Human Longevity: Nature *versus* Nurture, Survival *versus* Mortality

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Abstract: Numerous studies were published, investigating the associations of ApoE and ACE polymorphisms with longevity. In 1995, we conducted based cross-sectional study among 823 elderly Arab residents in Israel for studying the prevalence and genetics of dementia of Alzheimer's type (DAT). Epidemiological and genes results of (ApoE + ACE) were reported The ApoE frequency was the lowest in the world and no association was found between ACE and DAT. During 2008, we conducted a follow up study looking for ***survival and mortality*** rate among the same participants to endeavor if there is any relationship between genes and longevity. The total mortality rate among both genders after thirteen years was 66.34%, and the survival subjects of 88 years or older were 34 persons only out of the initial number 270 in 1995 (34/270 = 12.6%). We used the ***Chi-Square test*** to examine the null hypothesis (no difference between observed and expected results): When we compare the survival rate among both sexes in age group (60-74) in 1995 *vs.* age group (73-87) in 2008. We observed statistically significant high survival rate among males ***(p=0.003)*** comparing to females in the same age groups. In contrary, at advanced age, when we compare the survival rate in age group (75-85+) in 1995 *vs.* age group (88-98+) in 2008, we observed high and significant survival rate among females ***(p=0.033).***

193 Years old

Conclusion: High rate of mortality was observed among females in age groups (60-74) in1995 and age group (73-87) in 2008, and high rate of mortality at advanced age was observed among males at advanced age group (75-85+) in 1995 and age group (88-98+) in 2008. More females live longer than men and no association was found between ApoE, ACE genes and longevity, that seems that longevity is exceedingly dependent on other genes in addition to the environmental factors.

Keywords: Apolipoprotein, angiotensin-converting-enzyme, polymorphism, longevity, survival, mortality.

INTRODUCTION

Aging is a late terminal or final physiological process of life. It is a pervasive phenomenon affecting universally all the creatures. Hence, it is nerve-racking and miserable democratic regime. A station where everybody hopes to retrieve myriad things: Mainly rejuvenation, health, dreams and the unredeemed cognitive functions impairment if he still can.

Aging is deleterious, progressive and intrinsic complex disorder, charactarized as a summary term for a group of processes, which contribute to health deterioration, cumulative effects of cell loss over time, age-related cognitive impairment, decline in productivity, increased susceptibility to diseases, risk of system failure, and ultimately to death with the course of time.

Longevity is a collective ageing with extra dose of survival years. Plethora of studies, theories and empirical annotations and ideas on aging and longevity were carried out over the last three decades, and have become so numerous and abundant, and sometimes discrepant and requiring a special disciplines to organize these various and diverse observations into a comprehensive body of knowledge after further reevaluations and validations.

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Regardless of the substantial strides on the research on the underlying etiology of aging, and how we can postpone our monster senescence, the fundamental mechanisms that determine human longevity regardless the different theories, are still mysterious and more scrutinizers efforts still needto resolve this riddle [1].

Recently, multiple compelling evidence supports a role for important genetic and environmental interactions on longevity. Complex multifactorial trait like longevity results from an interaction between environmental factors and sets of epistatic alleles and they generally have pleiotropic age-dependent effects. Hence, gene-environment interactions are important because genes produce their effects in an indirect way (through proteins) and, therefore, the ultimate outcome of gene action may be different in various circumstances [2]. Although, genes do not change over the life course (creating impression of causal links) many traits in later life demonstrate very high environmental plasticity [2].

Most genes associated with complex disorders have low penetrance, which means that the likelihood of developing disease among genotype carriers is low. Substantial basic tenet especially in the era of genome ascribed genes as bearable cause that influences our longevity [3,4]. As a result it was observed upsurge interests in genetic and molecular biology and was thought that genomic damage is the most critically important force influencing human longevity (mutation theory of aging [5]).

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Enormous efforts throughout the last decades have been dedicated in identifying genes associated with longevity that protect from common diseases or slow the aging process. However, longevity possibly is a trait with a genetic background polygenic inheritance seems more likely than monogenic inheritance.

Candidates genes assumed to be involved in the aging process remain to be ascertained, but there are no doubts of their existence and the total genetic contribution to longevity is calculated around 25% [6-8]. Possibly the most widely studied gene in relation to longevity are Apolipoprotein-E (*APOE)*. Additionally, other genes were considered in different case-control longevity studies, one of these genes was the Angiotensin-Converting-Enzyme (ACE).

