact of drugs during their appropriate use in the early proliferative phase and to determine the efficacy and safety of topical treatment in newborn patients. The use of timolol in the very early stages of hemangioma growth may avoid the need for propranolol.

Timolol maleate is a drug used for more than 30 years for the treatment of open-angle glaucoma, both in children and adults.

In 2010, the first successful case was reported in the treatment of a superficial infantile hemangioma in the upper eyelid, with a complete improvement of the lesion after 7 weeks of use, without complications or side effects. To date, ≈ 300 patient cases have been reported, included in various case reports, case studies and controls, and a randomized clinical study, mostly for the treatment of superficial infantile hemangiomas. The current off-label indication is its use in the treatment of uncomplicated hemangiomas, however, within these case reports, they have been used in patients with periorificial, deep, ulcerated hemangiomas and a case with PHACE Syndrome (posterior fossa alterations cerebral, infantile facial hemangiomas, cerebral artery anomalies, coarctation of the aorta and other cardiac anomalies and ocular anomalies) being these subtypes of hemangiomas candidates for systemic treatment, all of them with excellent results and without any reported side effects.

To date, there are no studies that demonstrate the pharmacokinetics of this drug for its topical application. It has been reported that absorption through the eye in adult patients is around 50%. This route in pediatric patients has reported side effects as dizziness, bradycardia, burning sensation and itching, Cheyne Stokes breathing, apnea, and asthmatic exacerbations.

The effectiveness of the transcutaneous absorption of timolol maleate is attributed to two factors: the barrier function of the skin in infants, which matures up to one year of age, which favors the penetration of the drug, as well as the composition with lipophilic properties that favors absorption. Under this route, only two cases with adverse effects have been reported, a case with sleep disturbances that could no longer continue with the use of the drug and another case where the patient presented an irritating reaction of stinging and burning, due to the extensive surface of the lesion; however, we will continue with the treatment for improvement.

Infantile hemangioma is an exclusive disease of infants and young children. To date, an expectant therapeutic attitude is recommended in the treatment of innocent infantile hemangiomas. However, early intervention can reduce psychological stress in parents, reducing the consequences and aesthetic shadows in children.

It has been suggested as the probable mechanism of action for timolol maleate to cause an arrest in the growth of the hemangioma as well as a gradual reduction in coloration and size. To date, the pharmacokinetics of the drug in its topical application is not well known, nor has the appropriate vehicle or dosage been established. Some studies have used timolol concentrations at 0.5%, 0.25%, and 0.1%, as well as different vehicles, in gel, mineral oil (polyethylene glycol), and ophthalmic solution. The administration used is topically from 1 to 5 applications a day, with a treatment duration of 1 to 12 months, all this according to the improvement of the lesion before the use of the treatment.

No conclusion has been reached as to whether patient monitoring is necessary during treatment. Our experience with topical application is that it is safe, as we have not found any adverse effects to date. However, in studies carried out with systemic propranolol, it has been concluded to carry out heart rate and blood pressure monitoring in patients, which has also been followed in several studies with topical timolol. To date, no alterations have been

reported in the measurements of these constants, but it is useful to determine the safety of the treatment.

In one of the largest studies carried out, with a total of 73 patients, retrospective and multicenter, where two types of timolol maleate concentrations were evaluated at 0.5 and 0.1%, they were indicated with possible predictors of response to treatment: the age of onset early <6 months, duration of treatment for> 3 months, timolol concentration at 0.5% is better than the concentration at 0.1%, superficial infantile hemangioma subtype with a greater response, facial location (not statistically significant) and that To date, no relapse has been reported with the use of topical timolol.

In the only randomized clinical trial conducted to date, the regression rate in patients aged 1-6 months was also found to be higher than in patients> 6 months (P <0.05). Likewise, the regression rate and efficacy in the treated group improved significantly compared to the control group (P <0.05).

Based on what has already been established in these studies, we decided to carry out this study with the following guidelines: we will use the ophthalmic solution because it is the only one approved for use and commercialization in the Spanish territory, concerning concentration, we will use 0.5%, for the same reason and because it has already been shown that this concentration is more effective than if it is lowered. The route of application will be topical, distributing it over the entire lesion with an amount of 2 drops equivalent to 0.5 mg of topical timolol, reducing side effects with higher doses, with 12-hour intervals between each application, which is what is most reported since increasing the number of applications has not been related to a higher profit. The treatment period will be 6 months, because greater improvements are reported in patients who use the drug for more than 3 months and because we assume that at 6 months we will find the maximum degree of improvement.

Among the advantages that we find with the use of this treatment, it is a low cost, distribution, and bioavailability, as well as its easy administration and minimal adverse effects.

4.2. Study Objectives

4.2.1 Primary Objective

To analyze the efficacy of timolol maleate 0.5% solution in 10-60 day-old infants, which is expressed as the complete/ almost complete resolution of IH after 24 weeks of treatment.

4.2.2 Secondary Objective

1. Analyze the efficacy of timolol maleate 0.5% expressed as the decrease in thickness, color and volume of the IH at week 24 of treatment.

2. Analyze the efficacy of timolol maleate 0.5% through qualitative evaluations of IH improvement by parents or guardians.

3. Analyze the persistence of efficacy 12 weeks after the end of treatment.

5. Type of Study and Schematic Design of the Protocol

5.1 Study Type and Design

This is a prospective, randomized, double blind, placebo-controlled, parallel group, single-center, phase IIa study.

The protocol has been designed as a double blind, placebo-controlled study. The allocation of treatments will be done blindly. The products look similar and their packaging and labeling will be similar. Thus, researchers and parents or legal guardians will not know what product the patients receive during the study.

Participants will be randomized with a 1:1 ratio. Randomization will be managed by an independent randomization team conducted by the Department of Pharmacy. Since the hemangioma is a benign tumor, which does not carry a risk in the life of the patient, the opening of the blinded will not be performed until the end of the study (last visit of the last patient included).

Patients will receive study treatment twice daily for 24 weeks (from baseline to S24) and will then be followed for another 12 weeks (S36). If IH worsens during the treatment period and the investigator considers that, in the interest of the patient's well being, it is necessary to administer a new treatment of their choice, the study treatment will be permanently suspended. Likewise, if regrowth of IH occurs during the follow-up period, the patient can be given a treatment chosen by the investigator. In both cases, the concomitant medication must be fully documented in the case report form (CRF).

The dose that we will use of timolol maleate 0.5% ophthalmic solution/saline eye drops will be two drops on the lesion every 12 hrs. The equivalent of two drops of timolol maleate is 0.5 mg/timolol.

The end of the trial for each patient will take place after the 36 weeks of the study. The patients will be visited by the Dermatology Service to continue their evolution.

5.2 Description of Visits

Seven visits have been scheduled for the study. Whenever possible, patients should be selected and included on the same day. Thus, the maximum duration of the study for a given patient will be approximately 37 weeks.

5.2.1 Baseline Visit: W0 (inclusion)

The patient's parents or legal guardians will be informed about the study and given sufficient time to comment and think (together, if applicable) whether they consent to their child or ward to participate in the study. If they wish to give their informed consent, the patient can be enrolled and treatment started the same day.

Procedures to be carried out in this visit:

- Informed Consent Signature (Patient Information Sheet)
- Informed Consent Signing (Taking Pictures)
- Demographic data review
- Verification of inclusion/exclusion criteria
- Review of Clinical History/Concomitant Diseases
- Global Physical Exploration (present in the CRF, See Appendix 14.2)
- Taking Vital Signs (present in the CRF, See Appendix 14.2)
- Qualitative evaluations of the HI performed at the center (by the researcher) (present in the CRF, See Appendix (14.3)
- Review of concomitant medication
- Taking Photographs of the target HI: two basal photographs must be taken (a frontal projection and a lateral projection)
- Delivery of sufficient study drug to parents or guardians until next visit

If the patient continues to meet all the inclusion criteria and does not meet any of the exclusion criteria and they have agreed to participate in the study, the patient will be sent to the Pharmacy Service for randomization. Once the patient has the treatment, the medicine will be applied in the office and vital signs will be measured again one hour after the treatment is applied.

