

替療法である。この $\beta$ 細胞代替療法としては、膵臓移植および膵島移植といった移植医療がある。膵臓移植は、現時点において長期インスリン離脱率が最も高い治療法であり、生命予後改善効果も確認されているが、手術侵襲が高度である点が問題となる。

これに対し膵島移植は、膵臓移植と比較して、IVR手技を用いて局所麻酔下に完了する非常に低侵襲な治療法である。その一方で、膵臓移植と比較すると長期インスリン離脱率はいまだ不十分で、免疫抑制プロトコルのさらなる改善を要する。さらに、膵島移植のもう一つの問題点として、膵島分離法は確立したものの、分離時および移植直後の細胞喪失が多く、複数のドナーを必要とするケースが多い。加えて、欧米では、膵島収量が多く見込める肥満ドナーを用いているのに対し、本邦では肥満ドナーはごくわずかであるため、欧米の成績を本邦で再現するときのハードルとなっている。これらは、特に脳死および心停止ドナー提供数の少ない本邦において解決すべき問題である。

さて、この問題点を解決するために注目される一つの可能性として、脂肪由来間葉系幹細胞(adipose-derived mesenchymal stem cell, ADSC)がある。ADSCは、皮下脂肪などの脂肪組織より簡単に調製可能な幹細胞で、臨床膵島移植の成績を改善する可能性を有していることが報告されている。筆者らのグループでも、ADSCと膵島を共移植することで、移植に必要な膵島数を減量可能であることをマウスモデルで報告した<sup>1)</sup>。そこで、さらに膵島収量を増やすことを目的として、このADSCに対してリプログラミング手技を応用することを考えた。

### リプログラミング効率と幹細胞療法

人工多能性幹細胞(induced pluripotent stem cell, iPS細胞)は、2006年に京都大学の山中らのグループが最初に報告した多能性幹細胞で、再生医療を実現するための重要なツールとして研究が進んでいる<sup>2)</sup>。体細胞に数種類の遺伝子等を導入し、特定の条件下で培養することで得られる。以降iPS細胞の基礎医学・創薬への応用が期待されている。

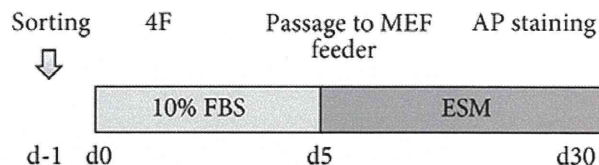


図1 プロトコール

(Kawamoto K et al. Disease Markers 35:573-579, 2013<sup>6)</sup>より)

血液幹細胞における研究において、未分化な細胞であるほど、リプログラミング効率にすぐれることが報告された<sup>3)</sup>。これらの知見に加えて、筆者らが膵島移植効率を高めるために応用しようと考えているヒトADSCにおいても、CD90high分画が分化誘導効率に関与するとの報告もある<sup>4)</sup>。このCD90(Thy-1)は、GPIアンカーの膜蛋白であり、当初は胸腺細胞の抗原として同定された。つづいて、幹細胞でも発現していることが報告された。このことは、リプログラミング効率にすぐれる分画が、幹細胞療法に適した分画であることを示唆している<sup>5)</sup>。そこで筆者らは、CD90high分画がリプログラミング効率にすぐれるか解析した<sup>6)</sup>。

### 研究の概要

8~12週齢のC57BL/6マウスの皮下脂肪より、通常法を用いてADSCを分離した。分離当日にFACSアリアセルソーターを用いて、ADSCをCD90high分画およびCD90low分画にソートした<sup>7)</sup>。ソート後に6ウエルプレートに播種した。翌日、レンチウイルスベクターを用いて、山中4因子(Oct3/4, Sox2, Klf4, c-Myc)を導入した。導入5日目に、1,000個の細胞を、マイトマイシンCで処理したマウス胎生線維芽細胞(MEF)のフィーダーに移し、以降はESメEDIUMで合計30日間培養した。リプログラミング効率はアルカリフォスファターゼ染色(AP染色)を用いて解析した。また免疫細胞染色にて、未分化マーカーの発現を確認した。

本研究における実験プロトコールを図1に示す。通常の単層培養条件下においてマウスADSCでは、コロニー形成は認めず、またAP染色においても陰性であった。一方、ADSCに山中4因子