While ApoE mediates the binding, internalization, and catabolism of lipoprotein particle, but in advancing age ApoE epsilon 4 allele plays a harmful role in accelerating fatal illnesses, whilst, ApoE, which is associated with increased coronary heart disease (CHD) [9-15], atherosclerosis and neurodegeneration, is inversely correlated with longevity [16].

Arousing curiosity, the ApoE2 allele is associated with type III and IV hyperlipidemia, and the ACE allele predisposes to coronary disease. These findings further suggest that genes can exert pleiotropic age-dependent effects upon longevity [17]. Perls *et al.* noted that support for a genetic contribution to human longevity is further provided by data demonstrating that siblings and parents of centenarians live longer [18,19].

In humans one of the forms of a gene coding apolipoprotein E (ApoE2) is associated with exceptional longevity and decreased susceptibility to Alzheimer's disease [20,21]. The survival rate for siblings of the centenarians steadily increased with age to the point that the siblings had a 4-fold greater probability of survival to age 91 [22].

It was estimated that individuals with E4/E4 genotype may have 5 years shorter life expectancy at age 65 compared to individuals with E2/E2 and E2/E3 genotypes [23].

Although, complex diseases such as (CHD and Alzheimer's disease (AD), which occur later in life, then become the major causes of mortality in western countries [24], are connected to ApoE- ε4 which appears to account for up to 40-50% of the genetic risk of AD [25-27].

Away from its various detriments, ApoE have assorted benefits in physiologically important mechanisms like its roles on the health effects of dietary fat intake, transport of cholesterol and in local lipid homeostasis. In addition, ApoE was attributed other credit on lengthen life span. The oxymoron (good and bad role of ApoE), and the above assumptions altogether may create a logical impression that ApoE is associated directly or indirectly to longevity and its effects are, collection of occurring or context dependant [28].

APOLIPOPROTEIN E (APOE)

The Apolipoprotein E (ApoE) is 299 amino acids long secreted glycoprotein that associates with lipoproteins and mediates uptake of these particles into target cells *via* receptor-mediated endocytosis by the low density lipoprotein (LDL) receptor family. ApoE plays an important role in plasma lipoprotein metabolism and in local lipid homeostasis [29] by transporting lipoproteins, fat-soluble vitamins, and cholesterol into the lymph system and then into the blood. Hence ApoE [30], is essential for the normal catabolism of triglyceride-rich lipoprotein constituents, the affinity of which depends on the APOE isoforms [31].

Eighty percent of apolipoprotein E (apoE) is synthesized principally in the liver, while the reminaining 20% is produced by the kidneys, spleen, adrenals, ovaries and brain, etc. [32-39].

APOE gene is polymorphic [40] consists of three alleles (epsilon2, epsilon3 and epsilon4) on the long arm of chromosome 19 at position 13.2 [41-43], and it consists of four Exons and three introns, totalling 3597 base pairs.

ApoE translate into three isoforms of the protein: normal - ApoE-ε3; dysfunctional - ApoE-ε2 and ApoE-ε4 [44]. The ApoE isoforms differ by single amino acid substitutions at one of two positions of the residue protein: The ApoE-ε3, has cysteine at position 112 and arginine at position 158 occurs at the highest frequency, followed by ApoE-ε4 has arginine at position 112; and ApoE- ε2 has cysteine at position 158. These changes cause significant differences in the structure and physical properties of the protein and are responsible for the association of ApoE protein variants with cholesterol metabolism and risk of atherosclerotic vascular disease.

The frequencies of ApoE genotypes vary substantially around the world [45], among Europeans *APOE ε*4 frequencies reveal a North to South cline [46-48]. Therefore, high relative *APOE* allele frequencies were observed in studies with Northern Europe participants (15–21%), and intermediate frequencies in Asia (8–13%) and North America (12–15%), and low frequencies in Southern Europe (4–6%).