5.2.2 Visit W2 (+/-1 day)

The following procedures will be performed on this visit:

- Photographs of the HI in two projections

- Global Physical Exploration (present in the CRF, See Appendix 14.2)
- Taking Vital Signs (present in the CRF, See Appendix 14.2)
- Qualitative evaluations of the IH performed at the center (by the researcher) (present in the CRF, See Appendix (14.3)
- Qualitative evaluations of effectiveness carried out at the center (by parents/guardians) (present in the CRF, See Appendix (14.3)
- Verification of study drug compliance assessment
- Review of concomitant medications
- Review of adverse events
- Local tolerability
- Delivery of sufficient study drug to parents or guardians until next visit

5.2.3 Visit W4 (+/- 3 days); Visit W8 (+/- 3 days); Visit W12 (+/- 3 days)

The following procedures will be performed at these visits:

- Photographs of the IH in two projections
- Global Physical Exploration (present in the CRF, See Appendix 14.2)
- Taking Vital Signs (present in the CRF, See Appendix 14.2)
- Qualitative evaluations of the IH performed at the center (by the researcher) (present in the CRF, See Appendix (14.3)
- Qualitative evaluations of effectiveness carried out at the center (by parents/guardians) (present in the CRF, See Appendix (14.3)
- Verification of study drug compliance assessment
- Review of concomitant medications
- Review of adverse events
- Local tolerability

- Delivery of sufficient study drug to parents or guardians until the next visit.

In case everything is correct, we observe improvement or the patient is stable, the patient will be summoned to his next visit for W24.

5.2.6 Visit W24 (+/- 7 Days) (End of Treatment)

The following procedures will be performed on this visit:

- Photographs of the IH in two projections
- Global Physical Exploration (present in the CRF, See Appendix 14.2)
- Taking Vital Signs (present in the CRF, See Appendix 14.2)
- Qualitative evaluations of the IH performed at the center (by the researcher) (present in the CRF, See Appendix 14.3)
- Qualitative evaluations of effectiveness carried out in the center (by parents/guardians) (present in the CRF, See Appendix 14.3)
- Verification of study drug compliance assessment
- Review of concomitant medications
- Review of adverse events
- Local tolerability
- Indication of the reason for the end of treatment in case of premature interruption of treatment.
- Each of the bottles with medicine will be collected by the Pharmacy Service.
- End of treatment

5.2.7 Visit W36 (+/- 7 days)

The following procedures will be performed on this visit:

- Photographs of the IH in two projections

- Global Physical Exploration (present in the CRF, See Appendix 14.2)
- Qualitative evaluations of the IH performed at the center (by the researcher) (present in the CRF, See Appendix (14.3)
- Qualitative evaluations of effectiveness carried out at the center (by parents/guardians) (present in the CRF, See Appendix (14.3)
- Review of concomitant medication

The end of treatment (ET) will occur in the following cases:

- Early failure of treatment (before W12) due to ineffective treatment according to the investigator's criteria. In particular, if the investigator considers that it is best to interrupt the study treatment prematurely because in his or her clinical evaluation she/he has found that the target IH has worsened compared to the previous visit or that it has not begun to improve from W4.
- Early treatment failure (before W12) due to treatment intolerance
- Refusal to complete the 24-week treatment period by the patient's parents or guardians. Details of the refusal to complete the 12-week treatment period should be noted in a comment field on the ET page.
- If the investigator determines that early suspension of treatment (before the ET) is necessary for safety reasons that are not reasonably considered related to the protocol treatment.
- Wrong inclusion according to the study protocol. The investigator and sponsor will jointly decide whether or not to keep the patient in the study.
- Early discontinuation of the study for a reason other than those just listed (for example, patients lost to follow-up).

The investigator or their representative may select one or more reasons for premature ET in the CRF. If more than one reason is selected, the choice of the main reason will be made in the following order for analysis purposes:

- Wrong inclusion according to the protocol
- Treatment intolerance
- Lack of efficacy of treatment

- Safety reason not related to protocol treatment
- The decision of the patient's parents or legal guardians
- Others

The investigator should not discontinue study treatment prematurely (before W24) due to treatment efficacy.

In the event that treatment is interrupted before W24, the procedures for reporting the cause must be carried out (parents decide to suspend treatment, due to ineffectiveness, prescription, or use of a prohibited treatment).

Whenever possible, all children should be followed up until the end of the study (W36), regardless of the reason for the ET.

6. Selection and Withdrawal of Subjects

6.1 Selection of subjects

Patients referred by primary care physicians and clinical pediatricians who are in the province of Barcelona will be included for evaluation in this study. Patients referred by the Pediatric Service of the Hospital de la Santa Creu i Sant Pau and patients who come to the dermatology consultation will also be assessed.

6.1.1 Inclusion criteria

- Informed Consent signature by the patient's parents or guardians, both for participation in the study and for taking photographs.
- The patient must be 10 to 60 days old at the time of inclusion.
- The patient must have at least:
  - A focal or segmental hemangioma, both superficial and mixed, between 0.3 and 5 cm in size, at any location on the body surface; or
  - A precursor of hemangioma defined as pinkish macules with a whitish halo in the periphery, clinically characteristic of the precursors of hemangiomas in childhood; or
  - An “abortive” or minimally proliferating hemangioma defined as telangiectatic angiomas showing proliferation in <5% of the hemangioma surface

6.1.2 Exclusion criteria

- Patients with less than 10 and more than 60 days of life at the time of inclusion.
- Patients with an indication for systemic therapy (ulcerated hemangiomas, on mucosal surfaces, disfiguring)
- Patients who are on another treatment modality for hemangiomas (beta-blockers, corticosteroids, interferon, cyclophosphamide, vincristine)
- Hemangiomas associated with Syndromes (PHACE, LUMBAR, SACRAL, PELVIS)
- Hemangiomas that affect any organ or airway
- Patients with any underlying disease (bronchial asthma, severe lung disease, sinus bradycardia, second and third-degree atrioventricular block, overt heart failure, or cardiogenic shock).
- Patients with congenital defects (patients with some chromosomal syndrome, patients with congenital heart disease (tetralogy of Fallot, transposition of the great vessels, Ventricular communication, Atrial septal defect, patent ductus arteriosus)
- Patients with oncological pathology (leukemias, sarcomas, neuroblastoma, retinoblastoma, etc)
- Hypersensitivity to the active principle or some of the excipients.

6. 2 Predetermination of Sample size

For this project, it is intended to recruit a total of 70 cases, distributed equally between both groups.

For this calculation, it is assumed that the percentage of regression in the control group will not exceed 10%, on the contrary in the treatment group it will be a minimum of 40%, this difference of 30% between treatments may seem high, but there are corroborating data in the literature.

For these calculations, the type I error has been set at the usual 5% (alpha = 0.05), a bilateral approximation. The type II error by 20% (beta = 0.80). Assuming a balanced design and with a loss percentage not exceeding 10%.

6.3 Patient Recruitment

Patients who attend the Dermatology Service of the Hospital de la Santa Creu i Sant Pau referred by pediatricians from the same hospital, by primary care physicians, and by clinical pediatricians who are in the province of Barcelona, as well as those from patients who come directly to the dermatology office.

On the other hand, although without any relation to the present study, an awareness campaign will be carried out regarding the pathology under study aimed at primary care physicians and pediatricians in the area, with the aim that patients with hemangiomas are referred in the phase of early proliferation, as it is when most can be affected with any type of treatment if necessary. We have found that currently, hemangiomas come to our office at around 5 months of age when they have completed growth. The actions we will carry out are yet to be determined: informative talks, information leaflets, etc. As already mentioned, this campaign will not include information about this study or anything related. The Pierre-Fabré laboratories will contribute to the realization of this campaign in a selfless manner.

6.4 Patient Identification

Patients will be identified using a unique 3-digit patient identification code: the number of the patient at the center, in chronological order of selection.

6. 5 Withdrawal criteria and planned analysis of withdrawals and dropouts

6.5.1 Withdrawal Criteria

All included patients will be followed up to S36 when possible. The reasons for premature withdrawal of a patient from the study may be the following:

- The decision of the parents or legal guardians of the patient. Parents or legal guardians of the patient who wish to withdraw their child or ward from the study for any reason can do so at any time but must inform the investigator. In all cases, the investigator should try to see the patient and his parents as soon as possible for a final evaluation with a view to:
  - Obtain the decision of the patient's parents or legal guardians to withdraw their child or ward in writing on the consent form.
  - Obtain, if possible, the reasons for withdrawal and write them down on the CRF
  - Assess the patient's clinical status
  - If necessary, take the appropriate therapeutic measures: treatment of an adverse reaction or concomitant disease, prescription of another treatment.
- By decision of the investigator for the benefit of the patient, for safety reasons (related or not related to the treatment) or insufficient response. Especially if a serious adverse event occurs and the investigator considers that it may endanger the patient's health or if a serious illness appears, which requires the prescription of a medication incompatible with the objective of the study. The monitor will be informed immediately by phone or fax. A letter or report explaining the withdrawal should be sent to the monitor as soon as possible.
- A wrong inclusion according to the protocol. The investigator and sponsor will jointly decide whether or not to keep the patient in the study
- Another reason

6.5.2 Substitution of Patients

Patients withdrawn prematurely or lost to follow-up will not be replaced.