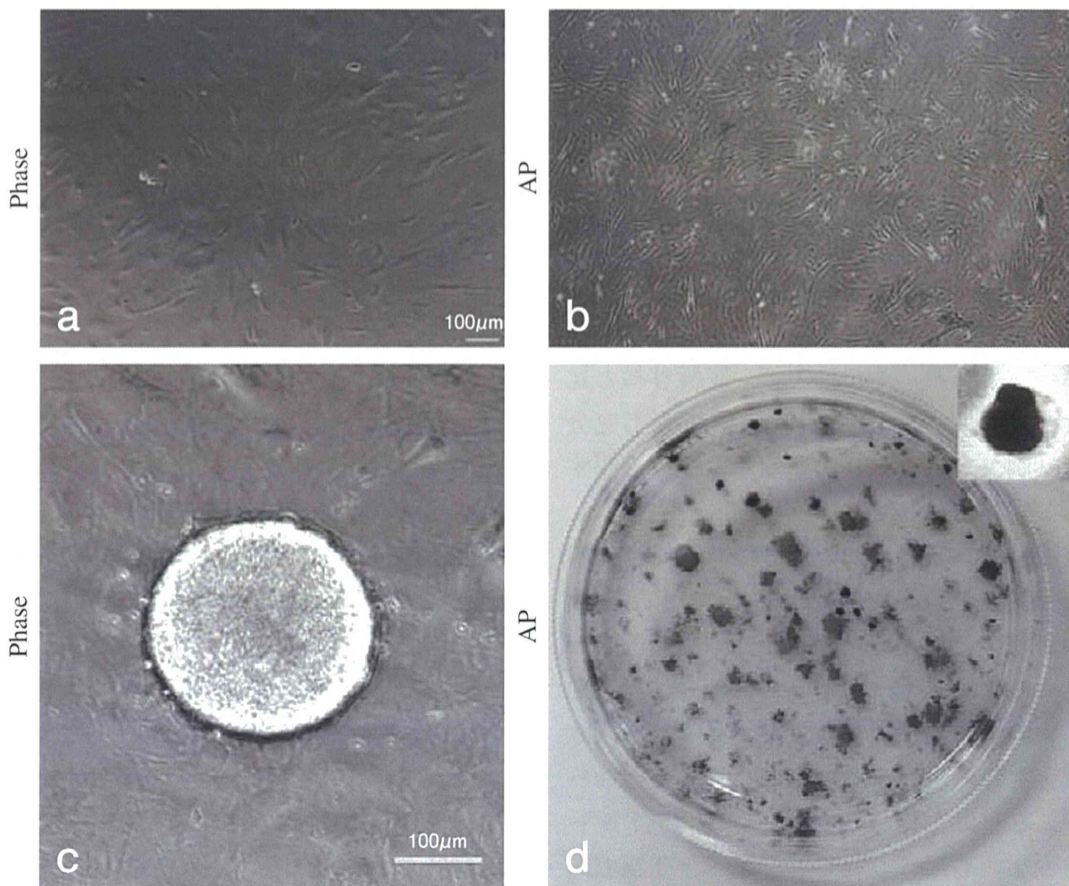


図2 マウス ADSC のリプログラミング前 (a, b), リプログラミング後 (c, d)  
(Kawamoto K et al. Disease Markers 35 : 573-579, 2013<sup>6)</sup>より)

を導入することで、iPS コロニーを誘導することができた(図2)。このことはマウス ADSC でもレンチウイルスベクターを用いたリプログラミングが可能であることを示している。また、FACS アリアを用いて doublet を除去、7-アミノアクチノマイシン D (7-AAD) 染色にて死細胞を除去、CD31-CD45-CD34-分画のうち、CD90<sup>high</sup> 分画と CD90<sup>low</sup> 分画にソートし、プレートに播種した(図3a)。翌日に観察したところ、いずれも形態学的には同様の細胞であった(図3b)。ソート後であっても、1週間培養すると、特に、CD90<sup>low</sup> 分画から、また CD90<sup>high</sup> 細胞が誘導されていた(図4)。したがって、ウイルスベクターを導入するのは、なるべくソート後早期がよいことが示唆される。また、このコロニーは、免疫細胞染色にて Oct3/4 および SSEA1 陽性であった(図5)。CD90<sup>high</sup> 分画および、CD90<sup>low</sup> 分画でソートした ADSC のリプログラミング効率、それぞれ  $1.48 \pm 0.24$ ,  $1.0 \pm 0.15$  と有意差を認め (p = 0.01) (図6, 7)。

## 細胞治療の今後

細胞療法は、ドナーのリンパ球輸注 (donor lymphocyte or leukocyte infusion, DLI) を皮切りに、最近では、樹状細胞、制御性 T 細胞、間葉系幹細胞 (mesenchymal stem cell, mSC) の効果が報告されている。なかでも mSC は、ある特定の細胞に分化誘導させることで、再生医療の細胞ソースとなる可能性に加えて、分化誘導していない mSC 自身にも、免疫修飾作用が知られている<sup>8)</sup>。このことは、分化誘導させた幹細胞の効果がいまだ確立していない現況を鑑みると、分化誘導せずとも、幹細胞自身を移植時に投与することで、成績改善が得られる可能性を示唆している。さらに最近、mSC が免疫抑制作用の可能性に基づき、免疫抑制剤の代替療法としての mSC を用いた臨床試験が、海外を中心に進行中である<sup>9-11)</sup>。また、このメカニズムに関しては、さまざまなサイトカインのオートクラインおよびパラクライン効果などが推察されている。本研究では、この mSC の一種で

