



Australian Government

Department of Health and Ageing
Therapeutic Goods Administration

Australian Public Assessment Report for Ingenol Mebutate

Proprietary Product Name: Picato 0.015% and
0.05% gel

Sponsor: LEO Pharma Pty Ltd

June 2013

TGA Health Safety
Regulation

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- An AusPAR is a static document, in that it will provide information that relates to a submission at a particular point in time.
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I. Introduction to product submission

Submission details

<i>Type of Submission:</i>	New Chemical Entity
<i>Decision:</i>	Approved
<i>Date of Decision:</i>	1 November 2012
<i>Active ingredient:</i>	Ingenol Mebutate
<i>Product Names:</i>	Picato 0.015% and 0.05% gel
<i>Sponsor's Name and Address:</i>	LEO Pharma Pty Ltd 25 Montpelier Rd, Bowen Hills QLD 4006
<i>Dose form:</i>	Gel
<i>Strengths:</i>	0.015% and 0.05% w/w
<i>Container:</i>	Single use tubes
<i>Pack sizes:</i>	3s (0.015%) and 2s (0.05%)
<i>Approved Therapeutic use:</i>	Picato gel is indicated for the topical treatment of solar (actinic) keratoses in adults.
<i>Route of administration:</i>	Topical
<i>Dosage:</i>	<i>Face and scalp:</i> Picato gel 0.015% should be applied to the affected area once daily for 3 consecutive days. <i>Body:</i> Picato gel 0.05% should be applied to the affected area once daily for 2 consecutive days.
<i>ARTG Numbers:</i>	190122 and 190113

Product background

LEO Pharma Pty Ltd has submitted a New Chemical Entity application for the registration of Picato gel, with the active ingredient ingenol mebutate, as a therapy for solar (actinic) keratoses. Solar keratosis is a cutaneous neoplasm, exhibiting as focal areas of epithelial dysplasia that occurs primarily on sun exposed skin areas. Because these lesions are unsightly and can progress to squamous cell carcinoma they should be treated. There are currently various forms of treatment available including surgical excision, radiotherapy, cryosurgery, chemotherapy (for example cream containing 5-fluorouracil) and photodynamic therapy.

Ingenol mebutate is extracted from *Euphorbia peplus* and is formulated in a gel base. According to the sponsor, "ingenol mebutate induces necrosis, resulting in debulking of locally affected tumour cells. Secondly, it induces a tumour specific immune response

characterised by antibody dependent cellular cytotoxicity (ADCC) which results in removal of residual disease. It also stimulates cluster of differentiation (CD)8+ T cells and CD4+ T cell responses, resulting in further anti-tumour activity and possible long term immunity.”

Ingenol mebutate is also referred to as PEP005 in this AusPAR.

Regulatory status

Picato gel is registered in USA and has the following indication: *The topical treatment of actinic keratoses.*

A summary of the current international regulatory status for Picato (ingenol mebutate) gel is presented in Table 1 below.

Table 1. Summary of International Regulatory Status

Country	Submitted	Status
USA	25 March 2011	Approved 23 January 2012
Europe	29 July 2011	Approved 15 November 2012
Brazil	10 November 2011	Approved 16 July 2012
Canada	08 February 2012	Approved 30 January 2013
Switzerland	14 March 2012	Evaluation ongoing
New Zealand	23 May 2012	Evaluation ongoing

The indication in all jurisdictions is for the topical treatment of solar (actinic) keratoses.

Product Information

The approved Product Information (PI) current at the time this AusPAR was prepared can be found as Attachment 1.

Abbreviations used in this AusPAR

Abbreviation	Definition
5-FU	5-fluorouracil
ADCC	antibody-dependent cell-mediated cytotoxicity
ADP	adenosine diphosphate
AK	Actinic Keratosis
AML	Acute myeloid leukemia
BCC	Basal cell carcinoma
CTL	cytotoxic T lymphocytes
CYP	cytochrome P450
DTT	dithiothreitol
EGFP	enhanced green fluorescent protein
ECM	extracellular matrix
EM	electron microscopy
HPLC	high-pressure liquid chromatography
LC	liquid chromatography
LLOQ	lower limit of quantification
MMP	matrix metalloproteinase
MS/MS	tandem mass spectrometry method
MTT	3-(4,5-Dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide
NK	natural killer cell
PBMC	peripheral blood mononuclear cell
PEP003	a mixture of three ingenol esters (3-angelyl ingenol, 3-angelyl 20-deoxyingenol, 3-angelyl-20-acetyl ingenol)
PEP005a	5-mebutate ingenol (isomer of PEP005b)
PEP005c	20-mebutate ingenol (isomer of PEP005b)
PEP006	3-angelyl 20-deoxyingenol
PEP008	3-angelyl-20-acetyl ingenol
PEP015	PEP005a (see above)

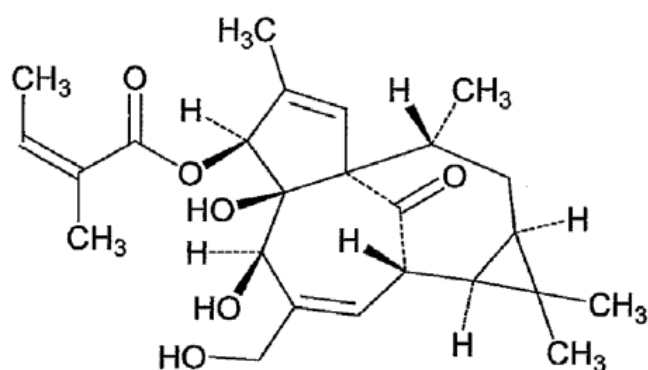
Abbreviation	Definition
PEP025	PEP005c (see above)
PHA	phytohemagglutinin
PI	propidium iodide
PI3K	phosphatidylinositol 3-kinase
PKC	protein kinase C
PMA	phorbol myristate acetate, tetradecanoylphorbol acetate
SCC	squamous cell carcinoma
SRB	sulforhodamine B

II. Quality findings

Drug substance (active ingredient)

Ingenol mebutate is a naturally occurring substance with an unknown mechanism that leads to local lesion cell death and promotes an inflammatory response characterised by infiltration of neutrophils and other immunocompetent cells. It is an ester which is active in itself and not a prodrug. The free alcohol (which is the product of hydrolysis) is only present in skin at levels of 1% that of ingenol mebutate.

Figure 1. Chemical structure of Ingenol Mebutate



Ingenol mebutate

C₂₅H₃₄O₆ MW = 430.53; CAS # = [75567-37-2]. Practically insoluble in water {<0.1 mg/mL}.

According to the Product Information document (PI) the gels are provided in single use tubes which come in packs of 3 tubes (0.015% strength) and 2 tubes (0.05% strength) to be used on 3 and 2 consecutive days, respectively. The amount in each tube is nominally 0.47 g (maximum 0.51 g) which is enough to cover an area of 25 cm² (5 cm x 5 cm). The

maximum daily dose is thus 76.5 or 255 µg of ingenol mebutate depending on the strength.¹

Drug product

The ingenol mebutate is isolated from the aerial parts of *Euphorbia peplus*. All aspects of the cultivation of the plants are controlled including that the only fungicide to be used is 'mancozeb'. The manufacturing process involves, drying and milling, extraction using a polymeric resin, desorption, chromatography, preparative high-performance liquid chromatography (HPLC) and crystallisation.

The specifications of the ingenol mebutate were generally acceptable.

The gels are different concentrations of ingenol mebutate made up in the same vehicle.

The manufacture is typical for gels.

The microbiological aspects of the manufacture were acceptable to the Microbiological evaluator of the TGA's Office of Laboratories and Scientific Services (OLSS).

The control of the gels was considered acceptable.

Stability data was provided that supported a shelf life of 2 years when stored in the refrigerator at 2-8°C. Freezing did not affect the product and the additional storage condition of 'Do not freeze' is not required, though it was stated on the carton labels.²

The chemistry and quality control aspects of the draft PI have been finalised to the satisfaction of the quality evaluator. As have the carton and tube labels and the Provisional ARTG Records³.

Biopharmaceutics

The products are for topical use. The quality evaluator was not required to evaluate any bioavailability data.

The draft PI included the statement "*the pharmacokinetic profile of ingenol mebutate and its metabolites has not been characterised in humans due to the absence of quantifiable blood levels following topical administration*" and this was accepted by the quality evaluator without evaluation. However, it is known that sunscreens may increase the bioavailability of drug substances and this was not investigated. It was suggested to the Delegate that it might be appropriate to include a statement to this effect in the PI.

Advisory committee considerations

This application was presented to the 144th meeting of the Pharmaceutical Subcommittee (PSC) of Advisory Committee on Prescription Medicines (ACPM) in March 2012. The PSC had no objections to approval of the submission provided all outstanding issues were addressed to the satisfaction of the TGA. The PSC indicated that it did not require to review this submission again. In particular the Committee agreed that it was difficult to unequivocally conclude that the proposed products were optimally formulated (based on the absence of biopharmaceutical data).

¹ Sponsor comment: "The maximum daily dose of Picato Gel is 0.25g/day which is extracted from a tube nominally containing 0.47g. For 0.05% strength, this gives a maximum daily dose of 125 microgram/day. For 0.015% strength, this gives a maximum daily dose of 37.5 microgram/day."

² Sponsor comment: "'Do not freeze' was removed from the carton labels prior to approval."

³ The GMP Clearances are valid until at least 11 March 2013.

Quality summary and conclusions

The quality evaluator had no objections to the approval of this submission with respect to chemistry and manufacturing control other than the lack of a test and limit for free ingenol in the drug substance. This issue, it was suggested should be resolved prior to presentation at ACPM.⁴

The quality evaluator did not evaluate any bioavailability data.

III. Nonclinical findings

Introduction

The nonclinical data submitted by the sponsor in support of the safety and efficacy of ingenol mebutate were generally of good quality and were performed by reputable laboratories. All pivotal, nonclinical safety studies were performed according to Good Laboratory Practice (GLP) standards.

Each treatment requires the patient to evenly spread the entire contents of one tube of Picato gel over an area of 25 cm² (5 cm × 5 cm) encompassing the lesion, allowing the area to dry and then leaving the area undisturbed for 6 h. A standard treatment area of 25 cm² is used even if the lesion is small. Hence, each treatment on the face or scalp uses approximately 70 µg of ingenol mebutate on an area of 25 cm² (that is, 0.028 µg/mm²) and each treatment on the trunk or extremities uses approximately 235 µg of ingenol mebutate on 25 cm² (that is, 0.094 µg/mm²).

Pharmacology

Primary pharmacology

An extensive range of studies were performed aimed at demonstrating the ability of ingenol mebutate to kill cells and defining the mechanism by which cell death is induced. These studies were mostly performed with a great variety of human (and some rodent) tumour-derived cell lines along with some studies using normal cells. It should be noted that there appear to be no practical animal models for solar keratosis. The lack of testing with cells that are presumably intermediate to normal and cells that are fully transformed seems unlikely to impact on the conclusions drawn from these studies (that is, the responses shown by solar keratosis cells to treatment with ingenol mebutate would presumably also be shown by normal and/or tumour-derived cells).

Ingenol mebutate is extracted and purified from cultivated crops of the plant *Euphorbia peplus*. Sap from *Euphorbia peplus* and related species has a long history of use as a folk remedy for skin ailments and other diseases⁵. The studies submitted by the sponsor demonstrated that ingenol mebutate is the major active component in the *Euphorbia peplus* sap and is approximately 10 fold more potent than crude sap at inhibiting the proliferation of human tumour-derived cell lines under *in vitro* conditions. Ingenol mebutate showed growth inhibiting activity of human tumour-derived cell lines of various

⁴ Sponsor comment: "A test and limit for free ingenol in the drug substance was included in the drug substance specification and was submitted to TGA during the marketing authorisation assessment procedure on 16 August 2012."

⁵ Adolf W., Chanai S. and Hecker E. (1983) 3-O-Angeloylingenol, the toxic and skin irritant factor from latex of *Euphorbia anitiquorum* L. (Euphorbiaceae) and from a derived Thai purgative and anthelmintic (vermifuge) drug. *Journal of the Science Society of Thailand*, 9: 81-88.

origins, including squamous cell carcinomas and melanomas. A feature of these results was the marked differences in responsiveness to ingenol mebutate treatment shown by different tumour lines (for example, there could be a 100 fold difference in the ingenol mebutate concentration required to produce a certain level of growth inhibition between melanoma lines). This finding raises the possibility of inter- and intra-individual differences in responsiveness of solar keratoses to ingenol mebutate. It is, however, not possible (based on the data provided by the sponsor) to state whether the therapeutic dosing regimen could eliminate all lesions encompassing such a range of intrinsic drug sensitivities. The sponsor's data were inconsistent regarding ingenol mebutate uptake following topical application, for example, an *in vivo* study indicated rapid uptake/metabolism by mouse skin whereas *in vitro* studies using dermatome specimens suggested that uptake was slow and variable between species.

In vitro studies involving normal human skin cell types indicated toxicity by ingenol mebutate towards fibroblasts and keratinocytes. Fibroblasts generally showed comparable responses to the most ingenol-resistant melanoma cell lines. At non-toxic concentrations ingenol mebutate could stimulate α -smooth muscle actin expression and induce differentiation of fibroblasts to myofibroblasts suggesting that the drug has pro-fibrotic activity.

In vivo studies examined the ability of topical applications of ingenol mebutate-containing gel or intra-lesional injections of ingenol mebutate to treat various murine SCC and melanoma tumours growing subcutaneously in mice. Three daily topical applications of approximately 18 μ g of ingenol mebutate (comparable with the suggested clinical dose per unit area) were shown to eliminate or induce marked growth delay of B16 mouse melanomas and of LK2 mouse squamous cell carcinomas at approximately 1-2 weeks post dosing. Some animals showed tumour recurrence even after the most effective dosing regimens. Other tumour types showed only partial regression following ingenol mebutate treatment. The results of some of the studies have been reported in the literature⁶. No studies were conducted to examine efficacy against human SCC or melanoma tumour xenografts.⁷

Topical treatment of tumours with ingenol mebutate produced an erythematous response that was associated with a prolonged infiltration of inflammatory cells, mostly neutrophils. *In vitro* studies showed that ingenol mebutate induced respiratory burst⁸ and phagocytosis in PBMCs and also induced the production of neutrophil-attracting cytokines such as interleukin isoforms (IL)-1 β , IL-6, IL-8 and Tumour necrosis factor (TNF)- α . Studies using systems in which neutrophil activity was compromised, for example by depletion of circulating neutrophils following anti-Ly-6G antibody administration, showed a decreased anti-tumoural efficacy of topical ingenol mebutate administration suggesting that neutrophils may have a significant role in eliminating residual tumour cells that escaped the direct cytotoxic activity of ingenol mebutate. Further such studies suggested that NK cells and macrophages did not have a significant role in the anti-tumour activity of ingenol mebutate. Some evidence was also presented to indicate that ingenol mebutate treatment can induce the production of tumour-specific antibodies capable of binding to residual tumour cells and potentially initiating immune cell-mediated cytotoxicity.

⁶ Ogbourne S.M., Suhrbier A., Jones B. *et al.* (2004) Antitumor activity of 3-ingenyl angelate: plasma membrane and mitochondrial disruption and necrotic cell death. *Cancer Research*, **64**: 2833–2839.

⁷ Sponsor comment: "Ogbourne 2004 - Table 2 lists human xenograft cell lines that were cured with PEP005; prostate, melanoma and cervical cancers."

⁸ Respiratory burst (sometimes called oxidative burst) the marked increase in metabolic activity that occurs in phagocytes and certain other cells following binding of particles resulting in an increase in oxygen consumption, formation of superoxide anion, formation of hydrogen peroxide and activation of the hexose monophosphate shunt.

The mode of cell death induced by ingenol mebutate exposure of various tumour-derived cell lines was examined *in vitro* using standard techniques (electron microscopic observation, annexin V and propidium iodide staining, DNA laddering). Studies with some cell lines (such as B16 and LK2) showed evidence that ingenol mebutate disrupts plasma membrane permeability resulting in calcium influx leading to mitochondrial swelling, membrane potential collapse and necrosis. However, studies with other cell lines showed evidence for induction of apoptosis by ingenol mebutate. Cells in S phase appeared to be the most sensitive to ingenol mebutate treatment with flow cytometric analysis which demonstrated a progressive loss of S phase cells with treatment time and an accumulation of cells in G1 and G2/M phases of the cell cycle.⁹

The activation of cellular signalling pathways and the induction of gene expression following ingenol mebutate exposure were examined in various cell lines using standard approaches. Consistent with the chemical relationship to the phorbol esters, ingenol mebutate was a strong activator of protein kinase C (PKC) and induced the translocation of classical PKC isoforms (α , β , and γ) to the plasma membrane in HeLa cells (an event typically associated with PKC activation). However, a novel PKC isoform (θ) and an atypical PKC isoform (ζ) did not show ingenol mebutate-induced plasma membrane translocation. PKC activation appears to be central to the biological effects of ingenol mebutate as indicated by the ability of PKC inhibitors to increase cellular resistance to apoptosis/growth inhibition by the test article. It was suggested that the effects of ingenol mebutate are primarily mediated through activation of the novel PKC isoform δ (see Figure 2). However, the evidence presented by the sponsor appears equivocal in this regard: (1) in one study examining total constitutive PKC protein levels (α , β , γ , δ , ϵ , and η) in ten human solid tumour-derived cell lines there was no clear correlation between ingenol mebutate sensitivity and expression of any PKC isoform, whereas in a study using human leukemic cell lines, PKC δ was expressed at high levels in ingenol mebutate-sensitive but was absent or at a much lower level in insensitive cell lines; (2) a study using a human colonic tumour line (Colo205) showed that ingenol mebutate induced down-regulation of PKC δ protein levels and translocation of PKC δ from cytosol to nucleus and membranes, however, results from a human melanoma (DO4) showed decreased phosphorylation of PKC δ after ingenol mebutate treatment (increased phosphorylation of PKC is associated with activation); and (3) ingenol mebutate treatment of Colo205 cells induced rapid phosphorylation of p38 MAP kinases that was asserted to be dependent on PKC δ due to its inhibition by rottlerin (a claimed "specific inhibitor of PKC δ "), however, data in the literature shows that rottlerin inhibits various protein kinases, but not PKC δ ^{10,11}. What is more convincing from the sponsor's data is that ingenol mebutate-induced activation of PKC leads to activation of the MAP kinase/ERK pathway¹² as demonstrated by changes in the phosphorylation status of members of the pathway and by the ability of MAP kinase pathway inhibitors to reduce ingenol mebutate-induced apoptosis. PKC activation was also linked to the induction of respiratory burst in ingenol mebutate-treated neutrophils, as indicated by the ability of a PKC inhibitor to block this response.

⁹ Sponsor comment: "From nonclinical work it does not appear that the cells in S phase are more sensitive. It appears more likely that they progressed to G2 and were then blocked. That is why S phase was lost. This was achieved at a cytostatic dose (as compared to cytotoxic dose)."