6.6 Exclusion Period after the Study

Patients will not be allowed to participate in another clinical trial while participating in the study and 1 month after the ET. This period is shortened to 1 week if the patient is not included or if, ultimately, does not receive study treatment.

7. Study Treatment

7.1 Dose, posology, route of administration, and pharmaceutical form

7.1.1 Active Treatment:

Timolol maleate 0.5% solution (eye drops)

Dosage	Dose	Route of administration
2 drops equivalent to 0.5 mg/timolol	Every 12 h	Topical

Dosage: 2 drops every 12 hours
Route of administration: Topical
Pharmaceutical formulation: ophthalmic eye drops

7.1.2 Placebo Treatment:

Ophthalmic eye drops with saline solution

Dosage: 2 drops every 12 hours
Route of administration: topical
Pharmaceutical formulation: ophthalmic eye drops

7.2 Special rules for handling and storage

7.2.1 Packaging and Labeling

The treatment kits will be packaged and labeled by the Department of Clinical Pharmacy of the Hospital de la Santa Creu i Sant Pau in compliance with local requirements and European directives.

7.2.2 Masking techniques of study treatments

Both topical timolol and placebo will be packaged in jars with the same characteristics; likewise, the labeling of the product will be carried out by the Hospital de Sant Pau Pharmacy Service, where only they will know how to distribute it.

7.3 Drug Administration

7.3.1 Duration of treatment

Patients will be treated for 24 weeks

7.3.2 Administration schedule

The administration of the product under study is twice a day (morning and evening) for 24 weeks.

7.3.3 Route and conditions of administration

Eye drops, solution for cutaneous use; 2 drops will be applied on the target hemangioma and a brief massage will be given

7.4 Substitution Procedure

Not applicable.

7.5 Concomitant treatments

Any previous or existing concomitant treatment at the baseline visit, any new treatment or dose change from a current concomitant treatment during the study should be noted on the CRF.

7.5.1 Prohibited treatments

The following medications are prohibited from being administered to the patient in the 14 days before randomization or at any time during the 24-week treatment period:

- Cardiovascular treatments:
  - Antiarrhythmics (amiodarone, disopyramide, etc)
  - Calcium antagonists (dihydropyridines, amlodipine, diltiazem, verapamil, etc)
  - ACEI (captopril, etc)
  - Vasodilators (hydralazine hydrochloride, etc)
  - Clonidine
- Antidiabetics (insulin, glitazones, etc.) or potentially hypoglycemia-inducing drugs (pentamidine, quinine, propoxyphene, indomethacin, oxytetracycline, quinidine, chlorpromazine, aluminum hydroxide, ritodrine, etc.).
- Oral non-steroidal anti-inflammatory drugs (NSAIDs) at autoinflammatory doses.
- Sympathomimetic medications and parenteral adrenaline
- Anesthetics, lidocaine (ban period shortened to 48 hours if anesthesia has been performed for diagnostic purposes (eg MRI, etc)
- Or cytochrome inhibitor substrates (amiodarone, cimetidine, fluoxetine, paroxetine, quinidine, imipramine, ciprofloxacin, fluvoxamine, isoniazid, theophylline, fluconazole, etc.).
- Antiulcer drugs (cimetidine, ranitidine, proton pump inhibitors other than omeprazole and lansoprazole.
- Metoclopramide
- Benzodiazepines
- Neuroleptics (chlorpromazine, sultopride hydrochloride, etc.)
- Other drugs: triptans, ergotamine, theophylline, warfarin, thyroxine, floctafenine

The patient is prohibited from taking the following medications at any time before the study or during the study until week 36:

- Corticosteroids, systemically (oral, intravenous or intramuscular), intralesional, or percutaneous.
- Imiquimod
- Vincristine
- Alpha interferon
- Propranolol or other beta blockers

The mother is also prohibited from taking beta-blockers (including propranolol) before the study and during the study (up to week 36) if she is breastfeeding the patient at the same time. In addition, the mother is prohibited from taking the following medications in the 14 days before randomization or during the study if she is breastfeeding the patient at the same time:

- Systemic corticosteroids (oral, intravenous, or intramuscular)
- Vincristine
- Alpha interferon

All IH treatments, as well as any surgical and/or medical procedures (eg laser treatment) before and during the study, are prohibited. If any IH treatment (other than study treatment) is deemed medically necessary during the study, the CRF must clearly state that it will be used to treat IH.

If during the 3-month treatment period it is considered medically necessary for the patient (and/or the mother while breastfeeding) to administer any other prohibited medication, the study treatment should be discontinued and the medication indicated in the CRF managed. However, patient follow-up should continue until W24 whenever possible.

7.5.2 Authorized treatments

All medicines other than the prohibited medicines indicated in the previous section are authorized. All medications administered during the study must be documented. The investigator will carefully analyze at each visit whether any concomitant medications taken by the patient (or nursing mother, if applicable) have been introduced or changed since the previous visit. All medications received by the patient (or nursing mother, if applicable) will be accurately described in the relevant section of the CRF with the following information:

- Treatment name
- Type of treatment (for example, syrup)
- The reason for the prescription
- The route of administration
- Daily dose
- The duration

7.6 Rescue Drug

There is currently no licensed treatment for IH in many countries, so it is appropriate to recommend a specific rescue medication in this protocol. Thus, if necessary, researchers are allowed to prescribe the rescue medication of their choice.

If medication is required due to a significant increase in hemangioma or distortion of body morphology, propranolol oral solution will be used in the following dose:

Propranolol oral solution 5 mg/ml
Dose 2 mg/kg/day, with stepwise start
Period: until improvement is observed, which can be 6 months

8. Response Evaluation

8.1. Efficacy Assessment

8.1.1 Main Criterion

The main variable to evaluate the efficacy of the treatment is the complete/almost complete resolution of the target IH at week 24 of treatment, where complete resolution is defined as complete improvement of the lesion and almost complete resolution is defined as the existence of minimal degree of telangiectasia, erythema, skin thickening, soft tissue swelling and/ or distortion of anatomical landmarks.

The primary binary endpoint (yes/no) will be assessed by comparing the blind centralized independent qualitative assessments of the W24 IH photographs with baseline period. Treatment success will be defined as a centralized “complete/near complete resolution” assessment of IH at W24 compared to baseline.

8.1.2 Secondary Criteria

8.1.2.1 Quantitative evaluations regarding the size, volume and coloration of the IH.

a) Size: in each visit will measure the size of the IH with a flexible tape measure. Two perpendicular measurements will be taken and the approximation of the thickness that protrudes from the skin surface.

b) Volume: the volume will be calculated according to the formula 0.07 x $m^{3.}$ Where m is the average of both perpendicular measurements.

c) Coloration: at each visit the change in coloration of the IH will be evaluated. The scale used will be: almost imperceptible, red with central or mottled clearance, dull red intense red. (See CRF).

8.1.2.2 Qualitative evaluations performed at the center by parents/guardians.

Qualitative evaluations of the degree of improvement in IH carried out by parents at W12 and W24 of treatment in relation to the previous visit, by applying the following scale: 0 complete resolution; 1 Improvement; 2 Stabilization; 3 Worsening.

8.1.2.3 Criteria for persistence of effectiveness.

The qualitative evaluation will be carried out by the researcher using categorical variables of the evolution of the IH, applying the 4-point scale (complete resolution, improvement, stabilization, worsening) at W36 (12 weeks after the end of treatment) and a comparison will be made with the data obtained at W24. Persistence success at W36 will be defined as the persistence between W24 and W36 of a complete/near complete resolution of IH, where "near complete resolution" is defined as a minimal degree of telangiectasia, erythema, skin thickening, soft tissue swelling and/ or distortion of anatomical landmarks.

8.1.2.2 Independent qualitative evaluations carried out at the center by the Researcher

Qualitative evaluations by the researcher at weeks 4, 12, and 24 compared with the baseline period and with the previous visit, by applying the following scale:

- Hemangioma resolution compared to onset:
  Complete/almost complete Yes _____
  Complete _____
  Almost complete _____

No _____

- Hemangioma evolution compared to the previous visit:
  0 Complete resolution
  1 Improvement
  2 Stabilization
  3 Worsening

- Hemangioma evolution compared to the onset:
  0 Complete resolution
  1 Improvement
  2 Stabilization
  3 Worsening

In the original data, photographs taken with standardized procedures uploaded in previous visits (also baseline) will be available to facilitate comparisons with the uploaded photos corresponding to the visit being evaluated.

8.1.2.3 Qualitative evaluations carried out at the center by parents/guardians

Qualitative evaluations by the parents at weeks 4, 12, and 24 in relation to the previous visit, by applying the following scale:

0 Complete resolution
1 Improvement
2 Stabilization
3 Worsening

8.1.2.4 Persistence of efficacy criteria

The qualitative evaluation will be carried out by the investigator using categorical variables of the evolution of the hemangioma, applying the 4-point scale (complete resolution, improvement, stabilization, worsening) at week 36 (12 weeks after the end of treatment) compared to week 24.