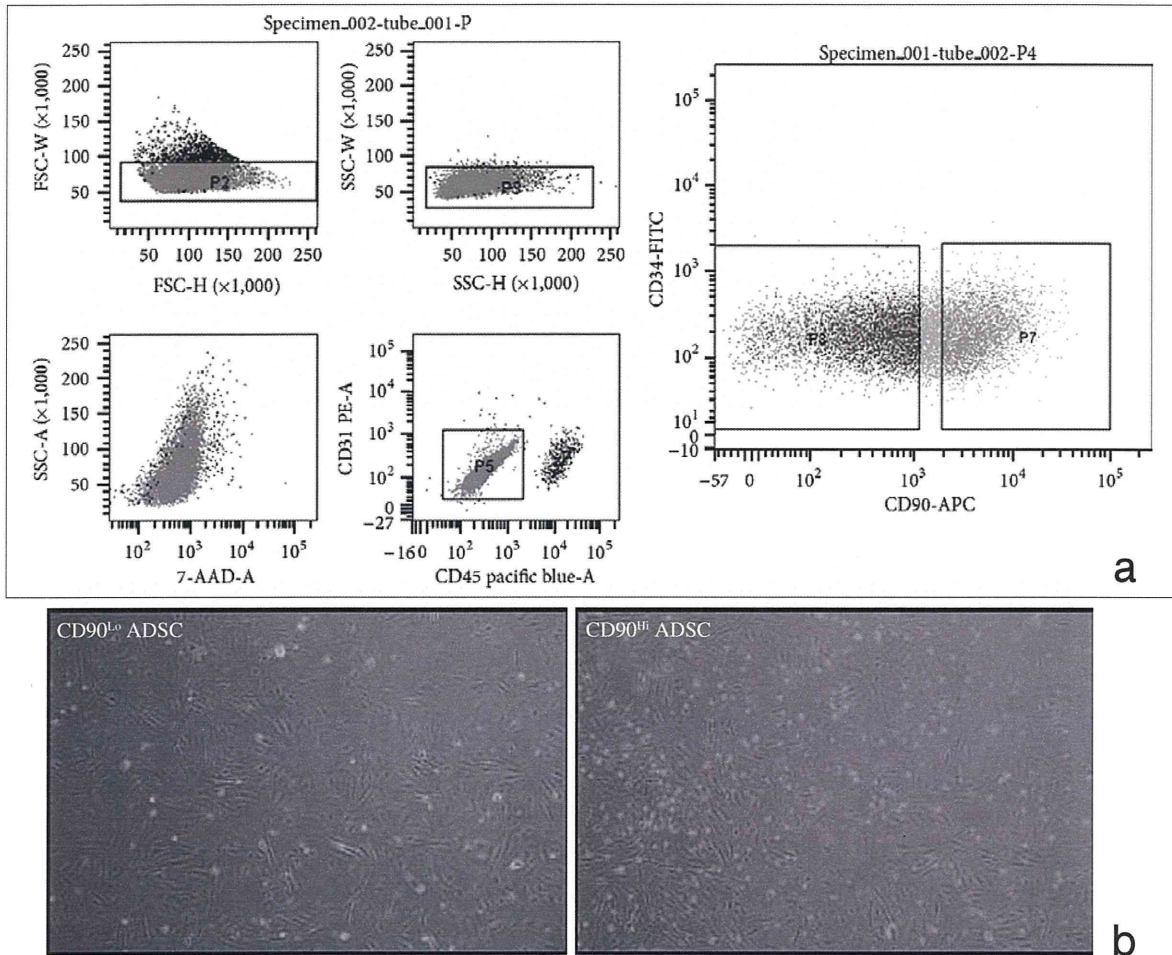


図3 ゲーティングによる分析  
(Kawamoto K et al. Disease Markers 35 : 573-579, 2013<sup>6)</sup>より)

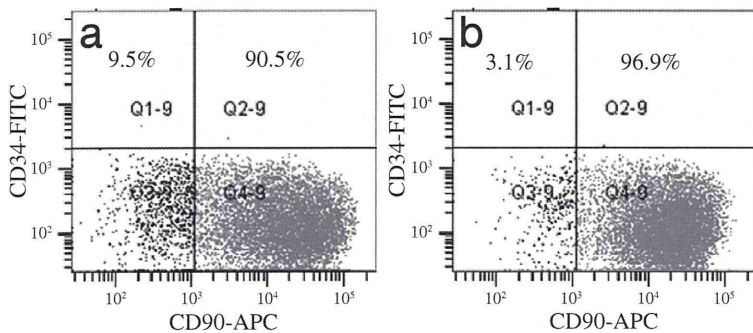


図4  
長期培養の影響  
a : CD90Lo ADSC  
b : CD90Hi ADSC  
(Kawamoto K et al. Disease Markers 35 : 573-579, 2013<sup>6)</sup>より)

ある ADSC を細胞ソースとして用いることで、1 型糖尿病に対する治療の可能性を検討した。

今回の研究で、レンチウイルスベクターを用いて山中 4 因子を導入することでマウス ADSC から iPS 細胞を誘導可能であることを、AP 染色および免疫細胞染色により証明した。細胞の初期化およびダイレクトリプログラミング(細胞の初期化を経ずに他の分化細胞に転換する現象)が、さまざまな転写因子により調節されていることは想

像にがたくないが、いまだにどのように細胞の初期化を制御しているかの詳細は明らかになっておらず、今後の研究の進捗が期待される。

幹細胞との共移植は、臨床移植を含む移植医療の成績を改善するテクノロジーとして期待されている。通常 ADSC そのものを使用するが、将来、移植療法により適した ADSC を峻別可能であれば、さらなる成績向上が期待できる。細胞療法施行時には、どのタイミングで、どれくらいの

