PS-8

Discovery of miRNA effecting myogenesis and studies on sarcopenia treatment based on the findings

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Background

❖ Exosomes are extracellular vesicles that are released from most cell types encapsulating specific molecular cargo. Exosomes serve as mediators of cell-to-cell and tissue-to-tissue communications. MicroRNAs (miRNAs) are small non-coding RNAs involved in post-transcriptional gene regulation. Circulating miRNAs may also modulate skeletal muscle function in physiological and pathological conditions.

❖ In this study, we aimed to conduct exosomal miRNA profiling after increasing muscle mass through exercise.

Methods

❖ We recruited 10 participants in our study. Participants were classified with two groups: the exercise group (n=5) and the control group (n=5). At the baseline, a DEXA scan was conducted, followed by resistance training and aerobic exercises for 8 weeks. After 8 weeks, a DEXA scan and blood sampling was performed.

❖ The DEXA scan was used to calculate the Axial Skeletal Muscle (ASM) and Skeletal Muscle Index (Skeletal Muscle Index). Blood was collected in 10-ml EDTA vacutainers and centrifuged immediately upon collection at 4°C. The supernatant plasma was stored at –80°C until analyses.

❖ To identify exosomes in serum samples, we performed scanning electron microscopy, nanoparticle tracking analysis, and immunoblotting showed that CD9, and CD63, were detected in exosomes.

❖ RNAs from each sample were used to construct sequencing libraries. We estimated the quantity of libraries using qPCR according to qPCR quantification protocol guide. The selection of miRNAs was based upon a Fold change criterion of 2, a p-value threshold of 0.1, and Normalized data (log2) value of 4.

❖ Data were analyzed using ExDEGA v.3.2.1 software (eBiogen, Seoul, Korea).

Results

❖ The control group showed no significant change in muscle mass over 8 weeks, whereas the exercise group displayed a significant increase in ASM (*P*-value=0.00) and SMI (*P*-value=0.004) after 8 weeks of exercise (Table 1).

❖ Exosomes were isolated from the blood collected from the subjects (Figure 1).

❖ In the exercise group, hsa-miR-1827 and hsa-miR-3652 were upregulated, while hsa-miR-221-3p and hsa-miR-502-3p were downregulated (Figure 2).

Table 1. Characteristics of participants

Exercise group																			
	Age	Body weight (kg)			upper limb muscle(kg)			lower limb muscle(kg)			Total(kg)			ASM(kg)			SMI(kg/m²)		
		pre	post	p-value	pre	post	p-value	pre	post	p-value	pre	post	p-value	pre	post	p-value	pre	post	p-value
1	44	67.00	65.40		6.65	7.14		16.91	17.57		25.26	25.44		23.56	24.71		7.61	7.98	
2	27	67.00	67.50		5.70	5.82		15.03	15.46		21.94	22.04		20.73	21.29		7.01	7.19	
3	29	72.20	72.90		6.14	6.28		17.58	18.86		24.70	24.97		23.72	25.14		8.06	8.61	
4	47	62.50	63.30		4.93	5.28		16.60	17.06		22.36	22.88		21.53	22.34		7.92	8.25	
5	30	75.00	74.70		7.45	7.58		17.77	19.11		28.21	26.47		25.22	26.69		8.24	8.61	
mean	35.4	68.74	68.76	0.97	6.17	6.42	0.03	16.78	17.61	0.01	24.49	24.36	0.76	22.95	24.03	0.00	7.77	8.13	0.0036
Control group																			
	Age	Body weight (kg)			upper limb muscle(kg)			lower limb muscle(kg)			Total(kg)			ASM(kg)			SMI(kg/m²)		
		pre	post	p-value	pre	post	p-value	pre	post	p-value	pre	post	p-value	pre	post	p-value	pre	post	p-value
1	38	70.9	69.7		6.52	6.36		18.25	18.03		53.21	51.69		24.76	24.39		8.34	8.18	
2	34	69.5	68.7		6.08	6.05		16.41	16.79		47.41	46.71		22.49	22.85		7.71	7.83	
3	54	69.4	69.3		6.39	6.35		15.72	15.19		49.32	48.76		23.12	21.54		7.45	7.32	
4	55	69.8	67.1		6.09	6.14		16.4	16.41		49.7	49.68		22.12	22.55		7.57	7.61	
5	27	66.9	67.0		6.26	6.11		16.16	16.13		49.5	48.95		22.41	21.84		7.72	7.67	
mean	41.6	69.3	68.4	0.1	6.27	6.20	0.17	16.59	16.51	0.55	49.83	49.16	0.05	22.98	22.63	0.40	7.76	7.72	0.53