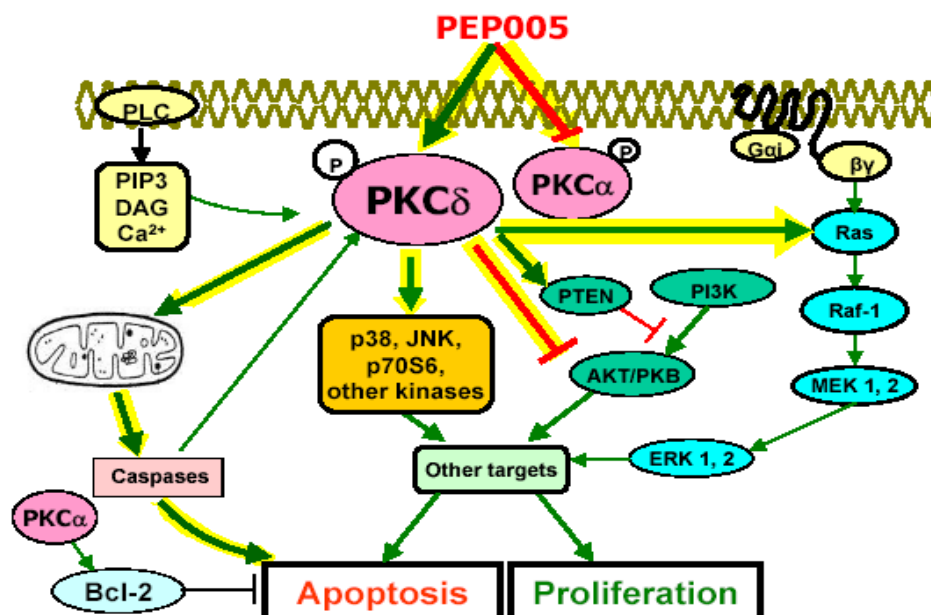
¹⁰ Davies S.P., Reddy H., Caivano M. and Cohen P. (2000) Specificity and mechanism of action of some commonly used protein kinase inhibitors. *Biochemical Journal*, 351: 95–105.

¹¹ Soltoff S.P. (2007) Rottlerin: an inappropriate and ineffective inhibitor of PKC δ . *Trends in Pharmacological Sciences*, 28: 453-458.

¹² The MAPK/ERK pathway is a chain of proteins in the cell that communicates a signal from a receptor on the surface of the cell to the DNA in the nucleus of the cell. The signal starts when a signaling molecule binds to the receptor on the cell surface and ends when the DNA in the nucleus expresses a protein and produces some change in the cell, such as cell division.

Although the above cellular responses may all contribute to the cytotoxic activity of ingenol mebutate the actual mechanism by which solar keratoses are eliminated remains unknown.

Figure 2. Proposed ingenol mebutate (PEP005)-induced cellular signalling cascade.



Secondary pharmacodynamics and safety pharmacology

Ingenol mebutate at 1 μM showed no significant activity in *in vitro* assays for 112 individual receptors and 41 enzymes. The test article was, however, a potent inducer of aggregation for both human platelet-rich plasma and for washed human platelets, with approximate half maximal effective concentration (EC_{50}) values of 6.46 ng/mL (15 nM) and 2.03 ng/mL (4 nM), respectively. This result is consistent with the finding from rodent systemic dosing studies of test article-induced thrombi (see below). This could be an issue of clinical concern if ingenol mebutate were to be misapplied, for example on wounds, ulcers *etc.*

Safety pharmacology studies were performed to GLP standards. Studies in rats and dogs given intravenous (IV) doses of ingenol mebutate showed transient increases in breathing frequency (respiratory rate) in both species at doses $\geq 7.5 \mu\text{g}/\text{kg}$, and dogs dosed at 10 $\mu\text{g}/\text{kg}$ showed a transient increase in mean arterial blood pressure. The study in dogs also indicated a negative inotropic effect at 1 or 3 $\mu\text{g}/\text{kg}$ of ingenol mebutate, although there were no changes in electrocardiographic rhythm or morphology that could be attributed to ingenol mebutate administration. Ingenol mebutate showed no statistically significant effect on hERG tail current density in hERG-expressing HEK293 cells. A tissue distribution study using radioactively-tagged ingenol mebutate indicated that the test article (and/or metabolite(s)) can cross the blood-brain barrier. Based on the very low systemic exposure occurring after topical application therapeutic exposure to ingenol mebutate is not predicted to produce adverse major organ effects.

Pharmacokinetics

Absorption: Ingenol mebutate was generally undetectable in blood following topical application to mice and pigs but was detectable at low levels following high level, topical application to rats. Despite the lack of systemic clearance, very little ingenol mebutate was recovered from mouse skin at times greater than 1 h after application. This result could be

explained by topically applied ingenol mebutate being predominantly metabolised in skin, however, such a suggestion appears inconsistent with the lack of ingenol mebutate-degrading metabolic activity in skin homogenate (see below). *In vitro* studies measuring ingenol mebutate absorption by rat, mini pig, and human dermatome specimens indicated maximum penetration rates (following application of ingenol mebutate at a dose of 15 $\mu\text{g}/\text{cm}^2$) of approximately 20 $\text{ng}/\text{cm}^2/\text{h}$. Such results, combined with the low percentages of drug found in the dermis/epidermis, suggest that the stratum corneum is an effective barrier to uptake of ingenol mebutate. These *in vitro* results appear inconsistent with the above-mentioned mouse result. Formulation appeared to affect uptake; the isopropanol-containing gel, to be used clinically, was best or equal best in these studies. No studies were conducted to assess whether co-administered topical products would affect uptake.

Distribution: Binding of radioactively labelled ingenol mebutate to the plasma proteins of rats (both sexes), dogs, mini pigs, and humans was very high ($\geq 99\%$) in the plasma of all the animal and human subjects investigated and showed no dependence on drug concentration over the range investigated (0.5-20 ng/mL). Intravenously administered, radioactively labelled ingenol mebutate showed a wide distribution in rat tissues suggesting that the drug and/or its metabolites can freely cross cell membranes. Initially, the highest levels were in lungs, liver, kidneys, adrenals, spleen and thyroid, and levels remained high in these tissues over the first few hours post dose.

Metabolism: There was little metabolism of ingenol mebutate following incubation with blood or with skin homogenate from various species including human. Acyl migration to produce ingenol mebutate isomers (PEP015 and PEP025) was the predominant reaction occurring. For example, after 3 h incubation of ingenol mebutate with human skin homogenate the reaction mixture contained approximately 65% ingenol mebutate and 30% 5-mebutate ingenol (PEP015). Hepatocytes from various species showed active metabolism of ingenol mebutate with the major products after incubation for 3 h with human cells being hydroxy-ingenol mebutate and ingenol representing approximately 40% and 20% of total radioactivity, respectively.

Excretion: Approximately 80% of radioactively labelled ingenol mebutate and its metabolites were eliminated via faeces (suggesting biliary excretion) following IV dosing of rats, with the remaining radioactivity appearing in urine. Around 90% of radioactivity was eliminated within 24 h post dosing.

Conclusion: The species chosen for toxicity studies were reasonable selections based on pharmacokinetics. However, a more comprehensive examination of *in vivo* metabolism and uptake of the test article by skin should have been performed. The data provided are incomplete and partly contradictory.

Pharmacokinetic drug interactions

In vitro studies using human hepatocytes and liver microsomal preparations exposed to ingenol mebutate concentrations many times higher than predicted systemic concentrations arising from dermal application showed little or no induction or inhibition of the major CYP isoforms. Based on the intended topical use of ingenol mebutate and its very low systemic levels no further pharmacokinetic drug interaction-type studies were presented by the sponsor.

Toxicology

Acute toxicity

Studies involved IV dosing of rats and rabbits and topical application to mini pigs. Dosing at 20 or 30 $\mu\text{g}/\text{kg}$ produced deaths (within 0.5 h of dosing) in rats, whilst dosing at 10

$\mu\text{g}/\text{kg}$ was not lethal. Decedents displayed ataxia, tachypnoea, nose bleeding and limp body posture, and necropsy showed darkening and inflation of all lobes of the lung. Topical application of ingenol mebutate to pigs at $500 \mu\text{g}/\text{site}$ ($0.833 \mu\text{g}/\text{mm}^2$) produced severe (Grade 4) erythema/eschar by 24 h after initiation of treatment.

Repeat-dose toxicity

IV dosing studies of up to four weeks duration were performed in dog, mouse and rat. More extensive studies, involving intermittent topical dermal or IV administration of ingenol mebutate, were performed using rats (SD) and mini pigs. The longest dermal application studies in rats (SD) and mini pigs were six cycles of three days of dosing over six months and eleven cycles of three days dosing over 41 weeks, respectively. The longest IV dosing study was two days of dosing per week over six months in rats. The studies performed were mostly GLP-compliant, were of appropriate length (relative to intended clinical dosing protocol), and used appropriate species and group sizes. (Note that the minipig has become an important research model, particularly for human skin diseases, because of the similarities between porcine and human organs¹³). The animal dermal application studies used the isopropanol gel formulation intended for clinical use.

Relative exposure

Exposure ratios for animals given topical applications of ingenol mebutate were calculated relative to the maximum proposed clinical dosing regimen for lesions on the trunk or extremities ($235 \mu\text{g}$ per 25 cm^2 , that is, $0.094 \mu\text{g}/\text{mm}^2$). Calculation of systemic exposure ratios, for both IV and topical dosing animal experiments, was more problematic as the sponsor has reported that ingenol mebutate concentrations in blood were below the lower limit of quantification (LLOQ; $0.10 \text{ ng}/\text{mL}$) following topical application to humans. Based on extrapolation of pharmacokinetic data from rats, it was predicted that a peak plasma concentration (C_{max}) in blood of $0.107 \text{ pg}/\text{mL}$ would occur at 2 h after application of ingenol mebutate at $2 \mu\text{g}/\text{kg}$ to human skin. It was suggested by the sponsor that the predicted human C_{max} be used for comparison with animal data. The various inconsistencies in the sponsor's uptake data (noted above) suggest, however, that the predicted human C_{max} value could be inaccurate. Accordingly, the LLOQ for the sponsor's ingenol mebutate assay ($0.10 \text{ ng}/\text{mL}$) has been used as the human reference value for relative exposure calculations. The use of the LLOQ means, of course, that calculated values likely underestimate true relative exposure values. In all mini pig, and in some rat, topical application experiments, systemic ingenol mebutate concentrations were below the LLOQ. Hence, systemic exposure ratios could not be calculated for those experiments. Note that calculations are for ingenol mebutate only and do not include values for its isomers (generally, approximately 5-10% of ingenol mebutate values).

The exposure ratio values at the No Observable Adverse Effect Levels (NOAELs) for IV dosing of Wistar rats and mini pigs with ingenol mebutate suggest that there is a significant margin (17 to 55 fold) between maximum human systemic levels following topical application and levels that cause adverse effects in these animal models (Table 2). The margin appears lower for Sprague Dawley rats and beagle dogs but is likely still greater than unity. Exposure ratios at the NOAEL values for topical application of ingenol mebutate to mini pigs and Sprague Dawley rats were, however, considerably less than unity (approximately 0.1 for mini pigs), indicating that rat and pig skin were markedly

¹³ Nunoya T., Shibuya K., Saitoh T., Yazawa H., Nakamura K., Baba Y. and Hirai T. (2007) Use of miniature pig for biomedical research, with reference to toxicologic studies. *Journal of Toxicologic Pathology*, 20: 125-132.

more sensitive than human skin to the toxic effects of ingenol mebutate application.¹⁴ In many cases, dosing in the topical studies was discontinued at the higher doses because of the severity of the effects.

Table 2. Relative exposure in ingenol mebutate repeat-dose toxicity studies. Table continued across two pages.

Species & strain	Study no.	Duration (day of sampling)	Route	Dose (IV = µg/kg/day; topical = µg/mm ² /day) ^a	C _{max} (ng/mL)	Exposure ratio ^b	
						Skin	Systemic
Rat (SD)	N106162	2 days of dosing per week over 6 months (182)	IV	1.5	1.89 ^c	- ^d	19
				7.5	5.59 (♀)	-	56
				15	17.5 (♀)	-	175
	2174-026	3 days (3)	Topical	0.025	0.14	0.27	1.4
				0.25	0.53	2.66	5
	457405	5 cycles of 6 days dosing over 13 weeks	Topical	0.0125	-	0.13	-
				0.025	-	0.27	-
	N106168A	6 cycles of 3 days dosing over 6 months	Topical	0.0125	< LLOQ	0.13	-
				0.025	< LLOQ	0.27	-
				0.05	< LLOQ	0.53	-
Rat (W1)	2174-020	1 week (7)	IV	1	0.55 ^c	-	6
				<u>5</u> (NOAEL for ♂)	2.57 (♂)	-	26
				<u>10</u> (NOAEL for ♀)	5.32 (♀)	-	53
				15	8.60 ^c	-	86
	2174-014	4 weeks (28)	IV	1.5	0.77 ^c	-	8
				<u>7.5</u>	5.54 ^c	-	55
				15	9.49 ^c	-	95
Mini pig (Göttingen ApS)	2174-022	1 week (1)	IV	<u>2.5</u>	1.7	-	17
		4 days (1)	IV	5	3.9	-	39

¹⁴ Sponsor comment: "The sponsor does not believe the skin of minipig is markedly more sensitive than human skin to toxic effects of ingenol mebutate. The skin of minipigs is probably the best mimic of human skin due to similarities in the skin structure between man and minipig. Concentrations in the interval 0.01% to 0.1% have been used in both rats and minipigs which indeed are in the clinical range. It is true that a NOAEL regarding local skin toxicity was not achieved in the majority of minipig (and rat) studies, but this is considered impossible due to the irritative nature of ingenol mebutate. The efficacy is clearly linked to the irritative properties and a NOAEL can only be obtained at concentration lower than efficacious concentration which is not meaningful. It is correct that dosing was discontinued in several minipigs, which probably reflect greater variation in skin response compared to what is seen in the clinical setting."

Species & strain	Study no.	Duration (day of sampling)	Route	Dose (IV = $\mu\text{g}/\text{kg}/\text{day}$; topical = $\mu\text{g}/\text{mm}^2/\text{day}$) ^a	C _{max} (ng/mL)	Exposure ratio ^b	
						Skin	Systemic
	2174-018	4 weeks (28)	IV	0.25	0.22 ^c	-	2.2
				1.5	1.44	-	14
				<u>3</u>	2.71	-	27
	2174-029	3 days	Topical	0.025	< LLOQ	0.3	-
				0.125	< LLOQ	1.3	-
				0.25	< LLOQ	2.7	-
	507450	3 days	Topical	0.0025	-	0.03	-
				<u>0.00625</u> (NOAEL for ♀)	-	0.07	-
				0.0125	-	0.13	-
				0.025	-	0.27	-
	509987	4 cycles of 3 or 5 days dosing over 13 weeks	Topical	0.0125	< LLOQ	0.13	-
				0.025	< LLOQ	0.27	-
				0.05	< LLOQ	0.53	-
				0.075	< LLOQ	0.80	-
	509992	11 cycles of 3 days dosing over 41 weeks	Topical	0.025	< LLOQ	0.27	-
0.05				< LLOQ	0.53	-	
0.075				< LLOQ	0.80	-	
Dog (beagle)	N106163A	1 week (7)	IV	2	-	-	-
				10	0.47 ^c	-	4.7
				20	1.07 ^c	-	11

a = NOAEL is bolded and underlined. No figure bolded and underlined indicates NOAEL for study is <LD.

b = See text for method of calculation.

c = Values are mean of ♂ and ♀.

d = Not relevant or not determined.

Major toxicities

Topical dermal application of ingenol mebutate-containing gel to rat or mini pig skin produced dose-dependent, mild to severe reactions of erythema, oedema, skin thickening, encrustations, desquamation and necrosis. Under the microscope, treatment sites were associated with lesions such as epidermal necrosis, epidermal hyperplasia, dermal inflammation, fibrosis, acanthosis and ulceration/erosion. Depending on their severity, many of these lesions could disappear during a post dosing recovery period. Dermal fibrosis was seen at significant frequency after a recovery period. These outcomes of ingenol mebutate application are presumably an exaggerated therapeutic effect reflecting the considerably higher sensitivity of rat and mini pig as compared with human skin.

Necropsy of decedents from rat IV repeat-dose studies suggested that ingenol mebutate-induced death was commonly associated with disseminated thrombi in lung vessels. The lungs of unscheduled decedents also showed interstitial inflammation, arterial inflammation, perivascular inflammation, haemorrhage and infiltration of alveolar macrophages. However, the latter findings were mild and lacked a consistent dose-related pattern. Cardiomyopathy was also seen in some studies, although the occurrence of the same lesions (though less severe) in control animals made their association with the test article problematic. The induction of cardiomyopathy is also potentially linked to thrombus formation by ingenol mebutate. Given the very low systemic concentrations of

ingenol mebutate produced by topical application, it is unlikely that these findings are of clinical relevance, except in the case of misapplication (for example to wound).

Genotoxicity

Ingenol mebutate gave negative results in appropriately performed, standard mutation assays (bacterial cell reverse mutation and mammalian cell forward mutation assays). A femoral bone marrow micronucleus assay, using rats given two IV doses of ingenol mebutate, hinted at an increase in micronucleus incidence. The increase was not, however, statistically significant and the assay was interpreted as giving a negative result. A Syrian hamster embryo cell *in vitro* transformation assay indicated a positive response for ingenol mebutate. The latter result was discounted by the sponsor as being indicative of carcinogenic potential for ingenol mebutate because the assay has been reported to have a poor predictive ability (sponsor's Nonclinical Overview document).

The nonclinical evaluator accepts that the sponsor's data indicates a lack of direct mutagenic potential for ingenol mebutate but is unconvinced by claims that the results show that the test article is not clastogenic. This is a significant concern and one worthy of careful attention because of the extensive evidence that phorbol esters (close chemical relatives of ingenol mebutate) can induce clastogenic activity (for example Kozumbo *et al.* 1987¹⁵). Indeed, the sponsor's own studies demonstrated that ingenol mebutate has an ability comparable with PMA (classical, tumour promoting phorbol ester) to induce a respiratory burst (that is, production of reactive oxygen species such as hydrogen peroxide) in neutrophils and monocytes under *in vitro* conditions. Species such as hydrogen peroxide can be converted (via the Fenton reaction) in biological systems into the hydroxyl radical, a potent clastogen. Furthermore, the sponsor's own studies demonstrated that ingenol mebutate administration to rodent skin induced an inflammatory response associated with large-scale recruitment of macrophages and neutrophils. A more useful series of genotoxicity studies might have included examination of chromosomal aberration frequencies in rodent skin cells at various intervals following repeat application of ingenol mebutate. Note that the sponsor's rat micronucleus induction study is of limited usefulness because the high systemic toxicity of ingenol mebutate severely limits the dose that can be administered and because the target cell for the assay is potentially in a protected niche.

Carcinogenicity

No carcinogenicity studies for ingenol mebutate were presented by the sponsor. It was stated that the lack of such studies was "*in agreement with regulatory authorities in the US and EU and according to ICH S1A*" (sponsor's Nonclinical Overview document). ICH S1A (1995)¹⁶ provides a waiver from carcinogenicity testing for ingenol mebutate. However, the document states that one of the factors supporting a need for such testing is: "*previous demonstration of carcinogenic potential in the product class that is considered relevant to humans*". The close chemical relationship between ingenol mebutate and the phorbol esters and their various commonalities of action (such as PKC activation) supports testing for carcinogenicity.

The sponsor gives four reasons in support of the decision not to test ingenol mebutate for carcinogenicity: "1) the demonstrated absence of mutagenic or clastogenic effects, 2) the acute nature of the clinical therapeutic PEP005 Gel regimen for the treatment of patients with AK, 3) lack of systemic exposure in man and 4) the irritative properties of PEP005

¹⁵ Kozumbo W.J., Muehlematter D., Jörg A. and Emerit I. (1987) Phorbol ester-induced formation of clastogenic factor from human monocytes. *Carcinogenesis*, 8: 521-526.