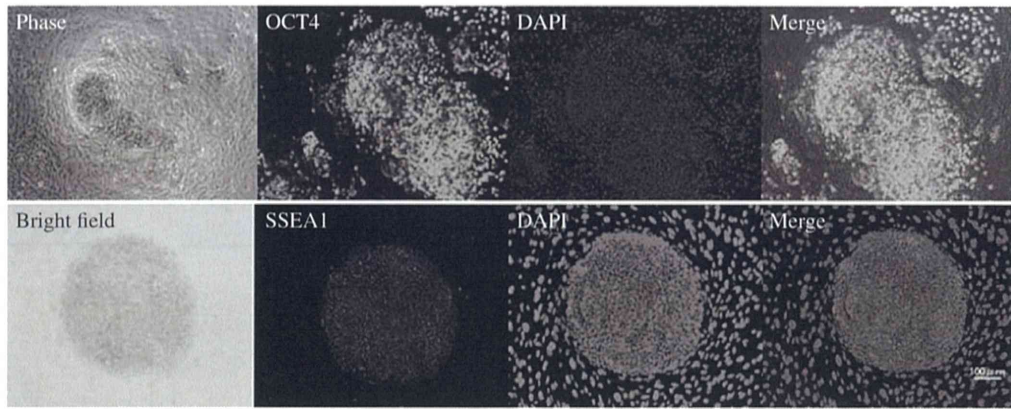


図5 細胞免疫染色

(Kawamoto K et al. Disease Markers 35 : 573-579, 2013<sup>6)</sup>より)

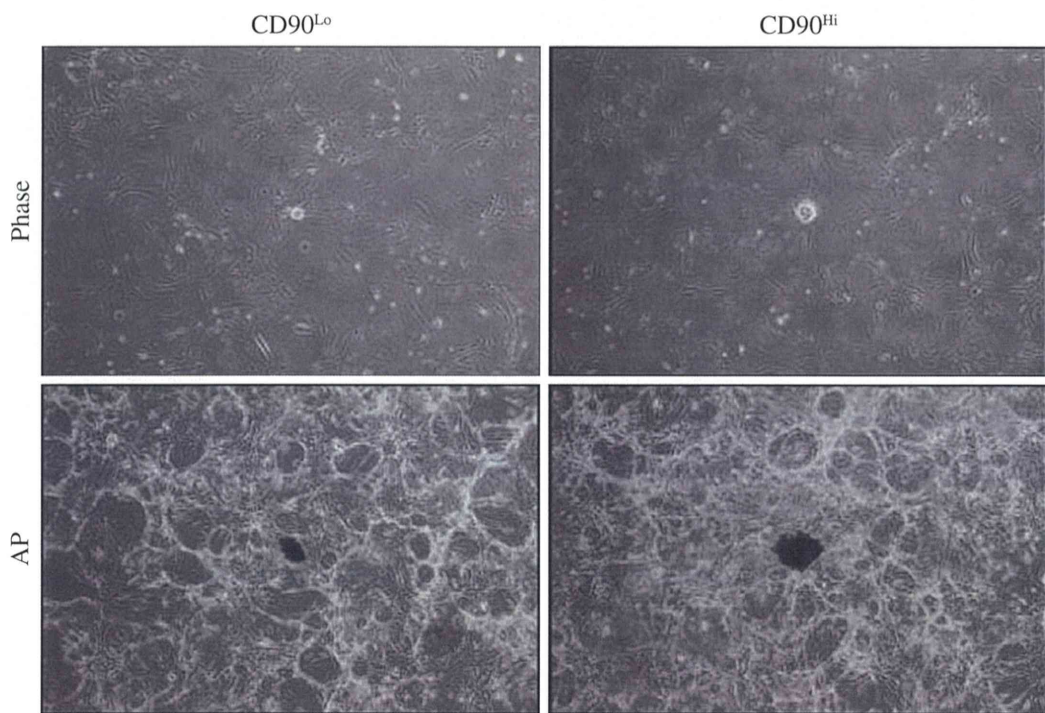


図6 ソート後のリプログラミング

(Kawamoto K et al. Disease Markers 35 : 573-579, 2013<sup>6)</sup>より)

質および量の細胞が必要とされるかは、マウスモデルでは同定不能であり、最終的にヒトでの臨床試験が必要となるが、コスト面から最適の条件を絞り切れていない点もハードルとなる。すなわち、ある細胞に移植成績を改善する効果があっても、使用方法が不適切であれば、ネガティブな結果しか得られない。筆者らの仮説どおりであれば、リプログラミング効率を、幹細胞を選択するときのマーカーとして利用することも可能である。今後は、マウス移植実験にて、実際の有効性を確認し、将来的には臨床検体を用いて同様の実験を計画している。また、臨床応用を考慮し、筆者らのグルー

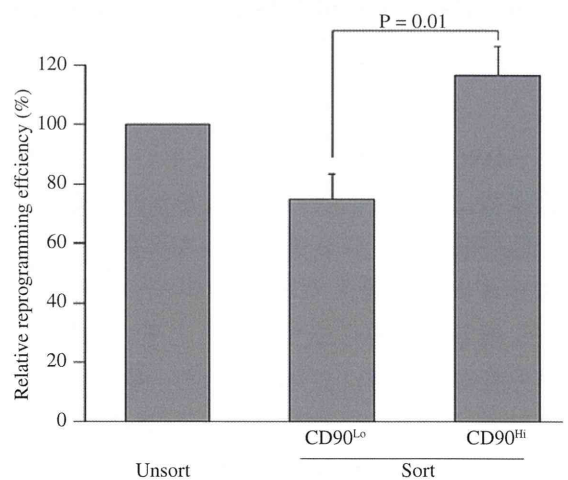


図7 リプログラミング効率

プを含めて、ウイルスベクターを用いない方法も報告されており、今後、ADSC から全能性を持つ細胞を誘導することで、難治性疾患に苦しむ患者への新規治療の可能性がみえてくる<sup>12,13)</sup>。