The relative high frequencies observed in north Europe have historically been accompanied by high consumption of saturated fat intake. Also immigration phenomenon has been considered as artifact: Migration from regions with relatively high allele frequency may overestimate the decrease in ε4 in aged relative to control, or in contrary, immigrants from areas with relatively low ε4 may underestimate the real initial allele frequency. This depends on the origin of the migrants; if they came from countries with high or low allele frequency. (For example: 31% as reported among Lapps [49], *vs*. 5% have been reported in Sardinians and Taiwanes) [50].

Among caucasian population ε3 allele is the most common, 95% of the population carries at least one ε3 allele, 27% an ε4 allele and 16% an ε2 allele [51]. Carriers of the ε4-allele have higher risk for chronic diseases, relative to people with the most common genotype, ε3/ε3, and to carriers of the ε2-allele who have lower risk, and is associated with exceptional longevity (which suggests, but does not prove, that those with the ε4 gene die younger (an overview of the literature by Gerdes *et al.* [52] found *APOE* ε4 to be associated with premature death), while those with the ε2 live longer [16].

Consequently, then, the frequency of ε4-carriers is lower, and the frequency of ε2-carriers is higher in octogenarians, nonagenarians, and centenarians than in control people [16, 53-56].

United Kingdom researchers have confirmed these results. Studying a population in Belfast who are prone to heart disease, they found that the ε2 variant was significantly more common among those over age 90 and the ε4 variant frequency was reduced in comparison to people aged 30-65 [57]. A Finnish study found that in looking at centenarians, the incidence of the ε2 version of the apolipoprotein gene was 9% among those aged 100-101, 21% for those 102-103, and 25% for those 104 and over [58].

However, different studies have reported mixed results in concerning the issue of longevity, with some documenting null findings for *APOE* e4 and longevity [59,60], and others showing negative associations in subgroups only [61].

Furthermore, human genetic studies have shown that common polymorphisms in one gene — apolipoprotein E (*APOE*) — influence lifespan, by exhibiting a significant influence on human longevity probably mainly through their association with disease [16,52,55].

ANGIOTENSIN CONVERTING ENZYME (ACE)

Angiotensin converting enzyme (ACE) is a zinc metallo-peptidase widely distributed on the surface of endothelial and epithelial cells. Several different names refer to this enzyme in the scientific literature (also known as peptidyl dipeptidase A, EC 3.4.15.1 or ACE) is a type-I membrane-anchored dipeptidyl carboxypeptidase that is essential, for blood pressure regulation by converting decapeptide angiotensin I to biologically active octapeptide angiotensin II, [62] and for electrolyte homeostasis through the renin–angiotensin–aldosterone system, which mediates extracellular volume (*i.e*. that of the blood plasma, lymph and interstitial fluid), and arterial vasoconstriction [63], Also ACE degrades bradykinin, a potent vasodilator, and other vasoactive peptides [64]. The ACE gene, 21 kilo bases (kb) long is located on chromosome 17q23 and consists of 26 exons and 25 introns; an insertion (I)-deletion (D) polymorphism of 287 base pairs has been identified in intron 16 [65].

There are two isoforms of ACE that are transcribed from the same gene in a tissue-specific manner. In somatic tissues it exists as a glycoprotein composed of a single, large polypeptide chain of 1,277 amino acids, whereas in sperm cells it is a lower-molecular-mass glycoform of 701 amino acids [66]. The Insertion (I) allele of the ACE gene has a reported frequency of approximately 0.47 in Caucasians [67]. Barley and colleagues [68] reported a considerably higher (I) allele frequency of 0.91 in Polynesian (Samoan) and 0.85 in South American native (Yanomami) populations. The frequency of the (I) allele in healthy Korean subjects (0.58) was very similar to that in healthy Japanese subjects (0.59) [69,70] and fell between the frequencies reported for Caucasians, Samoans, and Yanomami. The genotypes and (I/D) allele polymorphism of the ACE gene have been reported at different frequencies in different races. About half of the individual variation in plasma ACE depends on the *I/D* polymorphism of the ACE gene; average levels in *DD*, *ID*, and *II* genotypes are found to be approximately 5:4:3 [65].

The differences in the ACE polymorphism between populations may be due to differences in genetic background arising from genetic drift and a founder effect or from a selection process.

Most studies focused on an insertion/deletion (I/D) polymorphism in intron 16 of the *ACE* gene as a marker for a functional polymorphism, which has been related to a variation in the plasma level of the enzyme [65], and ACE activity [69,71] in a codominant pattern.

