azitra

NYSE: AZTR

Staphylococcus epidermidis for the topical treatment of epidermal growth factor receptor (EGFR) inhibitor-induced dermal toxicity

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Society of Investigative Dermatology

Precision dermatology powered by synthetic biology.

SPECIAL NOTE REGARDING FORWARD-LOOKING STATEMENTS

This document contains forward-looking statements concerning Azitra, Inc. ("Azitra", the "Company," "we," "us," and "our"). The words "believe," "may," "will," "potentially," "estimate," "continue," "anticipate," "intend," "could," "would," "project," "plan," "expect" and similar expressions that convey uncertainty of future events or outcomes are intended to identify forward- looking statements. These forward-looking statements include, but are not limited to, statements concerning the following:

- our future financial and operating results;
- our intentions, expectations and beliefs regarding anticipated growth, market penetration and trends in our business;
- the timing and success of our plan of commercialization;
- our ability to successfully develop and clinically test our product candidates.

These forward-looking statements are subject to a number of risks, uncertainties and assumptions, including (i) we are an early-stage clinical biopharmaceutical company with limited operating history, (ii) there are no drug products to date that incorporate our microbial library and genetic engineering platform and the clinical and commercial utility of our microbial library and genetic engineering platform is uncertain and may never be realized; (iii) we have only recently commenced Phase 1 clinical studies of our initial product candidates and our product candidates will require extensive additional preclinical and clinical testing; (iv) we expect we will need additional financing to execute our business plan and fund operations, which additional financing may not be available on reasonable terms or at all; and (v) those other risk described in "Risk Factors" section of the prospectus ("Prospectus") dated June 15, 2023 filed by Azitra with the Securities and Exchange Commission on June 21, 2023.

In light of these risks, uncertainties and assumptions, the forward-looking events and circumstances discussed in this document may not occur and actual results could differ materially and adversely from those anticipated or implied in our forward-looking statements. You should not rely upon forward-looking statements as predictions of future events. Although we believe that the expectations reflected in our forward-looking statements are reasonable, we cannot guarantee that the future results, levels of activity, performance or events and circumstances described in the forward-looking statements will be achieved or occur. Azitra does not undertake and specifically disclaims any obligation to update or revise our forward-looking statements to reflect new circumstances or unanticipated events as they occur, except as required by law.

Personal disclosures

Employment/compensation

Current

- Azitra Inc. (NYSE: AZTR) (Cofounder, COO)*
- Actuate Therapeutics Inc. (Cofounder)*
- Inspired Spaces LLC (Cofounder)*
- LetsImproveHealth LLC (Founder)*
- Umbrex LLC (Healthcare management consultant)*
- Yale University (Assistant Professor Adjunct)

Former

- Bios Partners LP (Former Partner)*
- Bios Research (Former Senior Analyst)
- Cue Biopharma (NASDAQ: CUE) (Former consultant)
- Encore Vision (Former consultant)
- Novartis (Former consultant)
- TFF Pharma (NASDSQ: TFFP) (Former consultant)

Shareholder

- Azitra Inc. (NYSE: AZTR)*
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- Opus Genetics*
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- Stream Biomedical*
- Trefoil Therapeutics*

Current Board of Directors

For-profit

- Azitra Inc. (NYSE: AZTR)*
- IN8bio (NASDAQ: INAB)*

Non-profit (No financial interest)

- International Network for Simulation-based Pediatric Innovation, Research, and Education (INSPIRE) (Treasurer)
- International Pediatric Simulation Society (IPSS) (Treasurer)

Grant/research support (PI)

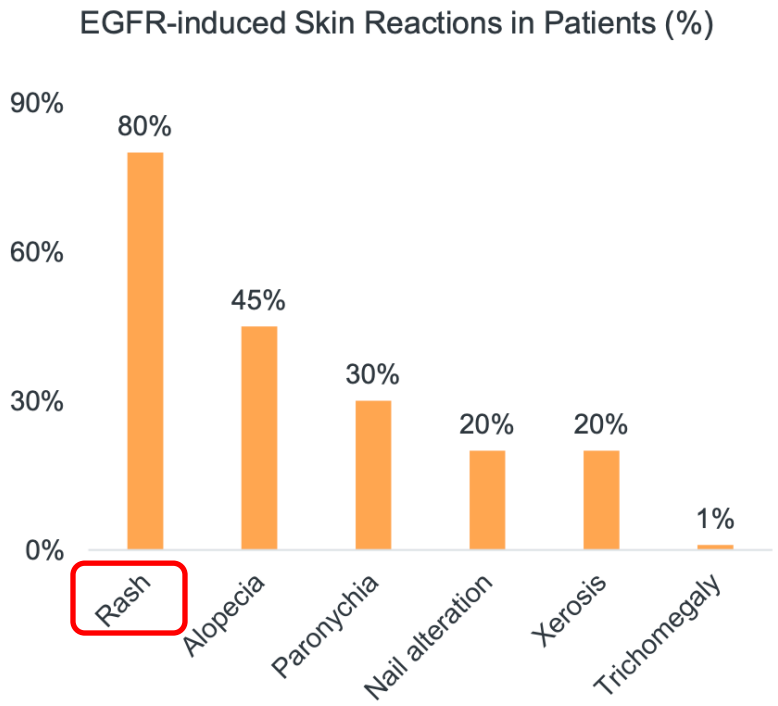
- Connecticut Innovations
- Connecticut Biosciences
- Defense Advanced Research Projects Agency (DARPA)
- Department of Defense (DoD)
- Indiana University
- Macy Foundation
- National Institutes of Health – NIAMS
- National Institutes of Health – NIAID
- National Science Foundation (NSF)
- Thiel Foundation
- Yale University

***Current financial interest**

EGFRi-induced dermal toxicity is highly prevalent with significant clinical impact

