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Potential Risk Factors for Down Syndrome in Libya

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عوامل الخطر المحتملة لمتلازمة داون في ليبيا

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Abstract

Down syndrome (DS) is a genetic condition of the live-born with an extra full or partial copy of chromosome 21. This extra genetic material has been linked to several risk factors. Some parents have a higher risk of having a child with DS. The current study aimed to evaluate the effect of potential risk factors for DS in Libya. This questionnaire-based study was conducted from September 2022 to December 2022 in different DS centers in Tripoli. One hundred fifty Libyan families (mother, father, and DS child) participated in the study. Data about demographic and health characteristics of the DS children and parental information about potential risk factors for having a DS child was obtained from questionnaires. The study findings indicated that higher parity and parental body mass index were important risk factors associated with DS. Additionally, based on descriptive statistics, maternal folic acid intake before pregnancy was low. Nevertheless, factor analysis results highlighted the fact that the risk of DS is multifactorial, with important contributions from parental age and birth order, parental health and medication use, genetic and environmental exposure, reproductive health and parental obesity. In conclusion, several risk factors for DS were identified in Libya, emphasizing the multifactorial nature of the condition.

Keywords: Down syndrome (DS), Risk factors, Trisomy, Chromosome 21 nondisjunction (Ch 21 NDJ).

الملخص
متلازمة داون هي حالة وراثية تصيب المواليد الأحياء الذين يحملون نسخة إضافية كاملة أو جزئية من الكروموسوم 21. وقد رُبطت هذه المادة الوراثية الإضافية بالعديد من عوامل الخطر. بعض الآباء أكثر عرضة لإنجاب طفل مصاب بمتلازمة داون. هدفت هذه الدراسة إلى تقييم تأثير بعض عوامل الخطر المحتملة لمتلازمة داون في ليبيا. أُجريت هذه الدراسة، التي اعتمدت على الاستبيان، في الفترة من سبتمبر 2022 إلى ديسمبر 2022 في مراكز متلازمة داون المختلفة في طرابلس. شاركت في الدراسة 150 أسرة ليبية (أم، أب، وطفل مصاب بمتلازمة داون). جُمعت البيانات المتعلقة بالخصائص الديموغرافية والصحية للأطفال المصابين بمتلازمة داون، بالإضافة إلى معلومات الوالدين حول عوامل الخطر المحتملة لإنجاب طفل مصاب بها، من خلال الاستبيانات. أشارت نتائج الدراسة إلى أن ارتفاع عدد مرات الحمل والولادة ومؤشر كتلة الجسم لدى الوالدين من عوامل الخطر المهمة المرتبطة بمتلازمة داون. بالإضافة إلى ذلك، وبناءً على الإحصاءات الوصفية، كان تناول الأم لحمض الفوليك قبل الحمل منخفضًا. ومع ذلك، أبرزت نتائج تحليل العوامل أن خطر الإصابة بمتلازمة داون متعدد العوامل، حيث تساهم عوامل مهمة في ذلك، منها اختلافات في عمر الوالدين وترتيب الولادة، وحالة الوالدين الصحية واستخدام الأدوية، والتعرض للعوامل الوراثية والبيئية، والصحة الإنجابية وسمنة الوالدين. وختامًا، تم تحديد عدة عوامل خطر للإصابة بمتلازمة داون في ليبيا، مما يؤكد الطبيعة متعددة العوامل لهذه المتلازمة.

الكلمات المفتاحية: متلازمة داون، عواما الخطر، التثلث الصبغي، عدم الانفصال لكروموسوم 21.

Introduction

Down syndrome (DS) is the most prevalent chromosomal disorder connected to intellectual disability, congenital malformations and unique physical features [1]. John Langdon Down, a physician from Cornwall, England, discovered it for the first time in 1866. Characteristics of DS are the result of the presence of an extra copy of chromosome 21 (Ch 21) due to nondisjunction (NDJ) [2]. The genetic processes that cause DS include meiotic NDJ, Robertsonian translocation and mosaicism. It is approximately that 95% of DS cases have a free trisomy of Ch 21 as a result of meiotic NDJ, Robertsonian translocation in 4% and mosaicism in 1% [3]. The prevalence of DS was estimated by the World Health Organization (WHO) to be between 1 in 1,000 and 1 in 1,100 live births worldwide [4]. There are between 3,000 and 5,000 newborns with this chromosomal abnormality every year. In Libya, the prevalence of DS was a rate of 1 in 516 live births [5].