¹⁶ CPMP/ICH/140/95

prohibiting repeat dermal administration for 2 years.” As noted under Genotoxicity above, there is insufficient evidence to properly appraise the accuracy of the first reason. The validity of the second reason is questionable. As noted under Introduction, ingenol mebutate is to be applied to a significant area of “normal” skin surrounding each solar keratosis. Given the known association between solar keratosis and sunlight exposure, it is likely that other pre-malignant cells could also occur within the ingenol mebutate treatment field. It is difficult then to predict whether or not the suggested clinical dosing protocol might serve to advance such cells down the malignancy pathway. The third reason can be viewed as of little relevance (given that skin carcinogenesis is likely to be the greatest concern) or as supporting carcinogenesis studies (given that it indicates that processing of ingenol mebutate may be predominantly in skin). The fourth reason and the associated issue of the low exposure ratios that were achieved in animal models (see Table 2) are certainly problems in trying to extrapolate the results to humans, nevertheless, some data in this area would be far preferable to mere speculation. In the pivotal 6 month IV study in rats, a few tumours were noted at the high dose of 15 µg/kg/day. Due to the small number of tumours, it is unclear if these are treatment-related or explicable as background. The sponsor reported the development of a protocol for examining ultraviolet light-induced p53-mutant patches in mouse skin but did not report the results at the time of submission of the application.¹⁷

The sponsor claims that the study of Adolf *et al.* (1983)⁵ demonstrates that ingenol mebutate lacks significant tumour promotion activity (sponsor’s Nonclinical Overview document). Again, such a claim is questionable. Adolf *et al.* (1983) treated mouse ear with ingenol mebutate following initiation with 7,12-dimethylbenz(a)anthracene. Their results, however, were compromised by high toxicity (that is, the ingenol mebutate doses used were too high) that resulted in many deaths and a shortened observation period.

The absence of carcinogenicity studies might be considered acceptable given the short duration of clinical use (2-3 days). However, actinic keratosis is a chronic condition and the possibility of re-application exists. Given the equivocal genotoxicity findings and the close structural relationship of ingenol mebutate with phorbol esters, known tumour promoters, there is clearly a need for continued monitoring of patient treatment fields for secondary tumours. In the longer term, the conduct of a carcinogenicity study should be considered, particularly if the clinical conditions of use should change.

Reproductive toxicity

An abbreviated group of studies was presented that examined possible effects on embryofetal development following IV dosing with ingenol mebutate of rats and rabbits from gestational day (GD) 6-16 and GD 6-18, respectively. All studies were GLP compliant and used appropriate group sizes. According to the sponsor, it was agreed with regulatory authorities in the US and the EU that (because of the very low systemic levels following topical application) no evaluation of potential effects of ingenol mebutate on fertility, early embryonic development, and pre- and postnatal development would be performed (sponsor’s Nonclinical Overview document).

¹⁷ Sponsor comment: “The results from these studies have since been published (see Cozzi SJ, Ogbourne SM, James C, Rebel HG, de Gruijl FR, Ferguson B, *et al.* Ingenol Mebutate Field-Directed Treatment of UVB-Damaged Skin Reduces Lesion Formation and Removes Mutant p53 Patches. *J Invest Dermatol.*2011).”

Table 3. Relative exposure to ingenol mebutate in developmental studies

Species & strain	Study	Dose ($\mu\text{g}/\text{kg}/\text{day}$) ^a	C _{max} (ng/mL)	Exposure ratio ^b
Rat (SD)	Embryofetal development	1.5	0.502 ^c	5
		<u>3</u> (maternal NOAEL)	-	-
		<u>5</u> (embryofetal NOAEL)	2.28 ^c	23
Rabbit (NZW)	Embryofetal development	<u>1</u> (embryofetal NOAEL)	0.233 ^d	2.3
		<u>2</u> (maternal NOAEL)	0.477 ^d	5
		4	0.931 ^d	9

a = NOAEL is bolded and underlined.

b = relative to the LLOQ for the ingenol mebutate assay (*i.e.* 0.10 ng/mL) (see above).

c = values for non-pregnant ♀ rats from study no. 2174-020

d = GD16 values from pilot experiment

The exposure ratios shown in Table 3 suggest that pregnant rats and rabbits were exposed to systemic concentrations of ingenol mebutate that were many times those anticipated to occur after topical application to humans. The rat studies indicated that ingenol mebutate dosing may increase early embryonic deaths. While the effect did not reach statistical significance, the consistency of this finding in two rat studies as well as one rabbit study, suggests a likely relationship with treatment. The rabbit studies suggested that ingenol mebutate may increase the incidence of several minor developmental abnormalities.

Pregnancy classification

The sponsor has proposed pregnancy Category B1¹⁸. Given that the sponsor presented only a limited dossier of studies (for example, no studies of test article effects on fertility and early embryonic development) and given that there were indications from the rat studies of an increase in early embryonic deaths and from the rabbit studies of an increase in minor developmental abnormalities, a B3 classification¹⁹ appears more appropriate.

Skin sensitisation

Repeat application of ingenol mebutate to mouse ear induced a marked increase in DNA replication in neighbouring lymph nodes, suggesting that the test article has the potential to cause skin sensitisation.

Phototoxicity

According to the sponsor's Product Information: "*Studies have been conducted to assess the effects of UV irradiation on the skin following single and multiple applications of ingenol mebutate gel, 0.01%. Ingenol mebutate gel did not demonstrate any potential for photo irritation or photo allergic effects*". Such studies were not submitted for evaluation by the nonclinical evaluator.

¹⁸ Category B1 = Drugs which have been taken by only a limited number of pregnant women and women of childbearing age, without an increase in the frequency of malformation or other direct or indirect harmful effects on the human fetus having been observed. Studies in animals have not shown evidence of an increased occurrence of fetal damage.

¹⁹ Category B3 = Drugs which have been taken by only a limited number of pregnant women and women of childbearing age, without an increase in the frequency of malformation or other direct or indirect harmful effects on the human fetus having been observed. Studies in animals have shown evidence of an increased occurrence of fetal damage, the significance of which is considered uncertain in humans.

Impurities

In aqueous solution, there is an equilibrium between three isomeric forms of ingenol mebutate (3-, 5-, and 20-mebutate ingenol) that are generated by acyl migration of the mebutate group between free hydroxyl groups on the ingenol moiety. (The sponsor's product predominantly contains the 3- isomer). All three isomers were shown to have comparable growth inhibitory activity in *in vitro* tests using human melanoma and other tumour-derived cell lines. A number of impurities/degradants in the drug substance and drug product were specified at levels above the qualification threshold. The sponsor's studies showed that ingenol mebutate spiked with elevated (compared to normal) levels of the 5- and 20-mebutate isomers and other impurities gave negative results in mutagenicity tests, and produced results comparable with 3-mebutate ingenol in repeat-dose toxicity studies using IV dosing of rats and dermal application to mini pigs. The proposed limits for impurities/degradants would result in levels below the threshold of toxicological concern (1.5 µg/day) and have been qualified by submitted toxicity studies.

Paediatric use

Solar keratoses do not occur on children, except for those with rare genetic diseases. Accordingly, ingenol mebutate is not proposed for paediatric use and no specific studies using juvenile animals were submitted.

Nonclinical summary and conclusions

- The nonclinical studies presented were of good quality and were performed in reputable laboratories.
- Primary pharmacology studies showed that ingenol mebutate has growth inhibitory/toxic activity towards a variety of tumour-derived cell lines (both human and rodent, and including squamous cell carcinomas and melanomas). There were marked differences in the activity of ingenol mebutate towards different tumour lines, raising the possibility of inter- and intra-individual differences in responsiveness of solar keratoses to test article treatment. Central to the actions of ingenol mebutate is its ability to activate PKC, which can lead directly to the induction of tumour cell death (via apoptosis or necrosis) or indirectly via the production of neutrophil-attracting cytokines and the induction of respiratory burst and phagocytosis in these cells.
- Ingenol mebutate was shown to be a potent inducer of platelet aggregation. This activity is, however, unlikely to be of clinical relevance because of a large difference between levels of ingenol mebutate inducing platelet aggregation and those present systemically following topical application to humans. No other clinically relevant hazards were identified from the secondary pharmacodynamics and safety pharmacology testing.
- Ingenol mebutate levels in blood were generally below the LLOQ following topical application to mice and pigs but the drug was detectable at low concentrations following high level, topical application to rats. Despite the apparent lack of systemic clearance, very little ingenol mebutate was recovered from mouse skin at times greater than 1 h after application. This result could be explained by skin metabolism of topically applied ingenol mebutate, however, skin homogenate showed a lack of ingenol mebutate-degrading activity. Ingenol mebutate showed little or no induction or inhibition of the major CYP isoforms.
- Ingenol mebutate-induced death in IV repeat-dose toxicity studies was commonly associated with disseminated thrombi in lung vessels (consistent with the demonstrated induction of platelet aggregation). Comparison of the exposure ratios at

the NOAELs for IV dosing of animal models suggested a very large margin between predicted human systemic levels following topical application and levels that cause adverse effects in these animal models. Topical application of ingenol mebutate to rat or mini pig skin produced dose-dependent, mild to severe reactions of erythema, oedema, skin thickening, encrustations, desquamation, and necrosis. In contrast to the systemic results, exposure ratios at the NOAEL values for topical application of ingenol mebutate to mini pigs and SD rats were considerably less than unity (approximately 0.1 for mini pigs).

- Standard mutation assays (using both bacterial and mammalian cells) gave negative results for ingenol mebutate. Carcinogenicity studies were not performed, even though an in vitro transformation assay gave positive results for ingenol mebutate. The lack of such studies is also worrisome because ingenol mebutate is closely related to the phorbol esters (potent PKC activators that are also potent tumour “promoters”).
- Pregnant rat studies indicated that ingenol mebutate dosing may increase early embryonic deaths. Rabbit studies suggested that ingenol mebutate may increase the incidence of several minor developmental abnormalities.
- Mouse studies, measuring DNA synthesis in lymph nodes near treatment sites, suggested that ingenol mebutate has the potential to cause skin sensitisation. This could limit the number of ingenol mebutate treatments per patient.

Conclusions and recommendation

- Primary pharmacology studies were performed with tumour-derived cell lines and (to a lesser extent) with normal cells. The absence of testing on solar keratosis cells (due to lack of suitable animal models) seems unlikely to impact on the conclusions drawn as to how ingenol mebutate induces cell death.
- Ingenol mebutate was shown to be a potent inducer of platelet aggregation however this activity is unlikely to be of clinical relevance unless the test article is mistakenly applied to cuts, skin ulcers, or similar lesions, or (possibly) if it enters the mouth. These possibilities may be cause for concern given the suggested treatment protocol, whereby ingenol mebutate-containing gel is applied to the solar keratosis by the patient.
- Ingenol mebutate was shown to be very toxic in IV repeat-dose studies and to be a potent irritant in topical application studies. The findings in topical studies are expected to be seen clinically and are associated with pharmacological action.
- Concerns about the possible genotoxicity and/or carcinogenicity of ingenol mebutate were not resolved by the sponsor’s studies. At the least, the sponsor should commit to an ongoing program of monitoring patient treatment fields for secondary tumours.
- Ingenol mebutate produced equivocal results in testing for possible reproductive toxicity. Such results are unlikely to be of clinical relevance given the large exposure ratios between animal testing and likely human systemic levels. Nevertheless, it is suggested that ingenol mebutate be given a B3 pregnancy classification.
- The nonclinical evaluator supports the registration of Picato gel for the intended clinical use for up to 3 days. A number of amendments to the draft Product Information were recommended but these are beyond the scope of this AusPAR.

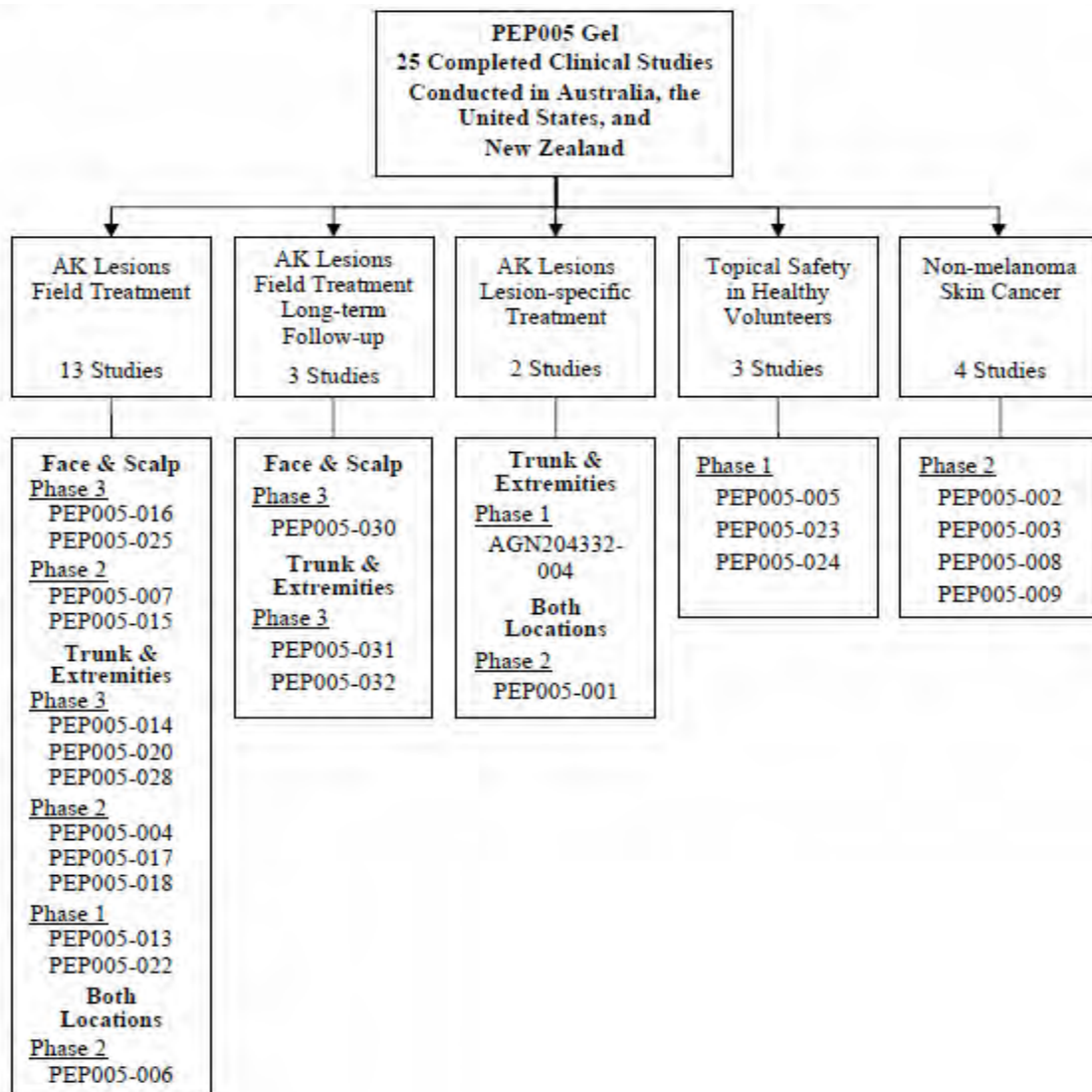
IV. Clinical findings

Introduction

A summary of the clinical findings is presented in this section. Further details of these clinical findings can be found in Attachment 2.

A total of 25 clinical studies were provided in support of this application as illustrated below. In these studies, Picato Gel was identified as PEP005 Gel. In addition, there were noted to be three ongoing safety studies, PEP005-033, PEP005-036 and LP0041-01. Minimal information was provided on these ongoing studies.

Figure 3. Overview of the Completed Clinical Studies for PEP005 Gel



Of these 25 completed studies, 18 were conducted in patients with SK lesions. The remaining 7 studies contributed data to the safety profile of Picato Gel and included 3 topical safety studies performed in healthy volunteers and 4 safety studies in patients with non malignant skin cancer (NMSC).

The minimum effective dose for both indications was identified in 2 dose-ranging studies, PEP005-015 for the face and scalp indication (0.015% Picato Gel) and PEP005-006 for the

trunk and extremities indication (0.05% Picato Gel). These studies are further described in sections 3.2.1 and 3.5.

Information provided on pharmacology focussed on demonstrating that PEP005 was not systemically absorbed. Pharmacokinetic information was obtained from a total of 32 subjects as well as predicted using allometric scaling from *in vivo* animal PK and *in vitro* percutaneous absorption data. This information is consistent with that provided in the draft PI. The precise mechanism of action of PEP005 gel is unknown, although possible mechanisms were postulated in the draft PI.

The dose proposed in the draft PI was used in the pivotal studies. The pivotal studies were all placebo-controlled, with a clinically acceptable superiority margin. There were no active controlled studies and the rationale for this was given by the sponsor. Data on recurrence was provided from 3 long term studies (PEP005-030, PEP005-031 and PEP005-032).

Limited information on histological confirmation of clearance was provided, including results from Study PEP005-001. Efficacy endpoints were also discussed and were consistently applied across all pivotal studies. Limited objective measures were used.

GCP aspects

No significant Good Clinical Practices (GCP) aspects were identified in the development of this product.

Pharmacokinetics

Introduction

Picato Gel is applied topically and not absorbed systemically. Ingenol mebutate, at the concentrations applied topically for treatment of SK lesions, has no detectable systemic absorption. The human PK profile was predicted using allometric scaling from *in vivo* animal PK and *in vitro* percutaneous absorption data. This profile suggested that the maximum intended clinical dose (2 micrograms/kg/day) would not produce measurable systemic blood levels of ingenol mebutate, and that a minimum topical dose of 2000 micrograms/kg/day would be required.

Evaluator's overall conclusions on pharmacokinetics

Picato Gel is applied topically and not absorbed systemically. Ingenol mebutate, at the concentrations applied topically for treatment of SK lesions, has no detectable systemic absorption. This is supported by the human PK profile from allometric scaling, as well as some limited PK data obtained from subjects in 4 clinical studies.

Pharmacodynamics

Introduction

No clinical studies on human pharmacodynamics (PD) were conducted. As such, human PD data was not available, and no PK/PD correlation studies were performed nor PK/PD relationship established. The mode of action of PEP005 has been established on nonclinical models using cell lines and animal models.

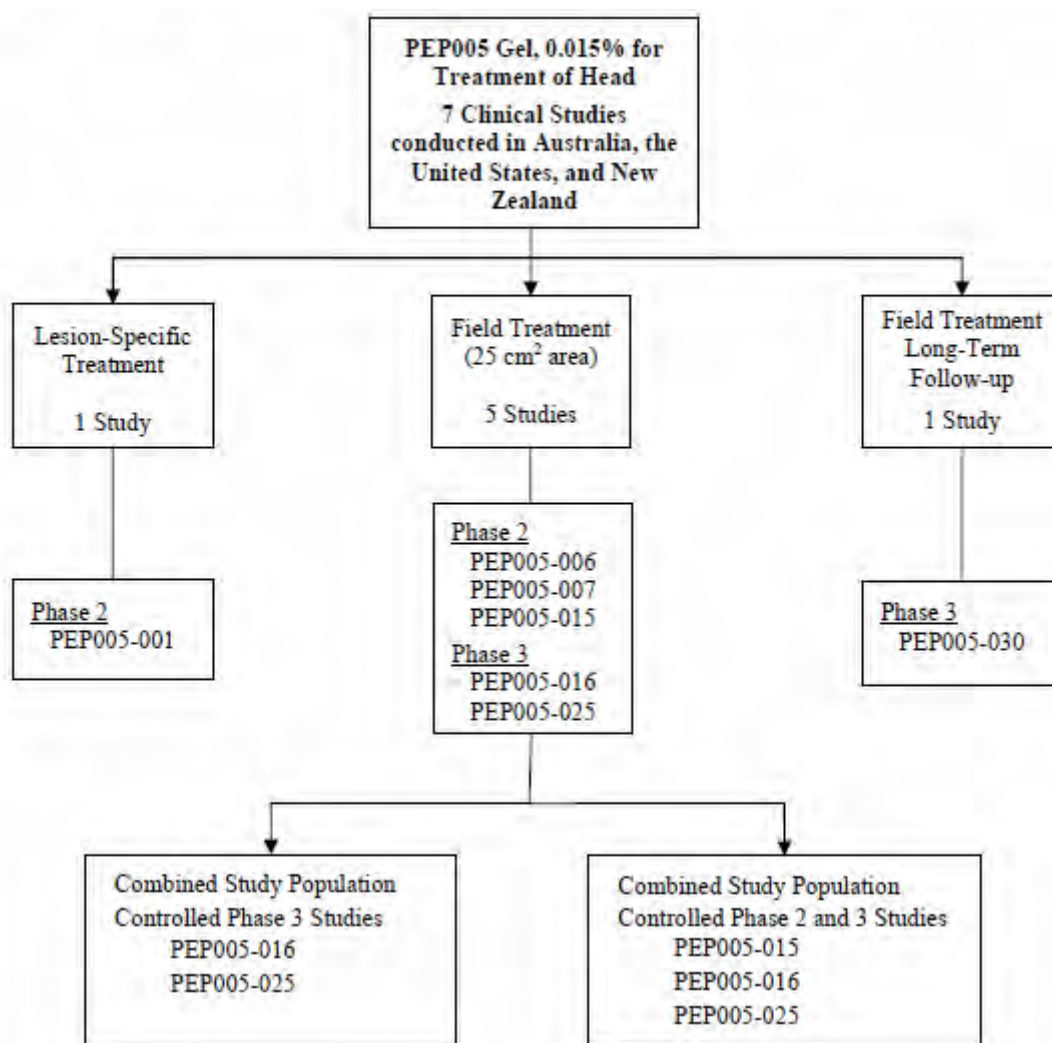
Evaluator's overall conclusions on pharmacodynamics

No clinical studies on human pharmacodynamics were conducted. The mechanism of action in SK is not fully understood but appears to be a combination of induction of local lesion cell death and promotion of an inflammatory response with neutrophils and other immunocompetent cells.