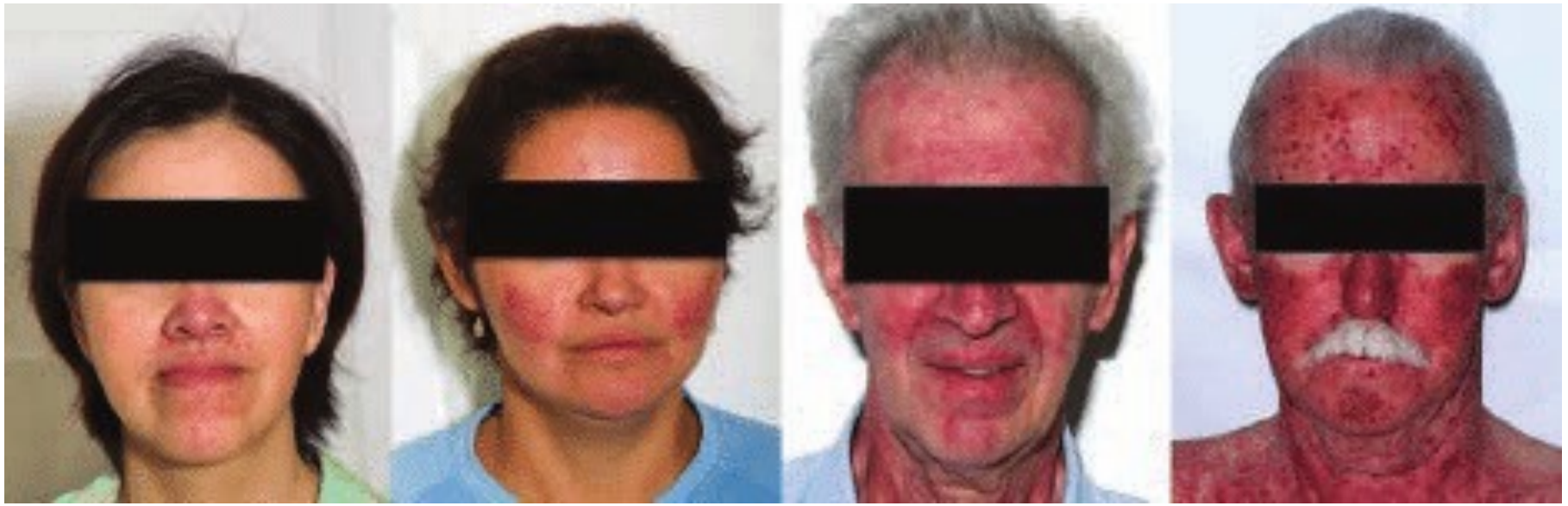
EGFR inhibitors cause dermal toxicity in many patients generating dysbiosis and inflammatory response; the rash is strongly linked to *S. aureus* colonization and elevated IL-36γ

- Dermatologic toxicities are the most prevalent side effects seen with the current EGFR inhibitors
- Similar toxicities occur in patients undergoing other cancer therapies, such as toxicity associated with immuno-oncology therapy, MEK inhibitors, or in radiation dermatitis
- The earliest to occur, most predictable, and most common adverse event of EGFR inhibitors is papulopustular rashes, affecting 60-90% of patients
- Management of the rash is essential to improve quality of life and avoid changes in needed treatment
- EGFRi-associated dermal toxicities cause suppression of cutaneous immunity are driven by elevations in *S. aureus* and IL-36γ, a pro-inflammatory cytokine
- **Concerns with the limited topical and oral treatments and their associated adverse events and interference with EGFRi efficacy have led to a clear need for effective and safe adjunct therapies**



Source: Fabbrocini G et al. Skin Appendage Disord, 2015

EGFRi-induced dermal toxicity is highly prevalent with significant clinical impact

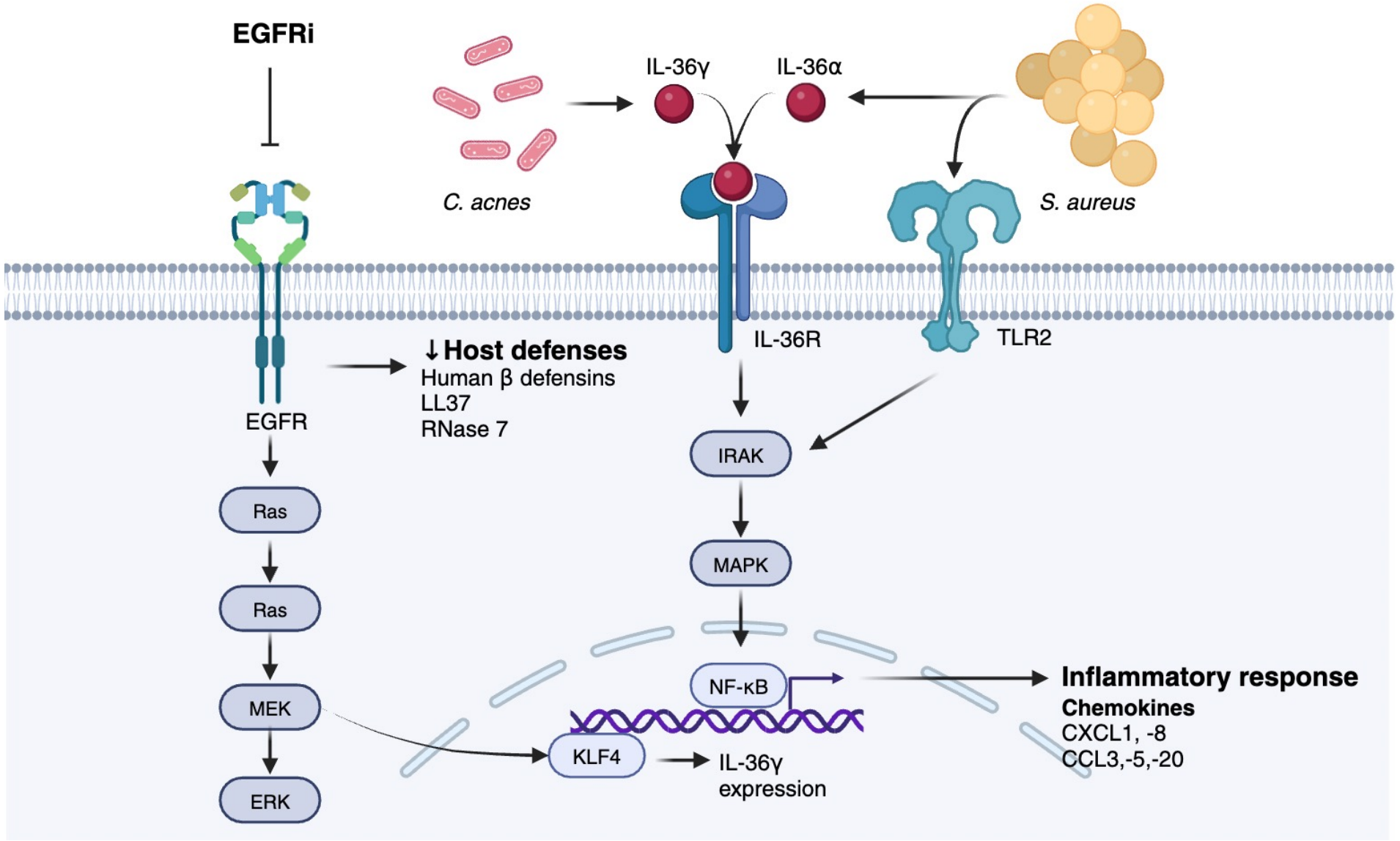


Grade 1	Grade 2	Grade 3	Grade 4

- Rash severity often linked to cancer drug dosing and correlates with *S. aureus* levels on the skin
- Rash can lead to significant changes in course of therapy and QOL
- Depending on generation of EGFRi, as many as 90% develop a rash, and **as many as 15-20% discontinue EGFRi therapy due to skin rash**
- ~150,000 patients on EGFR inhibitors in the US