Risk factors of DS are multifactorial, including biological, demographic, health, and environmental factors. The advanced maternal age (AMA) at conception is considered a significant risk factor for trisomy 21, with DS births being more common in mothers in their later reproductive years as is true for all human autosomal trisomies [6], [7], [8], [9]. While some contradicting reports regarding maternal age (MA) have no decisive influence on the manifestation of DS [10], [11], [12]. Research has demonstrated that older men produce more sperm with aneuploidy including trisomy 21 [13], [14], [15], [16]. However, other studies did not find a link between the increasing paternal age (PA) and the incidence of DS [17], [18], [19].

Some studies have proposed that the elevated risk of Ch 21 NDJ may be linked to a buildup of environmental pollutants that have harmed meiotic machinery and decreased oocyte quality [20], [21]. Thus, maternal occupational exposure may be a significant risk factor for DS [22]. This study was a first of its kind showed that the occupational solvent exposure is possible risk factors of MI NDJ affecting the oocyte. Moreover, the discovery of the exposure to endocrine-disrupting chemicals affects meiosis and raises the incidence of aneuploidy. For instance, exposure to endocrine-disrupting chemicals, including the common environmental pollutant bisphenol A (BPA), impacts the reproductive systems of both sexes (ovaries, testes and reproductive tract) [23], [24].

Moreover, paternal exposure to environmental chemicals such as pyrethroid insecticides was associated with a higher rate of spermatozoa aneuploidy, including Ch21 disomy [25]. In addition, sex chromosomal disomy was exacerbated by occupational factors found in contemporary workplaces, such as sitting for more than six hours at work or being exposed to mechanical vibrations [26]. Furthermore, a study found a correlation between a higher risk of having a child with DS and certain paternal occupations such as janitors, mechanics, farm workers, material-moving equipment operators, food processors, sheet-metal workers, ironworkers, other metalworkers and sawmill workers [27].

Pregnant women with a history of DS are always at a higher absolute risk than their MA-related risk of having a subsequent affected pregnancy [28], [29]. This absolute excess risk is higher for younger than for older women, based on the age at which the affected pregnancy occurred [28].

Some studies have confirmed that parental consanguinity is a risk factor for Ch21 NDJ [12], [30], [31]. However, contradictory study by Rezayat *et al* reported that nonconsanguineous marriages had a higher incidence of DS than consanguineous marriages [32].

Several studies have found a significant association between low maternal education (less than high school) and an increased risk of Ch21 NDJ, specifically during Meiosis II. A study noted that the risk of a DS pregnancy nearly doubled for women with low education [33]. Moreover, lower paternal education levels have also been associated with a slightly higher risk of DS, though the link is often considered less direct than maternal factors