Efficacy**Introduction**

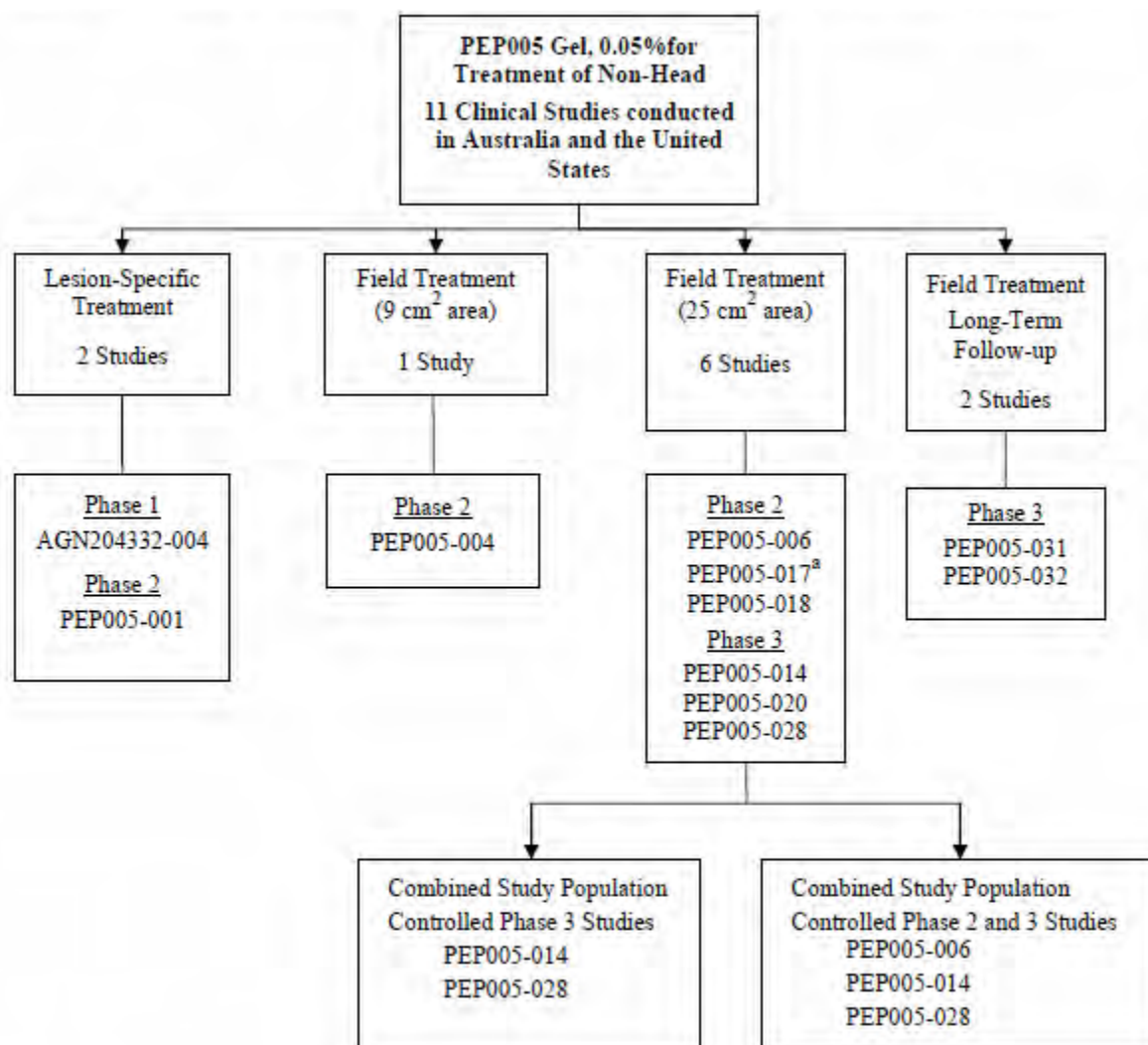
Information on clinical efficacy was provided for the 2 specific indications, the treatment of solar (actinic) keratoses (SK) on the face and scalp (0.015% Picato Gel), and for the treatment of SK on the body (non-head regions) (0.05% Picato Gel). For the first indication, there were 7 studies that evaluated Picato Gel. Of these studies, five (PEP005-016, PEP005-025, PEP005-015, PEP005-006 and PEP005-007) provided efficacy data for field treatment of Picato Gel to a defined 25 cm² skin area containing 4 to 8 SK lesions located on the head with efficacy assessed at Day 57 (study exit). The sixth trial (PEP005-030) was a long term follow-up study in patients who achieved complete clearance at Day 57 in studies PEP005-016 and PEP005-025. No study medication was administered during this study. The seventh trial was a lesion specific study (PEP005-001) which allowed up to five lesions to be treated on multiple anatomical sites.

Of these studies, PEP005-015, PEP005-016 and PEP005-025 were randomised, double-blind, vehicle-controlled, parallel-group studies and were the pivotal studies for this indication. PEP005-015 was a dose-ranging study that included the proposed dosage regimen treatment on head locations. PEP005-016 and PEP005-025 were Phase III studies that evaluated the proposed dosage regimen of Picato Gel for treatment of head locations. This study development program is summarised below.

Figure 4. Clinical studies located in the efficacy evaluation of head locations

For the second indication (trunk and extremities), 11 studies evaluated Picato Gel. Of these studies, six provided efficacy data for field treatment of Picato Gel to a defined 25 cm² skin area containing four to 8 SK lesions located on trunk and extremities with efficacy assessed at Day 57 (PEP005-014, PEP005-028, PEP005-006, PEP005-017, PEP005-018, and PEP005-020). Two trials (PEP005-031 and PEP005-032) were long term follow-up studies in patients who achieved complete clearance at Day 57 in previous trials. One study (PEP005-004) assessed efficacy where Picato Gel was applied to a small field of treatment (9 cm²) that included a single target lesion with efficacy endpoints assessed at Day 29. There were also two lesion-specific studies where Picato Gel was applied to individual SK lesions rather than to a field of skin (AGN204332-004 and PEP005-001).

Of these studies, PEP005-014 and PEP005-028 were well-controlled Phase III studies that evaluated the proposed dosage regimen application of Picato Gel for treatment of non-head locations and should be regarded as the pivotal studies for this indication. This study development is summarised below.

Figure 5. Clinical studies include in the efficacy evaluation of non head locations

^a For PEP005-017, an area of 100 cm² was treated and assessed for safety; efficacy was assessed in a 25 cm² area within the 100 cm² treatment area.

For the pivotal studies, a study design using an active comparator was not employed because of the potential to introduce bias with regard to the selection and timing of study endpoints. PEP005 Gel and any available active comparator used in the same patient population represent different modalities of treatment. As a result, efficacy endpoints would have been measured at different times during a comparator trial. The duration of treatment for PEP005 Gel is 2 or 3 days with efficacy assessment at Day 57 following study medication application. Other products have longer durations of treatment, may require repeat treatment periods and take longer to achieve efficacy. Therefore, the sponsor considered that an unbiased comparator trial would not be feasible.

For the pivotal studies, the majority of SKs were diagnosed clinically, not histologically. In a study where 271 lesions were biopsied to correlate SK and SCC, clinical diagnosis and histopathologic findings agreed in 91% of the biopsies further supporting the appropriateness of a clinical diagnosis for SK. Punch biopsies of 220 clinically diagnosed untreated AKs were performed at baseline plus 51 lesions unresponsive to treatment (total, 271). Clinical diagnosis and histopathologic findings agreed in 91% (246/271) of the lesions biopsied. The clinical diagnosis for the balance of the biopsied lesions were: (1)

benign changes 4% (11/271) and (2) occult cutaneous malignancy in 5% (14/271) of the cases, 12 squamous cell carcinomas and 2 basal cell carcinomas.

Early in this clinical development program, histological data were evaluated in a Phase I lesion-specific study (PEP005-001). Two single applications of study medication were applied directly to each selected lesion using PEP005 Gel at concentrations of 0.0025%, 0.01%, 0.05%, or vehicle gel. Punch biopsy samples were obtained pre-treatment to confirm the presence of SK and post-treatment on Day 85 to determine lesion clearance. All biopsies were reviewed by a central dermatopathologist. Results showed absence of SK lesions on post-treatment biopsy for approximately 50% of the biopsies performed across all treatment groups. Results from this study included proportion of pre- and post-treatment biopsies and the number who had complete clearance. Although based on few patients these findings provide evidence of histological clearance.

For the pivotal studies, the comparative efficacy analyses for head and non-head locations used the same efficacy endpoints. For the combined studies populations, the primary efficacy endpoint was complete clearance, defined as the proportion of patients at Day 57 with no clinically visible SK lesions in the selected treatment area. For the head location, this endpoint was pre-specified as the primary endpoint in the two adequate and well-controlled Phase III studies (PEP005-016 and PEP005-025) and the Phase II dose-ranging study (PEP005-015). For the non-head location, this endpoint was pre-specified as the primary endpoint in the two adequate and well controlled Phase III studies (PEP005-014 and PEP005-028) but was a secondary endpoint in the Phase II dose-ranging study (PEP005-006). The secondary endpoint was partial clearance rate, defined as the proportion of patients at Day 57 with a 75% or greater reduction in the number of clinically visible SK lesions identified at baseline in the selected treatment area. Percent reduction from baseline in the total number of SK lesions at Day 57 was an additional endpoint.

For the head locations, the primary efficacy analysis in the Phase III studies (PEP005-016 and PEP005-025) compared complete clearance rates across treatment groups (active vs. vehicle) using the Cochran-Mantel-Haenszel (CMH) test statistic. In each of these studies, in order to obtain at least eight patients per site per treatment group, study sites yielding fewer than 16 patients were combined in order of geographical proximity, referred to as "analysis sites". The exact composition of these "analysis sites" was determined and documented prior to breaking the study blind. The stratification for CMH analyses was based on the analysis sites, not on the actual study sites. For the non-head locations, the primary efficacy analysis in the Phase III studies (PEP005-014 and PEP005-028) also compared complete clearance rates across treatment groups (active vs. vehicle) using the CMH test statistic. The CMH analysis for PEP005-014 was stratified on anatomical location and the CMH analysis for Study PEP005-028 was stratified on analysis site. As part of this efficacy summary a CMH analysis for PEP005-014 stratified on analysis site is also presented.

For each Phase III study for the head and non-head locations, missing values were imputed using the last observation carried forward (LOCF) method. A sensitivity analysis of complete clearance assumed that all patients who missed the Day 57 visit or were outside the visit window (\leq Day 50 or \geq Day 85) did not achieve complete clearance. In addition, complete clearance rates were compared across treatment groups by location of treatment area (face or scalp for head and arm, back of hand, or "other" [back, shoulder, leg or chest] for non-head). Statistical tests were two-sided with a significance level of $\alpha = 0.05$. Analyses of study results were pre specified prior to database lock and unblinding. As part of this efficacy summary, two additional analyses of complete clearance were performed for each study. An additional sensitivity analysis was performed for complete clearance rates in which all active treatment patients who missed the Day 57 visit or were outside the visit window (\leq Day 50 or \geq Day 85) were considered as not achieving complete

clearance and all vehicle patients who missed the Day 57 visit or were outside the visit window were considered as achieving complete clearance. Furthermore, clearance rates were compared across treatment groups using a logistic regression model with terms for treatment, analysis site, and anatomical location.

Evaluator's overall conclusions on clinical efficacy

Information on clinical efficacy was provided for the 2 specific indications, the treatment of solar (actinic) keratoses (SK) on the face and scalp (0.015% Picato Gel), and for the treatment of SK on the body (non-head regions) (0.05% Picato Gel).

PEP005-015, PEP005-016 and PEP005-025 were randomised, double-blind, vehicle-controlled, parallel-group studies, and were the pivotal studies for the face and scalp indication. PEP005-015 was a dose-ranging study that included the proposed dosage regimen treatment on head locations.

PEP005-015 was a multi-centre, randomised, double-blind, vehicle-controlled, dose ranging study. The primary objective of the study was to evaluate the safety, tolerability and efficacy of PEP005 Gel (0.005%, 0.01% and 0.015%) compared to vehicle gel, administered once daily as either a two or three consecutive day treatment regimen, to a 25 cm² contiguous SK treatment area on the face or scalp. A total of 265 patients were randomised and 260 patients completed the study. Observed complete clearance rates were concentration and treatment regimen-dependent with the highest observed complete clearance rates in the 0.015% two-day and three-day groups. The observed complete clearance rates for PEP005 Gel at Day 57 for the Intent-to-treat (ITT) population ranged from 15% in the 0.005% two-day group to 50% in the 0.015% three-day group. Partial clearance rates were concentration and treatment regimen-dependent with the highest observed complete clearance rates in the 0.015% two-day and three-day groups. The observed partial clearance rates for PEP005 Gel at Day 57 for the ITT population ranged from 33.3% in the 0.005% two-day group to 71.9% in the 0.015% three-day group.

PEP005-016 was a multi-centre, randomised, double-blind, vehicle-controlled, Phase III study. The objective of the study was to evaluate the efficacy and safety of PEP005 Gel, 0.015%, compared to vehicle gel when administered once daily for three consecutive days to a contiguous 25 cm² area of skin on the head (face or scalp). A total of 269 patients were randomised (135 to PEP005 Gel 0.015% and 134 to vehicle gel); 259 patients completed the study. The PEP005 Gel group demonstrated a statistically significant, higher complete clearance rate versus vehicle gel (37% compared to 2%, $p < 0.001$, CMH test stratified by analysis site) based on the ITT population. For the secondary endpoint the PEP005 Gel group demonstrated a statistically significant, higher partial clearance rate versus vehicle gel (60% compared to 7%, $p < 0.001$, CMH test stratified by analysis site) based on the ITT population. The results of the per-protocol (PP) population were consistent with the results of the ITT population.

PEP005-025 was a multi-centre, randomised, double-blind, vehicle-controlled, Phase III study. The objective of the study was to evaluate the efficacy and safety of PEP005 Gel, 0.015%, compared to vehicle gel when administered once daily for three consecutive days to a contiguous 25 cm² area of skin on the head (face or scalp). A total of 278 patients were randomised (142 to PEP005 Gel 0.015% and 136 to vehicle gel); 277 patients completed the study. The PEP005 Gel group demonstrated a statistically significant, higher complete clearance rate versus vehicle gel (47% compared to 5%, $p < 0.001$, CMH test stratified by analysis site) based on the ITT population. The results for the PP population were consistent with the results for the ITT population. For the secondary efficacy endpoint, the PEP005 Gel group demonstrated a statistically significant, higher partial clearance rate versus vehicle gel (68% compared to 8%, $p < 0.001$, CMH test stratified by analysis site) based on the ITT population.

PEP005-014 and PEP005-028 were well controlled Phase III studies that evaluated the proposed dosage regimen application of Picato Gel for treatment of non-head locations, and were the pivotal studies for the trunk and extremities indication. Dose-ranging information for this indication was provided by PEP005-006, which involved subjects for both indications.

PEP005-014 was a multi-centre, randomised, parallel group, double-blind, vehicle-controlled, study. The objective of the study was to evaluate the efficacy and safety of PEP005 Gel, 0.05% compared to vehicle gel, when administered once daily for two consecutive days (Day 1 and Day 2) to a 25 cm² contiguous SK treatment area on non-head locations. A total of 255 patients were enrolled (126 to PEP005 Gel, 0.05% and 129 to vehicle gel). The observed complete clearance rate at Day 57 (ITT LOCF) overall was statistically significantly higher in the PEP005 Gel, 0.05% group (28%) than the vehicle group (5%) ($p < 0.0001$). Sensitivity analyses, including a multiple imputation method for handling missing data and analyses based on evaluable and per-protocol (PP) populations, all demonstrated a statistically significantly higher complete clearance rate in the PEP005 Gel, 0.05% group than in the vehicle group ($p < 0.0001$ for all comparisons). The observed partial clearance rate at Day 57 overall in the PEP005 Gel, 0.05% group was 44% (56/126) versus 7% (9/129) in the vehicle group ($p < 0.0001$).

PEP005-028 was a multi-centre, randomised, parallel group, double-blind, vehicle-controlled study. The objective of the study was to evaluate the efficacy and safety of PEP005 Gel, 0.05% compared to vehicle gel when administered once daily for two consecutive days (Days 1 and 2) to a 25 cm² contiguous SK treatment area on non-head locations. A total of 203 patients were enrolled (100 PEP005 Gel, 0.05%; 103 vehicle gel). The complete clearance rate at Day 57 (ITT LOCF) overall was statistically significantly higher in the PEP005 Gel, 0.05% group (42%) than the vehicle group (5%) ($p < 0.001$). The results of the analysis of the secondary efficacy endpoint, partial clearance ($\geq 75\%$ reduction) in SK lesions at Day 57, supported the results of the analysis of complete clearance. The partial clearance rate at Day 57 overall was statistically significantly higher in the PEP005 Gel, 0.05% group (55%) than the vehicle group (7%) ($p < 0.001$).

PEP005-006 was a multi-centre, randomised, double-blind, double dummy, vehicle-controlled sequential cohort study. The primary objectives of the study were to evaluate the safety and tolerability of 0.025% and 0.05% PEP005 Topical Gel compared to vehicle gel, administered according to 2 treatment schedules, Day 2 and Day 3 (0.05%) or Day 1, Day 2 and Day 3 (0.025% and 0.05%) to a 25 cm² contiguous SK treatment area, and to evaluate the efficacy of 0.025% and 0.05% PEP005 Topical Gel compared to vehicle gel when administered according to 2 treatment schedules, Day 2 and Day 3 (0.05%) or Day 1, Day 2 and Day 3 (0.025% and 0.05%) to a 25 cm² contiguous SK treatment area. One hundred sixty-one patients were treated on non-head locations: 43 patients received vehicle gel, 37 patients received 0.025% PEP005 Gel for three days, 42 patients received 0.05% PEP005 Gel for two days, and 39 patients received 0.05% PEP005 Gel for three days. For patients who received treatment on non-head locations, partial clearance rates by treatment group were 21%, 51%, 67% and 74%, respectively ($p \leq 0.0001$ for PEP005 Gel groups compared to vehicle gel). Complete clearance rates were 14%, 35%, 45% and 46%, respectively ($p < 0.006$ for PEP005 Gel groups compared to vehicle gel).

