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Are therapeutic effects of antiacne agents mediated by activation of FoxO1 and inhibition of mTORC1?

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Abstract: Acne pathogenesis has recently been linked to decreased nuclear FoxO1 levels and increased mTORC1 activity. This hypothesis postulates that antiacne agents either enhance nuclear FoxO activity or inhibit mTORC1. Benzoyl peroxide (BPO), by activation of oxidative stress-inducible kinases, increases nuclear FoxO levels promoting Sestrin3-mediated AMPK activation. Furthermore, BPO-derived ROS may activate AMPK via ataxia–telangiectasia mutated. Isotretinoin and *all-trans* retinoic acid may stimulate FoxO gene expression. Doxycycline may enhance FoxOs nuclear retention by inhibiting the expression of exportin 1. Suppression of TNFα signalling by tetracyclines, erythromycin and other macrolides may attenuate IKKβ-TSC1-mediated mTORC1 activation. Erythromycin attenuates ERK1/2 activity and thereby increases TSC2. Azelaic acid may decrease mTORC1 by inhibiting mitochondrial respiration, increasing cellular ROS and nuclear FoxO levels. Antiandrogens may attenuate mTORC1 by suppressing mTORC2-mediated Akt/TSC2 signalling. This hypothesis unmasks a common mode of action of antiacne agents as either FoxO enhancers or mTORC1 inhibitors and thus provides a rational approach for the development of new antiacne agents.

Key words: acne – antiacne drugs – FoxOs – mTORC1 – mTORC1 inhibitors

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Introduction

The recent viewpoint on the pathogenesis of acne (1) focused on decreased nuclear levels of the transcription factor FoxO1 and enhanced activity of the kinase mechanistic (or mammalian) target of rapamycin complex 1 (mTORC1). Notably, FoxOs are critical rheostats that inhibit mTORC1 (1,2). The hypothesis presented here suggests that antiacne agents either enhance nuclear FoxO1 activity or directly inhibit mTORC1. Improvement of acne by dietary and pharmacological intervention appears to be related to a common underlying mechanism: enhanced FoxO and attenuated mTORC1 signalling (3,4).

Benzoyl peroxide

Whereas benzoyl peroxide (BPO)-mediated extinction of *P. acnes* takes only minutes, acne improvement takes weeks of treatment. This points to another *P. acnes*-independent mode of BPO action, which apparently involves reactive oxygen species (ROS)-mediated increases in nuclear FoxO levels by ROS-inducible kinases, Jun-N-terminus kinase (JNK) and STE20-like protein kinase 1 (MST1) (5). Notably, JNK- and MST1-mediated FoxO phosphorylation is dominant to the inhibitory Akt-mediated FoxO phosphorylation (6). Addition of hydrogen peroxide to follicular granulosa cells stimulates FoxO1's nuclear translocation (7).

BPO-mediated FoxO upregulation may attenuate mTORC1, which controls the G1/S transition and G2/M progression of the cell cycle (8). BPO reduces the size of golden hamster ear sebaceous glands (SGs) and the number of sebocytes entering the S-phase (9). Antiproliferative effects of BPO have been confirmed in human SGs (10,11). In the rabbit ear microcomedo prevention assay, BPO decreases size and number of corneocytes and comedo formation (12,13) (Fig. 1).

Tetracyclines

Inflammatory events are associated with the initiation of acne (14). Tetracyclines exert anti-inflammatory effects. FoxOs are pivotal regulators of inflammation (15,16). FoxO transport through the nuclear pore requires its interaction with importins and exportins (17). Exportin 1, also known as *chromosomal region maintenance protein 1* (CRM1), recognizes specific nuclear export signals (NESs) present on FoxO1 including a leptomycin-sensitive NES (18). Intriguingly, in a mouse lung tumor model, doxycycline dramatically decreases CRM1 expression in comparison with untreated mice (19). Thus, the anti-inflammatory and possibly antiproliferative effect of doxycycline may derive from enhanced nuclear FoxO1 retention.

Tetracyclines inhibit NFκB activation and TNFα secretion (20). IKKβ, the crucial kinase of proinflammatory NFκB activation, inhibits TSC1 and thereby activates mTORC1 (21,22). As mTORC1-mediated phosphorylation of lipin1 controls the promoter access of *sterol response element-binding protein 1* (SREBP-1) to target genes of lipid synthesis (23,24), tetracycline-mediated suppression of IKKβ/mTORC1 signalling may attenuate sebaceous lipogenesis (25). In contrast, addition of TNFα to SZ95 human sebocytes increases lipid droplet formation and upregulates both fatty acid synthase and SREBP-1 expression (26).

Erythromycin and other macrolides

Erythromycin and other macrolides exert anti-inflammatory activity by attenuation of both TNFα-IKKβ and ERK1/2 signalling (27,28), thus increasing the inhibitory effect of TSC1/TSC2 towards Rheb.