[34]. Oppositely, women with higher education are more likely to have access to and opt for screenings that detect DS early in pregnancy [35].
Reproductive maternal health is an important factor in the occurrence of DS. It was reported that women who have had several spontaneous abortions are likely at a higher risk of aneuploidy [36]. Moreover, it was demonstrated that in comparison to women of the same age with no previous abortions, the relative risk of DS birth increases with the number of abortions and the mother's younger age [37]. In addition, many studies established that mothers of higher parity are at increased risk of DS birth [30], [38], [39], [40]. There is strong evidence that singletons have a higher prevalence of DS than twin fetuses [41]. Despite that, the prevalence of DS in twins overall has been reported by several research [41], [42], [43], [44].
Maternal obesity (BMI ≥30kg/m^2) is connected to an increased maternal risk of the birth of a child with DS compared with maternal normal-weight [45]. Furthermore, increased maternal weight can be an indicator of low socioeconomic status (SES), and Ch21 NDJ has been linked to low maternal SES, which may contribute to errors during maternal meiosis *II* (*MII*) [34], [46], [47]. Moreover, obesity (BMI 30–40 kg/m^2) has been associated with spermatozoa's Ch21 NDJ [48].
Numerous in *vitro* investigations have demonstrated that cells deficient in folate increase the rate of Ch21 NDJ [49], [50]. It was shown that the risk of maternal *MII* errors at AMA is increased when lack of folic acid supplementation during pregnancy [51]. Additionally, men who consumed more folate had fewer sperm with disomies X and 21, sex nullisomy and a lower total sperm aneuploidy measure as compared to men who consume less [52].
Numerous epidemiological investigations have examined preconception use of oral contraceptives (OC) [53], [54], [55], [56]. According to a study in 2001, the risk of DS in children born to mothers under 35 who became pregnant while using OC is comparable to the risk for mothers of DS who are older than 35 [57]. Additionally, it was proposed that the use of tobacco products (smoking cigarettes or chewing tobacco) may worsen by OC tablets used at peri-conception affecting particular types of mistakes that cause Ch21 NDJ during meiosis [56], [58].
The lifestyle factors of smoking, caffeine, alcohol, paternal drug and chemical exposure have been researched as potential causes of aneuploidy in spermatozoa [12], [59], [60].
There is evidence that the Chernobyl reactor accident increased DS cases in Europe, most likely as a result of low-dose irradiation in areas contaminated by radioactive clouds [61].
In respect to the Libyan population, there is no previous research about potential risk factors for DS. Therefore, this study aimed to investigate the effect of potential DS risk factors in Libya.

Material and methods

The population of this questionnaire-based study included 150 families (mother, father, and DS child) who participated in the study from September 2022 to December 2022. Questionnaires were collected from several DS centers in Tripoli including Libyan Society for DS, Children's Teaching Hospital, Tareq Almatar, Tripoli Center for Special Needs, Al-Rashad Center for People with Needs, Roah Alata School for People with Needs and Selah Center for People with Needs.
The questionnaire included demographic and health characteristics of the DS children and parental information about potential risk factors for having a DS child. It consisted of parental age, order of the child, parity, residence area, surrounds the residence area, parental education, parental occupation, consanguinity, family history of DS, twins, parental malnutrition, parental obesity, mother's reproductive function, father smoking, medications before pregnancy, folic acid supplements, oral contraceptives, exposure to X-rays, exposure to chemicals and parental diseases before pregnancy.

Data analysis

The questionnaire data were analyzed using descriptive statistics and the results were presented as percentages. In addition, factor analysis was performed using the SPSS v26 software (Statistical Package for the Social Sciences. Version 26, Chicago, IL, USA) to identify the potential risk factors for DS. Principal Component Analysis (PCA) was performed to explore the underlying factor structure of the data. The Kaiser-Meyer-Olkin (KMO) measure of sampling adequacy was calculated to assess data suitability, and Bartlett's Test of Sphericity was used to ensure sufficient correlations among variables. Factors were extracted based on eigenvalues greater than 1 and visual inspection of the scree plot. Varimax rotation was applied to maximize variance and improve factor interpretability.

Results

Among DS children (Table 1), 78 (52%) were male and 72 (48%) were female (M:F ratio 1.1:1). The diagnosis of DS is usually confirmed by a karyotype test, showing either partial or complete trisomy of Ch21. The questionnaire showed no karyotyping analysis results in 138 (92%), free trisomy 21 karyotypes in 10 (6.7%), mosaic trisomy karyotype in 2 (1.3%) and none with chromosome translocation (Table, 1). It was also found that the highest percentage for the order of the DS child in the family is the third at 30 (20%). The results of the

questionnaire of 150 families regarding prenatal diagnosis (Table, 1) showed that although 115 (76.7%) mothers received prenatal care, in 120 (80%) of the cases, the DS baby was diagnosed postnatally (Figure, 1).