The deletion (D) allele of the insertion/deletion polymer-phism of the *ACE* gene has been shown to be associated with a higher risk of CHD [72] and hypertension [73-76] even if other studies in contrary found no association with hypertension [77,78] Agarwal *et al.* [79] In 2005, in a review of the genetics of human hypertension, listed a completely new set of 26 association studies, of which 12 published positive and 14 published negative results [79], subsequently, homozygosity for the deletion (*D*) allele was reported to be associated with myocardial infarction, especially in low-risk subjects [80].

While the literature contains discrepant or even contrary reports [81], additional evidence, on balance, supports the latter association [82] and an interrelation between this polymorphism and one in the gene for the angiotensin II type 1 receptor and myocardial infarction has been described [83]. Other studies have found relations between the *D* allele and left ventricular hypertrophy [84], ischemic and idiopathic ardiomyopathy [85], and the expression of hypertrophic cardiomyopathy [86].

Furthermore, other studies [87] associated *ACE/D allele* with longevity. Blanché and other in subsequent studies did not confirm Schacter's finding on longevity association [88,89].

Whereas the insertion (I) allele has been found to be associated with a higher risk of Alzheimer's disease [90-92].

Furthermore, the I allele was associated with increased risk for ruptured intracranial aneurysms [93].

Impact of Apolipoprotein E (ApoE) and Angiotensin convertiong Enzyme (ACE) on longevity and chronic diseases: The Wadi-Ara study.

Since thirteen years ago (October 1, 1995), in an intention to dissect the dilemma of genetic components of multifactorial diseases, we have undertaken a population-based cross-sectional study for studing the prevalence of dementia of Alzheimer's type (DAT); the genetics and the environmental risk factors that may be underlying the etiology of the disease (Tables **1**-**4**).

We conducted a door-to-door survey of all people aged 60 years or older in a geographically defined region among an elderly Arab community in northern Israel. The study population comprised 823 persons (363 males) in the prevalence day, October 1, 1995; all were residents of the rural area of Wadi Ara [94-96] (Tables **1**,**2** and **4**).

In 1999- 2000, we performed a genetics study and we have collected 670 blood samples from the same participants

Table 1. Population Characteristics of Wadi-Ara Study (1995)

	n	Age mean (SD)	No Schooling*	Smoking**	DAT
Males	363	74.4 ± 8.4	41.6% (151/ 363)	54.5% (198/ 363)	14.6% (53/363)
Females	460	69.2 ± 7.7	92% (423/458)	5% (23/460)	25% (115/460)
Total	823	71.5 ± 8.3	70% (574/821)	27% (221/823)	20.4% (168/823)

*Variables for which the distribution among different groups were significantly different (P <.05). Schooling was unknown in 2 patients.
**Smoking refers to smokers (20 cigarettes/day or more) at present or those who had stopped smoking for less than 1 year.

Table 2. Risk Factors for Dementia of Alzheimer Type (DAT)

	Age		Schooling		Gender		Smoking	
	≤71y n=410	>71y n=413	None n=590	≥1 n=231	Females n=460	Males n=363	None n=602	Yes n=221
DAT n (%)	33(8%)	35(33%)	159(27%)	9(4%)	115(25%)	53(15%)	138(23%)	30(14%)
OR (CI)	5.6 (3.7-8.4)		9.0 (4.4-19)		1.9 (1.3-2.8)		0.7 (0.4-1.3)	
p	<.0001		<.0001		.0003		0.25	

Univariate analysis showing the effects of illiteracy, age, gender and smoking as risk factors for the development of DAT.
Smoking has a non-statistically significant effect (the analysis for smoking was done in men, because there were few smokers among females).
The schooling criteria were 1 year or more compared to no formal education.
The age comparison was 71 years or greater compared to 71 or lower (the chosen cut off of 71 years was the median age of the population).
Schooling was unknown in 2 subjects. Patients with vascular dementia (n=49) were excluded from the analysis, but were included in the denominator.

for studying the genetics of (DAT cases, vascular dementia, age related cognitive decline and controls). 441 cases were examined randomly by the polymerase chain reaction (PCR) based method for the determination of their ApoE genotype and for studing other possible candidate genes [97]. (Tables **5**, **6**, and Fig. **1**).