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本論文では、文献6: Kawamoto et al. *Disease Markers*, 2013<sup>6)</sup>の図を使用していますが、同誌の著作権ポリシー(<http://www.hindawi.com/journals/dm/guidelines/>)によれば適切な引用がなされていれば構わないとされております(Organ Biology 編集委員会)。

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Research paper

## Ethnic and geographic variations in muscle mass, muscle strength and physical performance measures



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### ABSTRACT

**Purpose:** While a universal definition of sarcopenia is desirable, ethnic diversity affects anthropometric measures, which in turn may affect the parameters used for the definition of sarcopenia. Other than Caucasian Asian differences, there may be diversity within different Asian populations. It is important to examine differences, if any, in the field of sarcopenia research. We compared available data (mean body mass index, muscle mass, grip strength, walking speed and chair stand times) for community living older people from different Asian locations and ethnicity to explore the extent of variation, and compared similar data from a Caucasian population.

**Subjects and methods:** Recent community studies which contain anthropometric and physical performance variables for men and women in the three age groups (65–74, 75–84, 85+) were identified from participants of the Asian Working Group on Sarcopenia and from other known longitudinal studies in the region. Caucasian values from the UK Hertfordshire Cohort Study were also used for comparison.

**Results:** There was considerable variation in mean values in body mass index, appendicular skeletal mass index (ASM/ht<sup>2</sup>), grip strength, walking speed between different Asian ethnic groups, and also between same ethnic groups living in different geographic locations. Differences in mean values were greater between the Asian groups compared with Caucasians. Comparison of ASM/ht<sup>2</sup> between Asian groups was limited by the use of different instruments.

**Conclusion:** A universal definition of sarcopenia that depends on absolute measurements may not be applicable to all ethnic groups and different geographic locations.

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## 1. Introduction

In the past two decades, sarcopenia has come to be recognized as an important geriatric syndrome, becoming a major focus of research covering from basic science to clinical management perspectives [1]. A universally accepted definition is thus of great importance, to facilitate the conduct of clinical trials of a preventive or interventional nature. In particular, clinical trials involving pharmaceutical agents would need to fulfil regulatory requirements [2]. There is current consensus worldwide that the working definition has evolved from the original one covering muscle mass only [3] to including measures of muscle strength as

well as physical performance measures [4,5]. Three consensus groups have met and essentially arrived at the same conclusions, although there are minor variations. These are the International Working Group on Sarcopenia (IWGS) which has a predominantly North American input [6], the European Working Group on Sarcopenia in Older People (EWSOP) [7], and the Asian Working Group for Sarcopenia (AWGS) [8]. While there is broad consensus on the choice of measurement of each of these parameters (appendicular muscle mass divided by height<sup>2</sup>, grip strength and walking speed), the classification of what values are normal so that cut-off values may be determined is unclear, there being variations between studies [6]. Among Caucasians, consensus cut-off values appear to have been accepted. Proposed cut-off values are not necessarily based on population studies, which have been limited. However, since all these measures are likely dependent on body

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size and shape, and to a certain extent lifestyle habits [9–11], it is not surprising that studies among Asian populations yield different values. The AWGS agreed on a consensus regarding the cut-off values appropriate for Asians, based on available published studies [8].

However there is ethnic diversity even within Asia with respect to anthropometry and lifestyle, factors which may affect the parameters used for the definition of sarcopenia. Existing data are sparse; yet a comparison of available age group and sex specific mean values used for sarcopenia definition between different Asian populations may begin to address this question. In this study we compared available data (mean body mass index, muscle mass, grip strength, walking speed and chair stand times) for community living elderly people aged 65–74, 75–84, and 85+ for Chinese in Beijing, Hong Kong, Singapore, Japanese, and Malays and Indians in Singapore, to explore the extent of variation. Similar data from a Caucasian population [12] are listed for comparison.

## 2. Subjects and methods

Recent community studies which contain anthropometric and physical performance variables for men and women in the three age groups (65–74, 75–84, 85+) were identified from participants of the Asian Working Group on Sarcopenia, consisting of researchers from Taipei, Beijing, Hong Kong, Japan, Malaysia, Thailand, Korea; and from other known longitudinal studies in the region (Singapore Longitudinal Aging Study [SLA]). The SLA consists of predominantly Chinese, but also smaller numbers of Malays and Indians [13]. Participants from each country and the Principal Investigator of the SLA were contacted to see if they can provide data for comparison. Data from Japan, China (mainland), China (Hong Kong), and Singapore (Chinese, Malays and Indians) were available, although not all parameters were available from all cohorts. Caucasian values from the UK Hertfordshire Sarcopenia Study (HSS), a sub-study of the Hertfordshire Cohort Study (HCS) were also used to compare Asian Caucasian differences [12].

The Hong Kong cohort was the Mr. and Ms Os dataset collected between 2001–2003 as part of a bone health survey, and consisted of two thousand community-dwelling Chinese men and women aged 65 and older recruited by placing recruitment notices in community centers for older adults and housing estates. Participants were volunteers, and excluded those who were unable to walk independently, had had bilateral hip replacement, and not competent to give informed consent [14].

