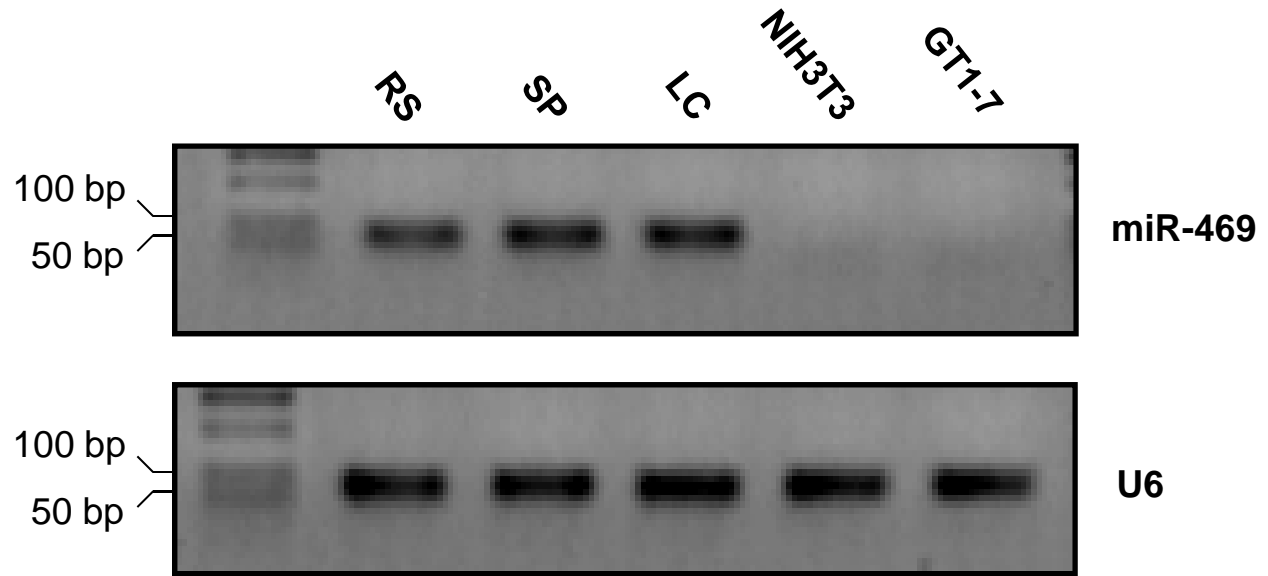


S1.



SUPPLEMENTAL FIGURE LEGENDS

Figure S1. Expression of miR-469 in testicular germ and Leydig cells. Enriched germ cells (round spermatids and spermatocytes) and Leydig cells were obtained by centrifugal elutriation. Total RNA was prepared followed by microRNA RT-PCR. U6 small nuclear RNA: endogenous control. NIH3T3 (mouse embryonic fibroblast cells (ATCC) and GT1-7 (Immortalized hypothalamic neuronal cells) (58) were used as a negative control. RS: round spermatids. SP: spermatocytes. LC: Leydig cells. Results from previous studies implied that miR-469 was expressed solely in male germ cells specifically meiotic spermatocytes and round spermatids, since no expression was found in Sertoli cells and tissues of other organs (17). We have now determined that miR-469 is also present in WT Leydig cells by RT-qPCR. The finding of miR-469 in Leydig cells is of much interest since the Leydig cell is a site of expression of GRTH protein and recently functions of GRTH in steroidogenesis have been demonstrated in our laboratory (59).