Three clinical studies provided information on long term efficacy. PEP005-030 was a long term follow-up study in patients who achieved complete clearance at Day 57 in studies PEP005-016 and PEP005-025 for the head and scalp indication. Two trials (PEP005-031 and PEP005-032) were long term follow-up studies in patients who achieved complete clearance at Day 57 in previous trials for the trunk and extremities indication. No study medication was administered during these studies.

PEP005-030 was a 12 month, long term follow-up study of patients with SK on the head (face or scalp) who completed Day 57 in studies PEP005-016 or PEP005-025. The

objectives of the study were to summarise treatment area recurrence of SK lesions, in the selected treatment area, during a 12 month follow-up period for patients who achieved complete clearance of SKs at Day 57 in studies PEP005-016 and PEP005-025, and to summarise long term safety data, in the selected treatment area over a 12 month follow-up period for patients who completed Day 57 in studies PEP005-016 and PEP005-025. A total of 117 patients who had demonstrated complete clearance of SK lesions in either Study PEP005-016 or PEP005-025 were enrolled in this long term follow-up study. Of these 117 patients, 108 had received 0.015% PEP005 Gel and 9 had received vehicle in the previous study. At 12 months of follow-up, 53.9% of patients who had been treated with PEP005 Gel in the previous Phase III studies (N=108), had at least one new or recurrent SK lesion within the selected treatment area. The estimated median time to lesion recurrence was 365 days. Based on the number of lesions observed within the treatment area during 12 months of follow-up relative to the number of lesions at baseline (determined prior to treatment with PEP005 Gel in the Phase III studies), the mean lesion-based recurrence rate was 12.8%. At 12 months of follow-up, 72.2% of vehicle-treated patients (N=9) had a new or recurrent SK lesion, with a median time to recurrence of 183 days. For this group of patients, the mean lesion-based recurrence rate at 12 months was 16.3%.

PEP005-031 was a 12 month, long term follow-up study of patients with actinic keratosis on non-head areas (trunk and extremities) who have completed Day 57 in Study PEP005-020. The objectives of the study were to summarise treatment area recurrence of actinic keratosis lesions, in the selected treatment area, during a 12 month follow-up period for patients with complete clearance who completed Day 57 in Study PEP005-020, and to summarise long term safety data, in selected treatment area over a 12 month follow-up period for patients who completed Day 57 in Study PEP005-020. A total of 38 patients who had demonstrated complete clearance of SK lesions in Study PEP005-020 were enrolled in the study. At 12 months of follow-up, 62.5% of patients in the CC57 population treated with PEP005 Gel, 0.05% in Study PEP005-020 (N=38) had at least one new or recurrent AK lesion within the selected treatment area. The estimated median time to lesion recurrence was 274 days. Based on the number of lesions observed within the treatment area during 12 months of follow-up relative to the number of lesions at baseline (determined prior to treatment with PEP005 Gel in the Phase III study), the mean lesion-based recurrence rate was 11.3%.

PEP005-032 was a 12 month, long term follow-up study of patients with actinic keratosis on non-head locations (trunk and extremities) who completed Day 57 in Study PEP005-028. The objectives of the study were to summarise treatment area recurrences of actinic keratosis lesions, in the selected treatment area during a 12 month follow-up period for patients with complete clearance, who completed Day 57 in Study PEP005-028, and to summarise long term safety data, in selected treatment area over a 12 month follow-up period for patients with complete clearance, who completed Day 57 in Study PEP005-028. A total of 43 patients were enrolled (38 received PEP005 Gel 0.05% and 5 received vehicle gel in the previous study [PEP005-028]). At 12 months of follow-up, 50% of patients treated with PEP005 Gel, 0.05% in Study PEP005-028 (N=38) had at least one new or recurrent SK lesion within the selected treatment area. The estimated median time to recurrence was >183 days. Based on the number of lesions observed within the treatment area during 12 months of follow-up relative to the number of lesions at baseline (determined prior to treatment with PEP005 Gel in the Phase III studies), the mean lesion-based recurrence rate was 14.9%. At 12 months of follow-up, 80% of patients treated with vehicle gel in the previous study (N=5) had a new or recurrent SK lesion, with a median time to recurrence of 183 days and the mean lesion-based recurrence rate at 12 months was 19.2%.

In addition to the studies mentioned above, a further 9 clinical studies provided supporting information on efficacy. These studies were PEP005-007, PEP005-020,

PEP005-004, PEP005-017, PEP005-018, PEP005-013, PEP005-022, AGN204332-004 and PEP005-001

PEP005-007 was an open-label, multi-centre, dose-escalation, cohort study. The primary objective was to determine the optimal tolerated regimen of PEP005 Topical Gel when administered to patients once daily as a two or three consecutive day application schedule to a 25 cm² contiguous SK treatment area on the face or face and scalp. The study was not statistically powered to evaluate efficacy and the resulting groups had a small number of patients.

PEP005-020 was a multi-centre, open-label study to evaluate the safety and efficacy of PEP005 (ingenol mebutate) Gel, 0.05% in patients with actinic keratoses on non-head locations (trunk and extremities). The objectives of the study were to evaluate the safety of PEP005 Gel, 0.05% when administered once daily for two consecutive days (Days 1 and 2) to a 25 cm² contiguous actinic keratosis treatment area on non-head locations and to evaluate the efficacy of PEP005 Gel, 0.05% when administered once daily for two consecutive days (Days 1 and 2) to a 25 cm² contiguous SK treatment area on non-head locations. No hypotheses were tested and no inferential analyses were performed in this study. Overall, 40/102 (39.2%) patients had a complete clearance of SK lesions at Day 57. The overall partial clearance rate was 54.9%.

PEP005-004 was an open-label, dose-escalation, cohort study to determine the maximum tolerated dose and safety of PEP005 Topical Gel when applied on Day 1 and Day 2 to actinic keratoses on the shoulders, chest, back or arms followed by a post-treatment follow-up period lasting at least four weeks. The primary objective of this study was to determine the maximum tolerated dose (MTD) for PEP005 Topical Gel, administered once daily for two consecutive days, by applying 90 µL of PEP005 Topical Gel over a 3 cm x 3 cm field surrounding a target actinic keratosis lesion comprising both diseased and perilesional skin. No hypothesis/inferential testing was conducted for this study. At the Day 29 (End of Study) assessment, complete clearance was reported in two (66.7%) patients in the 0.01% PEP005 Topical Gel cohort, one (33.3%) patient in the 0.025% PEP005 Topical Gel cohort, six (60.0%) patients in the 0.05% PEP005 Topical Gel cohort, and three (50.0%) patients in the 0.075% PEP005 Topical Gel cohort.

PEP005-017 was a randomised, double-blind, vehicle-controlled study to evaluate the pharmacokinetics of PEP005 (ingenol mebutate) Gel, 0.05%, when applied in a maximal use setting to the dorsal aspect of the forearm in patients with actinic keratosis. The primary objective was to evaluate the potential for systemic exposure of ingenol mebutate when applied in a maximal use setting to the dorsal aspect of the forearm in patients with actinic keratosis. The secondary objectives were to evaluate the safety and efficacy of PEP005 Gel, 0.05%, when applied in a maximal use setting to the dorsal aspect of the forearm in patients with actinic keratosis. Of the patients who received treatment with PEP005 Gel, 0.05%, 77% (10/13) had complete clearance of all SK lesions and all patients had partial clearance.

PEP005-018 was a multicentre, open-label study to examine the safety and tolerability of 0.05% PEP005 Topical Gel in patients with actinic keratoses on the dorsum of the hand. The objectives of the study were to examine the safety and toleration of PEP005 Gel, 0.05%, administered on two consecutive days, to a 25 cm² contiguous SK treatment area on the dorsum of a single hand and to examine the efficacy of PEP005 Gel, 0.05%, administered on two consecutive days, to a 25 cm² contiguous SK treatment area on the dorsum of a single hand. At Day 57, the complete clearance rate was 27.3% (3/11 patients) and the partial clearance rate was 45.5% (5/11 patients).

PEP005-013 was a Phase I, pharmacokinetic study to evaluate the extent of systemic absorption of PEP005, when applied as 0.05% PEP005 Topical Gel to a 100 cm² (5 cm x 20

cm) contiguous actinic keratosis treatment area on the extensor (dorsal aspect) forearm. No efficacy results were provided.

PEP005-022 was a multicentre, open-label, dose-area escalation, cohort study to evaluate the safety and tolerability of 0.05% PEP005 Topical Gel applied for two consecutive days to treatment area(s) of up to a total of 100 cm² in patients with actinic keratoses on the extensor (dorsal aspect) forearm(s). The objective of the study was to evaluate the safety and tolerability of two, once-daily, consecutive applications of PEP005 Gel, 0.05%, when applied to 25, 50, 75, or 100 cm² actinic keratosis treatment area(s) on the dorsal forearm(s). No efficacy results were provided.

AGN204332-004 was a multicentre, double-blind, parallel, randomised, vehicle-controlled study of the safety of a single application of up to 0.2 ml of 0.01% PEP005 gel to actinic keratoses on the shoulders, chest, back and/or arms followed by a post-treatment follow-up period lasting at least 2 weeks. A total of 80 lesions were treated (55 with 0.01% PEP005; 25 with PEP005 vehicle). Complete clearance in all five lesions at last available follow-up was reported for one patient treated with 0.01% PEP005. Another 0.01% PEP005 patient had complete clearance in 4/5 treated lesions. Extent of lesion clearance according to individual lesions showed that at Day 14, 8/55 lesions (15%) treated with active gel had complete clearance and by the last available follow-up, this had increased to 16/55 lesions (29%) treated with active gel.

PEP005-001 was a multi-centre, randomised, double-blind, parallel-group, vehicle-controlled study conducted in Australia between March and October 2005. The primary objective of the study was to determine the safety of PEP005 Topical Gel at 0.0025%, 0.01%, and 0.05% administered as two applications to patients with SK on the arms, shoulders, chest, face, and/or scalp under the following two treatment schedules: Day 1 and Day 2 or Day 1 and Day 8. The study was not statistically powered to conduct formal hypothesis/inferential testing.

Safety

Introduction

Information on safety was available from all clinical studies detailed above. In addition, a further 7 studies provided additional safety information. Of these studies, three assessed safety in healthy volunteers (PEP005-005, PEP005-023, and PEP005-024) and four provided safety data in patients receiving treatment for non-malignant skin cancer (NMSC) (PEP005-002, PEP005-003, PEP005-008 and PEP005-009). These studies are described below.

Evaluator's overall conclusions on clinical safety

- Information on safety was available from all clinical studies. A total of 1774 patients/subjects received PEP005 Gel across the 25 completed studies in the clinical development program. The safety overview provided focused on the 13 studies in which patients received PEP005 Gel across a selected area of skin (that is, field) for treatment of SK lesions. Within this group of patients, 1165 received PEP005 Gel and 632 received vehicle gel. Long term safety from 3 observational studies was also presented. In addition, a further 7 studies provided additional safety information. Of these studies, three assessed safety in healthy volunteers (PEP005-005, PEP005-023, and PEP005-024) and four provided safety data in patients receiving treatment for non-malignant skin cancer (NMSC) (PEP005-002, PEP005-003, PEP005-008 and PEP005-009).

- Of the supporting studies in healthy volunteers, PEP005-005 was a randomised, controlled study to evaluate the sensitizing potential of PEP005 Topical Gel (0.01% concentration) using a repeat insult patch test design. A total of 238 subjects were enrolled. There was no evidence of a sensitization potential or significant irritation following repeated applications of PEP005 Topical Gel (0.01% concentration). PEP005-023 was a 4 day, randomised, controlled, open application study to evaluate the photo-irritation potential of PEP005 (ingenol mebutate) Gel, 0.01%, using a phototoxicity test design. A total of 34 subjects were enrolled. There were no statistically significant differences between these irradiated treatment areas with respect to signs of photo-irritation. There was no significant irritation observed at the non-irradiated areas treated with the study medication or vehicle, and there was no statistically significant difference in signs of the photo-irritation between non-irradiated treatment areas. PEP005-024 was a randomised, controlled study to evaluate the photo-allergic potential of PEP005 (ingenol mebutate) Gel, 0.01% in healthy volunteers using an open application photo-allergic test design. A total of 60 subjects were enrolled. Neither PEP005 Gel, 0.01% nor the vehicle showed any potential for photosensitization.
- Of the supporting studies in patients receiving treatment for non-malignant skin cancer (NMSC), PEP005-002 was a multi-centre, randomised, double-blind, parallel-group, vehicle-controlled study to determine the safety of PEP005 Topical Gel, 0.0025%, 0.01%, and 0.05%, with two treatment schedules, Day 1 and Day 2 or Day 1 and Day 8 applications to nodular basal cell carcinoma. A total of 58 patients were enrolled. The study was not statistically powered to conduct formal hypothesis/inferential testing. No statistically significant differences were observed among treatment groups. PEP005-003 was a multi-centre, randomised, double-blind, parallel-group, vehicle-controlled study to determine the safety of PEP005 0.0025%, 0.01%, and 0.05% gel with two treatment schedules, Day 1 and Day 2 or Day 1 and Day 8 applications to superficial basal cell carcinoma. The study was not statistically powered to conduct formal hypothesis/inferential testing. Overall, the incidence of adverse events (AEs) was low. No patients enrolled in this study had any serious adverse events (SAEs). Of the 60 patients enrolled in this study, four patients were recorded to have experienced wound infections and three patients experienced a fall. PEP005-008 was a multi-centre, open-label study to determine the safety and efficacy of PEP005 0.05% Topical Gel in patients with cutaneous Squamous Cell Carcinoma in situ (SCCIS, Bowen's Disease). A total of 34 subjects were screened and 25 were enrolled in the study. Safety parameters were assessed using summary statistics. All 25 subjects experienced at least one expected local skin reaction (LSR) greater than baseline within the first week after receiving PEP005 0.05% Topical Gel. Of these, the most frequently reported responses were erythema (25/25; 100%), skin desquamation (20/25; 80%) and swelling (14/25; 56%). PEP005-009 was an open-label, multicentre, dose-escalation, cohort study to determine the maximum tolerated dose and safety of PEP005 Topical Gel given as either a single application (on Day 1) or as two applications (on Day 1 and Day 8) to a superficial basal cell carcinoma (sBCC) on the trunk. The most common treatment-related AEs among 47 patients treated at the 0.25% concentration were application site reactions (36% of patients in Arm 1 and 50% in Arm 2); these included pruritus, irritation and, less frequently, pain. Other possibly treatment-related AEs in the overall study population were limited to influenza and arthralgia in one patient each.
- From the overall safety database (across all SK field treatment studies), 42.5% of PEP005 Gel-treated patients had an AE compared with 24.2% of vehicle-treated patients. For PEP005 Gel-treated patients, the most frequently reported AEs considered related to study medication included application site pruritus (10.7%), application site pain (7.8%), and application site irritation (7.0%). Differences were

noted with respect to the incidence of treatment-related AEs between treatment locations. In the controlled Phase III studies, patients treated with PEP005 Gel on the face or scalp had a higher incidence of application site pain than patients treated on the trunk or extremities (13.9% versus 1.8%, respectively). Similarly, patients treated on the face or scalp had eye-associated disorders, such as eyelid oedema (1.1%) and periorbital oedema (2.6%), whereas patients treated on the trunk or extremities had no reports of these events. The majority of patients had an AE with maximum severity of mild or moderate intensity. Severe AEs were reported by 3.2% of PEP005 Gel treated patients and 1.6% of vehicle treated patients.

- From the overall safety database (across all SK field treatment studies), SAEs were identified for 4.2% of patients in the PEP005 Gel group and 3.6% of patients in the vehicle group. Of these SAEs, BCC (occurring in 1.5% of PEP005 Gel-treated patients and 1.1% of vehicle-treated patients) and SCC (0.9% of PEP005 Gel-treated patients and 0.8% of vehicle-treated patients) were the most frequently reported for both treatment groups. Three patients (all treated with PEP005 Gel, 0.05%) had an SAE that was assessed as treatment-related; 1 patient had Bowen's disease, graded as mild and 2 patients had SCC, 1 graded as mild and the other graded as moderate.
- Across all studies, there was one death, which occurred in a patient who received PEP005 Gel, 0.005% in Study PEP005-015. The patient, a 58-year-old White male, died of coronary artery atherosclerosis and hypertension approximately 4 months after receiving study treatment. There were no safety issues identified with regard to abnormal laboratory findings. No significant safety issues were identified as a result of discontinuations.
- Safety results from pivotal studies were consistent with the overall safety database, and did not raise any significant safety issues.
- Long term safety was assessed in 3 prospective, longitudinal, observational studies (PEP005-030, PEP005-031, and PEP005-032) that were designed to evaluate lesion recurrence and safety within the selected treatment area over a 12 month follow-up period in patients who had achieved complete clearance of AK lesions in studies PEP005-016, PEP005-020, PEP005-025, and PEP005-028. A total of 198 patients had demonstrated complete clearance of AK lesions (following treatment with PEP005 Gel [184 patients] or vehicle [14 patients]) at the Day 57 visit of the prior study and were enrolled in the long term, follow-up studies. During follow-up, no patient received PEP005 Gel, and 14 patients prematurely discontinued due to: withdrawal of consent (9 patients), protocol violation (2 patients), lost to follow-up (1 patient), investigator decision (1 patient) and inability to return to the study site for the 12 month visit (1 patient). Over 12 months of follow-up, 3 of the 198 patients had an AE within the selected treatment area that consisted of a mild sun burn, a moderate haematoma and a mild rash. All 3 AEs occurred approximately 8 to 9 months after the start of follow-up; all events resolved within 2 weeks of onset and all were considered not related to the study drug received during the prior study.

List of questions

There were no questions posed by the clinical evaluator to the sponsor.

Clinical summary and conclusions

Clinical aspects

Pharmacokinetics

- Ingenol mebutate, at the concentrations applied topically for treatment of SK lesions, has no detectable systemic absorption. This is supported by the human PK profile from allometric scaling as well as some limited PK data obtained from subjects in 4 clinical studies.

Pharmacodynamics

- No clinical studies on human pharmacodynamics were conducted. The mechanism of action in SK is not fully understood but appears to be a combination of induction of local lesion cell death and promotion of an inflammatory response with neutrophils and other immunocompetent cells.

Clinical efficacy

Dose-response studies and main clinical studies

- Information on clinical efficacy was provided for the 2 specific indications, the treatment of solar (actinic) keratoses (SK) on the face and scalp (0.015% Picato Gel), and for the treatment of SK on the body (non-head regions) (0.05% Picato Gel). For the first indication, PEP005-015, PEP005-016 and PEP005-025 were randomised, double-blind, vehicle-controlled, parallel-group studies and were the pivotal studies. PEP005-015 was a dose-ranging study that included the proposed dosage regimen treatment on head locations. PEP005-016 and PEP005-025 were Phase III studies that evaluated the proposed dosage regimen of Picato Gel for treatment of head locations. For the second indication, PEP005-014 and PEP005-028 were well-controlled Phase III studies that evaluated the proposed dosage regimen application of Picato Gel for treatment of non-head locations and should be regarded as the pivotal studies for this indication. Additionally, PEP005-006 was a Phase II, randomised, double-blind, double dummy, vehicle-controlled sequential cohort study which provided dose-ranging information for the second indication, although it included subjects with both face and scalp as well as body (non-head regions) SK.
- For the pivotal studies, a study design using an active comparator was not employed because of the potential to introduce bias with regard to the selection and timing of study endpoints. This was reasonable, as any available active comparator used in the same patient population represent different modalities of treatment. As a result, efficacy endpoints would have been measured at different times during a comparator trial. As an example, Imiquimod is approved for treatment on the face and scalp, and is available in two strengths. The first is a 5% cream (Aldara™) which in Australia is approved for cyclic treatment; 3 times per week for 4 weeks. After a 4-week no-treatment period another 4 week course can be applied if required. A continuous treatment period of 3 times per week for 16 weeks is also approved in Australia. All pivotal studies were vehicle-controlled.
- For the pivotal studies, the majority of SKs were diagnosed clinically, not histologically. In a study where 271 lesions were biopsied to correlate SK and SCC, clinical diagnosis and histopathological findings agreed in 91% of the biopsies. Punch biopsies of 220 clinically diagnosed untreated AKs were performed at baseline plus 51 lesions unresponsive to treatment (total, 271). Clinical diagnosis and histopathological findings agreed in 91% (246/271) of the lesions biopsied. The clinical diagnosis for the balance of the biopsied lesions was: (1) benign changes 4% (11/271) and (2) occult cutaneous malignancy in 5% (14/271) of the cases, 12 squamous cell

carcinomas and 2 basal cell carcinomas. Histological data were evaluated in a Phase I, lesion-specific study (PEP005-001). While only small numbers were involved, it did appear to support histological confirmation of clearance.