Source: Melosky et al. (2015). Grade 1, gefitinib; grade 2, erlotinib; grade 3, erlotinib; grade 4, erlotinib

Etiology of EGFRi-driven rash: suppression of host defenses and concurrent inflammatory response



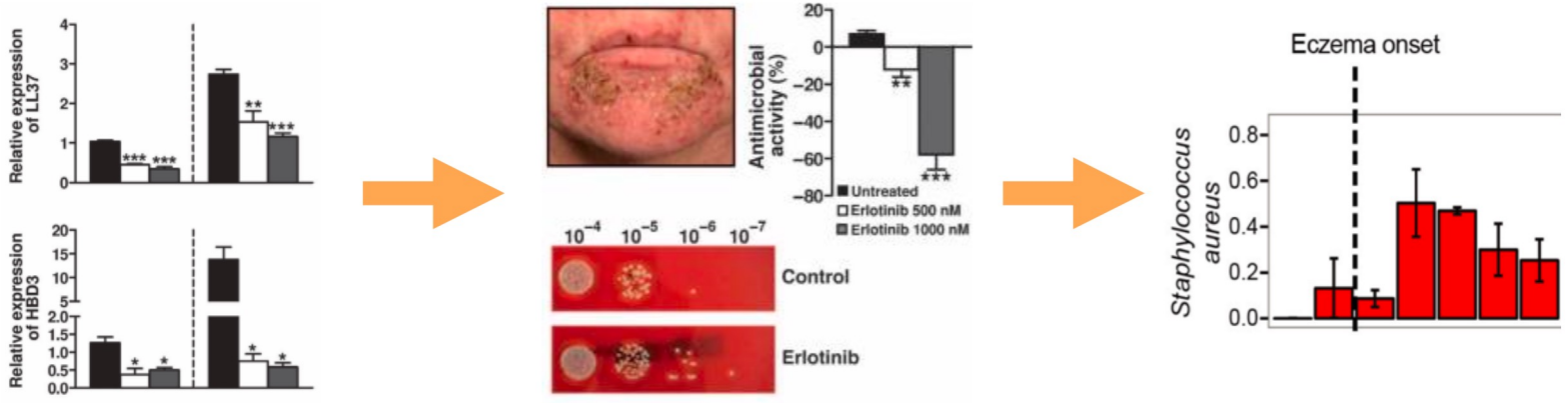
Clinical evidence of *S. aureus* and IL-36γ elevations in patients experiencing EGFRi-related skin toxicity

EGFR inhibition impairs the cutaneous immune defense of keratinocytes allowing *S. aureus* colonization

Decreased antimicrobial peptides in the skin

Decreased antimicrobial activity

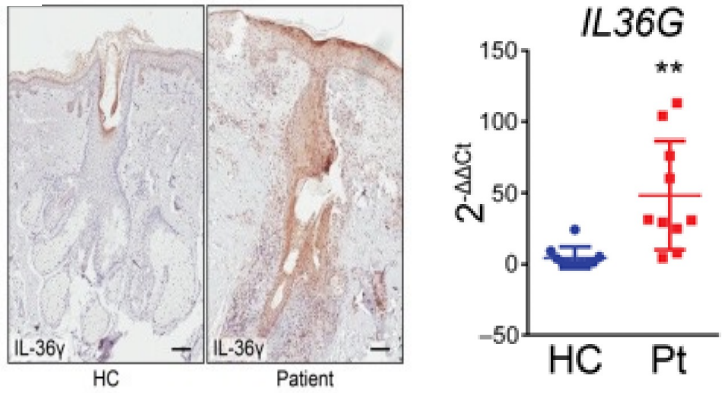
Increased *S. aureus*



Lichtenberger et al., 2013 Sci Translational Med

EGFR inhibition leads to significant increases in IL-36γ

Increased IL-36γ in patient skin vs. healthy control



Satoh et al., 2020 JCI

ATR-04: auxotrophic *S. epidermidis* for EGFR inhibitor-associated rash

ATR-04 Summary

- Severity of the dermal toxicity is linked to IL-36γ signaling as well as correlations to *S. aureus* increases
- **ATR-04,** a drug product containing *S. epidermidis* strain **SE484,** is topically administered and inhibits IL-36γ and *S. aureus* with the following properties:
  - Isolated from healthy volunteer and engineered to be auxotrophic to control growth with D-alanine
  - Inhibits IL-36γ
  - Reduces *S. aureus*
  - Increases human beta defensin 2
- Pre-IND meeting with FDA held in April 2024 with IND filing expected mid-2024