Table 3. Risk Factors for Dementia of Alzheimer Type (Prevalence Study)

Factor	OR (Exp β)	CI	P
Illiteracy	7.9	3.8 -16.9	<.0001
Age	7.6	4.8 -11.8	<.0001
Gender	1.7	1.1 - 2.7	0.0006
Smoking	0.7	0.4 -1.2	0.2

Mutivariate analysis (logistic regression) indicating that the effect of illiteracy on the development of dementia is the highest, exceeding even that of age. Statistically significant effect was also found regarding female gender, while no significant effect was found for smoking.

Table 4. Population Characteristics of Wadi-Ara Study (1995)

Age	Males	Females	Total
60-74	196	357	553
75-84	116	78	194
85+	51	25	76
Total	363	460	823

Table 5. The Frequency of Apolipoprotein E- ε4 Allele in Wadi-Ara Study. (Total Number = 441)

	Age Mean ± SD (years)	Number of subjects	ε4 alleles	ε4 allele frequency
Healthy Subjects	73.5 ± 13	173	10	0.029
AD patients	81.8 ± 9	92	5	0.027
AAMI	74.4 ± 6	136	9	0.033
Others	74 ± 7	40	4	0.05

ApoE- ε4 allele was tested in a random sample of 441 subjects from the total of 823.

Table 6. The Association between Apolipoprotein E Genotype and AD in Different Ethnic Groups: ε4 Allele Frequency

Population (Ethnic groups)	Control	AD Patients
Caucasians	0.137	0.367
Hispanics	0.11	0.31
Canadians	0.05	0.33
US (Mayo Clinic)	0.13	0.35
Israel	0.11	0.27
Africans-Nigerians	0.205	0.167
African-Americans	0.139	0.403
Oriental		
Chinese	0.067	0.169
Japanese	0.093	0.276
Arabs		
Wadi-Ara (current)	*0.029*	*0.027*

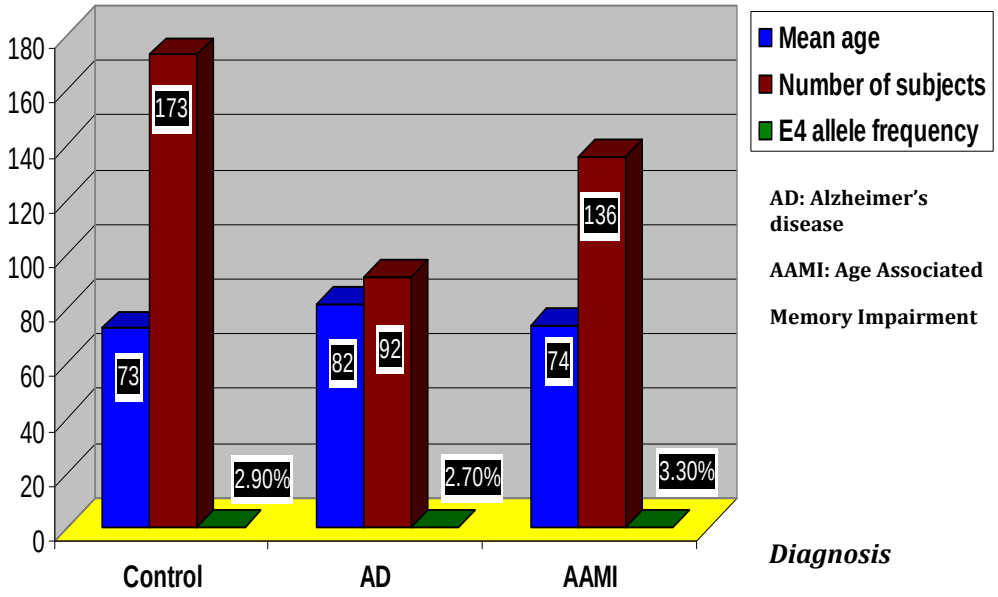


Fig. (1). The frequency of ApoE-E4 allele in Wadi-Ara.

The totals survival subjects 88 years or older today are only 34 persons out of the initial number 270 in1995 (34/270 = 12.6%); (Tables **8A**, **8B** and Fig. **2**).