- For the pivotal studies, the comparative efficacy analyses for head and non-head locations used the same efficacy endpoints. For the combined studies populations, the primary efficacy endpoint was complete clearance, defined as the proportion of patients at Day 57 with no clinically visible SK lesions in the selected treatment area. For the head location, this endpoint was pre-specified as the primary endpoint in the two adequate and well-controlled Phase III studies (PEP005-016 and PEP005-025) and the Phase II dose-ranging study (PEP005-015). For the non-head location, this endpoint was pre-specified as the primary endpoint in the two adequate and well controlled Phase III studies (PEP005-014 and PEP005-028) but was a secondary endpoint in the Phase II dose-ranging study (PEP005-006). The secondary endpoint was partial clearance rate, defined as the proportion of patients at Day 57 with a 75% or greater reduction in the number of clinically visible SK lesions identified at baseline in the selected treatment area. Percent reduction from baseline in the total number of SK lesions at Day 57 was an additional endpoint. These efficacy endpoints were clinically appropriate. All pivotal studies were adequately powered to produce statistically significant results.
- For the head locations, results of the primary efficacy analyses were statistically significant for both pivotal studies. At Day 57, 37% and 47% of the PEP005 Gel-treated patients achieved complete clearance compared to 2% and 5% of vehicle gel-treated patients in Study PEP005-016 and PEP005-025, respectively (Study PEP005-016, $p < 0.001$; Study PEP005-025, $p < 0.001$). Findings from the secondary analysis were consistent between the studies; 60% and 68% of the PEP005 Gel-treated patients achieved partial clearance compared to 7% and 8% of vehicle gel-treated patients in Study PEP005-016 and PEP005-025, respectively (Study PEP005-016, $p < 0.001$; Study PEP005-025, $p < 0.001$). The additional efficacy endpoint of percent reduction from baseline in the number of SK lesions was also positive and consistent across the two studies (median of 83% and 87% in the PEP005 Gel groups for PEP005-016 and PEP005-025, respectively versus 0% in the vehicle gel group for each study).
- For the non-head locations, results of the primary efficacy analyses were statistically significant for both controlled studies. At Day 57, 28% and 42% of the PEP005 Gel-treated patients achieved complete clearance compared to 5% in each of the vehicle gel groups in Study PEP005-014 and PEP005-028, respectively (Study PEP005-014, $p < 0.001$; Study PEP005-028, $p < 0.001$). Findings from the secondary analysis were consistent between the studies; 44% and 55% of the PEP005 Gel-treated patients achieved partial clearance compared to 7% in each of the vehicle gel groups in Study PEP005-014 and PEP005-028, respectively (Study PEP005-014, $p < 0.001$; Study PEP005-028, $p < 0.001$). The additional efficacy endpoint of percent reduction from baseline in the number of SK lesions was also positive and consistent across the two studies (median of 69% and 75% in the PEP005 Gel groups for PEP005-014 and PEP005-028, respectively versus 0% in the vehicle gel group for each study).

Clinical studies in special populations

Not applicable.

Analysis performed across trials (pooled analyses and meta-analysis)

- For the face and scalp indication, data were pooled across studies and referred to as “combined studies populations”. Two combined studies populations were presented. One combined studies population pooled data from PEP005-016 and PEP005-025 and another pooled data PEP005-016, PEP005-025, and PEP005-015. For the trunk and extremities indication, data were pooled across studies and referred to as “combined

studies populations". Two combined studies populations were presented. One combined studies population pooled data from PEP005-014 and PEP005-028 and another pooled data from PEP005-014, PEP005-028 and PEP005-006.

- For the combined face and scalp data, 42% of PEP005 Gel patients achieved complete clearance compared with 4% of vehicle gel patients ($p < 0.001$). In addition, 64% of PEP005 Gel patients achieved partial clearance compared to 7% of vehicle gel patients ($p < 0.001$). For the combined trunk and extremities data, 34% of PEP005 Gel patients achieved complete clearance compared with 5% of vehicle gel patients ($p < 0.001$). In addition, 49% of PEP005 Gel patients achieved partial clearance compared to 7% of vehicle gel patients ($p < 0.001$).

Supportive studies

- Three clinical studies provided information on long term efficacy. PEP005-030 was a long term follow-up study in patients who achieved complete clearance at Day 57 in studies PEP005-016 and PEP005-025 for the face and scalp indication. Two trials (PEP005-031 and PEP005-032) were long term follow-up studies in patients who achieved complete clearance at Day 57 in previous trials for the trunk and extremities indication. No study medication was administered during these studies. In PEP005-030 at 12 months of follow-up, 54% of patients who had been treated with PEP005 Gel in the previous Phase III studies (N=108), had at least one new or recurrent AK lesion within the selected treatment area. The estimated median time to lesion recurrence was 365 days. Based on the number of lesions observed within the treatment area during 12 months of follow-up relative to the number of lesions at baseline (determined prior to treatment with PEP005 Gel in the Phase III studies), the mean lesion-based recurrence rate was 13%. For the combined PEP005-031 and PEP005-032 at 12 months of follow-up, 56% of patients who had been treated with PEP005 Gel in the previous Phase III studies (N=76), had at least one new or recurrent AK lesion within the selected treatment area. The estimated median time to lesion recurrence was 274 days. Based on the number of lesions observed within the treatment area during 12 months of follow-up relative to the number of lesions at baseline (determined prior to treatment with PEP005 Gel in the Phase III studies), the mean lesion-based recurrence rate was 13%. A further 9 clinical studies provided supporting information on efficacy. These studies were PEP005-007, PEP005-020, PEP005-004, PEP005-017, PEP005-018, PEP005-013, PEP005-022, AGN204332-004 and PEP005-001. These studies were either open-label, involved small numbers, or had limited statistical power.

Clinical safety

Patient exposure

- A total of 1774 patients/subjects received PEP005 Gel across the 25 completed studies in the clinical development program. The safety overview provided focused on the 13 studies in which patients received PEP005 Gel across a selected area of skin (that is, field) for treatment of SK lesions. Within this group of patients, 1165 received PEP005 Gel and 632 received vehicle gel.

Adverse events

- A total of 42.5% of PEP005 Gel-treated patients had an AE compared with 24.2% of vehicle-treated patients. For PEP005 Gel-treated patients, the most frequently reported AEs considered related to study medication included application site pruritus (10.7%), application site pain (7.8%), and application site irritation (7.0%). Differences were noted with respect to the incidence of treatment-related AEs between treatment locations. In the controlled Phase III studies, patients treated with PEP005 Gel on the face or scalp had a higher incidence of application site pain than

patients treated on the trunk or extremities (13.9% vs. 1.8%, respectively). Similarly, patients treated on the face or scalp had eye-associated disorders, such as eyelid oedema (1.1%) and periorbital oedema (2.6%), whereas patients treated on the trunk or extremities had no reports of these events.

- The majority of patients had an AE with maximum severity of mild or moderate intensity. Severe AEs were reported by 3.2% of PEP005 Gel-treated patients and 1.6% of vehicle-treated patients. Severe events occurred at a higher frequency for patients treated with PEP005 Gel compared to vehicle in the SOC of general disorders and administrative site conditions, with application site reactions (such as irritation, pain and pruritus) attributed for this difference between treatment groups.

Serious adverse events and deaths

- SAEs were identified (by both the investigator and Applicant) for 4.2% of patients in the PEP005 Gel group and 3.6% of patients in the vehicle group. Of these SAEs, BCC (occurring in 1.5% of PEP005 Gel-treated patients and 1.1% of vehicle-treated patients) and SCC (0.9% of PEP005 Gel-treated patients and 0.8% of vehicle-treated patients) were the most frequently reported for both treatment groups. Across all studies, there was one death, which occurred in a patient who received PEP005 Gel, 0.005% in Study PEP005-015. The investigator judged the death as not related to study medication.

Laboratory findings

- There were no meaningful shifts or trends in any of the clinical laboratory parameters.

Safety in special populations

Not applicable.

Immunological events

Not applicable.

Safety related to drug-drug interactions and other interactions

Not applicable.

Discontinuation due to adverse events

- In the 13 studies that evaluated field treatment of PEP005 Gel for SK lesions, a total of 3 patients (1 treated with PEP005 Gel and 2 treated with vehicle) discontinued from the study due to one or more AEs. In the 13 SK field treatment studies, a total of 37 patients (37 treated with PEP005 Gel and 0 treated with vehicle) discontinued study medication due to one or more AEs. No significant safety issues were identified as a result of these discontinuations.

Benefit risk assessment

Benefits

From the clinical study information provided, the complete clearance rates of PEP005 Gel, 0.015% on head locations were 37% and 47%, and for PEP005 Gel, 0.05% on non-head locations, the complete clearance rates were 28% and 42%. These results were statistically significant in comparison to vehicle gel ($p < 0.001$, for each study and for the pooled Phase III data for each location [head and non-head]). These results appear comparable with currently marketed products (figures provided by the applicant were 15-58% for 5-FU; 34-47% for diclofenac; and 26-46% for imiquimod). A table summarising results for Aldara (imiquimod) is shown below.

Table 4. Trials of topical treatments of actinic keratoses

<i>Trials*</i>	<i>Treatment regimen</i>	<i>No. of participants</i>	<i>Complete response (%)</i>	<i>Partial response† (%)</i>
Diclofenac 3% (Solaraze) in hyaluronan 2.5% gel				
Wolf, et al., 2001 ³⁸	Twice daily for 90 days	120	Treatment: 47.0 Vehicle: 19.0	—
Imiquimod 5% cream (Aldara)				
Korman, et al., 2005 ³⁵	Once daily, three times per week, for 16 weeks	492	Treatment: 48.3 Vehicle: 7.2	Treatment: 64.0 Vehicle: 13.6
Lebwohl, et al., 2004 ³³	Once daily, two times per week, for 16 weeks	436	Treatment: 45.1 Vehicle: 3.2 (95% CI, 34.9 to 49)‡	Treatment: 59.1 Vehicle: 11.8 (95% CI, 39.5 to 55.1)‡
Szeimies, et al., 2004 ³⁴	Once daily, three times per week, for 16 weeks	286	Treatment: 57.1 Vehicle: 2.2	Treatment: 72.1 Vehicle: 4.3

CI = confidence interval.

*—Randomized, double-blind, vehicle-controlled, phase III trials.

†—75 percent reduction in actinic keratoses.

‡—CIs are for the treatment group minus the vehicle group.

Information from references 33 through 35 and 38.

McIntyre WJ et al. Treatment options for actinic keratoses. *Am Fam Physician* 2007;76(5):667-71

PEP005 Gel treatment, however, is efficacious after being applied for a substantially shorter duration. For head (face and scalp), PEP005 Gel, 0.015% is applied for three days and for non-head (trunk and extremities), PEP005 Gel, 0.05% is applied for only two days. In contrast, the applicant noted that other products have longer durations of treatment. Treatment with 5-FU requires at least 3 to 4 weeks diclofenac requires 8 to 12 weeks and imiquimod requires 4 weeks of treatment with efficacy assessment after the following 4 week off treatment period or 16 weeks of continuous treatment. Longer treatment durations reduce patient compliance. Treatment compliance with the PEP005 Gel dosing regimen in the Phase III studies was noted to be 99%. It must be noted, however, that the safety and efficacy of PEP005 Gel treatment was not directly compared with any active comparators.

PEP005 Gel applied topically at the concentrations used for treatment of SK lesions, has no systemic absorption, whereas, the applicant noted that measurable plasma concentrations occur with use of 5-FU, diclofenac and imiquimod.

A follow-up period of 12 months was selected for each of the long term studies. At the 12 month follow-up, patient-based recurrence was 54% for head (face and scalp) locations and 56% for non-head (trunk and extremities) locations. The applicant noted that these results are consistent with patient-based recurrence for imiquimod (42-67%). Lesion-based recurrence at 12 month follow-up was 13% for PEP005 Gel treated patients in each location (head and non-head). These findings are also consistent with lesion-based recurrence of 9% observed for imiquimod.

Risks

Adequate information was provided to establish a side-effect profile for this product. A total of 43% of patients treated with PEP005 Gel experienced an AE. Most AEs could be attributed to application site reactions and were typically considered related to treatment. Application site pruritus, application site pain, and application site irritation were the most frequently reported. Only 3.2% of PEP005 Gel-treated patients had a severe AE. Discontinuation of treatment due to an AE occurred in only 3% of patients who received PEP005 Gel; discontinuation from the study occurred in 0.1% of patients. Serious AEs occurred at a low incidence (4% of PEP005 Gel patients) and most were unrelated to study medication. One death occurred in a PEP005 Gel-treated patient (due to coronary

atherosclerosis and hypertension) and was considered unrelated to study medication. Neoplasms such as BCC and SCC of the skin were infrequent and occurred at similar incidences for PEP005 Gel and vehicle patients. This profile appeared to compare favourably with currently marketed products. While therapeutic failure could present a possible risk, results from the clinical studies program suggest that this would be comparable to any of the active comparators currently marketed. The nature of the treatment of AK using any medication requires clinical follow-up for monitoring of both therapeutic failure and possible recurrence, especially in chronically solar-damaged skin.

Four studies provided data in patients receiving treatment for non-malignant skin cancer (NMSC) (PEP005-002, PEP005-003, PEP005-008 and PEP005-009). While these studies were included primarily to contribute to the safety database, they did provide some efficacy data on utility in alternative diagnoses. However for the majority of these studies, the analyses were primarily descriptive in nature, and were not statistically powered to conduct formal hypothesis/inferential testing. Conditions treated included nodular BCC, superficial BCC, and SCCIS.

Eye disorders, however, were seen more frequently in patients treated with PEP005 Gel (2.3% versus 0.3% in vehicle patients). Local skin responses were common with PEP005 Gel treatment; 95% of PEP005 Gel-treated patients showed an increase in LSR scores relative to baseline whereas most patients treated with vehicle showed no change from baseline LSR score. For patients treated on the face or scalp, the maximum LSR score occurred on Day 4, which returned to baseline values (or below) by Day 15. For patients treated on the trunk or extremities, the maximum LSR score occurred between Days 3 and 8 and returned to baseline values (or below) by Day 29. For both treatment locations (face/scalp and trunk/extremities), erythema and flaking/scaling were the most common LSRs, followed by crusting and swelling. Local skin responses on the face and scalp generally were of greater intensity than responses on the trunk and extremities. The majority of patients who received PEP005 Gel showed no signs of hypopigmentation, hyperpigmentation or scarring at baseline or the end-of-study assessments.

No clinically meaningful changes in laboratory parameters, vital signs or electrocardiogram (ECG) assessments were seen with PEP005 Gel treatment. Localised application site disorders (pruritus, pain, irritation) and local skin responses, particularly erythema, flaking and scaling are the main characteristics of the safety profile of PEP005 Gel. These local adverse events are transient and typically resolve without sequelae within 2-4 weeks of application. Additionally, PEP005 Gel is not systemically absorbed. The applicant noted that comparator products also report local skin responses but these appear more intense and several products recommend "rest-periods" when severe skin reactions are experienced (imiquimod and 5-FU). This is consistent with current clinical experience. No tabular summary comparing adverse event rates based on clinical trial data was provided. The applicant also noted that comparator products also report systemic absorption when applied topically and systemic adverse events such as flu-like symptoms and fatigue (imiquimod).

The formulation proposed for marketing was used in the pivotal studies (0.015% Picato Gel in studies PEP005-016 and PEP005-025, 0.05% in studies PEP005-014 and PEP005-028). A valid rationale for the lack of active comparators was provided. While the data set provided only shows that Picato Gel is better than placebo, adequate information was provided to suggest that efficacy was comparable to currently available comparators. While a non-inferiority study would be preferred, this risk would be mitigated by adequate postmarketing surveillance. The superiority margins in the pivotal studies were appropriate and clinically meaningful.

Balance

On balance, the benefits of PEP005 Gel appear to be at least comparable to other currently marketed comparators. In addition, other benefits of PEP005 Gel include a shorter duration of treatment with probable improved compliance as a result. The risks associated with the safety profile of PEP005 Gel appear reduced compared to that of marketed treatments. Overall the risk-benefit profile of PEP005 Gel is considered favourable.

Conclusions

PEP005 Gel appears to be efficacious in the treatment of SK with complete clearance rates of 42% for the face and scalp (95% confidence interval of 36.4, 48.3) and 34% for the trunk and extremities (95% confidence interval of 27.9, 40.6). The short duration of treatment (2 to 3 days) appears to contribute to improved treatment compliance. PEP005 Gel has no detectable systemic absorption even at concentrations applied topically which are effective for complete clearance of SK lesions. At the 12 month follow-up, patient-based recurrence was 54% for head (face and scalp) locations and 56% for non-head (trunk and extremities) locations. Lesion-based recurrence at 12 month follow-up was 13% for each (head and non-head locations).

Localised application site disorders (pruritus, pain, irritation) and local skin responses, particularly erythema, flaking, and scaling are the main characteristics of the safety profile of PEP005 Gel. These local adverse events are transient and typically resolve without sequelae within 2 weeks of application to the face or scalp and within 4 weeks of application to the trunk or extremities.

PEP005 Gel has a shorter duration of treatment compared to other products and it is considered to offer comparable short term efficacy. With the shorter duration of treatment, efficacy appears to be achieved more rapidly following study medication application. The risks associated with the safety profile of PEP005 Gel appear reduced compared to that of marketed treatments, based on information provided by the applicant and clinical experience. Overall the risk-benefit profile of PEP005 Gel is considered favourable.

Recommended conditions for registration and Product Information

Conditions for registration

On the basis of the information provided by the applicant, the application for registration of Picato Gel is supported based on the proposed indications.

V. Pharmacovigilance findings

Risk management plan

The sponsor submitted a Risk Management Plan which was reviewed by the TGA's Office of Product Review (OPR).

Safety specification

The summary of the Ongoing Safety Concerns as specified by the sponsor is as shown in Table 5 below.

Table 5. Ongoing Safety Concerns

Important identified risks	Local skin responses Eye disorders
Important potential risks	Actinic keratoses (AK) to squamous cell carcinoma (SCC) progression Overdose after treatment of multiple locations
Important missing information	Retreatment with ingenol mebutate gel Populations that are Immunocompromised/immunosuppressed

OPR reviewer comment

It is recommended that the above summary of the Ongoing Safety Concerns is considered acceptable. It is noted that with the updated RMP (Version 2) the summary of Ongoing Safety Concerns has been updated. *Accidental eye exposure* and *Ingestion* have been removed from Important potential risks. However, both of these risks are now listed under the *Potential for Medication error* section (in RMP). Overdose after treatment of multiple locations has been added as an Important potential risk. *'Retreatment with ingenol mebutate gel'* and *'Populations that are Immunocompromised/immunosuppressed'* have been added as two areas of Important missing information. The sponsor states the reason for including these safety concerns is due to review of the Risk Management Plan by the European medicines Agency (EMA).