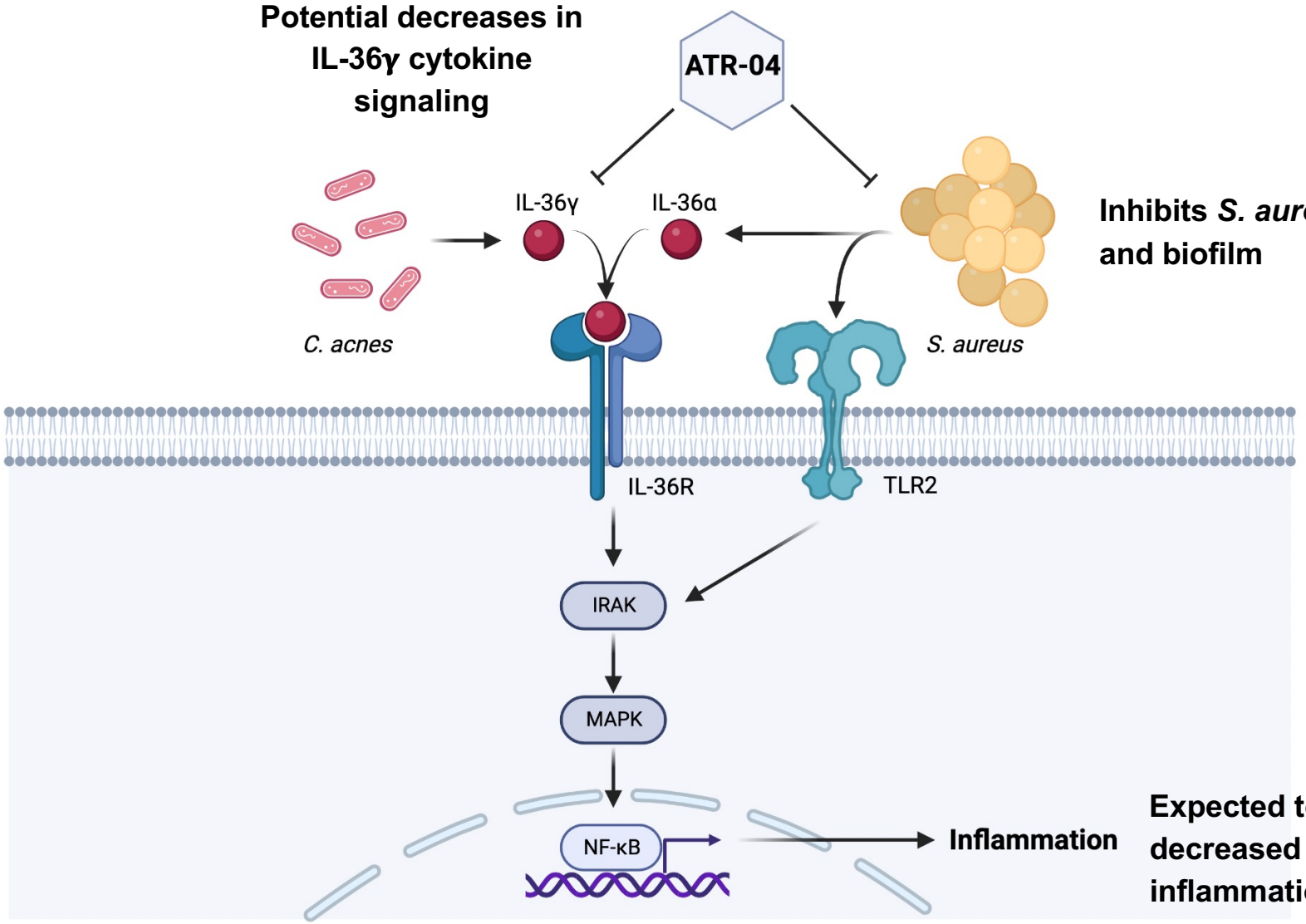
ATR-04 Key Facts

Primary Mechanism:
IL36γ inhibition, *S. aureus* control

Clinical Status:
IND filing expected mid-2024

US Prevalence:
~150,000 patients

Proposed mechanism of action of ATR-04



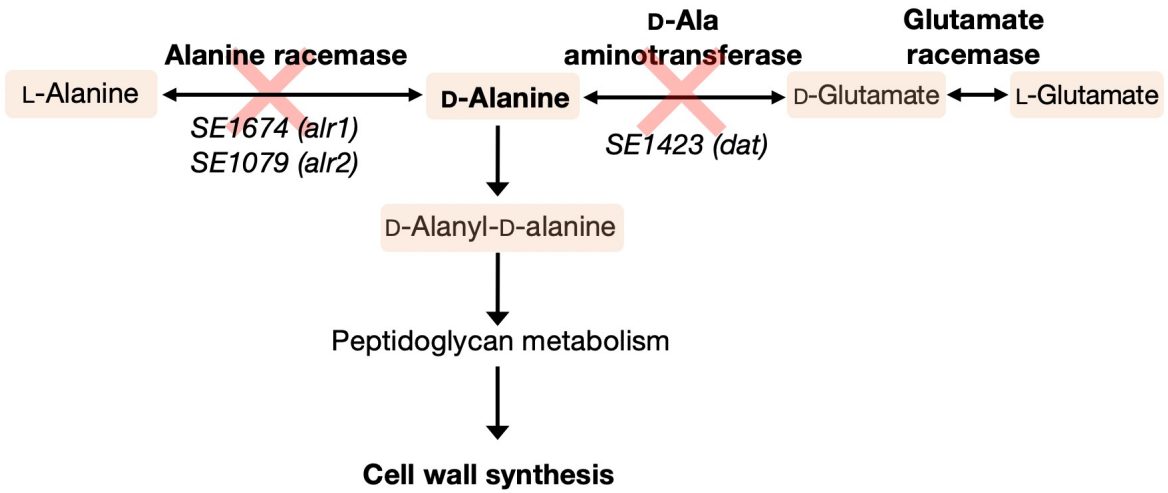
Inhibits *S. aureus* growth and biofilm

Expected to lead to decreased inflammation

Collectively, ATR-04 addresses dermal toxicity driven by EGFR inhibition

Engineering of *S. epidermidis* to create candidate ATR-04

D-alanine auxotrophy in *S. epidermidis*



SE484: Auxotrophic *S. epidermidis* with deleted mupirocin resistance and putative bacteriophage genes

Parent strain SE398
wild type-skin commensal
D-alanine Prototroph

SE484
D-alanine Auxotroph

SE398

aka: SE25, MM_{50}-SE25, *AZS-SE_25*
Genotype: SE398 mupA^{R}, *capsid*$^{+}$ and *epiA*$^{+}$
Wild type S. epidermidis, isolated from healthy volunteer

↓ *alr1*, *alr2* and *dat* deleted

SE464

Genotype: SE398 Δalr1Δalr2Δdat mupA^{R}, *capsid*$^{+}$, and *epiA*$^{+}$
Auxotrophic for D-alanine, mupA^{R}