Table *1.* Demographic characteristics of the DS children

Demographic characteristics	No (%)
Gender	
Male	78 (52%)
Female	72 (48%)
DS type	
Free Trisomy 21	10 (6.7%)
Translocation	0 (0%)
Mosaic	2 (1.3%)
No karyotype	138 (92%)
Order of the child	
1	25 (16.7%)
2	22 (14.6%)
3	30 (20%)
4	29 (19.3%)
5	20 (13.3%)
6	16 (10.7%)
7	3 (2%)
8	2 (1.3%)
9	1 (0.6%)
10	1 (0.6%)
11	1 (0.6%)
DS discovered	
During pregnancy	30 (20%)
Immediately after birth	118 (78.7%)
After a period of birth	2 (1.3%)
Prenatal examination	
Yes	115 (76.7%)
No	35 (23.3%)

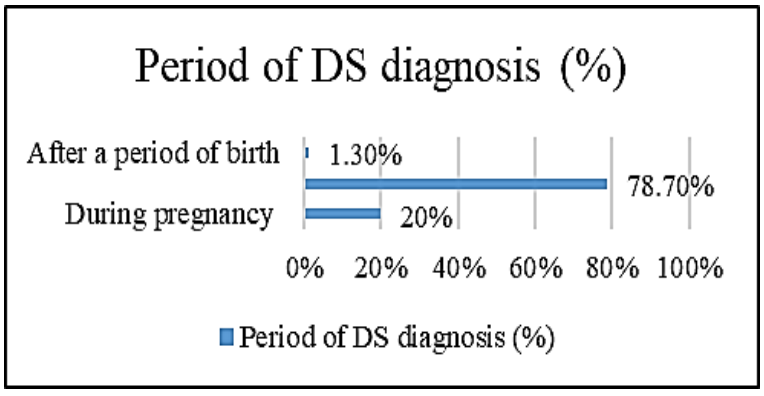

Figure 1. Period of DS diagnosis.

The majority of diagnoses (78.70%) occurred after birth, while 20.00% were made during pregnancy. A small fraction (1.30%) was identified in the period following birth. Data labels indicate the specific percentage for each category.

Among the health problems of children with DS, congenital heart defects (CHDs) were the most common, observed in 31 (20.7%) cases, followed by immunological dysregulation in 6 (4%), combined CHDs and immunological dysregulation in 6 (4%), respiratory system in 3 (2%), vision problems in 4 (2.6%) and nervous system in 2 (1.3%) (Table 2).

Table 2. Health problems risk factors of the DS children

Health problems/ Disease	No (%)
Yes	56 (37.3%)
Congenital heart defects	31 (20.7%)
Immunological dysregulation	6 (4%)
CHDs and Immunological dysregulation	6 (4%)
Respiratory System	3 (2%)
Nervous System	2 (1.3%)
Kidney disease	1 (0.6%)
Digestive	1 (0.6%)
Vision problems	4 (2.6%)
Flat feet	1 (0.6%)
Deformity of the finger	1 (0.6%)
No	94 (62.7%)

The results of this study showed that the percentage of mothers age <35 years old when giving birth to a DS child was 51 (34%) and mothers aged ≥35 years was 99 (66%) with the mean age of the mother was (35.9 ± 5.9), with a range (18–48) years (Table, 3). In addition, the highest percentage of mothers, 55 (36.7%), were between 36-40 years of ages (Table, 3). The percentage of fathers <40 years old was 30% (n=45), and the percentage of fathers aged ≥40 years was 70% (105). The mean age of the fathers was 42.3 ± 6.2, with a range of 26-60 years. Moreover, the highest percentage of fathers between the ages of (41-45) years was 40.7% (61) (Table, 4).

Table 3. Parental age of DS children

Risk factor	No (%)
Maternal age	
<35	51 (34%)
≥35	99 (66%)
Paternal age	
<40	45 (30%)
≥40	105 (70%)

Table 4. Distribution of parental ages in DS cases (N =150).

Age range (years)	No. of mothers (%)	No. of fathers (%)
<20	3 (2%)	-
20-25	8 (5.3%)	-
26-30	14 (9.3%)	5 (3.3%)
31-35	37 (24.7%)	17 (11.3%)
36-40	55 (36.7%)	27 (18%)
41-45	31 (20.7%)	61 (40.7%)
46-50	2 (1.3%)	27 (18%)
>51	-	13 (8.7%)
Total	150 (100%)	150 (100%)

Table 5, presents the distribution of participants according to residence-related risk factors. The majority of participants were from the Tripoli region (88%). The study clearly showed that a greater number of children with DS were born in urban areas (84%) compared to rural areas (16%). Regarding environmental risk factors, the highest percentage of exposure factors surrounding the residential region was for transmission towers (12%).