Isotretinoin and *all-trans*-retinoic acid

Circumstantial evidence supports the view that isotretinoin (*13-cis* retinoic acid) increases nuclear FoxO1 levels (29,30). Isotretinoin

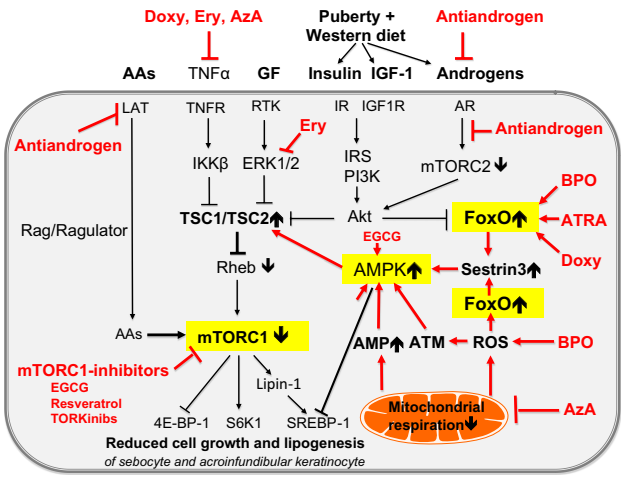

Figure 1. Upregulation of FoxO1 and inhibition of mTORC1 by antiacne agents: oral isotretinoin, *all-trans* retinoic acid (ATRA), doxycycline (Doxy) and benzoyl peroxide (BPO) increase nuclear FoxO levels, which stimulate the expression of Sestrin3. Sestrin3 activates AMPK and augments the inhibitory function of TSC2 towards Rheb, thus suppressing mTORC1. BPO may stimulate ROS-mediated activation of ATM, a further stimulator of AMPK-mediated mTORC1 inhibition. Azelaic acid (AzA) via inhibition of mitochondrial respiration may increase ROS-mediated upregulation of FoxOs and Sestrin3 expression as well as cellular AMP levels, all activating AMPK. Antiandrogens inhibit mTORC2-dependent activation of Akt, thus increasing TSC1/TSC2-mediated inhibition of Rheb. Antiandrogens suppress the expression of L-type amino acid (AA) transporter (LAT), thus interfering with AA-mediated activation of mTORC1. Erythromycin and other macrolides inhibit IKKβ and ERK1/2 signalling and thereby increase the inhibitory activity of the TSC1/TSC2 complex towards Rheb/mTORC1. Natural mTORC1 inhibitors like resveratrol and epigallocatechin-3-gallate (EGCG) as well as synthetic mTOR inhibitors inhibit the ATP-dependent kinase domain of mTOR, thereby directly reducing mTORC1 activity. Thus, all antiacne drugs may directly or indirectly impair downstream mTORC1 signalling and may attenuate growth and proliferation of acroinfundibular keratinocytes, sebocytes and sebaceous lipogenesis.

isomerizes to *all-trans* retinoic acid (ATRA), which interacts with retinoic acid receptors (RARs) (31). ATRA/RAR signalling induces secondary responses in gene expression involving FoxO proteins (32). ATRA increases the expression of FoxO3a in neuroblastoma cells (33). FoxO3a mediates ATRA-induced granulocytic differentiation and apoptosis in acute promyelocytic leukaemia (APL) cells (34). Notably, FoxO3a is a strong inducer of FoxO1 (35). ATRA treatment for APL cells translocates FoxO3a into the nucleus (34). In this regard, the ATRA-induced protein *stimulated by retinoic acid 8* (STRA8), which physically interacts with CRM1, may modify CRM1-mediated nuclear FoxO1 export (36).

Isotretinoin induces cell cycle arrest in SEB-1 sebocytes with increased expression of the cell cycle inhibitor p21 (37). Upregulation of the FoxO1 target gene p21 could thus explain isotretinoin-induced sebocyte apoptosis (38). Furthermore, isotretinoin/ATRA-induced FoxO upregulation may stimulate autophagy by FoxO-mediated suppression of mTORC1 (2,39).

Azelaic acid

Azelaic acid (AzA) dose-dependently suppresses mitochondrial respiration in rat liver by competitive inhibition of mitochondrial enzymes including succinic dehydrogenase decreasing ATP generation and increasing mitochondrial ROS release (40,41). Another mitochondrial toxin, 3-nitropropionic acid (3-NP), selectively inhibits mitochondrial succinic dehydrogenase (42,43). 3-NP treatment-induced mitochondrial ROS leakage induces apoptosis in neuronal cells (42,43). In mouse follicular granulosa cells, 3-NP increases ROS formation, raises nuclear FoxO1 levels and induces apoptosis (7). AzA-mediated mitochondrial ROS release may upregulate FoxO expression and inhibit mTORC1, thus mimicking ROS-stimulated effects of BPO treatment.

Antiandrogens

Androgen-mediated stimulation of mTORC2 and consecutive mTORC2-mediated activation of Akt inhibit FoxO1 activity (44) and stimulate androgen receptor transcriptional activity (45). The therapeutic effectiveness of antiandrogens in female acne patients may thus be related to impaired androgen/TORC2/Akt-mediated suppression of TSC2 inhibition as well as reduced Akt-mediated nuclear FoxO1 export, thus attenuating mTORC1 activity.

Conclusion

Translational evidence supports the hypothesis that antiacne agents may either attenuate mTORC1 activity indirectly by upregulating nuclear FoxO levels or by direct inhibition of mTORC1 (Fig. 1). This hypothesis may allow the rational development of new antiacne drugs such as FoxO enhancers or synthetic and natural mTORC1 inhibitors like resveratrol and epigallocatechin-3-gallate (Table S1).

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Conflict of interests

The authors have declared no conflicting interests.

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Supporting Information

Additional Supporting Information may be found in the online version of this article:

Data S1. Full-length hypothesis.

Figure S1. FoxO shuttling between nucleus and cytoplasm.

Figure S2. Drug-mediated modifications of nuclear FoxO trafficking.

Figure S3. Inhibition of mTORC1 activity by anti-acne agents.

Table S1. Action of commonly used and potential new antiacne drugs correcting imbalanced FoxO1-/mTORC1 signalling in acne.