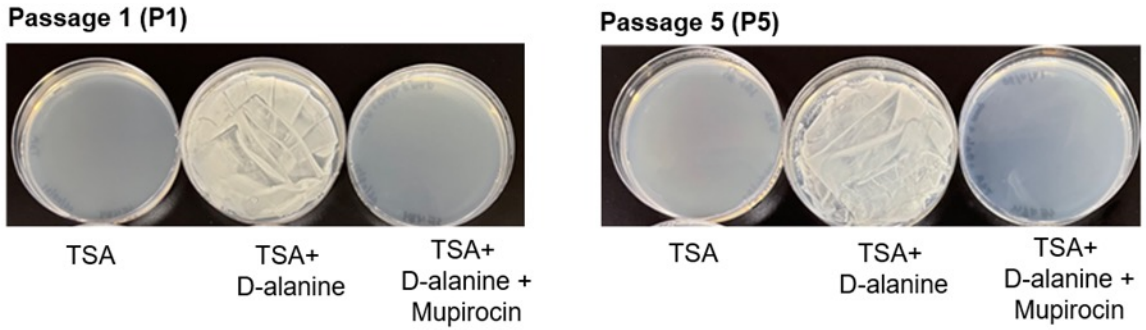
↓ *mupA* and *capsid* deleted

Strain SE484
ATR04-484

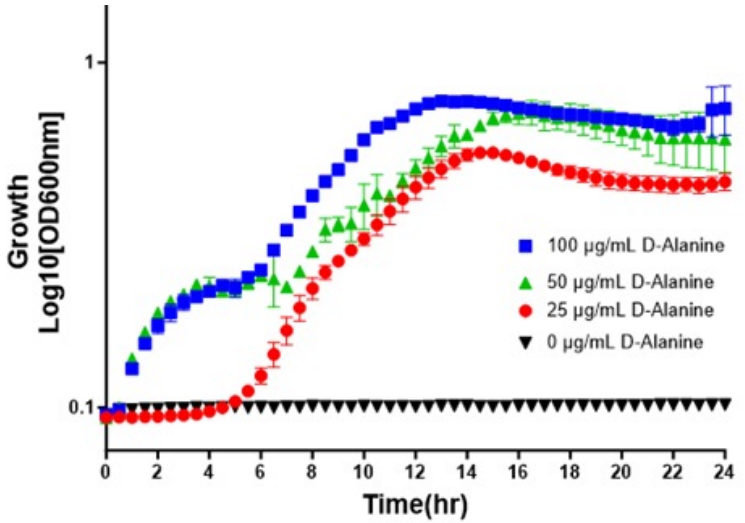
Genotype: SE398 Δalr1 Δalr2 Δdat ΔmupA Δcapsid and *epiA*$^{+}$
Auxotrophic for D-alanine, mupA^{S}

Confirmation of auxotrophy and growth characteristics of SE484

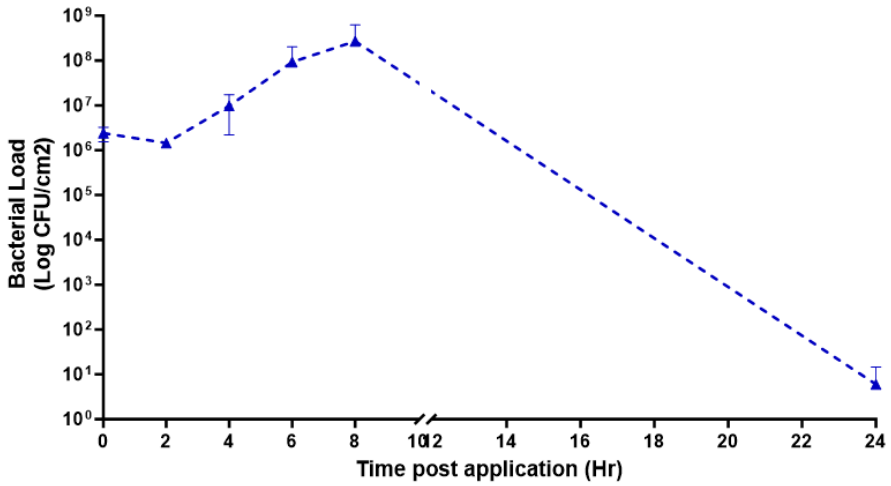
Confirmation of D-alanine auxotrophy and mupirocin sensitivity on agar plates



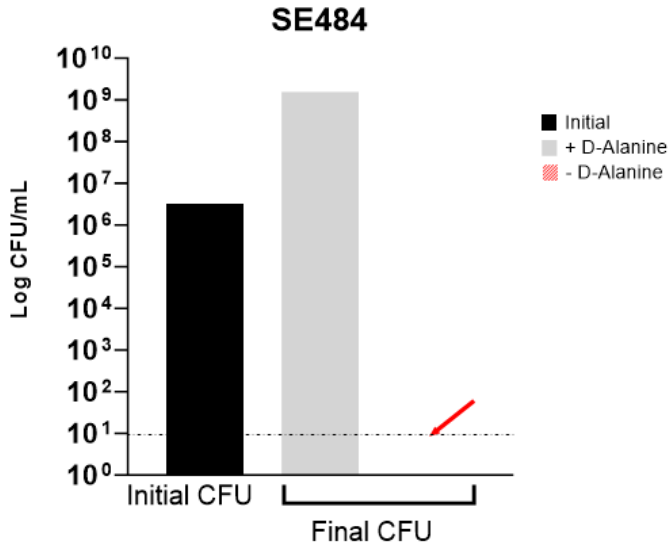
SE484 does not grow without D-alanine *in vitro*



Single dose of formulated SE484 (ATR-04) on *ex vivo* pig skin dies within 24 hours