Persistent success at week 36 will be defined as the persistence between week 24 and week 36 of a complete/near-complete resolution of IH, where "near-complete resolution" is defined as a minimal degree of telangiectasia, erythema, skin thickening, soft tissue swelling, and/or distortion of anatomical landmarks.

8.1.2.5 Other secondary variables

At all scheduled visits, investigators will also perform other clinical evaluations of the target HI (color intensity, tightness, superficial component, deep component, distortion of local anatomical landmarks).

8.1.3 IH Photographs and Centralized Assessments

In order to perform the evaluations, the researchers at the center will take at least two digital photographs of each patient's IH at the baseline visit (S0), week 2, week 4, week 8, week 12, week 24, and week 36.

The first photograph will be taken with the image plane parallel to HI (front projection) and the second will be taken with the image plane at a different angle from the first (side projection) so that the thickness of the lesion can be clearly seen. The photographs will be used to identify the HI and to make a qualitative evaluation of the evolution of the target HI between S0 and S24 and between S0 and S4, S12, and S24. The evolution of the target HI will not be evaluated with centralized evaluations between S0 and S2 and S8.

The image evaluation sessions will be carried out by independent evaluators under blind conditions. Assessors will be asked to assess, under blinded conditions, whether or not the target HI has been completely/nearly completely resolved in the photographs, where “near-complete resolution” is defined as a minimal degree of telangiectasia, erythema, skin thickening, soft tissue swelling and/or distortion of anatomical landmarks.

8.2. Safety Assessment

8.2.1 Record of Adverse Events (AE)

At each study visit, the appearance of any AE from the last visit or from baseline visit will be determined through the following mechanisms: spontaneous notifications by parents or legal guardians, questions by researcher that will not suggest answers, and investigator clinical evaluations.

All AE will be registered in the CRF, regardless of their severity or casual relationship.

8.2.2 Local Tolerability

At each visit from week 2 to week 24, local tolerability will be assessed as follows:

- Very good tolerance: the absence of subjective signs or physical signs of local side effects.

- Good tolerance: transitory subjective signs, without physical signs or the need to modify the frequency of application of the product.

- Poor tolerance: persistence of subjective signs or physical signs of local side effects, which make it necessary to modify the frequency of application of the product but not to suspend treatment.

- Very poor tolerance: subjective and/or physical signs that make it necessary to suspend treatment.

8.2.3 Global physical examination

A comprehensive physical examination will be performed at each visit to detect any relevant clinical signs. If anomalies are observed, these must be commented on in writing in the CRF.

At each visit, measurements of height, weight, lung auscultation, and liver palpation will be recorded.

8.2.4 Vital Signs

The following vital signs will be determined at each visit: heart rate (HR), blood pressure (BP), and respiratory rate (RR).

The fact that the infant cries or moves can influence the values of heart rate, respiratory rate and blood pressure. These vital signs should be assessed when the child or ward is resting, not moving, or sleeping, if possible. If he is agitated or crying, this should be noted on the CRF.

To determine HR and blood pressure, a stethoscope and cuff of appropriate sizes should be used to avoid false readings.

8.3 Compliance

At each post-basal visit during the treatment period, parents or legal guardians will be asked to return the used/unused Research Product (RP) vial.
The researcher will carry out a qualitative evaluation of compliance (bad/fair/good) in each post-basal visit except S2 and at the global level. In the event of poor or regular compliance, the investigator will record the reasons for his evaluation in the CRF.

<u>Scale for assessing compliance with treatment with respect to the previous visit:</u>

0 Poor or bad
1 Average or regular
2 Good

Where good is defined as the application of the treatment as the original indication (twice a day), Average or Regular is the application of the treatment at least once a day a week or at least 5 times a week and poor or regular is that have suspended treatment for more than a week in the time period of each scheduled visit

8.3.1 Concomitant Treatments

Concomitant treatments will be evaluated at each visit, and recorded in the CRF.

9. Statistical analysis

Once the database has been closed and the randomization code has been opened, Dr. Ignasi J. Gich Saldrich, from the UCICEC-Sant Pau statistics unit, will be in charge of the statistical analysis of this study.

Databases: with all the clinical results obtained and collected in CRF (RedCAP databases) were created with the Excel 2010 program.

Descriptive statistics: For categorical variables, the percentage (%) was calculated based on the number of patients (n); for quantitative variables the arithmetic mean was provided, as well as the range and standard deviation (SD).
Inferential statistics: To evaluate the possible change in response to treatment, Fisher's test was used and the odds ratio (OR) and the confidence interval (CI) were calculated. To evaluate the rest of the variables of the secondary objective that correspond to volume, thickness, and color, as well as the changes observed by the parents, the comparison was repeated with the non-parametric equivalent (Wilcoxon test) for paired data. For the comparison of the treatment time between the two groups (treatment / placebo), the analysis of variance (ANOVA) and test for non-parametric data (Friedman test) were used. Statistical significance: If the statistical analysis showed p <0.05%, the differences detected were considered statistically significant.

Statistical program: The program used for all the analyzes has been the statistical package IBM-SPSS (V. 25.0)

9.1.1 General considerations

The main objective of the study is to evaluate the efficacy of treatment with timolol maleate 0.5% versus placebo in infants with localized uncomplicated proliferative IH. Only the primary analysis of the primary effectiveness measure, on which the sample size justification

is based, can cause a causal interpretation. The other statistical results will have to be viewed from a descriptive perspective. The level of statistical significance of the different two-tailed tests for all analyzes will be 5%.

9.1.2 Stratification

The objective of stratification is to minimize the imbalance between the number of patients in each treatment group associated with the age factor, which may make interpretation difficult. It is not planned to stratify by age, since the inclusion age is only 10 to 60 days and IH at this age is in the proliferation phase equally. Nor will it be stratified by type or location of hemangioma since all hemangiomas will present changes with treatment regardless of type or location.

9.1.3 Sample size

This is a proof of concept study. There are no data on the evolution at 24 weeks of localized infantile hemangiomas. We postulate complete or near complete resolution in 10% of patients in the placebo group and expect 40% success in the timolol group, based on previously published open-label studies. Based on these hypotheses, a sample size of 70 patients will suffice, distributed equally between both groups (with a risk of α of 5% (0.05%) and a β error of 20% (0.80) and a power of 90%.
A loss percentage of no more than 10% is assumed.

9.1.4 Protocol deviations

Before closing the database, the members of the validation committee will review the protocol deviations and classify them as minor or significant. A deviation will be considered significant when it is likely to exert a significant bias in the calculation of the treatment effect based on the primary measure of effectiveness. Patients with significant deviations will be excluded from the analyzed data set. The data set analyzed corresponds to the randomized patients who have received at least one dose of the study treatment. It is the one that will be used to perform the safety and efficacy analyzes. The persistence analysis set corresponds to all randomized patients with a follow-up period, not including patients who have received an unauthorized drug for IH between visits of week 4-24 identified by the validation committee. It is the one that will be used to perform the analysis of the persistence of effectiveness criteria.

9.1.5 Treatment of dropouts or incomplete data

The number and percentage of all treated patients (full set of analyzes) who have prematurely stopped taking study drug will be reported by treatment group. For patients who have stopped taking the study drug prematurely, the time elapsed until discontinuation and the reasons for

premature discontinuation of study drug treatment will also be indicated. Special attention will be paid to the description of adverse events (serious or not) that have caused the suspension of treatment with the study drug. In the event of premature withdrawal during the treatment period, the end-of-treatment visit evaluations will implement the first scheduled visit not performed during the treatment period, regardless of its actual dates (OC observed case analysis). Regarding efficacy criteria, the principle of last observation (not including baseline) considered (LOCF) will be applied to evaluations of OC visits to impute efficacy values not available. In the case of patients who have prematurely stopped taking study drug before the 24 week visit, the last available post-baseline assessment before week 24 will be considered and will be analyzed as "the last visit" during the treatment period for the purposes of efficacy analyzes.

9.1.6 Demographic and baseline characteristics

At the time of selection or inclusion (before the first application of the trial drug), the full set of analyzes will describe the history of the patients, their medical and surgical history, their demographic data, epidemiological data and baseline criteria. The results will be expressed in the form of descriptive statistics in the form of percentages (%), means and standard or median deviations and interquartile ranges.