Table 7. ACE- Genotypic and Allelic Frequencies among AAMI, DAT and Normal Populations

ACE- ID	Normal	AAMI	DAT
II	8(7.14)	3(6.82)	5(5.88)
DI	56(50)	22(50)	35(41.18)
DD	48(42.86)	19(43.18)	45(52.94)
f(I)	72(32.14)	28(31.82)	45(26.47)

Genotypic p-Value:

	AAMI	Normal
DAT	0.5745	0.3731
AAMI		0.9973

Allelic p-Value:

	AAMI	Normal
DAT	0.3660	0.223
AAMI		0.9559

In 2004, supplementary research came to our awareness to investigate the ACE polymorphism as a possible potential risk factor for AD in 75 patients with AD and 75 healthy controls from the same sample. We examined 6 single base polymorphism (SNPs) and an insertion/deletion polymerphism in or near the ACE gene in a series of AD cases and control. For genotyping, we used the homogeneous mass extend reaction (Sequenom) [98] (Table **7**).

2008: Data and Results

Recently, I performed a follow up study, looking for survival and mortality rate among our previous participants (n=823) in Wadi Ara study that was performed in 1995. Reminding the readers that the population characteristics were as follows: [Males number = 363; mean age = (74.4 ± 8.4) years; Females number = 460; mean age = (69.2 ± 7.7) years] (Tables **1**, **2** and **4**). Updated data were collected by personal communication, in addition new lists were provided based on death certificates from the municipalities and local councils in the area.

RESULTS

The mortality rate among males was the same as women [240/363 (66.11%)] *vs.* 306/460 (66.52%)], respectively); the total mortality rates among both genders were [546/823 (66.34%)]. The survival rate among both gender was [277/823 (33.66%)]; (Tables **8A**, **8B** and Fig. **2**).

ANALYSIS AND DISCUSSIONS

One of the triumphs of the 20th century is the increase in lifespan. Substantial improvements in medical and health systems increase the knowlegade of the underlying etiology of diseases *via* new discoveries in different disciplines of medical science, and answer to unsettled challenges of heath problems, altogether these innovations create new equation and strategies of prevention, deductions and early intervention to delay the onset, or cure various diseases, particularly complex diseases.

Actually, the human reap big benefits from these inventions, early diagnosis using sophisticated technology and molecular biology can increase the understanding how to fight different diseases, and by wisdom he accomplishes relatively capability of adaptation to the environmental hazardous condition.

Table 8A. Survival Subjects of Wadi-Ara Study after 13 Years: (2008)

Age	Males (2008)	Females (2008)	Total
73 - 87	108	135	243
88 - 97	13	16	29
98+	2	3	5
Total Survivals	123/363 (33.88%)	154/460 (33.48%)	277/823 (33.66%)

The total number of our participant in Wadi Ara study in 1995 was 823 subjects, generally, every group age increased by 13 years by 2008. For example the age group of (60 -74 in 1995) in (Table **2**), today this group represents the new one of (73 -87 in 2008) in (Table **5**). In 2008; I observed that 546 subjects out off the initial total participants (823) passed away, (546/823 = 66.34%), the survival from different age group (Table **8A** and Table **8B**) were (277/823 = 33.66%).

Table 8B. The Survival Rate (1995 – 2008), after 13 Years of Follow Up in Wadi Ara Study

Age 1995	Age 2008	Males 1995	Males 2008	Females 1995	Females 2008	Total 1995	Total 2008
60-74	+13 (yr)	196	108	357	135	553	243
75-85+	73-87	176	15	103	19	270	29
Total	88-98+	363	123	460	154	823	277

We observe the number of death among males & females after 13 years.
The number of death among males: 363 – 123 = 240
The number of death among females: 460 – 154 = 306
The total death rate in both gender: 546/823 = 66.34%
The total survival rate in both gender: 277/823 = 33.66%
We used the statistical *Chi-square test* to compare observed data with data we would expect to obtain according to a specific hypothesis.
By the chi-square test, which always test what call the null hypothesis, and states there is no significant difference between the expected and observed result. In our study we expect that the number of survival in both gender will be similar, but as we observed that the deviations (differences between observed and expected), were different: When we compare the survival rate among both gender at age group (60-74) at 1995 *vs.* (73-87) at 2008, we observed more survival among males comparing to females (p=0.003), this means that the rate of mortality was higher among females, and the rate of survival was higher among males *(p=0.003).* Indeed, in 2008 we expected to have 86 males alive but we were surprise to found 108 males alive, in contrary we expected to have more females alive (157) but we only found 135 alive at 2008. In contrary at advanced age, when we compare the survival rate at age Group (75-85+) at 1995 *vs.* (88-98+) at 2008, we observed high rate of survival among females *(p=0.033).* Indeed, we expect 21 males alive instead of only 15, and we expected 13 females alive instead of 19 females. In conclusions, males survival was higher at age group (60-87), and females survival was higher at advanced age group (75-98+).