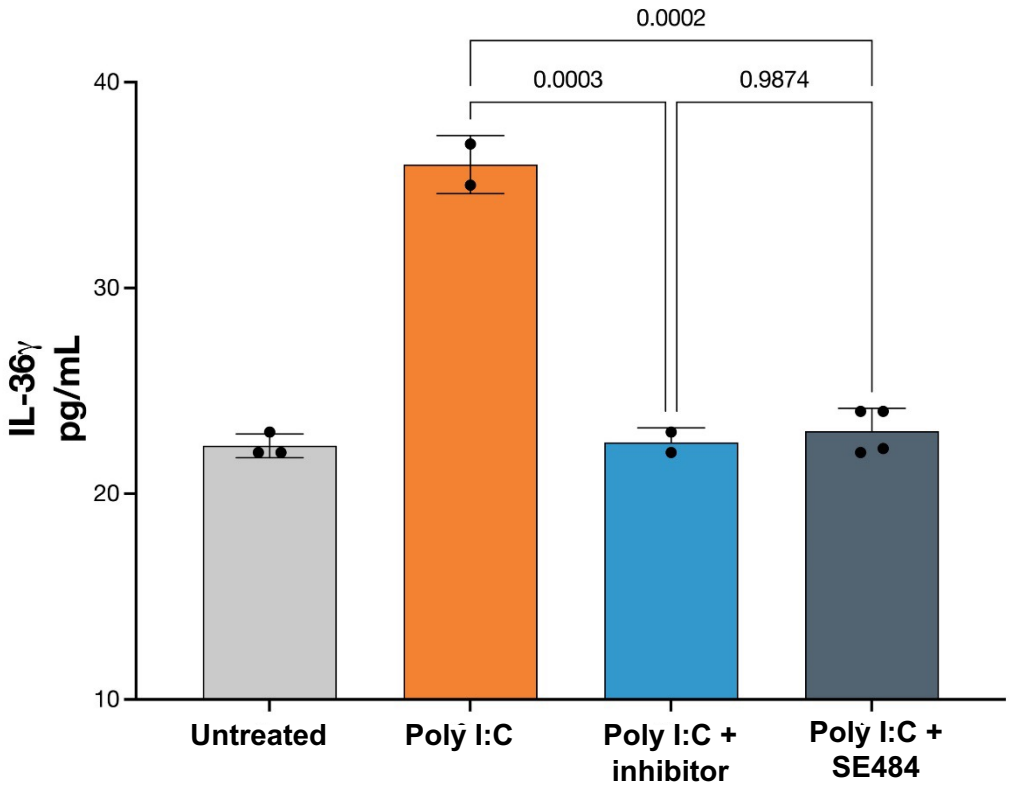


SE484 does not grow in human blood

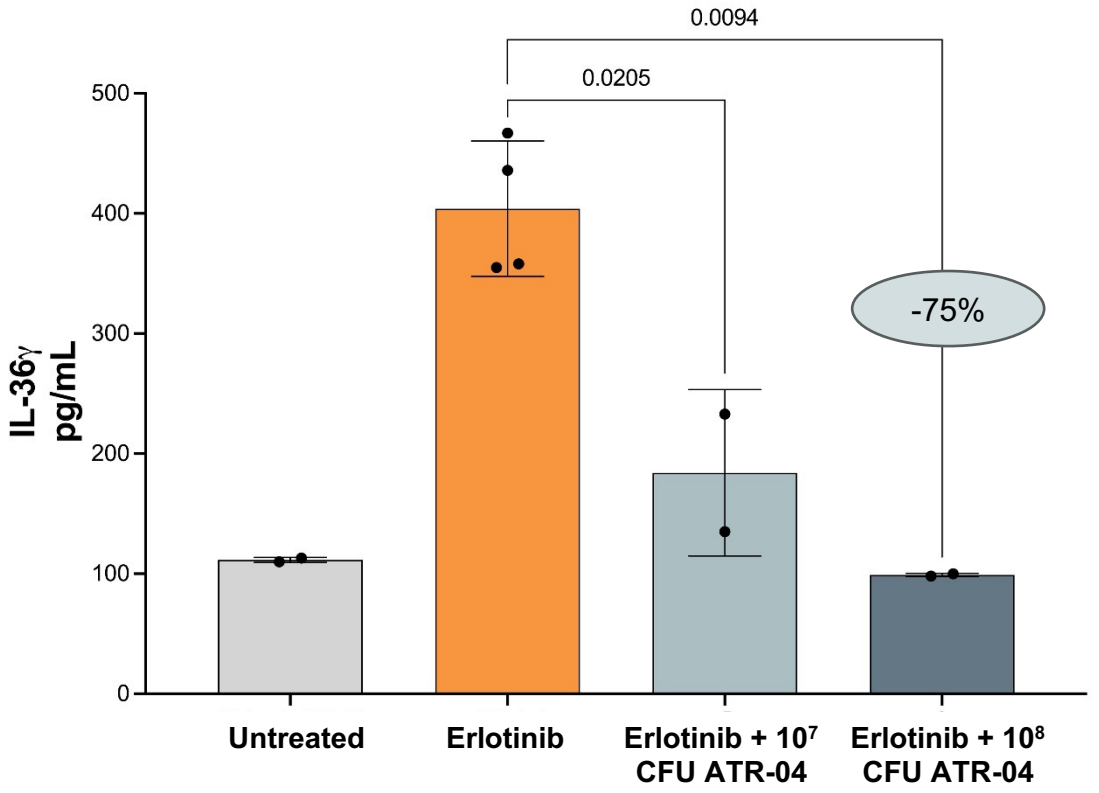


In vitro data show ATR-04 significantly reduces IL-36γ

HaCaT cells



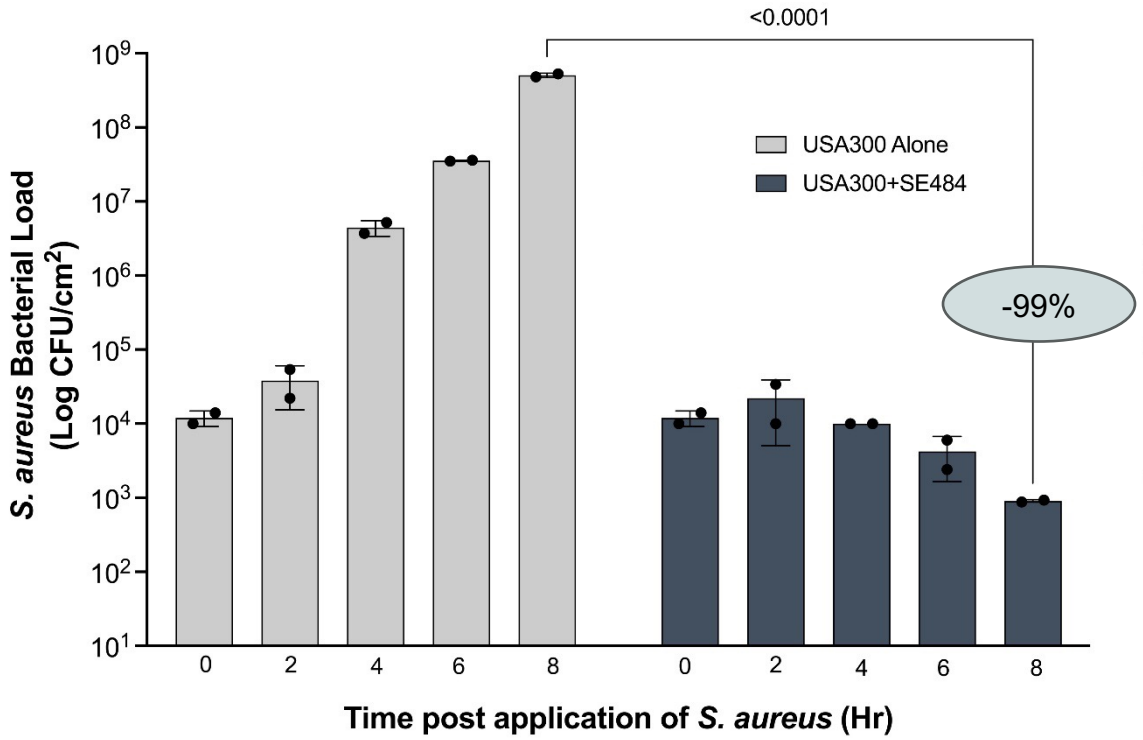
Reconstructed human epidermis



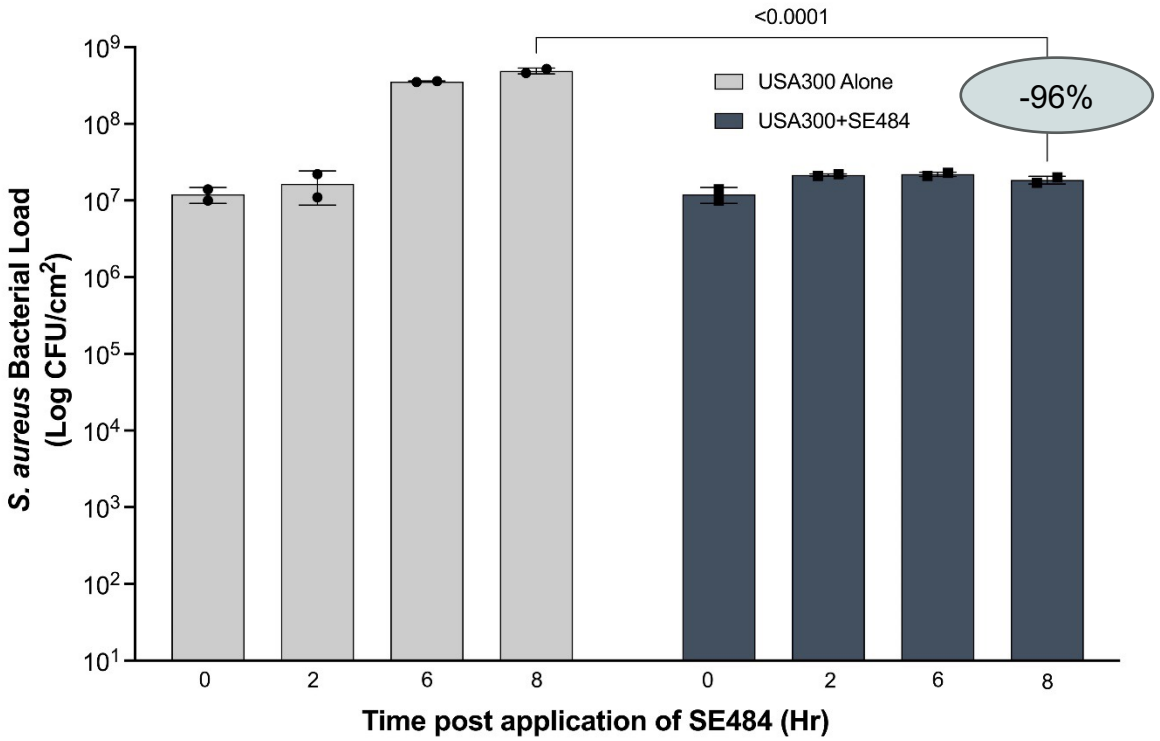
- ✓ IL-36γ is elevated in reconstructed human epidermis following erlotinib exposure
- ✓ ATR-04 reduces IL-36γ induced by erlotinib
- ✓ Dose-dependent effect observed