Table 5. Residental and surrounded exposure characteristics of the DS children parents

Risk factor	No (%)
Residence area	
Tripoli Region	132 (88%)
Western Region	8 (5.3%)
Middle Region	3 (2%)
Eastern Region	4 (2.7%)
Southern Region	3 (2%)
Residence	
Urban region	126 (84%)
Rural region	24 (16%)
Surrounds the area	
Transmission towers (high pressure towers)	18 (12%)
Nuclear energy source	1 (0.6%)
Factory	7 (4.6%)
Oil fields	3 (2%)
Steam station	2 (1.3%)
Camp	4 (2.7%)
Other	3 (2%)
Nothing	112 (74.7%)

Maternal and paternal education results showed that the highest percentages were for high school education and higher, at 123 (82%) and 128 (85.3%), respectively. Furthermore, in terms of maternal occupation, 78 (52%) of mothers were employed and 72 (48%) were housewives. Occupations were classified according to the 2018 Standard Occupational Classification (SOC). The majority 44 (29.3%) of the employed mothers were working in education, training, and library occupation (Figure, 17). Moreover, the most common exposure factors surrounding the workplace including transmission towers, factories or oil fields. The results also showed that the majority of paternal occupations, 43 (28.7%), were in office and administrative support (Table, 6).

Table 6. Education and occupation characteristics of the DS children's parents

Risk factor	No (%)
Maternal education	
High school and higher	123 (82%)
Less than high school	25 (16.7%)
Uneducated	2 (1.3%)
Paternal education	
High school and higher	128 (85.3%)
Less than high school	22 (14.7%)
Uneducated	0 (0)
Maternal occupation	
Housewife	72 (48%)
Employee	78 (52%)
Education, Training and Library	44(29.3%)
Farming ,Fishing, and Forestry	1 (0.6%)
Legal	1 (0.6%)
Office and Administrative Support	15 (10%)
Healthcare Practitioners and Technical	6 (4%)
Healthcare Support	3 (2%)
Protective Service	2 (1.3%)
Community and Social Service	6 (4%)
Mother's job near	
Transmission towers	5 (3.3%)
Factory	2 (1.3%)
Oil fields	2 (1.3%)
High-pressure towers and Factory	1 (0.6%)
Roads and bridges company	1 (0.6%)
Car workshop area	1 (0.6%)
Nothing	138 (92%)
Paternal occupation	
Transportation & Material Moving	15 (10%)
Farming ,Fishing, and Forestry	4 (2.7%)
Architecture and Engineering	13 (8.7%)
Business and Financial Operations	14 (9.3%)
Office and Administrative Support	43 (28.7%)
Construction and Extraction	4 (2.7%)
Healthcare Practitioners and Technical	2 (1.3%)
Healthcare Support	5 (3.3%)
Protective Service	16 (10.7%)
Education ,Training, and Library	10 (6.6%)
Sales and Related	6 (4%)
Military Specific	5 (3.3%)
Management	1 (0.6%)
Building and Grounds Cleaning and Maintenance	3 (2%)
Installation ,Maintenance & Repair	2 (1.3%)
Production occupation	4 (2.7%)
Arts, Design, Entertainment, Sports and Media	1 (0.6%)
Community and Social Service	1 (0.6%)
Personal Care and Service	1 (0.6%)
Father's job near	
Transmission towers	9 (6%)
Factory	11 (7.3%)
Oil fields	12 (8%)
Steam station	1 (0.6%)
Roads and bridges company	1 (0.6%)
electricity company	1 (0.6%)
Nothing	115 (76.7%)

The questionnaire results showed that 116 (77.3%) of the cases had nonconsanguineous marriages, while 34 (22.7%) were the result of consanguineous marriages, most of which were fourth-degree. In addition, there was no family history of the child having DS in 103 (68.6%) cases, while it was found in 47 (31.3%) in different proportions from the mother's and father's side or both. There was DS recurrence in the same family in 3 (2%) of cases. Additionally, the ratio for parity was ≥5 at a rate of 96 (64%) and <5 at a rate of 54 (36%). There were four cases of DS born as twins with another normal child. The majority of pregnancies were normal except for one case where the pregnancy type was IVF (Table, 7).

