文献分享 | ECHO 显微镜在抗穆勒氏管激素研究中的应用

在 2017 年 12 月 27 日，由 Jing Xu，Fuhua Xu，Maralee S Lawson，Olena Y Tkachenko 等作者在 Biology of Reproduction 中发表一篇了《Anti-Mullerian hormone is a survival factor and promotes the growth of rhesusmacaque pre antral follicles during matrix-free culture》的文章。

Biology of Reproduction, 2018, 98(2), 197–207
doi:10.1093/biolre/iox181
Research Article
Advance Access Publication Date: 27 December 2017

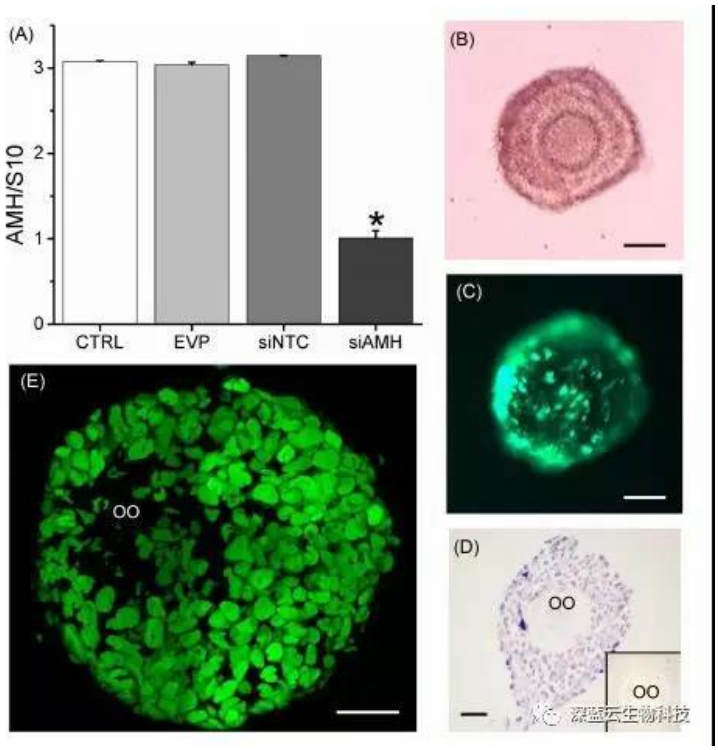
OXFORD

Research Article

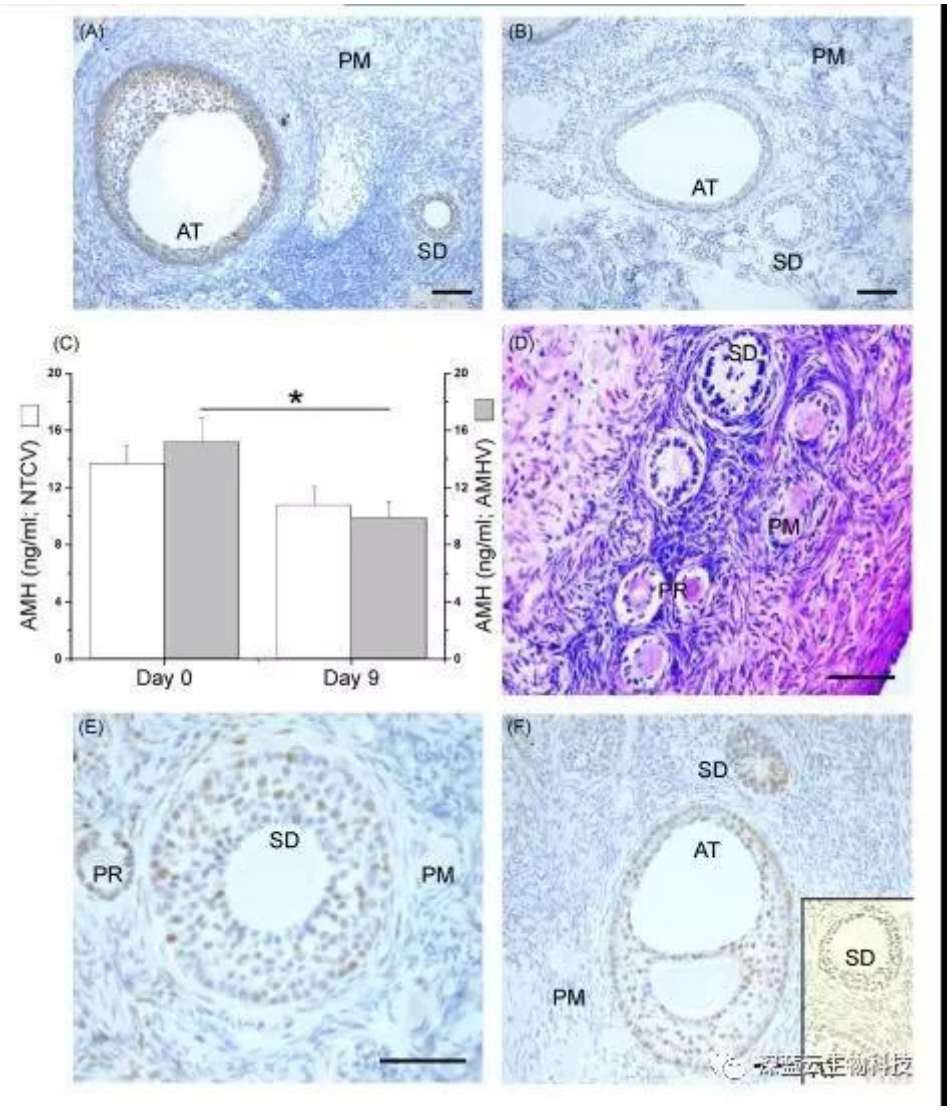
Anti-Müllerian hormone is a survival factor and promotes the growth of rhesus macaque preantral follicles during matrix-free culture†