9.1.7 Main Variable

The primary measure of effectiveness is complete or near complete resolution of the target IH between the first assessment at the week 2 visit and the last visit at week 24 of treatment. The primary endpoint (whether complete / nearly complete or not) will be assessed by comparing the blind independent qualitative assessments of the W24 HI photographs with the baseline assessment. Success rates, which will be based on centralized independent qualitative assessments, will be described in the full set of analyzes by treatment group and time of assessment (W12 and W24). The evaluation of the qualitative variables will be carried out using Fisher's exact test, the OR and the 95% CI will be provided, as well as the X2 test. Any value of $p < 0.05$ will be considered significant. Patients in whom the treatment period has been prematurely interrupted due to ineffectiveness or safety reasons (with a related AE that led to the definitive discontinuation of the study drug) between the 4-24 week visits will be classified as failures. . "A related AE" is an AE that is related to the study drug. No patient will be excluded in the final statistical analysis. Regarding patients who have taken a concomitant treatment not allowed before the 24 week visit, the validation committee will decide whether they are classified as failures.

9.1.8 Secondary Variable

All secondary efficacy endpoints will be discussed in the full set of analyzes. Descriptive statistics will be provided for all criteria, by treatment group and time of evaluation.

a) Decreased size of the IH
Evaluations will be made regarding the volume and thickness of the IH in W2, W4, W8, W12, W24 and W36. The data obtained will be expressed in percentages (%). The mean of both measurements will be taken into account to determine the evolution of IH with respect to the treatment and the evaluation time, using the Wilcoxon test. Any value of P <0.05 will be considered significant. To determine if there is a difference between the two groups with respect to volume, it will be assessed using mixed linear models and an autoregressive structure of covariance (ANOVA). The autoregressive covariance structure was used to allow the nearby volumes at each evaluation point to be more correlated than the further ones. A time-per-group interaction was included to allow groups to vary differently by time.

b) Decreased thickness of IH
It will be evaluated using categorical variables (flat, slight elevation, moderate elevation and marked evaluation) according to the thickness of the surface component of the IH. Evaluations will be carried out in W12 and W24. The data obtained will be expressed in percentages (%). At each evaluation point, the Wilcoxon test will be used to compare the two independent samples. Any value of P <0.05 will be considered significant. To determine if there is a difference between the two groups with respect to volume, it will be assessed using mixed linear models and an autoregressive structure of covariance (ANOVA). The autoregressive covariance structure was used to allow the nearby volumes at each evaluation point to be more correlated than the further ones. A time-per-group interaction was included to allow groups to vary differently by time.

c) Decreased color of IH
It will be evaluated using categorical variables (imperceptible, dull red, red with central clearance or mottled, intense red). Evaluations will be carried out in W12 and W24. The data obtained will be expressed in percentages (%). At each evaluation point, the Wilcoxon test will be used to compare the two independent samples. Any value of P <0.05 will be considered significant.

d) Efficacy reported by parents
It will be evaluated using categorical variables (complete resolution, improvement, stabilization, worsening). Evaluations will be carried out in W12 and W24. At each evaluation point, the Wilcoxon test will be used to compare the two independent samples. Any value of P <0.05 will be considered significant.

e) Persistence of efficacy at W36

It will be evaluated using categorical variables (complete resolution, improvement, stabilization, worsening). Evaluations will be carried out in W12 and w24. At each evaluation point, the Wilcoxon test will be used to compare the two independent samples. Any value of P <0.05 will be considered significant.

f) Local tolerability
Vital sign measurements (systolic and diastolic pressure and heart rate) will be taken at each evaluation visit. At the baseline visit, the treatment will be administered in the hospital and vital signs will be taken before and one hour after the administration. Bradycardia, tachycardia, and hypotension will be tabulated. The results will be expressed in the form of descriptive statistics in the form of means and standard deviations. The X2 test or Pearson's test will be used to evaluate the differences. Any value of P <0.05 will be considered significant. To determine if there is a difference between the two groups with respect to the measurements of SP, DP and HR, it will be assessed using mixed linear models and an autoregressive structure of covariance (ANOVA). The autoregressive covariance structure was used to allow the nearby SP, DP, and HR measurements at each evaluation point to be more correlated than those further away.

g) Side effects reported by parents
Serious adverse events will be described individually: treatment group, patient code, sex and age, term reported by the investigator, preferred term, starting day based on the date of the first administration of study treatment, duration, measure taken in relation to the administration of the study treatment use of a corrective treatment, outcome and relation to the study treatment in the opinion of the investigator. AEs causing the definitive suspension of study drug treatment will also be described on an individual basis.

9.1.9 Concomitant treatments
Concomitant treatments will be tabulated from a descriptive perspective. They will be classified by therapeutic area according to the WHO drug dictionary.

9.1.10 Compliance
Study treatment utilization will not be assessed by treatment group, as returned vials will not be weighed. Researchers' compliance assessments (poor / fair / good) at each visit and globally will be presented by treatment group. Any deviations or other administration related problems reported in the CRF will be provided in individual data listings.

9.1.11 Intermediate analysis and data monitoring
No interim analysis is planned. In addition, taking into account the low systemic exposure to the study product, no safety problems are expected. Thus in this study there will be neither an independent data monitoring committee nor a data and safety monitoring council.

10. Assessment of safety

10.1. Adverse Events. Definitions

Adverse Event (AE)
Any incidence harmful to health in a patient or clinical trial subject treated with a drug, even if it does not necessarily have a causal relationship with said treatment.
An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom or disease temporarily associated with the use of an investigational drug, whether or not it is related to the investigational drug.

Adverse Reaction (AR)
An AR is any unintended and harmful reaction to an investigational drug, regardless of the dose administered.
Unlike an AE, in the case of an adverse reaction there is a suspected causal relationship between the investigational drug and the adverse event.

10.2. Severity

The investigator will assess the severity of each AE using his/her clinical expertise and judgment based on the most appropriate description below:

1. Mild
   - The AE is a transient discomfort and does not interfere in a significant manner with the subject's normal functioning level
   - The AE resolves spontaneously or may require minimal therapeutic intervention
2. Moderate
   - The AE produces limited impairment of function and may require therapeutic intervention
   - The AE produces no sequela/sequelae
3. Severe
   - The AE results in a marked impairment of function and may lead to temporary inability to resume usual life pattern
   - The AE produces sequela/sequelae, which require (prolonged) therapeutic intervention

These severity definitions will also be used to assess the severity of an AE with a study-related procedure(s), if necessary.

Severity:
According to the severity of the process, AEs and RAs are classified as:

Severe: Any adverse event or adverse reaction that, at any dose: causes death, threatens the life of the subject, requires hospitalization of the patient or prolongs an existing hospitalization, causes permanent or significant disability or disability, or results in an abnormality or congenital malformation.
For the purposes of its notification, those suspected adverse events or adverse reactions that are considered important from a medical point of view, even if they do not meet the above criteria, would also be treated as serious, including important medical events that require intervention to prevent one of the consequences described above from occurring. Likewise, all suspicions of transmission of an infectious agent through a drug will be reported as severe.

Non-severe: that adverse event that does not meet the previous severity criteria.

Causality:
The causal relationship of an adverse event with the study medication will be established according to the following definitions:

Certain: A clinical event or laboratory test disturbance, which appears in a reasonable time sequence after drug administration and cannot be explained by concurrent disease or other drugs or chemicals. The response to drug discontinuation must be clinically plausible. The event should be pharmacologically or phenomenological definitive using, if necessary, an appropriate re-exposure procedure.

Likely: A clinical event or laboratory test disturbance, which appears in a reasonable time sequence after drug administration and which is unlikely to be explained by concurrent disease or other drugs or chemicals. The response to drug discontinuation is clinically plausible. Information on re-exposure is not required to meet this definition.

Possible: A clinical event or laboratory test disturbance, which appears in a reasonable time sequence after drug administration but which could also be explained by concurrent disease or other drugs or chemicals. Information about the outage may be missing or unclear.

Unlikely: A clinical event or laboratory test alteration, with a temporal relationship with the administration of the drug that makes a causal relationship unlikely (but not impossible). The underlying disease, other drugs, or chemicals provides plausible explanations.

Conditional/not classified: A clinical event or laboratory test disturbance, reported as an adverse event, for which more data is essential for a proper evaluation or additional data is being evaluated.

Not assessable/Not classifiable: Report that suggests an adverse event, which cannot be assessed due to insufficient or contradictory data, and which cannot be supplemented or verified.

In a simplified way, for the purposes of notification to regulatory authorities, the following definitions regarding causation will be adopted:

Related/Suspicious: The temporal relationship of an AE with the study medication indicates a possible causal relationship and cannot be explained by other factors such as the patient's clinical status, therapeutic interventions, or concomitant medication.

Unrelated/Not Suspicious: The temporal relationship of AE with study medication indicates an unlikely causal relationship, or other factors (medication or concomitant conditions), other therapeutic interventions provide a satisfactory explanation for AE.