In regards to the Important potential risk *'Retreatment with ingenol mebutate gel'* the sponsor states that treatment of multiple locations could occur. There has been no experience in the clinical setting of overdose after simultaneous administration of ingenol mebutate gel to multiple locations of the body. In addition, the sponsor states that the product information describes the proper use of the product. This is considered acceptable.

Pharmacovigilance plan

Routine and additional pharmacovigilance activities are proposed to monitor all safety concerns. The following additional pharmacovigilance activities have been proposed (Table 6):

Table 6. Additional pharmacovigilance activities.

Additional pharmacovigilance activity	Assigned safety concern	Study Design	Sample size	Duration	Status
1. Risk of Squamous Cell Carcinoma on skin area treated with Ingenol Mebutate Gel, 0.015% and Imiquimod	Actinic keratoses (AK) to squamous cell carcinoma (SCC) progression	RCT	480	36 months	Proposed
2. Ingenol Mebutate Gel, 0.015% Repeat Use for Multiple Actinic Keratoses on Face and Scalp	Retreatment with ingenol mebutate gel	RCT	454	12 months	Ongoing (includes Australian patients)

RCT=Randomised control trial

1. Risk of squamous cell carcinoma on skin area treated with Ingenol Mebutate Gel, 0.015% and Imiquimod

In the Pharmacovigilance Plan (Page 40 of the RMP Version 1) the sponsor states that they commit to initiating a study in order to measure the long term outcome with respect to development of SCC (Important potential risk - AK to SCC Progression) but no study details were provided. In the sponsor's response to the TGA request for information, the sponsor has provided a study protocol for this study (RMP, annex 5):

This is a Phase III multi-centre, randomised, parallel, open label, controlled, 36-month trial to compare the cumulative incidence of SCC after treatment with ingenol mebutate 0.015% and imiquimod 5% cream for multiple AKs on the face and scalp.

OPR reviewer's comments in regard to the pharmacovigilance plan and the appropriateness of milestones

The routine activities that the sponsor has outlined are consistent with the activities outlined in 3.1.2 Routine pharmacovigilance practices. Note for Guidance on Planning Pharmacovigilance Activities (CPMP/ICH/5716/03).

The proposed long term outcome study is considered acceptable. The sponsor states in the study protocol that the first subject visit is planned for the second quarter of 2013 and the last subject visit is planned for second quarter of 2018. However, it is recommended that the sponsor provide milestones for the planned date for submission of final data to the TGA.

2. Ingenol Mebutate Gel, 0.015% Repeat Use for Multiple Actinic Keratoses on Face and Scalp

This ongoing study is considered acceptable. The sponsor states in the study protocol that the first subject visit is planned for second quarter of 2012 and the last subject visit is planned for fourth quarter of 2013. However, it is recommended that the sponsor provide milestones for the planned date for submission of final data to the TGA.

Important potential risks - Overdose after treatment at multiple locations

In the proposed pharmacovigilance plan the sponsor states that routine pharmacovigilance will be used to monitor 'Overdose after treatment at multiple locations'. However, the sponsor describes in the Summary of the Risk Management Plan that "Additional pharmacovigilance activities include a planned trial will investigate the

possibility of overdose after treatment in multiple locations". No further information about this planned study is provided. It is recommended that the sponsor confirm if routine or additional pharmacovigilance is planned for '*Overdose after treatment at multiple locations*'. If additional pharmacovigilance is planned then the sponsor should provide a protocol for this study and a justification to explain how this study is appropriately designed to provide a greater understanding of this Important potential risk. This justification may include, but not be limited to, factors such as study design, outcome measurements (primary and secondary), sample size, duration of treatment exposure and inclusion/exclusion criteria.

Risk minimisation activities

No additional risk minimisation activities are planned by the sponsor for Picato gel.

OPR reviewer comment

In regard to the proposed routine risk minimisation activities, it was recommended to the Delegate that the draft Product Information and Consumer Medicine Information documents were satisfactory.

Summary of recommendations

The OPR provides these recommendations in the context that the submitted RMP is supportive to the application;

It is recommended that the Delegate:

- Implement RMP Version 2, data lock point [01 February 2012], dated 20 April 2012, including the sponsor's response to the TGA's section 31 request for information/documents and any future updates as a condition of registration.

It is recommended to the Delegate that the sponsor:

1. Provide milestones for the planned date for submission of final data to the TGA for the studies:
 - Risk of Squamous Cell Carcinoma on skin area treated with Ingenol Mebutate Gel, 0.015% and Imiquimod
 - Ingenol Mebutate Gel, 0.015% Repeat Use for Multiple Actinic Keratoses on Face and Scalp
2. Confirm if routine or additional pharmacovigilance is planned for '*Overdose after treatment at multiple locations*'. If additional pharmacovigilance is planned then the sponsor should provide a protocol for this study and a justification to explain how this study is appropriately designed to provide a greater understanding of this important potential risk. This justification may include, but not limited to, factors such as study design, outcome measurements (primary and secondary), sample size, duration of treatment exposure and inclusion/exclusion criteria.

VI. Overall conclusion and risk/benefit assessment

The submission was summarised in the following Delegate's overview and recommendations:

Quality

There are no outstanding chemistry and quality control issues that preclude registration.

The stability data support the proposed shelf life of two years when stored in the refrigerator at 2-8°C. Freezing did not affect the product and the additional storage condition of 'Do not freeze' is not required.

No bioavailability data were submitted and this was acceptable as the product is for topical use.

It was also considered by PSC at its 144th meeting; all issues raised have been resolved.

The evaluator recommended registration from a chemistry point of view.

Nonclinical

The nonclinical evaluator recommended approval.

Primary pharmacology: The mechanism of action is not clearly understood. Studies indicate that it could be due to cytokine induction or associated inflammatory response that is responsible for its efficacy in actinic keratosis (AK)/solar keratosis (SK). Though its efficacy was seen towards a variety of tumour derived cell lines, there were marked differences observed in different tumour cell lines suggesting different intra and inter-individual responsiveness.

No other clinically significant effect was seen.

Pharmacokinetics: Ingenol mebutate levels in blood were generally below the lower limit of quantification (LLOQ) after topical application in mice and pigs. It was present in low concentration in rats. There was little or no induction or inhibition of major CYP isoforms.

Repeat dose toxicity studies: IV ingenol mebutate induced death in repeat dose toxicity studies due to thrombi in lung vessels. Exposure ratios at the NOAELs suggested a "very large margin" between human predicted systemic exposure and levels that causes AEs in animal models. The evaluator mentions that, "*topical application of ingenol mebutate to rat or minipig skin produced dose-dependent, mild to severe reactions of erythema, oedema, skin thickening, encrustations, desquamation and necrosis.*"

The evaluator mentions that standard mutation assays (using both bacterial and mammalian cells) produced negative results. Carcinogenicity studies were not performed even though there was a positive result in the SHE cell transformation assay. The evaluator expresses concern as ingenol mebutate is closely related to phorbol esters which are potent activators that are also potent tumour promoters. The sponsor should state whether it plans to undertake a carcinogenicity study, in its pre-ACPM response. If not, there should be an adequate justification.

There was an increased incidence of embryofetal mortality in rats and a few foetal visceral and skeletal variations noted in an intravenous embryofetal developmental toxicology study.

Several amendments to the PI were recommended and adopted, except for the following:

"Ingenol mebutate was not mutagenic in an in vitro Ames test or a mouse lymphoma assay but gave a positive response in the Syrian hamster embryo cell in vitro transformation assay." The evaluator recommends that this statement be included. The reason for this is that, "relative to the concurrent control, an increase in micronucleated PCE was seen at IV doses of 5 and 10 µg/kg/day, which was not statistically-significant but the numbers were higher than historical control data and there was an indication of a dose-related increase".

Overall, the evaluator recommends approval from a nonclinical perspective.

Clinical

Pharmacokinetics

The evaluator mentions that ingenol mebutate at the concentrations applied topically does not result in systemic absorption. This was confirmed in pharmacokinetic data obtained in clinical studies. Levels of ingenol mebutate or its metabolites (acyl isomers) were not detected, with a lower limit of quantification at 0.1 ng/mL. Of note:

Study PEP005-017 was a randomised double blind vehicle controlled study where 0.05% ingenol mebutate was applied in a maximal use setting on the dorsal aspect of the forearm in patients with AK. No systemic absorption was detected.

Study PEP005-013 was also a pharmacokinetic study where ingenol mebutate 0.05% was applied to a 100 cm² area of solar keratosis in the dorsal aspect of the arm in 6 patients on two consecutive days. No systemic absorption was observed.

Pharmacodynamics

The evaluator mentions that no human pharmacodynamic studies were conducted. Thus, there are no pharmacokinetic/ dynamic correlation studies performed.

The evaluator also mentions that the mode of action is established based on nonclinical studies.

Efficacy

Studies relating to the two indications, treatment of SK on the face and scalp (0.015% Picato Gel) and for the treatment on the body (non-head regions, 0.05% Picato Gel) are considered separately.

Face and scalp indication:

The clinical studies to support the indication (extracted from the evaluation report) are as shown in Figure 4 above.

Of these, PEP005-015 and PEP005-006 were dose response studies. Two other studies (PEP005-016 and 025) were pivotal, Phase III studies. Study PEP005-030 was a long term follow-up study in patients who achieved complete clearance at Day 57 in studies PEP005-016 and PEP005-025.

Dose response studies (PEP005-015 and PEP005-006):

PEP005-015: This is a Phase II dose ranging study which was randomised double blind, vehicle controlled study. Three dose strengths (0.005%, 0.01% and 0.015%) were compared to placebo in treatment groups of 30 patients each. The treatment was administered once daily for 2 or 3 consecutive days to a 25 cm² SK treatment area on the face and scalp. Patients were evaluated on Day 57 for complete clearance (see *Efficacy* studies).

The evaluator presented the study findings. There were statistically significant changes seen versus placebo. A dose response was seen with the 2 day regimen in relation to complete clearance (not with the three day regimen). The numbers were too small to detect significant differences between dose strengths; also, the study was not powered to do this. However, the study provides a crude measure to support 0.015 % as the preferred dose strength.

PEP005-006: This is also a Phase II dose finding study. It was a multicentre randomised double blind vehicle controlled study where 0.025% and 0.050% were used for three consecutive days. There was a two treatment arm of 0.050% also. Details are included in

the account of the study by the evaluator. This study included both head/scalp and trunk/extremities AK subjects. The complete clearance rate (with the 95% CI) is included on page 39. The 0.05% treatment group fared better than the vehicle and the 0.025% treated groups. The two day treatment was better than three day treatment.

Other studies:

Study PEP005-007 was an open label multicentre dose escalation study on 94 patients. 0.0025%, 0.005%, 0.0075%, 0.0125%, 0.0175% or 0.025% ingenol mebutate gel were used once daily for 2-3 consecutive days. The analyses were descriptive in nature and did not provide meaningful results as the numbers were too small.

Histological evidence of clearance:

Study PEP005-001 was a multicentre randomised double blind parallel group vehicle controlled study conducted in Australia. 0.0025%, 0.01% and 0.05% were applied as two applications using two different schedules: Day 1 and Day 2 (treatment arm A) and Day 1 and Day 8 (treatment arm B). Subjects were to have at least 5 solar keratosis lesions on the arms, shoulders, chest, face or scalp. Histological clearance was assessed on Day 85 on the ITT population.

The analysis of this was of a descriptive nature and is as follows:

Table 7. Histological response to treatment (punch Biopsy) at the end of study (Day 85). ITT population.

Treatment	Vehicle Gel N=12		PEP005 0.0025% N=17		PEP005 0.01% N=16		PEP005 0.05% N=18	
	Arm A N=6	Arm B N=6	Arm A N=9	Arm B N=8	Arm A N=8	Arm B N=8	Arm A N=9	Arm B N=9
End of Study								
Number of 2 mm Punch Biopsies	6	6	8	7	7	8	8	7
Presence of AK	3	4	7	4	3	5	6	3
Absence of AK	3	2	1	3	4	3	2	4
Difference PEP005 vs. Vehicle	-	-	0.3750	-0.0952	-0.0714	-0.0417	0.2500	-0.2381
95% CI ^a	-	-	(-0.1332, 0.8832)	(-0.6800, 0.4895)	(-0.6747, 0.5318)	(-0.5981, 0.5147)	(-0.3012, 0.8012)	(-0.8228, 0.3466)
P-value ^b	-	-	0.2448	1.0000	1.0000	1.000	0.5804	0.5921
P-value ^c Treatment Arm A	0.2637							
P-value ^c Treatment Arm B	0.9126							

Note: Percentages are based on the number of non-missing biopsies within each treatment group.

^a Two-sided confidence interval from normal approximation to the binomial.

^b P-value from Fisher's exact test comparing individual doses to vehicle gel.

^c P-value from Fisher's exact test comparing all treatment groups.

As seen in the table above, the numbers having punch biopsy were small. The clinical clearance rate however was statistically significantly superior to vehicle a.

It appears that no other studies examined histological clearance. The sponsor should confirm this in its pre-ACPM response.

Efficacy studies:

In these studies complete clearance was defined as the proportion of subjects at Day 57 after treatment with no visible AK lesions in the 'selected treatment area'. Selected treatment area was defined as a continuous 25 cm² treatment area of skin with 4 to 8 clinically typical, visible and discrete AK lesions. Recurrence was defined as any number of clinically visible AK lesions 12 months after complete clearance in the selected area at Day 57.

Two studies are pivotal (PEP005-016 and PEP005-025).

Study PEP005-016 was a multicentre (USA and Australia) randomised parallel group double blind vehicle controlled study where ingenol mebutate 0.015% gel was used to treat actinic keratosis on the face or scalp. The treatment was to be applied to a contiguous 25cm² area on the face or scalp. Those with 4-8 clinically typical, visible and discrete AK lesions within a contiguous 25 cm² treatment area on the head region (face or scalp) were eligible to enrol.

Those who had cosmetic or therapeutic procedures within two weeks and within 2 cm of the selected area were excluded. Similarly, those treated with immunomodulators, interferon/interferon inducers or systemic medications that suppress the immune system within 4 weeks, treatment with 5-FU, imiquimod, diclofenac or photodynamic therapy within 8 weeks and within 2 cm of the selected treatment area. Female subjects must be of either non-child bearing potential or childbearing potential providing negative serum and urine pregnancy test and using effective contraception.

Efficacy assessments were conducted at baseline and on Day 57 (end of study). The evaluator mentions that "*the primary efficacy endpoint was complete clearance rate of SK lesions at the Day 57 visit. A patient with no clinically visible SK lesions in the selected treatment area was defined to have complete clearance. The secondary efficacy endpoint was the partial clearance rate of SK lesions at the Day 57 visit. A patient with a 75% or greater reduction in the number of clinically visible SK lesions identified at baseline, in the selected treatment area was defined to have partial clearance. The primary efficacy analysis was based on the intent-to-treat (ITT) population. All treatment comparisons were tested with two-tailed tests and a 0.05 significance level.*"

Sample size calculations were adequate and the study was of adequate power to detect a clinically significant difference between the active and placebo groups.

A total of 269 patients were randomised (135 to PEP005 0.015% and 134 to vehicle gel) and 259 patients completed the study.

The following results were extracted from the evaluation report.

Table 8. Complete clearance rate of actinic keratoses lesions on Day 57, Overall and by anatomical location. ITT population.

	PEP005 Gel 0.015% (N = 135)	Vehicle Gel (N = 134)	P value
Overall			
Complete Clearance Rate [n/N (%)]	50/135 (37.0)	3/134 (2.2)	<0.001 ^a
95% Confidence Interval ^b	28.9, 45.8	0.5, 6.4	
Breslow Day P value ^c			0.574
Face			
Complete Clearance Rate [n/N (%)]	46/109 (42.2)	3/109 (2.8)	<0.001 ^d
95% Confidence Interval ^b	32.8, 52.0	0.6, 7.8	
Scalp			
Complete Clearance Rate [n/N (%)]	4/26 (15.4)	0/25	0.110 ^d
95% Confidence Interval ^b	4.4, 34.9	0.0, 13.7	

^a P values are from Cochran-Mantel-Haenszel test, stratified by analysis site. The P values ≤ 0.05 are considered statistically significant.

^b Confidence intervals are calculated using the exact binomial distribution (Clopper-Pearson).

^c P values ≤ 0.10 are considered statistically significant.

^d P values are from Fisher's Exact test treatment group comparison. The P values ≤ 0.05 are considered statistically significant.

Statistically significant superiority was demonstrated in relation to complete clearance rate; it was also demonstrated with partial clearance rate.

Study 025 was similar in design to PEP005-016 where a total of 278 patients were randomised (142 to PEP005 0.015% and 136 to the vehicle). The following results were obtained in relation to the primary efficacy endpoint (complete clearance).

Table 9. Complete clearance rate of actinic keratoses lesions on Day 57, Overall and by anatomical location. ITT population.

	PEP005 Gel 0.015% (N = 142)	Vehicle Gel (N = 136)	P value
Overall			
Complete Clearance Rate [n/N (%)]	67/142 (47.2)	7/136 (5.1)	<0.001 ^a
95% Confidence Interval ^b	38.8, 55.7	2.1, 10.3	
Breslow Day P value ^c			0.306
Face			
Complete Clearance Rate [n/N (%)]	58/111 (52.3)	6/111 (5.4)	<0.001 ^d
95% Confidence Interval ^b	42.6, 61.8	2.0, 11.4	
Scalp			
Complete Clearance Rate [n/N (%)]	9/31 (29.0)	1/25 (4.0)	0.031 ^d
95% Confidence Interval ^b	14.2, 48.0	0.1, 20.4	

^a P values are from Cochran-Mantel-Haenszel test, stratified by analysis site. The P values ≤ 0.05 are considered statistically significant.

^b Confidence intervals are calculated using the exact binomial distribution (Clopper-Pearson).

^c P values ≤ 0.10 are considered statistically significant.

^d P values are from Fisher's Exact test treatment group comparison. The P values ≤ 0.05 are considered statistically significant.

Partial clearance rates also showed statistically significant superiority.

Recurrence of AK

PEP005-030 is a 12 month extension study for AKs recurrence and local safety, see page 43. This was a prospective longitudinal follow up study of subjects with AKs who

demonstrated complete clearance by Day 57 in Studies 016 and 025. Subjects were not re-treated. All who completed the Day 57 visit and demonstrated complete clearance of AK lesions were eligible to participate. A total of 117 patients demonstrated complete clearance and 9 of these were in the vehicle treated at Day 57. At 12 months follow up, 54% of subjects (58/108) had at least 1 or more recurrent AK lesions.²⁰

Trunk and extremities indication

The number and type of studies are shown in Figure 5 above.

Pivotal efficacy studies: (PEP005-014 and 028)

These studies were Phase III vehicle controlled studies conducted in the trunk/extremities. The design was similar to the pivotal studies of face and scalp.

PEP005-014

A total of 126 patients were enrolled to PEP005 0.05% and 129 to the vehicle group.

The results in the ITT population by anatomical location were shown in the clinical report.

Of note:

Table 10. Results by anatomical location. ITT population.

Clinical assessment	PEP005, 0.05% (N=126)	Vehicle (N=129)	p-value
Observed complete clearance n (%)	35 (27.8%)	6 (4.7%)	<0.0001
95% CI	(20.2%, 36.46%)	(1.7%, 9.9%)	
Difference between treatment groups [95%CI]	23.13% [14.5%, 31.8%]		

The evaluator mentions that, “the observed partial clearance rate at Day 57 overall in the PEP005 Gel, 0.05% group was 44% (56/126) versus 7% (9/129) in the vehicle group ($p < 0.0001$). Overall, the complete clearance rate at Day 57 was significantly higher in the PEP005 Gel, 0.05% group relative to the vehicle group. In addition, the other efficacy variables supported the results of the primary efficacy endpoint.”