SE484 both reduces and prevents *S. aureus* growth in reconstructed human epidermis

Prophylactic setting



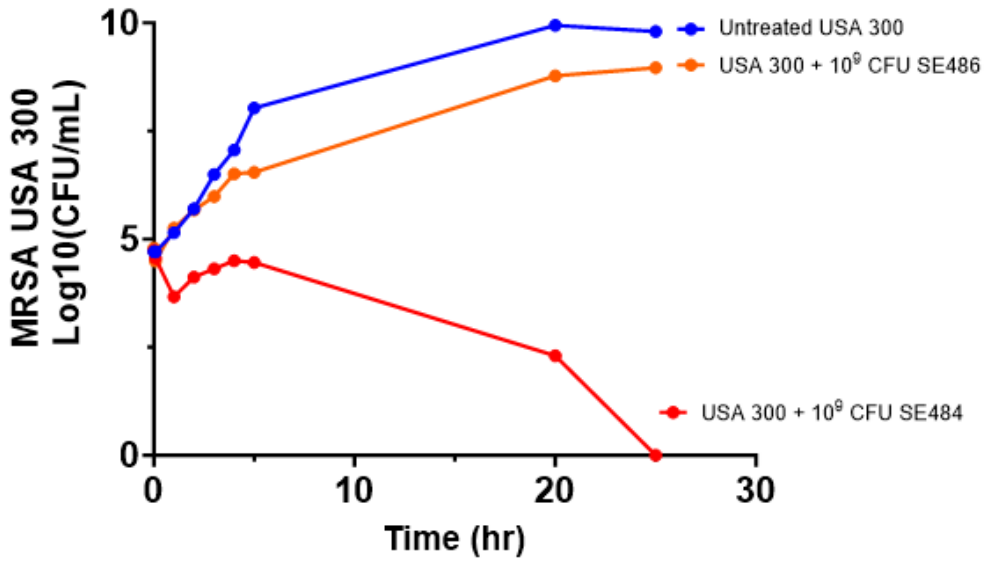
Treatment setting



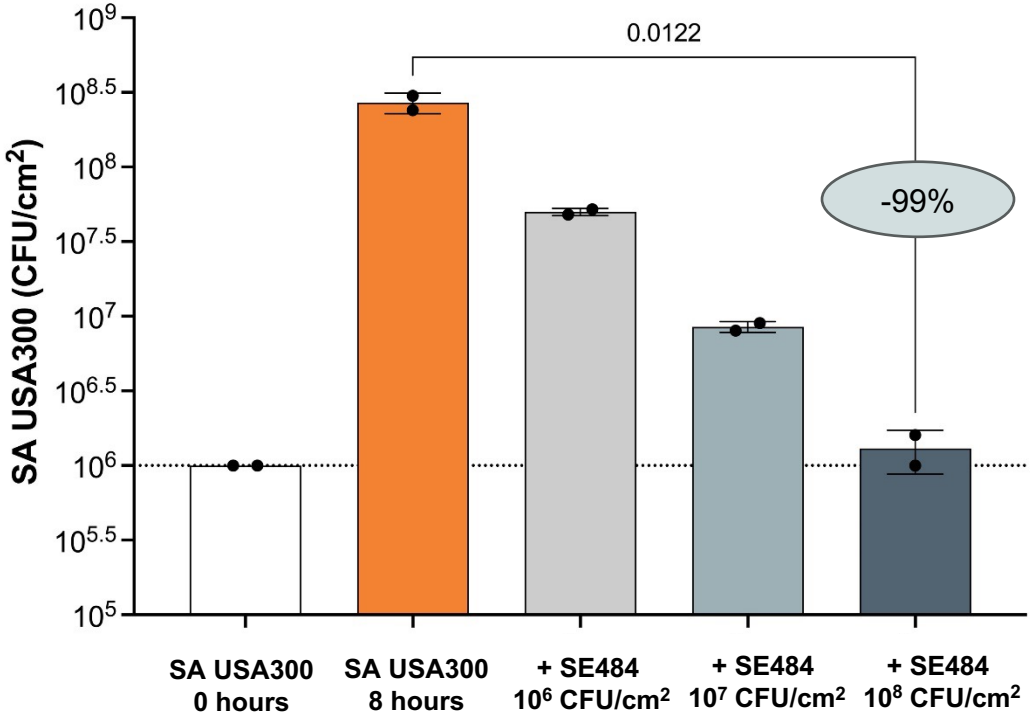
- ✓ In reconstructed human epidermis models, methicillin-resistant *S. aureus* (MRSA) was reduced by >99% in the prophylactic setting (pretreated with SE484 then MRSA added) and by 96% in the treatment setting (pretreated with MRSA then SE484 added)

