Stimulation of vascular network formation by fraction D of date palm fruit extract-conditioned media: Implications for healthy ageing and cardiac repair

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ABSTRACT

Introduction: Cardiovascular disease (CVD) is the leading cause of death globally, with risk rising sharply with age, making it a major barrier to healthy ageing. Ageing reduces the heart's regenerative capacity, particularly angiogenesis and vascular repair. While mesenchymal stem cells (MSCs) hold promise for cardiac repair via their paracrine secretome, hypoxia within injured myocardium limits their function. Natural bioactives offer a means to enhance MSC function. Date palm fruit contains bioactives with regenerative potential. We fractionated an aqueous extract by solid-phase extraction and, based on BMMSC proliferation (MTT), selected Fraction D (FD) for functional testing of angiogenic paracrine activity. **Materials and Methods:** Bone marrow-derived mesenchymal stem cells (BMMSCs) were co-cultured with hypoxia-injured cardiomyocytes for 24 h under the following conditions: (i) basal medium (negative control), (ii) complete growth medium supplemented with 10% fetal bovine serum (FBS; positive control), (iii) 10% FBS with Fraction D (FD), or (iv) 2% FBS with FD. Following incubation, conditioned media (CM) were collected from each group and applied to endothelial cells seeded on Matrigel for tube formation assays. Tube formation parameters — including number of junctions, number of branches, total tube length, and total branching length — were quantified at 4, 8, 12, and 24 h. Statistical analysis was performed using the Mann–Whitney U test, with $p < 0.05$ considered significant. **Results:** FD markedly potentiated angiogenesis compared with both controls, with the largest effects at 4–12 h. Relative to basal CM, 10% FBS + FD increased junction number by ~8–17-fold and branches by ~6–21-fold; total and branching lengths rose ~10–16-fold. Versus complete CM, FD provided consistent gains: junctions +1.3–2.0×, branches +1.2–1.4×, total length +1.2–1.4×, and branching length +1.2–1.4× (4–12 h; all $p < 0.05$). Reducing serum to 2% while keeping FD preserved or enhanced performance: at 4–12 h, 2% FBS + FD was comparable to 10% FBS + FD (0.95–1.1× across metrics), and at 24 h it maintained network stability more strongly, exceeding complete CM by ~1.3–2.6× and 10% FBS + FD by ~1.5–2.2× ($p < 0.05$). **Conclusion:** CM from BMMSC–injured cardiomyocyte co-cultures supplemented with Fraction D robustly enhances endothelial network formation, with serum-sparing (2% FBS) conditions retaining or even improving durability of tubes at 24 h. These data support Fraction D as a pro-angiogenic modulator that amplifies the reparative BMMSC-cardiomyocyte secretome and may enable lower-serum formulations for cardiac tissue repair strategies. As cardiovascular disease is an ageing-related disorder marked by impaired vascular repair, FD shows promise as a natural adjunct to regenerative cardiology, supporting vascular resilience and promoting healthy ageing.

Keywords: Ajwa date palm fruit, proliferation, angiogenesis, mesenchymal stem cells, hypoxic cellular mode