Unexpected Adverse Reaction (UAR):

An Unexpected Adverse Reaction (UAR) is defined as any adverse reaction whose nature, intensity or consequences do not correspond to the reference information for the drug (eg, the investigator's manual in the case of an investigational drug not authorized for marketing , or the technical data sheet of the product in the case of an authorized medicine)

10.3. Safety Reporting

Any adverse event that occurs during a study, and that is voluntarily reported by the subject or observed by the investigator, must be recorded on the adverse events form included in the data collection notebook, regardless of the opinion of the researcher regarding its relationship to treatment. The investigator will determine the relationship between the adverse event and the drugs under study, and will record their conclusions in the corresponding section of the data collection notebook.

AE Collection and Assessment Methods

Every AE should, in principle, be documented in the section of the data collection notebook reserved for this purpose, and not as a comment collected anywhere in said data collection notebook. The following aspects will be collected:

the start;
the duration and, where applicable,
the completion of the AE;
a description of the AE;
Any factor considered as a possible causal agent of the AE;
Concomitant medication; and
An assessment of the relationship of intensity, severity, causality, and expected condition made by the researcher.

10.4. Notification

Any serious adverse event must be notified to the monitor and the promoter, by telephone or fax within 24 hours of the occurrence of the event. The investigator will complete the Adverse Events form of the CRF and Appendix D and will send it to the monitor and the promoter via fax and email within a period not exceeding 24 hours. This communication will be made within said period even if not all the information provided for in the form is available, which must be completed within 10 days. The form should include an assessment of the intensity, severity, causality, and the expected condition between the investigational drug and/or a concomitant treatment and the AE.

The reports to the sponsor on the evolution of the adverse event will continue until the event in question has disappeared or the clinical situation has stabilized. If necessary, any additional information will be provided.

The investigator is obliged to report each serious adverse event immediately, by phone or fax to:

Head of pharmacovigilance:

Claudia E. Delgado Espinoza
Research Institute of the Hospital de la Santa Creu i Sant Pau
C/ Sant Antoni Maria Claret, 167
08025 Barcelona
Phone: 93 553 76 34
Fax: 93 553 78 12
e-mail: cdelgadoe@santpau.cat

Research Monitor:

Pablo Bros/ Nàdia Llavero
Research Institute of the Hospital de la Santa Creu i Sant Pau
C/ Sant Antoni Maria Claret, 167
08025 Barcelona
Phone: 93 553 76 35
Fax: 93 553 78 12
e-mail: PBros@santpau.cat/ nllavero@santpau.cat

Notification will be carried out using the CRF Adverse Event Reporting Form and Appendix D.

The fact that a serious AE is considered related or not related to the investigational drug (s), and is considered expected or not expected, will be determined by the person in charge of pharmacovigilance of the UCICEC Sant Pau according to the reference documents, which for this study will be the technical data sheet of the study drug/investigator manual.

If it is a suspicion of RAGI, the promoter will notify the Spanish Agency for Medicines and Health Products, the competent body in matters of Pharmacovigilance of the Autonomous Community where it occurred, the respective Ethics Committee and the researchers involved in the trial. The maximum notification period is 7 days if the event is fatal or has threatened the life of the subject (any event that would have resulted in the subject's death without therapeutic intervention). Any other serious adverse event will be reported within 15 days.

Adverse events that are not serious or that are considered unrelated to the treatments under trial or that are considered expected should be reported in tabulated form in the final report of the clinical trial.

10.5. Specific aspects for safety assessment

To evaluate the safety of the drug, important vital signs will be taken: blood pressure and heart rate, under the measurements already established as normal for the age range.

a) Heart rate:
   Newborns (<1 month): 70 beats/min
   Infants 1-12 months: 80 beats/min
   Children (>12 m): 70 beats/min

b) Systolic pressure:
   It is difficult to establish the parameters for the classification of systolic hypotension in infants, as a general rule, one that is below the 5th percentile or 2 SD on normal auscultation is described:

   Newborn: 57 mmHg (5th percentile) or 64 mmHg (2 SD auscultation)
   6 months: 85 mmHg (5th percentile) or 65 mmHg (2 SD auscultation)
   1 year: 88 88 mmHg (5th percentile) or 66 mmHg (2 SD auscultation)

To obtain an accurate measurement, the patient must be in a warm room at rest, asleep, or awake. A special cuff of adequate size must be used, a minimum covering 75% of the limb and a length that occupies 2/3 of the length of the upper limb.

These measurements will be carried out at baseline, before starting the treatment, and one hour after the first application, and then they will be carried out in all the control appointments, at week 2, 4, 8, 12 and 24.

At each appointment, data will be taken in case there are adverse effects due to the use of the drug, likewise, if they present intercurrent diseases and the treatment they administered, as well as vaccination data and if they presented any systemic reaction and will be documented in each clinical history of the patients. In the event of an adverse event that leads to the suspension of treatment, the parents will be given a contact telephone number to report it, both with the principal investigator or co-investigators and the date and time of the report will be documented. A detailed assessment will be carried out to determine if the adverse event is

related to the drug under study, in case known adverse events of the drug are met, according to the technical sheet attached.

To date, no life-threatening adverse events have been reported from topical administration of this drug. However, in the event of any adverse event, the patient will be asked to suspend the drug and then a visit will be given to determine the severity of the event or if it can be reintroduced in a short period of time.

Regarding the distribution of treatment by an external pharmaceutical agent, errors could be made with respect to the distribution of the drug or control; in the event of a mistake, the event should be reported urgently to the study promoter.

Most of the published adverse effects are minimal, and can be resolved quickly by the investigator. In the event that a patient is excluded from the study, the sponsor should be notified of the cause of said adverse event.

Despite the fact that this is a double-blind clinical trial, at no time in the study will opening of the blinding be required, since the underlying pathology does not carry any risk to the life of the patient, in the event of extensive growth of the significant lesion, ulceration, or disfigurement, the patient will be given the option to switch to systemic propranolol therapy.

This clinical trial does not include the inclusion of diseases with great morbidity or mortality, nor the risk of pregnancy or congenital defects. It will be carried out in healthy infants, without any other associated pathology. The drug does not carry any added medical risk.

It is difficult for errors to occur due to overdose or administration of the drug used. Parents will be explained in detail about the topical application of the drug, that it is not used near mucous membranes so as not to increase the absorption of the drug, likewise, they will have a telephone number and close contact with the researcher to report any abnormality.

11. Ethical aspects

11.1. General considerations

The study will be carried out in strict accordance with international ethical recommendations for research and clinical trials in humans. The researcher will be responsible for ensuring that the clinical trial is carried out in accordance with the standards set out in the Declaration of Helsinki and following the recommendations of the Spanish Ministry of Health regarding clinical trials.

Before including any subject in the study, the Ethics Committee of the participating centers and the Spanish Agency for Medicines and Health Products must approve the study protocol, the information that will be given to the subject and the informed consent model that will be used.

The study must be carried out in accordance with the protocol and the Standard Work Procedures (SWPs) that ensure compliance with the Good Clinical Practice (GCP) standards, as described in the ICH Tripartite Harmonized Standards for Good Clinical Practice nineteen ninety six.

The CEIC must be informed of any subsequent amendment to the protocol and its opinion must be requested in the event that a new evaluation of the ethical aspects of the trial is necessary.

11.2. Patient information sheet and consent

It is the responsibility of the researcher to obtain the informed consent of the parents or legal guardian. The patient cannot participate in any specific study procedure before obtaining such consent.
Before the start of the trial, and before obtaining informed consent, the researcher or the person designated by the same, will explain to the legal guardian/parents of the potential participant, the objectives, methods and potential risks of the study and any discomfort that this may cause.
The explanation about the nature, scope and possible consequences of the study will be made in understandable language.
The legal guardian/parents should have time to reflect on their participation in the study, and have the opportunity to ask questions. After this explanation, and before entering the trial, consent must be properly recorded by the signature of the legal guardian/family member.
The information provided must include:

- An explanation that the test involves an experimental procedure.
- An explanation of the purpose of the test.
- A description of the drug to be studied and random assignment. A statement that the treatment may not be the study drug (eg in cases where placebo or another comparator drug is given in a randomized trial).
- Description of the procedures to follow, including invasive ones. Duration of the subject's participation. Approximate number of subjects who will participate in the trial.
- Responsibilities of the subject
- Reasonably foreseeable risks and discomforts for the subject (if applicable for the embryo or fetus) and planned remedial measures.
- Description of the benefits for the subject/society.
- Availability of alternative treatments with their potential risks and benefits.
- Compensation to subjects: coverage of risks, medical treatment of possible damages, financial compensation.
- Knowledge of any additional cost for the subject that may arise from their participation in the research.