Table *7.* Consanguinity and family history characteristics of the DS children.

Risk factor	All cases No (%)
Consanguinity	
Yes	34 (22.7%)
Fourth degree	31 (20.7%)
Fifth degree	3 (2%)
Non consanguineous	116 (77.3%)
Family history of DS	
Yes	47 (31.3%)
Mother	17 (14.6%)
First degree	2 (1.3%)
Second degree	7 (4.6%)
Third degree	5 (3.3%)
Fourth degree	3 (2%)
Fifth degree	0 (0)
Father	24 (16.6%)
First degree	1 (0.6%)
Second degree	11 (7.3%)
Third degree	7 (4.6%)
Fourth degree	4 (2.6%)
Fifth degree	1 (0.7%)
Both of them	6 (4%)
No family history	103 (68.6%)
More than one DS child in the family	
Yes	3 (2%)
No	147 (98%)
Parity	
<5	54 (36%)
≥5	96 (64%)
Twins	
Yes	4 (2.6%)
No	146 (97.3%)
Type of pregnancy	
Normal	149 (99.3%)
In Vitro Fertilization (IVF)	1 (0.6%)
Intracytoplasmic Sperm Injection (ICI)	0

The results indicated that 26 (17.3%) of parents had different diseases and health problems in preconception. The study found that 68 (45.3%) of mothers with a BMI of ≥30Kg/m^2 and 95 (63.3%) of fathers with a BMI of ≥30Kg/m^2. Of the 150 DS families studied, spontaneous abortions were observed in 49 (32.7%) families with 30 (61.2%) occurred prior to the birth of a child with DS, while stillbirths were reported in 8 (5.3%). Analysis also found that only one mother used oral contraceptives. A small number of mothers who were interviewed indicated the intake of folic acid supplements at 4 (2.7%), while parents who took certain medications in preconception were 29 (19.3%). Additionally, different environmental exposures such as exposure to X-ray and chemicals whether from the mother, father or both, were 20 (13.3%) and 9 (6%). The results indicated that 58 (38.7%) of fathers were smokers (Table, 8).

Table 8. Parental health condition in preconception of DS parents

Risk factor	All cases No (%)
Malnutrition before pregnancy	
Yes	5 (3.3%)
No	145 (96.7%)
Diseases before pregnancy	
Yes	26 (17.3%)
Mother	7 (4.6%)
Father	16 (10.6%)
Both of them	3 (2%)
No	124 (82.7%)
Maternal obesity (BMI ≥30 Kg/m^2)	
<30 Kg/m^2	82 (54.7%)
≥30 Kg/m^2	68 (45.3%)
Paternal obesity (BMI ≥30 Kg/m^2)	
<30 Kg/m^2	55 (36.7%)
≥30 Kg/m^2	95 (63.3%)
Mother's reproductive function	
Abortion	49 (32.7%)
1	19 (12.6%)
≥2	30 (20%)
Stillbirth	8 (5.3%)
Transplantation	2 (1.3%)
Nothing	91 (60.7%)
Father smoking cigarettes	
Yes	58 (38.7%)
Ex-smoker	4 (2.7%)
No	88 (58.7%)
Medications before pregnancy	
Yes	29 (19.3%)
Mother	17 (11.3%)
Father	9 (6%)
Both of them	3 (2%)
No	121 (80.7%)
Folic acid supplements	
Yes	4 (2.7%)
No	146 (93.3%)
Oral contraceptives	
Yes	1 (0.6%)
No	149 (99.3%)
Exposure to X-rays	
Yes	20 (13.3%)
Mother	15 (10%)
Father	4 (2.6%)
Both of them	1 (0.6%)
No exposure	130 (86.7%)
Exposure to chemicals	
Yes	9 (6%)
Mother	3 (2%)
Father	6 (4%)
I don't know	5 (3.3%)
No exposure	136 (90.7%)
Type of chemical:	
Clorox	3 (2%)
Hydrogen sulfate gas	3 (2%)
Other	3 (2%)
Exposed to radioactive materials	
Yes	1 (0.6%)
Mother	1 (0.6%)
Father	0
Both of them	0
No exposure	149 (99.3%)

In the current study, forty items were identified for factor analysis that represent potential risk factors for the birth of a child with DS. After conducting the initial analysis, it was found that the KMO (0.499) was very low and there were some items with low communalities (<0.30) and high cross-loadings. For this reason, some items were combined into one variable due to conceptual similarity, and items with high cross-loadings or weak loadings were excluded. After adjustments, the final model retained 13 items and 5 factors (Table, 9), and the KMO measure of sampling adequacy was 0.708. Bartlett's test of sphericity was significant ($\chi^2 = 530.038$, $p < .001$). The factors were extracted using PCA and varimax rotation, explaining 65.98% of the total variance.