Appendicular muscle mass was measured by DEXA using a Hologic Delphi W4500 densitometer (Hologic Delphi, auto whole body version 12.4, Hologic Inc, Bedford, Massachusetts, USA). ASM was calculated as the sum of appendicular lean mass minus bone mineral content of arms and legs. ASM index (ASMI) was calculated as ASM divided by height in meters squared ( $ASM/ht^2$ ). Grip strength was measured using a dynamometer (JAMAR Hand Dynamometer 5030JO, Sammons Preston Inc, Bolingbrook, IL, USA). Two readings were taken from each side, and the average value between right and left was used for analysis. Gait speed was measured using the average time in seconds to complete a walk along a straight-line 6 meters long. A warm up period of < 5 minutes was followed by two walks, and the average time recorded. Chair stand was measured by asking the participant to rise from a chair (seat height 54 cm) with arms folded across the chest, five times as quickly as possible. The time taken was recorded on a stopwatch.

The Japanese cohort consisted of community-dwelling older people living in both rural and urban areas [15]. Exclusion criteria

were classification as frail according to the long term care insurance certification in Japan; artificial implants such as cardiac pacemakers or joints which precluded the use of bioimpedance for measurement of muscle mass; severe cognitive impairments; severe cardiac, pulmonary or musculoskeletal disorders; comorbidities associated with greater risk of falls such as Parkinson's disease or stroke. Appendicular muscle mass was measured using bioimpedance [Inbody 720, Biospace Co. Ltd, Seoul, South Korea]. Participants stood on two metallic electrodes and held metallic grip electrodes. Grip strength was measured using a hand held dynamometer with the arm by the side of the body. Participants were instructed to squeeze as hard as they can use the dominant hand. The better of two performances was used. Walking speed was measured as the best time taken to walk 15 metres at a comfortable pace. The time required to reach the 10 m point (marked in the course) was recorded using a stopwatch. For the chair stand, participants were asked to stand up and sit down five times as quickly as possible, and the time taken from the initial sitting position to the final standing position at the end of the fifth stand was recorded. The better performance of two trials was taken.

The China (mainland) cohorts consist of volunteers  $\geq 65$  years, being part of a nation-wide survey of the health status of older people carried out from 2010–2013 in different regions of China. Some parameters were only available from the Beijing urban and rural cohorts, which consisted of retired teachers, workers and farmers [unpublished data]. Appendicular muscle mass was not available from this cohort. Grip strength was measured using a hand dynamometer (WCS-II, Beijing), with the highest of two readings for each hand being chosen; walking speed was measured over 6 m.

The Singapore data were from the Singapore Longitudinal Ageing Study [13], which consists of whole population samples from several contiguous small areas in the South East and South West Region of Singapore, covering Singapore citizens and permanent residents who were aged 55 years and above, not physically or mentally incapacitated, able to provide informed consent, participate in face-to-face interviews and carry out physical performance tests. The response rate was 75%. Muscle strength was assessed as knee extension strength. This was measured isometrically in the dominant leg, with the angles of the hip and knee at 90 degrees with the participant seated, using Lord's strap and strain gauge assembly component of the Physiological Profile Assessment (PPA). The best of three trials was recorded in kg [16]. The 6-meter fast gait speed test used the average of two measurements of the participants walking across a distance of 6 m as fast as possible [17]. Single timed chair rise was measured as the time in seconds taken for the participant to complete 5 stands from a seated position on a hard back chair with arms folded [18].

For the Hertfordshire cohort, only data for men were available. Body composition was assessed by anthropometry in all participants and validated using DEXA (Hologic Discovery, software version 12.5) in a sub-set; the walking speed measured as the customary paced walk over 3-m; chair rise time as the time to move from a seated position to fully standing five times unaided, and grip strength measured using a Jamar dynamometer with the maximum value attained from three attempts in both the right and left hands derived as the best grip strength (Promedics, Blackburn, UK) [19].

For each parameter, the mean (SD) values and the lowest 20th percentile values with the exception of chair stand time were listed according to the three age groups for men and women separately. Linear trend ANOVA test was used to examine changes with age within each cohort, and Student's unpaired *t*-test to examine between cohort differences by age and gender, using HK Chinese as a reference group.