- Conditions of participation: Consent expressed according to their free will, right to abandon the trial at any time, right to refuse to participate without prejudice to the subject.
- Explanation that the identity of the subject is confidential but that the histories can be reviewed by the trial monitor, the auditors and can be made known to the health authorities.
- Statement that new relevant findings will be made available to the subject.
- Identification of whom and to which service they can go to obtain answers regarding any aspect of the trial or the rights of the subject (name and telephone number).
- Description of the circumstances in which the investigator may discontinue a subject's participation in a trial.
- In the case that the subject is female, if deemed necessary, the sponsor will provide additional information to prevent the possibility of pregnancy during the process of selection, development and follow-up of a trial.

As an Appendix, the Model Information Sheet is presented to the Legal Guardian/Parents and the Consent Form.

11.3. Assessment of foreseeable benefits and risks for trial subjects and other potential patients.

With regard to the evaluation of the benefits in this study, a dermatological evaluation of the patients will be maintained in a close manner; those patients who experience an exaggerated growth of the lesion, ulceration or disfigurement, will be given the option of systemic management with propranolol continuing close monitoring, probably before the risk of sequelae has been reached.
Regarding the foreseeable risks, to date no serious side effects or those that put the patient at risk have been reported. The effects that may occur are secondary to an increase in systemic absorption, hypotension, hypoglycemia, bradycardia, sleep disturbances, which will be evaluated at each visit to avoid risk, parents will be instructed about possible side effects and what they can do to prevent them or be in close contact with them if you have any questions.
Patients may not benefit from participating in this study, however the results of the study will allow us to establish whether timolol is effective in involution of early proliferative phase infantile hemangioma.

11.4. Data Confidentiality

To preserve the confidentiality of the subjects' personal data, only the principal investigator, his collaborators, and the technical personnel participating in the study will have access to their identity. For the same reason, the complete affiliation data and the written consent will be kept in the file of the center researcher.
Regarding the confidentiality of the study data, the provisions of Organic Law 15/1999 of December 13, on "Protection of Personal Data" will be followed. In accordance with Law 15/1999 on the Protection of Personal Data, the personal data required from patients are those

necessary to meet the objectives of this study. Your name will not appear in any of the study reports and your identity will not be revealed to anyone except to fulfill the purposes of the study, and in the case of medical emergency or legal requirement. Any personal information that may be identifiable will be kept and processed by computerized means under secure conditions by the study researchers. Access to such information will be restricted and will always be done under conditions of confidentiality. The results of the study may be communicated to the health authorities and, eventually, to the scientific community, without the identity of the participating subjects being recorded in any case.
In accordance with current law, the subject participating in the study has the right to access their personal data and, if justified, has the right to request its rectification or cancellation.
Information regarding the identity of patients will be considered confidential for all purposes. The identity of the patients cannot be revealed or disclosed. The data of the patients collected in the Case Report Form during the study must be documented anonymously and dissociated, linking to a code (patient number), so that only the researcher can associate such data with an identified person or identifiable.

The database generated by the study will not contain any identification of the patient, other than a numerical code by which it will not be possible to reveal their identity. The information collected in the study will always be treated as grouped data and never as individual or personal data, thus maintaining anonymity and confidentiality.

11.5. Insurance policy

In accordance with Royal Decree 223/2004 of February 6, which regulates clinical trials with drugs, the promoter has signed an insurance policy to insure against possible damages arising from the research.
Studies or experiments carried out in this study on behalf of the sponsor are specifically and expressly guaranteed.
It is advisable to point out that failure to comply with the legal conditions of investigation is a reason for exclusion of the guarantee.

12. Practical Considerations

12.1. Responsibility of Participants in the Trial

Researchers
The principal investigator will be responsible for carrying out the trial in accordance with the regulations for clinical trials in force in Spain (Drug Law 25/1990 and Royal Decree 223/2004), being solely responsible for the execution of the trial. The principal investigator and his collaborators undertake to practice each and every one of the examinations and complementary tests that are specified in the clinical judgment criteria of the protocol on all subjects included in the trial.

The auxiliary personnel collaborating in the study must be informed by the principal investigator of their responsibilities towards the subject.

The Hospital de la Santa Creu i Sant Pau Pharmacy Service will be in charge of preparing, packaging and distributing the medication to the patients participating in this study.

Auxiliary staff responsibilities
The auxiliary personnel collaborating in the study will comply with the general rules established for conducting the trial and will follow the instructions of the investigator at all times.

12.2. Data File Conditions and Corrections

The data obtained must be transcribed in the Case Report Form (CRF) and these data will be considered valid information for the subsequent evaluation of the efficacy and safety data of the treatments under study. The Logs must be completed correctly and in the event that data already transcribed must be corrected, these will be crossed out, noting the correct value alongside. Corrections must always be dated and validated by the signature of the principal investigator or his collaborators.

The documents corresponding to this trial will be kept with the investigator for up to five years after the end of the clinical trial. In any case, an identification list of the participants will always be kept. The promoter, the Institut de Recerca, will maintain a main file of the study for a period of five years.

12.3. Monitoring, Audits and Inspections

A) Monitoring
The study is expected to be monitored by the monitor designated by the sponsor.
The monitoring will include visits to the center and telephone communication with the research team in order to ensure correct compliance with the protocol, the GCP and the regulations of the health authorities.
During the monitoring visits, the most relevant aspects of the study will be reviewed, such as the procedure followed to obtain informed consent, a check of the documentation in the investigator's file, and a percentage of the data registered in the CRF (these data must be correctly completed and be truthful with the source documents), inclusion/exclusion criteria and adverse events that have occurred to date.
The findings of the monitoring will be reported to the promoter or to the delegated structure so that the appropriate measures can be taken, through the reports of the monitoring visits. A follow-up letter will be sent to the investigator, where the most significant findings and any pending issues will be reported.
At the end of the study, a report will be prepared with the global results regarding the quality and reliability of the data, as well as adherence to the protocol and to the observed good clinical practice procedures.
At the end of the study, the data collection logs will be sent to the Study Sponsor or to the corresponding delegated structure for filing.

B) Audits
The clinical trial will be included in the IR-HSCSP's Quality Assurance Program, in relation to Clinical Research, analyzing in each case the criticality of the trial and the audits to be performed.

C) Inspections
Both the Researcher and the Sponsor will allow direct access to the data or source documents for the performance of the monitoring, the audit, the review by the CEIC, as well as the inspection of the trial by the health authorities in case they are required.

12.4. Corrections to the Clinical Trial Protocol

Any change made to the study protocol will always take the form of a written amendment or addendum. For its formalization, the approval of all the people responsible for the study who also signed the protocol is required. And if they are relevant modifications, the express approval of the Ethics Committee and the health authorities is necessary.

12.5. Deviations from the Study Protocol

Deviations from the study protocol are not allowed, especially regarding the prescription of drugs or doses not programmed in the study, as well as other modes of administration, other indications, or longer treatment periods.

Major deviation is understood to be those that involve changes that may interfere with the results and conclusions of the study.

12.6. Drug Control

The entry record of the samples received, as well as the inventory of their use, must be kept by the hospital pharmacy, observing the following rules at all times:

a) the researcher should keep the study medication in the hospital pharmacy, accessible only to the persons authorized to administer said drugs.

b) The inventory of the hospital pharmacist. The inventory will include details about the material received, and clear indications about when it was administered and on which individual it was used. This record will also indicate the amount and type of material that is available at any time during the trial, throughout the duration of the trial.

c) The pharmacist will carry out an inventory of the surplus material at the end of the clinical trial, and will record the results on the Drug Accounting Form. The hospital pharmacist will return any excess material and its packaging to the study promoter, either empty or still containing some study medication.

c) The investigator agrees that he will not provide study drugs to anyone, with the exception of his research collaborators and patients under study.

12.7. Identification and Labeling of Samples

The different types of labels designed for this phase of the study will be printed using the ETIK program, before starting the final packaging.
The person responsible for preparing the medication will have the randomization list. And each day the number of treatments to be used that day will be prepared according to the randomization list. The medication will be labeled and identified according to the randomization sequence.

Code
xxxxxxxx/xxxxxxxxx
Treatment nºXXX
Lot : XXX
Expiration: XX/XX/XX (refrigerator)
Exclusive sample for clinical trials

12.8. Assignment of study treatments

The randomization list will be generated by the Methodological and Statistical Support Unit of the Institut de Recerca de l'HSCSP. The study sponsor will not have access to this list. The randomization list will be generated in such a way that both treatments have an equal probability of being assigned.
The patients will be included in the study consecutively from the inclusion of the first eligible patient according to the selection criteria. When a patient is included in the study, the researcher will assign a patient code, this code must be consecutive with respect to the previous assigned code, and taking as a reference the date and time when the intervention is planned.
The randomization codes will be assigned in a correlative way, as the patients are included in the study.
In the study SOP, the HSCSP pharmacy service has included the conditioning procedure for the study medication.
The center is recommended to keep a register of both included and non-included patients in order to ensure that no bias is introduced into the selection process. This registry will also provide information on the reasons for not including patients who have undergone intervention.
If the randomized treatment is discontinued, the investigator may prescribe any other treatment based on his clinical judgment. The patient will no longer be able to contribute to the study drug exposure phase for the main analysis, but the patient will remain in the follow-up phase.