Study PEP005-028

A total of 203 patients were enrolled (100 to 0.05% and 103 to vehicle). The complete clearance rate was 42 (42%), [95% CI 32.2, 52.3%] in the active group versus 5 (4.9%) [1.6%, 11.0%] in the placebo group, $p < 0.001$. Details by anatomical site were given in the clinical report. Clearly the numbers were too small to yield conclusive results. The partial clearance rate at Day 57 overall was statistically significantly higher in the PEP005 Gel, 0.05% group (55%) than the vehicle group (7%) ($p < 0.001$).

²⁰ Sponsor comment: “This is a Kaplan-Meier estimate and cannot be directly translated into a number of subjects having recurrence in the study, since both recurrent (n=56) and censored (n=52) subjects are taken into consideration.”

Long term studies (PEP005-031 and 032)**Study PEP005-031**

This is a 12 month follow up study of patients who completed Day 57 assessment in Study PEP005-020. No study medication was administered. The event rates, that is SK recurrence is summarised using KM methods, as per the long term face and scalp follow up study (PEP 005-030). The evaluator mentions that *“At 12 months of follow-up, 50% of patients treated with PEP005 Gel, 0.05% in Study PEP005-028 (N=38) had at least one new or recurrent SK lesion within the selected treatment area”*.

Study PEP005-032

This was a 12 month follow up of those who completed Day 57 in PEP005-028 and was similar in design to the other long term studies. A total of 42 patients were enrolled (38 to the active and 5 to the vehicle). The estimated recurrence rates were included in the clinical report. It is stated that *“At 12 months of follow-up, 62.5% of patients in the CC57 population treated with PEP005 Gel, 0.05% in Study PEP005-020 (N=38) had at least one new or recurrent AK lesion within the selected treatment area. The estimated median time to lesion recurrence was 274 days.”*

Supporting studies

Studies PEP005-018, 020, 040, 022 and AGN204 204332-004 also were considered supporting data because of the nature of design (mostly open label), different dosing regimen (some studies) and small number of patients. They in essence confirmed the results of the pivotal studies in relation to efficacy. These studies were summarised in the CER and will not be discussed further.

Safety

The summary of patient exposure is discussed under *Clinical Findings*. The details of this (extracted from the US Summary basis for approval) is given below.

Table 11. US Summary of Patient Exposure

Population	No (N) of subjects who received at least one dose of study medication	
	PEP005 gel	Vehicle Gel
AK lesions, field treatment (13 trials)	1165	632
Phase III pivotal vehicle controlled trials: face/ scalp (2 trials)	274	271
Phase III pivotal vehicle controlled: trunk/extremities (2 trials)	225	232
AK lesions, lesion specific treatment (2 trials)	57	17
Topical safety, healthy volunteers (3 trials)	332	*
NMSC (4 trials)	220	24

*All subjects in the dermal safety trials received PEP005 Gel and vehicle gel; NMSC-nonmelanotic squamous cell carcinoma

Adverse events: There were 42% in the active groups versus 24.2% in the vehicle treated groups. This increase was attributed to higher incidence of application site reaction (22.7% versus 2.8%).

There were also pruritus, application site pain and irritation. In the controlled clinical Phase III studies these were reported more in the face and scalp regions (13.9% versus 1.8%). The face and scalp studies also reported eyelid oedema (1.1%) and periorbital oedema (2.6%).

The evaluator reports that most were mild to moderate in severity. Some 3.2 % and 1.6% of the patients reported severe adverse events.

SAEs occurred in 4.2% (in the active) versus 3.6% in the vehicle treated group. BCC and SCC were the most frequently reported SAEs for both treatment groups. Serious BCC occurred in 1.5% of PEP005 Gel group and 3.6% in the vehicle group. Serious SCC occurred in 0.9% of PEP005 Gel treated group and in 0.8% of vehicle treated patients. There was one death reported in a patient who received PEP005 Gel; this was a 58 year old man who died of coronary artery atherosclerosis 4 months after receiving study treatment. The investigator judged the death as not study related.

Across the 13 field application AK studies, 37 (3.2%) subjects in the active treatment group discontinued treatment versus 0% in the vehicle group. They were essentially application site reactions, application site pain, erythema and skin exfoliation.

The evaluator mentions that the “*vast majority of clinical laboratory parameters*” were within normal limits.

Overall conclusion: The evaluator recommends registration.

Risk management plan

The RMP evaluator mentions additional pharmacovigilance activities, that is, two studies that are to be conducted;

1. Risk of Squamous Cell Carcinoma on skin area treated with Ingenol Mebutate Gel, 0.015% and Imiquimod and
2. Ingenol Mebutate Gel, 0.015% Repeat Use for Multiple Actinic Keratoses on Face and Scalp.

The study protocols are considered acceptable and the evaluator recommends that the mile stones for the planned date of submission to the TGA should be submitted.

Other routine pharmacological activities and statements in the PI and CMI are considered satisfactory.

Risk-benefit analysis

Delegate considerations

1. Efficacy studies are placebo controlled studies only. The comparative efficacy versus registered products is not known.
2. The PI should stipulate that the target population selected in the studies has 4 to 8 clinically typical, visible and discrete AK lesions in a 25 cm² area of continuous skin. There are no data on applying to greater areas of affected skin.

Similarly, the durability of effect is not known. The long term studies on head/face and trunk areas report recurrence at 3 months. These data should be included in the

PI. At present, there are no studies submitted on re-treatment. This should also be stated in the PI.

3. Histological confirmation of efficacy is lacking. This should be stated in the PI.

Delegate's actions

The Delegate proposed to approve ingenol mebutate Picato 0.015% and 0.05% Gel in packs containing 3 or 2 Tubes (respectively) of 0.47 g for the topical treatment of solar (actinic) keratoses (AK) in adults.

The Committee's advice was sought.

Response from sponsor

LEO Pharma is pleased to acknowledge that Picato gel has been recommended for approval by the Delegate following positive recommendations from all scientific evaluators.

Pharmaceutical chemistry issues

The Delegate commented that the stability data support the proposed shelf life of two years stored in refrigerated conditions at 2-8°C and that freezing did not affect the product. The sponsor confirms that this is correct and agrees to remove the "Do not freeze" statement from the packaging material.

Nonclinical issues

The Delegate requests clarification as to whether a carcinogenicity study is planned. Due to lack of systemic exposure in the clinical setting, the risk of tumour formation in man is regarded as negligible. The question of local skin tumours has been assessed in 6 and 9 months studies in rats and mini pigs and no neoplastic changes were noted in the skin. In addition, a recent study in SKH-1 mice was completed.²¹ The study demonstrated that prophylactic treatment of ultraviolet B (UVB)-damaged skin with 2 x 0.05% ingenol mebutate resulted in a significant reduction (approximately 70%) in the number of p53+ keratinocyte patches and it was concluded that the eradication of UV-damaged keratinocytes combined with the obliteration of cutaneous immunosuppressive mast cells contributed to the overall reduction in UVB lesions following ingenol mebutate treatment. The mentioned dermal studies in mice, rats and mini pigs clearly indicate that ingenol mebutate is not a strong tumour promoter in these species which is in line with the other investigations performed that is, PKC modulation, ODC inducing activity and structural-activity relationship (sponsor's Nonclinical Overview and Report no. HCPEP-005-001). Also, findings are consistent with the low tumour response elicited in the Adolf et al. mouse skin tumour promotion study²², although the doses used in this study were toxic and somewhat interfered with data interpretation. Ingenol mebutate was negative in two *in vitro* and one *in vivo* standard genotoxicity assays and positive in the Syrian hamster embryo (SHE) cell transformation test at concentrations $\geq 0.1 \mu\text{g/mL}$ (24h) and $\geq 0.025 \mu\text{g/mL}$ (7d). However, as systemic exposure is below 0.1 ng/mL (LLOQ) in man and the usefulness of the SHE assay as a predictive tool is debatable, it may be difficult to assess the relevance of the SHE assay in carcinogenicity risk assessment (sponsor's Nonclinical Overview).

²¹ Cozzi SJ, Ogbourne SM, James C, Rebel HG, de Gruijl FR, Ferguson B, et al. Ingenol Mebutate Field-Directed Treatment of UVB-Damaged Skin Reduces Lesion Formation and Removes Mutant p53 Patches. *J Invest Dermatol.* 2011

²² Adolf W, Chanai S, Hecker E. 3-o-Angeloylingenol, the toxic and skin irritant factor from latex of *Euphorbia antiquorum* L. (Euphorbiaceae) and from a derived Thai purgative and anthelmintic (vermifuge) drug. *J Sci Soc Thailand.* 1983;9:81.

Numerous *in vitro* cytotoxicity assays have demonstrated ingenol mebutate activity against more than 100 different tumour cell lines with active inhibition of cell growth at varying degrees of potency (Study Reports P093; P115A; P115B). In nonclinical studies, topical application of ingenol mebutate gel once daily for 3 days cured a number of different murine and human tumours implanted subcutaneously in mice including mouse melanoma cell line, B16.²³ These *in vitro* and *in vivo* studies clearly demonstrate that ingenol mebutate has potent anti-cancer properties.

Due to the irritative properties of ingenol mebutate a traditional 2 year dermal carcinogenicity study in rats or mice with repeated daily administration is not considered feasible and dosing with concentrations much below clinical strengths is not considered meaningful. As ingenol mebutate gel is only to be used 2-3 times, an application pattern of 3 consecutive treatment days once monthly is feasible (and has been done for 6 months in rats and 9 months in mini pigs as mentioned above). Two-three applications are however unlikely to induce tumours probably even with TPA. Normally, a tumour promoter needs to be applied repeatedly to have a tumour promoting effect. The clinical regimen for ingenol mebutate gel consists of daily dosing for only 2 or 3 consecutive days which is not considered sufficient to drive tumour promotion even in the event of retreatment.

Chronic tissue injury is generally recognised as a risk factor for human carcinogenesis and there is a strong association between chronic inflammatory conditions in a particular tissue and cancer specific to that tissue.²⁴ Ingenol mebutate gel does not induce chronic tissue injury if used as prescribed. The skin irritation observed in repeat dose toxicity studies in rats and mini pigs was of an acute nature and did not progress over time. Also, in the clinical setting, ingenol mebutate gel demonstrated favourable cosmesis with no signs of chronic skin irritation.

Hyperplasia seen in nonclinical species in dermal toxicology studies was associated with inflammation indicating the hyperplasia is due to irritation. Hyperplasia, by definition, remains responsive to control and in the absence of the causative stimulus it will not progress. Hyperplasia seen in animals treated dermally with ingenol mebutate demonstrated no progressive potential and was reversible in both rats and mini pigs.

Ingenol mebutate is a short chain ingenane ester. Structural-activity relationship studies conducted to assess the relative tumour promoting potential of ingenol-3-esters has demonstrated that long chain esters were tumour promoting, whereas short chain esters were not. Specifically, esters of six carbons or less were considered "very weak to weak promoters".

Ingenol mebutate is considered a weak tumour promoter more comparable to the nonpromoting phorbol prostratin and 12-deoxyphorbol 13-phenylacetate (dPP) than the classical tumour promoter TPA. Also, "phorbol esters" with unsaturated side chains tend to be inflammatory but not tumour promoting. The sponsor believes that the potential skin tumour formation is more easily and accurately monitored in the clinical setting. The sponsor has thus decided to initiate a study in order to measure the long term outcome with respect to development of SCC (Study Protocol LP0041-63). A clinical study is considered to be of more value than non-clinical studies.

The Delegate requests a recommendation to include the following statement in the PI:

²³ Ogbourne SM, Suhrbier A, Jones B, Cozzi SJ, Boyle GM, Morris M, et al., Antitumour activity of 3-ingenyl angelate: plasma membrane and mitochondrial disruption and necrotic cell death. *Cancer Res.* 2004;64(8):2833-9.

²⁴ Alden CL, et al. A critical review of the effectiveness of rodent pharmaceutical carcinogenesis testing in predicting for human risk. *Vet Pathol* 2011 May;48(3):772-84.

"Ingenol mebutate was not mutagenic in an in vitro Ames test or a mouse lymphoma assay but gave a positive response in the Syrian hamster embryo cell in vitro transformation assay":

Some text seems to be missing in the Delegate's comment and it is not completely clear what text is being proposed. The sponsor acknowledges the positive response of the SHE assay and agrees this should be reflected in the Product Information (PI). However, the sponsor is not of the opinion that the result of the *in vivo* micronucleus assay is equivocal as suggested in the Nonclinical Assessor's evaluation report. Therefore, the sponsor proposes the text below based on the clarification previously provided in the "errors and omissions" document for the evaluation reports submitted by the sponsor in July 2012, as it was considered a misinterpretation of the data. Proposed PI text:

"Ingenol mebutate was not mutagenic in an in vitro Ames test, mouse lymphoma assay, and in vivo rat micronucleus test, and gave a positive response in the Syrian hamster embryo cell in vitro transformation assay".

Clinical issues

The sponsor acknowledges the Delegate's comment on histological clearance data. The sponsor confirms that at present histological clearance has not been examined in studies other than PEP005-001. Therefore, the sponsor accepts the Delegate's request to this information in the PI, the proposed text to go into the *Clinical Trial Section* is:

"These studies did not assess clearance by histology".

Please be informed that a histology study is planned as an EU Post Authorisation Measure. Data from this study is expected to be available for submission to TGA in April 2014.

The Delegate commented that the efficacy studies are placebo controlled studies only and that the comparative efficacy versus registered products is not known. It is acknowledged that comparative studies have not been carried out.

However, a systematic literature review and meta-analysis have been made by the sponsor showing a risk-benefit ratio for ingenol mebutate comparable to other therapies for SK (the review can be forwarded upon request). In addition, the sponsor has agreed to conduct a clinical study investigating ingenol mebutate versus imiquimod in an active control setting as a Post Authorisation Measure in the EU (see section below).

The Delegate requested to include information in the PI on the population selected in the studies: The sponsor would like to refer to the PI, *Clinical Trial Section*, where the following is stated for face and scalp:

"Patients had 4 to 8 clinically typical, visible, non-hyperkeratotic, non-hypertrophic discrete SK lesions on the face or scalp within a contiguous 25 cm² treatment area."

and for body:

"Patients had 4 to 8 clinically typical, visible, nonhyperkeratotic, non-hypertrophic discrete SK lesions on the trunk or extremities within a contiguous 25 cm² treatment area."

The sponsor therefore finds that the Delegate's request to describe the target population of the clinical studies is addressed in the PI.

The Delegate commented that there are no data on application to greater area of affected skin. The sponsor kindly refers to the *Clinical Trial Section*, Experience of treatment of a larger area, where the following is stated:

"In a double-blind, vehicle-controlled study to evaluate systemic exposure, ingenol mebutate 0.05% gel, from 4 single dose tubes, was applied to a 100 cm² contiguous treatment area daily for 2 consecutive days. Results demonstrated no systemic absorption. Picato gel, 0.05%

was well tolerated when applied to a contiguous treatment area of 100 cm² on trunk and extremities”

This statement is based on results from Clinical Study PEP005-017.

The Delegate commented that the durability of effect is not known and that the long term studies on head/face and trunk areas report recurrence at 3 months. These data are requested to be included in the PI. The durability of the effect has been studied in the long term studies. The median time to recurrence was 365 days for patients treated on the face and scalp, and 274 days for patients treated on the trunk and extremities (sponsor’s Summary of Clinical Efficacy). The sponsor suggests to include the median time to recurrence in the PI.

The Delegate comments that at present there are no studies submitted on retreatment and that this should be stated in the PI. The sponsor confirms that at present no studies on re-treatment have been conducted, however as presented in the section below, the sponsor has initiated a study investigating re-treatment.

The sponsor accepts to include the following under the *Clinical Trial Section* of the PI: For face and scalp:

“Clinical data on treatment for more than one Picato gel 0.015% treatment course for 3 days is not available”

and for body:

“Clinical data on treatment for more than one Picato gel 0.05% treatment course for 2 days is not available.

Risk management plan issues

The Delegate requests that milestones for the planned dates of submission of the SCC study and the Repeat use study are submitted to the TGA.

Milestones to planned dates of final data to the TGA:

1. Risk of Squamous Cell Carcinoma on skin area treated with ingenol mebutate gel, 0.015% and imiquimod (LP0041-63).

The sponsor plans to submit the CSR to TGA October 2018.

2. Ingenol mebutate gel, 0.015% Repeat Use for Multiple Actinic Keratoses on Face and Scalp (LP0041-22).

The sponsor plans to submit the CSR to TGA April 2014

The RMP evaluator requests the sponsor to confirm if routine or additional pharmacovigilance is planned for *“Overdose after treatment at multiple locations”*. The sponsor would like to confirm that routine pharmacovigilance is planned as management for the potential risk of *“Overdose after treatment at multiple locations”*. Routine pharmacovigilance will likewise be used to assess this potential risk in any future studies investigating ingenol mebutate. For awareness, non-regulatory studies are planned in the future to investigate the use of ingenol mebutate on more than one treatment area.

The RMP was at the time currently under review by the Committee for Medicinal Products for Human Use (CHMP) and the sponsor proposes to submit an updated version to TGA once the CHMP opinion has been adopted. The target date was 20 September 2012.

Summary

The Product Information and Consumer Medicine Information has been updated as discussed above and recommended by the Delegate.

Notes to the ACPM: The US Product Information (USPI) is generally in line with the Australian PI and is based on the same data package. There are a few differences in the information stated in the USPI compared to the Australian PI, such as the squamous cell carcinoma (SCC) data from the long term studies which are not presented in the USPI. In addition, there are a few warnings not present in the USPI following the FDA request to remove these during evaluation.

Note that there have been no serious unexpected adverse drug reactions which have not been submitted to the TGA previously.

Advisory committee considerations

The Advisory Committee on Prescription Medicines (ACPM), having considered the evaluations and the Delegate's overview, as well as the sponsor's response to these documents, advised the following:

The ACPM, taking into account the submitted evidence of efficacy, safety and quality, agreed with the delegate and considered these products to have an overall positive benefit–risk profile for the following indication;

Picato gel is indicated for the topical treatment of solar (actinic) keratoses (AK) in adults

In making this recommendation the ACPM noted that the products have demonstrated sufficient evidence of efficacy balanced by a predictable and acceptable safety profile. The ACPM were also of the view that the products offered increased efficacy due to increased likelihood of patient compliance with the treatment regimen.

The ACPM agreed with the Delegate's proposed amendments to the Product Information (PI) and Consumer Medicine Information (CMI); however, expressed concern that patients may self or misdiagnose lesions and the patient information and education should caution against unsupervised use.

The ACPM agreed with the Delegate on the proposed conditions of registration and specifically advised on the inclusion of the following:

- Submission of the additional studies agreed by the sponsor, particularly on re-treatment, in view of the nature of the medical condition.

The ACPM advised that the implementation by the sponsor of the recommendations outlined above to the satisfaction of the TGA, in addition to the evidence of efficacy and safety provided would support the safe and effective use of these products.

Outcome

Based on a review of quality, safety and efficacy, TGA approved the registration of Picato gel (ingenol mebutate 0.015% and 0.05%) for topical administration, indicated for:

Picato gel is indicated for the topical treatment of solar (actinic) keratoses in adults.

Specific conditions applying to these therapeutic goods

1. The implementation in Australia of the Picato gel (Ingenol mebutate) 150 mcg/g (0.05%) and 500 mcg/g RMP, (Version 5.0, dated 14 August 2012), included with submission PM-2011-02307-3-5, and any subsequent revisions with any accompanying caveats and requests for pharmacovigilance activities as agreed with the TGA and its Office of Product Review.

Attachment 1. Product Information

The Product Information approved at the time this AusPAR was published is at Attachment 1. For the current Product Information please refer to the TGA website at <http://www.tga.gov.au/hp/information-medicines-pi.htm>.

Attachment 2. Extract from the Clinical Evaluation Report

Therapeutic Goods Administration

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<http://www.tga.gov.au>

Reference/Publication #