SE484 significantly reduces *S. aureus* in liquid culture and on *ex vivo* pig skin

Liquid cultures



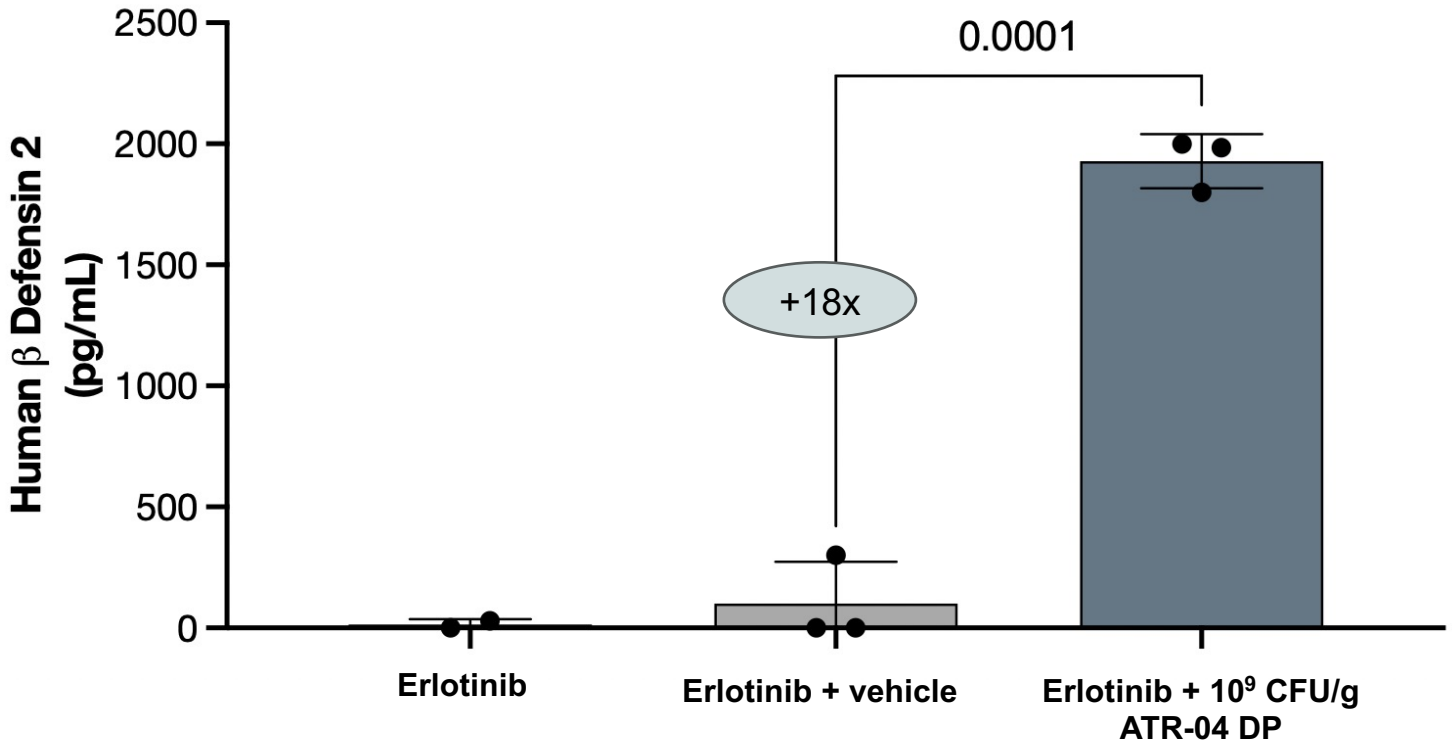
***Ex vivo* pig skin**



- ✓ In liquid cultures, SE484 **reduced** methicillin-resistant *S. aureus* (MRSA) in a **dose-dependent** effect
- ✓ In *ex vivo* pig skin, *S. aureus* was **reduced by >99%** in the treatment setting (pretreated with SE484 then MRSA added) and was **dose dependent**

ATR-04 significantly increases human beta defensin 2 on reconstructed human epidermis

Reconstructed human epidermis after 48 hours



✓ In reconstructed human epidermis, 10^9 CFU / g ATR-04 treatment led to significantly higher levels of human **β** defensin 2 compared to vehicle

Summary

- ✓ EGFRi-associated dermal toxicity is a **large, unmet need** that interferes with EGFRi effectiveness and leads to **discontinuations and poor quality of life** in cancer patients
- ✓ ATR-04, a drug product containing SE484, an auxotrophic strain of *S. epidermidis* in development for EGFRi-associated dermal toxicity, has demonstrated key proof of concept in preclinical studies:
  - ✓ ATR-04 is an auxotroph controlled by D-alanine
  - ✓ SE484 **significantly reduces** IL-36**γ** induced by erlotinib
  - ✓ ATR-04 **significantly reduces *S. aureus*** in multiple models, including *ex vivo* pig skin
  - ✓ ATR-04 **significantly increases human beta defensin** compared to vehicle
- ✓ Azitra plans to file an IND for a Phase 1b in patients with a rash due to EGFR inhibitors in mid-2024; study start expected in Q4 2024

Acknowledgements

Azitra, Inc.
David Dodds PhD
Trudy Grossman PhD
Roger Leger PhD
Dorival Martins PhD
Sara Mootien PhD
Marie-Aude Pinoteau PhD
Francisco Salva
Mark Sampson PhD
Mary Spellman MD
Alex Tikhonov PhD

University of Connecticut
Technology Incubation Program

Yale University
Tara Bancroft PhD
Leonard Milstone MD
Jonathan Levanthal MD

Funding sources:
Department of Defense
CT Innovations
CT Biosciences New Haven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THANK YOU
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