**Table 1a**  
Descriptive statistics and comparison by populations on body mass index (BMI).

Ethnic groups	Male						Female							
	n	65–74	n	75–84	n	≥85	P-value <sup>c</sup>	n	65–74	n	75–84	n	≥85	P-value <sup>c</sup>
Mean ( $\pm$ SD), lowest 20th percentile														
Chinese (Hong Kong) <sup>d</sup>	1295	23.62 ( $\pm$ 3.06), 21.25	543	23.08 ( $\pm$ 3.23), 20.2	42	21.98 ( $\pm$ 3.02), 18.91	<0.001	1292	24.19 ( $\pm$ 3.42), 21.37	583	23.53 ( $\pm$ 3.44), 20.68	57	22.5 ( $\pm$ 3.71), 18.71	<0.001
Chinese (Beijing) <sup>d</sup>	1759	24.1 <sup>e</sup> ( $\pm$ 3.1), 21.5	1581	24 <sup>f</sup> ( $\pm$ 3.2), 21.2	347	23.7 <sup>g</sup> ( $\pm$ 3.2), 21	0.002	1950	24.3 ( $\pm$ 3.5), 21.5	1193	23.8 ( $\pm$ 3.6), 20.8	177	23.1 ( $\pm$ 3.6), 20	<0.001
Chinese (Singapore) <sup>d</sup>	353	23.6 ( $\pm$ 3.55), 20.6	148	23 ( $\pm$ 3.68), 19.8	165	22.9 ( $\pm$ 3.76), 19.5	0.043	541	24.2 ( $\pm$ 3.99), 20.9	178	23.8 ( $\pm$ 4), 20.4	204	23.6 ( $\pm$ 3.98), 20.3	0.062
Japanese <sup>d</sup>	266	23.2 <sup>e</sup> ( $\pm$ 3.2), 20.7	254	23.1 ( $\pm$ 2.5), 21	48	21.9 ( $\pm$ 3.6), 19.3	<0.001	650	23 <sup>h</sup> ( $\pm$ 3.4), 20.3	594	22.7 <sup>i</sup> ( $\pm$ 3.11), 20.3	70	22 ( $\pm$ 2.3), 20.5	<0.001
Malays and Indians (Singapore) <sup>d</sup>	41	25 <sup>e</sup> ( $\pm$ 3.57), 21.3	27	23.2 ( $\pm$ 2.38), 21	29	23.2 ( $\pm$ 2.4), 20.5	0.016	67	28 <sup>h</sup> ( $\pm$ 4.25), 24.9	14	24.9 ( $\pm$ 5.17), 18.7	15	25 <sup>j</sup> ( $\pm$ 4.99), 18.9	0.009
UK – HSS <sup>a,d</sup>	81	27.1 <sup>e</sup> ( $\pm$ 3.8), 24.0	24	27.6 <sup>f</sup> ( $\pm$ 2.6), 26.1		<sup>b</sup>			<sup>b</sup>		<sup>b</sup>		<sup>b</sup>	

Lower 20th percentile values are shown in italics.

<sup>a</sup> Hertfordshire Sarcopenia Study (HSS) cohort is only comprised of male participants, and is classified into 2 age groups: 68.3 years–74.9 years and 75.0 years–77.4 years, respectively.

<sup>b</sup> Figures not available.

<sup>c</sup> ANOVA test for linear trend was used to examine any significant difference by age group.

<sup>d</sup> Independent 2-sample *t*-test (2-tailed) was used to examine age-specific difference in mean values, with Chinese (Hong Kong) as reference. Only significant difference ( $P$ -value < 0.05) is reported.

<sup>e</sup> Significantly different from Chinese (Hong Kong) – Chinese (Beijing) (mean difference 0.48,  $P$  < 0.001), Japanese (mean difference –0.42,  $P$  = 0.042), Malays and Indians (Singapore) (mean difference 1.38,  $P$  = 0.005), and UK Caucasian (mean difference 3.68,  $P$  < 0.001).

<sup>f</sup> Significantly different from Chinese (Hong Kong) – Chinese (Beijing) (mean difference 0.92,  $P$  < 0.001), and UK Caucasian (mean difference 4.12,  $P$  < 0.001).

<sup>g</sup> Significantly different from Chinese (Hong Kong) – Chinese (Beijing) (mean difference 1.72,  $P$  = 0.001).

<sup>h</sup> Significantly different from Chinese (Hong Kong) – Japanese (mean difference –1.19,  $P$  < 0.001), and Malays and Indians (Singapore) (mean difference 3.81,  $P$  < 0.001).

<sup>i</sup> Significantly different from Chinese (Hong Kong) – Japanese (mean difference –0.83,  $P$  < 0.001).

<sup>j</sup> Significantly different from Chinese (Hong Kong) – Malays and Indians (Singapore) (mean difference 2.5,  $P$  = 0.035).



**Table 1b**  
Descriptive statistics and comparison by populations on appendicular skeletal mass index (ASM)/height<sup>2</sup> (kg/m<sup>2</sup>).

Ethnic groups	Male						Female							
	<i>n</i>	65–74	<i>n</i>	75–84	<i>n</i>	≥ 85	<i>n</i>	65–74	<i>n</i>	75–84	<i>n</i>	≥ 85	<i>P</i> -value <sup>e</sup>	
Mean (±SD), lowest 20th percentile														
Chinese (Hong Kong)	1295	7.3 (±0.8), 6.66	543	7.01 (±0.82), 6.31	42	6.64 (±0.79), 5.93	<0.001	1292	6.13 (±0.74), 5.51	583	5.94 (±0.68), 5.35	57	5.89 (±0.77), 5.3	<0.001
Chinese (Beijing) <sup>d</sup>	<i>b</i>		<i>b</i>		<i>b</i>			<i>b</i>		<i>b</i>		<i>b</i>		
Chinese (Singapore) <sup>d</sup>	<i>b</i>		<i>b</i>		<i>b</i>			<i>b</i>		<i>b</i>		<i>b</i>		
Japanese <sup>d</sup>	266	6.99 <sup>e</sup> (±1.1), 6.22	254	6.73 <sup>f</sup> (±1.08), 5.57	48	6.28 (±1.71), 4.93	<0.001	650	5.57 <sup>g</sup> (±1.03), 4.6	594	5.17 <sup>h</sup> (±1.01), 4.12	70	4.64 <sup>i</sup> (±0.86), 3.77	<0.001
Malays and Indians (Singapore) <sup>d</sup>	<i>b</i>		<i>b</i>		<i>b</i>			<i>b</i>		<i>b</i>		<i>b</i>		
UK – HSS <sup>a,d</sup>	81	7.99 <sup>e</sup> (±0.91), 7.30	24	7.89 <sup>f</sup> (±0.74), 7.18	<i>b</i>			<i>b</i>		<i>b</i>		<i>b</i>		

Lower 20th percentile values are shown in italics.