Jing Xu[1,2,*], Fuhua Xu[2], Maralee S Lawson[1], Olena Y Tkachenko[1], Alison Y Ting[1], Christoph A Kahl[3], Byung S Park[4], Richard R Stouff and Cecily V Bishop[1]

文章使用 Revolve 正倒置一体显微镜获得了如下结果:



该图中 B-E 是通过 Revolve 正倒置一体显微镜成像得到的结果 ↑



该图中 A-B、C-F 是通过 Revolve 正倒置一体显微镜成像得到的结果 ↑

中文摘要：

抗穆勒氏管激素（AMH）在卵泡发育过程起着至关重要的作用，其局部作用于卵泡生长过程中的动态分泌有关。目前不同物种的 AMH 在腔内卵泡生长和功能的影响已有所验证，但对腔前卵泡发育的作用仍待进一步验证。因此，我们通过实验研究 AMH 在腔前卵泡发育过程中所起到的作用。利用 AMH-shRNA 重组腺病毒载体降低猕猴卵泡中 AMH 基因的表达。将次生卵泡从成年猕猴卵巢中分离出来后用超低附着培养皿单独培养，且培

养基中加入促卵泡激素和胰岛素，培养 5 周。将卵泡随机分配给治疗组，分别为 a.对照组；b.非靶向对照组 shRNA-vector；c.AMH shRNA-vector；d.AMH shRNA-vector + 重组人源 AMH；e.重组人源 AMH。评估卵泡的存活和生长情况，并分析培养基中的类固醇激素和旁分泌因子的浓度。在体内研究中，将非靶向对照 shRNA-vecotor 和 AMH shRNA-vector 分别注入猕猴的卵巢，术后 9 天对卵巢进行形态学及免疫组化检测。结果发现，在非人灵长类动物中，AMH 表达下调抑制腔前卵泡的存活和生长。体外实验发现，在培养基中添加 AMH 则可促进腔前卵泡生长发育至小腔窦期，类固醇激素和旁分泌因子表达增加，并促进卵母细胞成熟。这些数据表明，AMH 是一个关键的卵泡旁分泌/自分泌因子，促进灵长类的腔前卵泡存活和生长。

英文摘要：

Anti-Mullerian hormone (AMH) plays a key role during ovarian follicular development, with local actions associated with a dynamic secretion profile by growing follicles. While results for AMH effects on antral follicle growth and function are consistent among studies in various species, any effects on preantral follicle development remain controversial. Therefore, experiments were conducted to investigate the direct actions and role of AMH during follicle development at the preantral stage. Macaque-specific short-hairpin RNAs (shRNAs) targeting AMH mRNA were incor- porated into

adenoviral vectors to decrease AMH gene expression in rhesus macaque follicles. Secondary follicles were isolated from adult macaque ovaries and cultured individually in the ultra-low-attachment dish containing defined medium supplemented with follicle-stimulating hor- mone and insulin for 5 weeks. Follicles were randomly assigned to treatment groups: (a) control,(b) nontargeting control shRNA-vector, (c) AMH shRNA-vector, (d) AMH shRNA-vector + recombi-nant human AMH, and (e) recombinant human AMH. Follicle survival and growth were assessed. Culture media were analyzed for steroid hormone and paracrine factor concentrations. For in vivo study, the nontargeting control shRNA-vector and AMH shRNA-vector were injected into macaque ovaries. Ovaries were collected 9 days postinjection for morphology and immunohistochemistry assessment. Decreased AMH expression reduced preantral follicle survival and growth in non- human primates. Supplemental AMH treatment in the culture media promoted preantral follicle growth to the small antral stage in vitro with increased steroid hormone and paracrine factor production, as well as oocyte maturation. These data demonstrate that AMH is a critical follicular paracrine/autocrine factor positively impacting preantral follicle survival and growth in primates.

ECHO Revolve 正倒置一体显微镜展现了其非凡的灵活性，可以轻松地实现正置和倒置显微镜转换，创新性地把正倒置显微镜合二为一，开启了显微镜 Hybrid 时代。

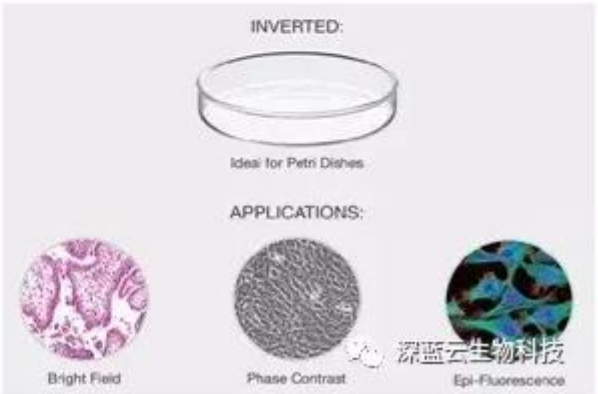
> 视网膜屏显示技术---比拟目镜人眼观察效果

> 全视野观察---更清晰，更方便

> 多通道荧光---多达 4 个 EPI 荧光通道，通过 iPad Pro 实现了完全自动化操作，轻松快速地完成多色荧光显微分析

> App 应用软件---基于 IOS 的 Echo App 是与 Apple 团队合作研发的专业显微镜软件

> 精湛的工艺尽显高端品质---实现非凡的性能



ECHO Revolve 正倒置一体显微镜兼具正置和倒置显微镜的优势，方便小巧，一机多能，可以非常便利地通过旋转实现正倒置配置切换；以 iPad 桌面替代目镜设计，获得前所未有的视觉体验；同时基于 iOS 的 Echo app，使软件操作更人性化。