ASM, axial skeletal muscle; SMI, skeletal muscle index

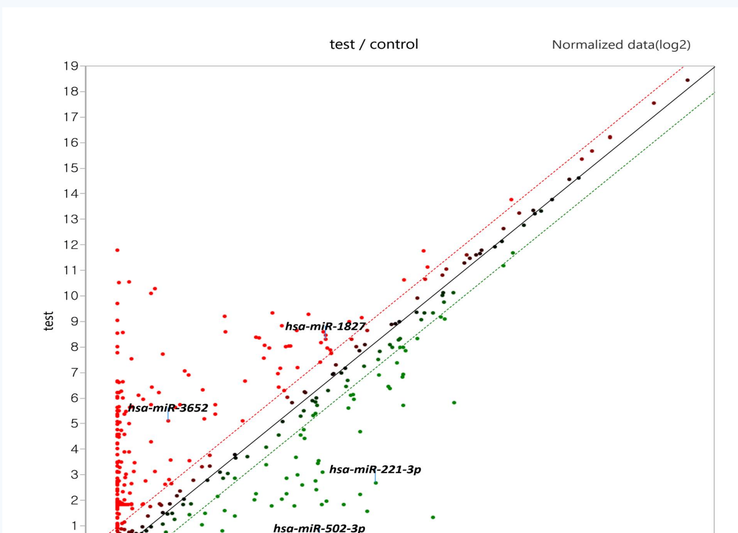

Figure 2. Scatter plots of expressed serum exosomal miRNAs in the two groups using ExDEGA for data analysis.

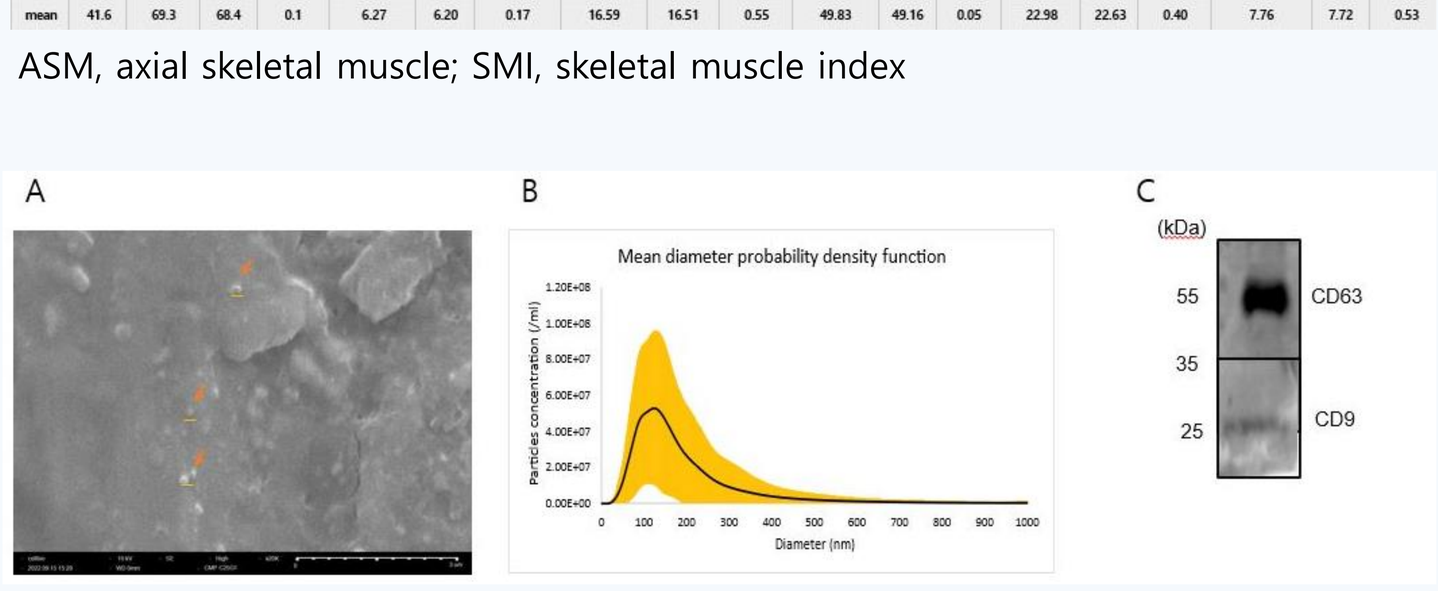

Figure 1. Characterization of exosomes isolated from plasma. A) Representative SEM images of exosomes isolated from plasma. Scale Bar = 300 nm. B) Nanoparticle tracking analysis revealed a size distribution of the exosomes. C) Western blot analysis for exosomes marker in exosomes.

Conclusion

❖In the group where muscle mass was increased through exercise, exosomal microRNA was isolated, and upregulated and downregulated miRNAs were identified. Further research is needed to determine the actual impact on myogenesis through in vivo studies.