<sup>a</sup> Hertfordshire Sarcopenia Study (HSS) cohort is only comprised of male participants, and is classified into 2 age groups: 68.3 years–74.9 years and 75.0 years–77.4 years, respectively.

<sup>b</sup> Figures not available.

<sup>c</sup> ANOVA test for linear trend was used to examine any significant difference by age group.

<sup>d</sup> Independent 2-sample *t*-test (2-tailed) was used to examine age-specific difference in mean values, with Chinese (Hong Kong) as reference. Only significant difference (*P*-value < 0.05) is reported.

<sup>e</sup> Significantly different from Chinese (Hong Kong) – Japanese (mean difference –0.31, *P* < 0.001), and UK Caucasian (mean difference 0.7, *P* < 0.001).

<sup>f</sup> Significantly different from Chinese (Hong Kong) – Japanese (mean difference –0.28, *P* < 0.001), and UK Caucasian (mean difference 0.89, *P* < 0.001).

<sup>g</sup> Significantly different from Chinese (Hong Kong) – Japanese (mean difference –0.56, *P* < 0.001).

<sup>h</sup> Significantly different from Chinese (Hong Kong) – Japanese (mean difference –0.77, *P* < 0.001).

<sup>i</sup> Significantly different from Chinese (Hong Kong) – Japanese (mean difference –1.25, *P* < 0.001).

**Table 1c**  
Descriptive statistics and comparison by populations on appendicular skeletal mass index (ASM)/weight.

Ethnic groups	Male							Female						
	n	65–74	n	75–84	n	≥ 85	P-value <sup>c</sup>	n	65–74	n	75–84	n	≥ 85	P-value <sup>c</sup>
Mean ( $\pm$ SD), lowest 20th percentile														
Chinese (Hong Kong) <sup>d</sup>	1295	0.31 ( $\pm$ 0.02), <i>0.29</i>	543	0.31 ( $\pm$ 0.03), <i>0.28</i>	42	0.3 ( $\pm$ 0.03), <i>0.28</i>	< 0.001	1292	0.25 ( $\pm$ 0.02), <i>0.24</i>	583	0.26 ( $\pm$ 0.03), <i>0.23</i>	57	0.27 ( $\pm$ 0.03), <i>0.24</i>	< 0.001
Chinese (Beijing) <sup>d</sup>	<i>b</i>		<i>b</i>		<i>b</i>			<i>b</i>		<i>b</i>		<i>b</i>		
Chinese (Singapore) <sup>d</sup>	<i>b</i>		<i>b</i>		<i>b</i>			<i>b</i>		<i>b</i>		<i>b</i>		
Japanese <sup>d</sup>	266	0.3 <sup>e</sup> ( $\pm$ 0.04), <i>0.27</i>	254	0.29 <sup>f</sup> ( $\pm$ 0.05), <i>0.24</i>	48	0.28 <sup>g</sup> ( $\pm$ 0.05), <i>0.23</i>	< 0.001	650	0.24 <sup>h</sup> ( $\pm$ 0.04), <i>0.19</i>	594	0.22 <sup>i</sup> ( $\pm$ 0.04), <i>0.18</i>	70	0.21 <sup>j</sup> ( $\pm$ 0.04), <i>0.17</i>	< 0.001
Malays and Indians (Singapore) <sup>d</sup>	<i>b</i>		<i>b</i>		<i>b</i>			<i>b</i>		<i>b</i>		<i>b</i>		
UK – HSS <sup>a,d</sup>	81	0.30 <sup>e</sup> ( $\pm$ 0.03), <i>0.28</i>	24	0.29 <sup>f</sup> ( $\pm$ 0.02), <i>0.26</i>	<i>b</i>			<i>b</i>		<i>b</i>		<i>b</i>		

Lower 20th percentile values are shown in italics.

<sup>a</sup> Hertfordshire Sarcopenia Study (HSS) cohort is only comprised of male participants, and is classified into 2 age groups: 68.3 years–74.9 years and 75.0 years–77.4 years, respectively.

<sup>b</sup> Figures not available.

<sup>c</sup> ANOVA test for linear trend was used to examine any significant difference by age group.

<sup>d</sup> Independent 2-sample *t*-test (2-tailed) was used to examine age-specific difference in mean values, with Chinese (Hong Kong) as reference. Only significant difference (*P*-value < 0.05) is reported.

<sup>e</sup> Significantly different from Chinese (Hong Kong) – Japanese (mean difference –0.01, *P* < 0.001), and UK Caucasian (mean difference –0.02, *P* < 0.001).

<sup>f</sup> Significantly different from Chinese (Hong Kong) – Japanese (mean difference –0.02, *P* < 0.001), and UK Caucasian (mean difference –0.02, *P* < 0.001).

<sup>g</sup> Significantly different from Chinese (Hong Kong) – Japanese (mean difference –0.05, *P* < 0.001).

<sup>h</sup> Significantly different from Chinese (Hong Kong) – Japanese (mean difference –0.01, *P* < 0.001).

<sup>i</sup> Significantly different from Chinese (Hong Kong) – Japanese (mean difference –0.04, *P* < 0.001).

<sup>j</sup> Significantly different from Chinese (Hong Kong) – Japanese (mean difference –0.06, *P* < 0.001).