In this concern, the manuscript understudy gave review of ApoE and ACE genes and investigated and critiqued their role in longevity.

Notwithstanding, the facts that carriers of the ε4-allele, which appear to account for up to 40% -50% of the genetic risk of AD [25-27], and have higher risk for other complex diseases occurring predominantly later in life (CHD), and then it become the major causes of mortality in western countries [24]. Aside it was estimated that individuals with E4/E4 genotype may have 5 years shorter life expectancy at age 65 compared to individuals with E2/E2 and E2/E3 genotypes [23]. And because myriad of article reviews and manuscripts through the literature, support the ApoE ε4 association with multifactorial disorders, relative to people with the most common genotype, ε3/ε3, and to carriers of the ε2-allele who have minimum risk for AD. They are associated with exceptional longevity (which suggests, that those with the ε4 gene die younger or at high risk for premature death, while those with the ε2 live longer) [16, 52].

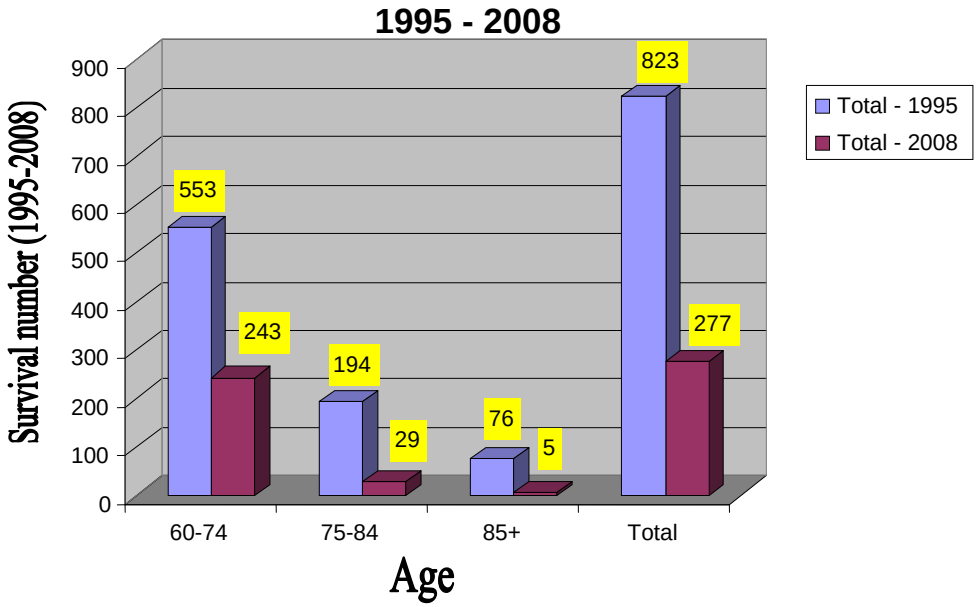
In spite of other different studies that have reported mixed results in concerning the issue of longevity, with some documenting null findings for *APOE* ε4 and longevity [59, 60], and others show negative associations in subgroups only [61,99,100].

In addition, immense numbers of studies were published, investigating associations of Insirtion/Deletion polymerphism of ACE enzyme and ApoE polymorphism with different pathophysiological conditions (especially AD, CHD and Longevity). Several studies found significant associations, whereas others did not confirm those findings; the motive may be related to chance observations, publication bias accounted for the positive associations in the different studies and misclassifications in both genotypic and phenotypic data may lead to false-negative or false-positive results.