12.9. Trial Interruption

The clinical trial can be interrupted by the principal investigator and/or by the sponsor in any of the following cases:

- Inefficacy of the studied treatment.
- Appearance of adverse events unknown to date, as well as known adverse events whose nature, severity, duration or incidence are not as expected.
- Insufficient number of patients included in the study.

12.10. Publishing conditions

The results obtained as a consequence of the clinical research with the product under study will be reviewed and discussed by the research team and the sponsor for subsequent publication.

The data obtained will not be disclosed to third parties until an agreement is reached with the promoter for its disclosure, either in the form of a conference, communication to congress, or publication. As an exception to the previous paragraph, researchers may include the title of the essay in their respective Curriculum Vitae, as long as it does not include any information about the promoter.

12.11. Preparation of the Final Report

After obtaining the conclusions of the study, a final report will be prepared in collaboration with the promoter. Said report will include the statistical analysis and a medical evaluation of the results. This report will be based on the objectives stated in the study protocol.

12.12. Funding

This study will be financed with the funds from the Dermatology service deposited in the Research Institute.

13. Bibliography

1. Dickison P, Christou E, Wargon O. A prospective study of infantile hemangiomas with a focus on incidence and risk factors. Pediatr Dermatol 2011; 28(6):663-669
2. Tollefson MM, Frieden IJ. Early growth of infantile hemangiomas: what parents' photographs tell us. Pediatrics. 2012 Aug;130(2):e314-20
3. Chang LC, Haggstrom AN, Drolet BA, Baselga E, Chamlin SL, Garzon MC, Horii KA, Lucky AW, Mancini AJ, Metry DW, Nopper AJ, Frieden IJ; Hemangioma Investigator Group. Growth characteristics of infantile hemangiomas: implications for management. Pediatrics. 2008 Aug;122(2):360-7.
4. Drolet BA, Frommelt PC, Chamlin SL, Haggstrom A, Bauman NM, Chiu YE, Chun RH, Garzon MC, Holland KE, Liberman L, MacLellan-Tobert S, Mancini AJ, Metry D, Puttgen KB, Seefeldt M, Sidbury R, Ward KM, Blei F, Baselga E, Cassidy L, Darrow DH, Joachim S, Kwon EK, Martin K, Perkins J, Siegel DH, Boucek RJ, Frieden IJ. Initiation and use of propranolol for infantile hemangioma: report of a consensus conference. Pediatrics. 2013 Jan;131(1):128-40.
5. Marqueling AL, Oza V, Frieden IJ, Puttgen KB. Propranolol and infantile hemangiomas four years later: a systematic review. Pediatr Dermatol. 2013 Mar-Apr;30(2):182-91.
6. Guo S, Ni N.Topical treatment for capillary hemangioma of the eyelid using betablocker solution. Arch Ophthalmol. 2010;128(2):255–256
7. Chan H, McKay C, Adams S, Wargon O. RCT of timolol maleate gel for superficial infantile hemangiomas in 5- to 24-week-olds. Pediatrics. 2013 Jun;131(6):e1739-47.
8. Yu L, Li S, Su B, Liu Z, Fang J, Zhu L, Huang M, Shan W, Song D, Ye B, Luo C. Treatment of superficial infantile hemangiomas with timolol: Evaluation of short-term efficacy and safety in infants. Exp Ther Med. 2013 Aug;6(2):388-390.
9. Semkova K, Kazandjieva J. Rapid complete regression of an early infantile hemangioma with topical timolol gel. Int J Dermatol. 2013 Nov 21.
10. Thomas J, Kumar P, Kumar DD. Ulcerated infantile haemangioma of buttock successfully treated with topical timolol. J Cutan Aesthet Surg. 2013 Jul;6(3):168-9.
11. Yu L, Li S, Su B, Liu Z, Fang J, Zhu L, Huang M, Shan W, Song D, Ye B, Luo C. Treatment of superficial infantile hemangiomas with timolol: Evaluation of short-term efficacy and safety in infants.Exp Ther Med. 2013 Aug;6(2):388-390
12. Rizvi SA, Yusuf F, Sharma R, Rizvi SW. Managment of Superficial Infantile Capillary Hemangiomas with Topical Timolol Maleate Solution. Semin Ophthalmol. 2013 Sep 27.
13. Chan H, McKay C, Adams S, Wargon O.Related citations RCT of timolol maleate gel for superficial infantile hemangiomas in 5- to 24-week-olds. Pediatrics. 2013 Jun;131(6):e1739-47.
14. Berk DR, Lehman PA, Franz TJ, Blunt JR, Bayliss SJ. On topical timolol gel-forming solution for infantile hemangiomas.Pediatr Dermatol. 2013 Jan-Feb;30(1):160-1.
15. Chantasart D, Hao J, Li SK. Evaluation of skin permeation of β-blockers for topical drug delivery. Pharm Res. 2013 Mar;30(3):866-77.
16. Xue K, Hildebrand GD.Topical timolol maleate 0.5% for infantile capillary haemangioma of the eyelid. Br J Ophthalmol. 2012 Dec;96(12):1536-7.
17. Ye XX, Jin YB, Lin XX, Ma G, Chen XD, Qiu YJ, Chen H, Hu XJ. Topical timolol in

the treatment of periocular superficial infantile hemangiomas: a prospective study]. Zhonghua Zheng Xing Wai Ke Za Zhi. 2012 May;28(3):161-4. Chinese.
18. Semkova K, Kazandjieva J.Topical timolol maleate for treatment of infantile haemangiomas: preliminary results of a prospective study. Clin Exp Dermatol. 2013 Mar;38(2):143-6.
19. Jha AK, Mallik SK, Raihan M. Topical ophthalmic solution in infantile hemangioma. J Postgrad Med. 2012 Apr-Jun;58(2):163-5.
20. Moehrle M, Léauté-Labrèze C, Schmidt V, Röcken M, Poets CF, Goelz R. Topical timolol for small hemangiomas of infancy. Pediatr Dermatol. 2013 Mar-Apr;30(2):245-9.
21. Calvo M, Garcia-Millán C, Villegas C, Fueyo-Casado A, Burón I. Topical timolol for infantile hemangioma of the eyelid. Int J Dermatol. 2013 May;52(5):603-4.
22. Chambers CB, Katowitz WR, Katowitz JA, Binenbaum G. A controlled study of topical 0.25% timolol maleate gel for the treatment of cutaneous infantile capillary hemangiomas. Ophthal Plast Reconstr Surg. 2012 Mar-Apr;28(2):103-6.
23. McMahon P, Oza V, Frieden IJ. Topical timolol for infantile hemangiomas: putting a note of caution in "cautiously optimistic". Pediatr Dermatol. 2012 Jan-Feb;29(1):127-30.
24. Cante V, Pham-Ledard A, Imbert E, Ezzedine K, Léauté-Labrèze C. First report of topical timolol treatment in primarily ulcerated perineal haemangioma. Arch Dis Child Fetal Neonatal Ed. 2012 Mar;97(2):F155-6.
25. Oranje AP, Janmohamed SR, Madern GC, de Laat PC. Treatment of small superficial haemangioma with timolol 0.5% ophthalmic solution: a series of 20 cases. Dermatology. 2011;223(4):330-4. doi: 10.1159/000334778. Epub 2011 Dec 16.
26. Chakkittakandiyil A, Phillips R, Frieden IJ, Siegfried E, Lara-Corrales I, Lam J, Bergmann J, Bekhor P, Poorsattar S, Pope E. Timolol maleate 0.5% or 0.1% gel-forming solution for infantile hemangiomas: a retrospective, multicenter, cohort study. Pediatr Dermatol. 2012 Jan-Feb;29(1):28-31.
27. Khunger N, Pahwa M. Dramatic response to topical timolol lotion of a large hemifacial infantile haemangioma associated with PHACE syndrome. Br J Dermatol. 2011 Apr;164(4):886-8.
28. Pope E, Chakkittakandiyil A. Topical timolol gel for infantile hemangiomas: a pilot study. Arch Dermatol. 2010 May;146(5):564-5.
29. Guo S, Ni N. Topical treatment for capillary hemangioma of the eyelid using beta-blocker solution. Arch Ophthalmol. 2010 Feb;128(2):255-6.