2008 Follow Up

I reported results from our previous studies on two genes (ApoE + ACE) [97,98], which were chosen for their impact on Alzheimer's disease (AD), Coronary Heart disease (CHD) and Hypertension, encoding apolipoprotein E (ApoE) and angiotensin−converting enzyme (ACE). Recently (2008) I conducted a new follow up study looking for ***survival and mortality*** rate among the same study participants (n=823) that was conducted in 1995 to endeavor if there are any relationship between these two genes and longevity as were reported in the international literature, and this is indeed the crucial aim of this study:

However, in effort to address the puzzle of complex polyfactorial process like longevity in Wadi Ara community, and to explore if there is any relation between ApoE and ACE genes with longevity, were the frequency of ApoE is the lowest in the world, We found that the frequency of ε4



The figure show the comparison of the survival rate on different aged groups during (1995- 2008).
Age group 60-74 was 553 in 1995 *vs*. 243 in 2008
Age group 75-84 was 194 in 1995 *vs*. 29 in 2008
Age group 85+ was 76 in 1995 *vs*. 5 in 2008
- All survival on 1995 (823) *vs*. 277 in 2008.

Fig. (2). Survival Rate of Wadi Ara Population.

allele of *APOE*, is the lowest in the world [97] (Tables **5** and **6**). Notably, the frequency of ApoE ε4 among DAT, Age Associated Memory Impairment (AAMI) and control was 2.7%, 3.3%, and 2.9% respectively [97] (Table **5** and Fig. **1**). Regarding ACE polymorphism, we did not observe any association between the gene and DAT or AAMI in the elderly Arab population of Wadi Ara [98] (Table **7**).

Despite the involvement of ApoE-ε4 and ACE polymerfisms in different perilous mechanisms that accuses them directly or indirectly in pathology of various diseases in late age and influence lifespan, by exhibiting a significan influence on human longevity probably mainly through their association with disease [16,52,55], which suggests, that those with the ε4 gene die younger (an overview of the literature by Gerdes *et al*. [16,52].

In the light of these statements, in this study the results should be encouraged, because, the effect of both genes (ApoE and ACE) found to be insignificant, neutral and have null effect in this community. The lack of correlation observed should be translated to advantage and reveals protection against different diseases, mainly by enhancing the lifespan in this community [97,98] (Table **5** and Fig. **1**). Indeed, my initial expectation was to found high number of aged people still alive and benefit from circumstances of shortage ApoE gene and lack of association with ACE gene.

Unfortunately, the greater part passed away (66.52%), and the totals survival subjects who are 88 years or older on 2008 are only 34 persons out of the initial number 270 at 1995 (34/270 = 12.6%); (Tables **8A**, **8B** and Fig. **2**).

We used the statistical *Chi-square test* to compare observed data with data we would expect to obtain according to a specific hypothesis.

By the chi-square test, which always test what call the null hypothesis, and states there is no significant difference between the expected and observed result. In our study we expect that the number of survival in both gender will be similar, but as we observed that the deviations (differences between observed and expected), were different. When we compare the survival rate among both gender at age group (60-74) in 1995 *vs*. (73-87) in 2008, we observed more survival among males comparing to females (p=0.003), this means that the rate of mortality was higher among females, and the rate of survival was higher among males *(p=0.003)*. Indeed, in 2008 we expected to have 86 males alive but we were surprised to find 108 males alive, in contrary we expected to have more females alive (157), but we only found 135 alive at 2008. In contrary at advanced age, when we compare the survival rate at age Group (75-85+) in 1995 *vs*. (88-98+) in 2008, we observed high rate of survival among females *(p=0.033)*. Indeed, we expected 21 males alive instead of only 15, and we expected 13 females alive instead of 19 females (Table **8B**).

In conclusion, congruent with previous negative studies our study confirms the lack of association between ApoE and ACE genes with longevity, taking in consideration that the total genetic contribution to longevity is calculated internationaly around (25%). The high mortality rate observed among elderly in this community maybe linked to extrinsic environmental risk factors or to unknown genes which need to be ascertained to reply at least partially to this phenonmenon. Environmental risk factors: as high meat consumption, high rates of smoking, lack of physical activities, stress, depression and inadequate health system may be the underlying cause of speedy mortality in this community.

However, no matter how dissimilar the reasons of death on the globus, death is one inevitable and is similar everywhere. We all need to remember that anywhere something lives; there is up on skies, open in some place, a register in which time is inscribed and our maximum life span is calculated.

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