Table 9. Rotated component matrix.

Items		Component				
		1	2	3	4	5
1	Mother's age of 6 groups	.882				
2	Father's age of 6 groups	.857				
3	Father's age group of two groups	.830				
4	Mother's age group of two groups	.812				
5	Order of the child	.605				
6	The parents tacked certain medications		.806			
7	The parents suffered from diseases		.797			
8	The parents exposed to radioactive materials		.604		.447	
9	Family history			.706		
10	The parents exposed to X-rays			.673		
11	Mother's reproductive function				.842	
12	Parental obesity					.836
13	The parents exposed to chemicals			.518		

Factor analysis was performed with 1 as the Eagan value to improve the strength of the factors. These five distinct factors (components) composed of first factor which is parental age and birth order of the child, with high factor loadings (mother's age = 0.882, father's age = 0.857 and older of the child = 0.605). The second factor is parental health and medication use encompassed parental medical history and medication use before pregnancy (taking certain medications before pregnancy = 0.806). The third facto is genetic and environmental exposure including family history of DS and exposure to X-rays/chemicals (family history = 0.706, parental exposure to X-rays = 0.673 and parental exposure to chemicals = 0.518). The fourth factor is Reproductive Health included the mother's reproductive function (0.842). Finally, the fifth Factor is parental obesity included parental BMI risks (obesity = 0.836).

Discussion

In the present study, the overall gender ratio of DS males and females with M:F ratio of 1.1:1. These results are similar to those found in Indonesia with DS M:F ratio of 1.1:1 [62]. Additionally, the proportions in current study were slightly similar to those reported in the study conducted in Qatar, with DS M:F ratio 1.2:1 [63]. The increased number of males than females is universal and was reported in many studies in different countries [11], [38], [64], [65]. This result may be related to gender bias in DS children. An NDJ mechanism, in which the additional Ch21 preferentially segregates with the Y chromosome, has been implicated in the higher male-to-female sex ratio among children with DS. As a result, Y-bearing sperm have a higher frequency of Ch21 disomy than X-bearing sperm [66]. In contrast, These findings were in disagreement with the study conducted in Libya and reported DS M: F ratio of 1:1.4 [67]. In addition, present results contradicted a Tunisian study found that DS M: F ratio of 1:1.1 [68]. However, in the current study, there was no difference in the proportions of DS M:F ratio for the gender of children with DS of maternal origin (Data not shown). On the contrary, this study reported that all paternal Ch21 NDJ origin were DS males (4 males) (Data not shown). Thus, these findings suggest that disomy 21 sperm are more likely to be Y-bearing than X-bearing, providing the first molecular proof that paternal NDJ may be partially responsible for the more males in DS. In this study, CHDs were the most common health defect in DS children, occurring in 20.7% of cases. This finding was close to that reported in previous studies [11], [62]. In contrast, it was lower than previously reported in Libya and Egypt [67], [69]. Additionally, other health defects appeared in DS children in current study at varying rates and differed from what was reported in previous studies, which may be due to the sample size and population difference [11], [62], [64], [70].

Despite a great deal of study has been done on DS in the past few decades, the majority of the findings about demographic and etiological factors were based on data from Western countries. The circumstances in Libya are completely different, thus, this study insisted to highlight the potential risk factors of DS childbirth. The potential risk factors for the birth of a DS child in all cases (150 cases). Having children later in life is becoming more

prevalent [71]. Most likely as a result of women's greater access to education and their growing focus on their careers [72]. In Libya, between 2015 and 2020, the majority of births occurred to women aged 30 to 34, totaling approximately 190,000 births [73], followed by women aged 35 to 39, who accounted for around 156,000 births during the same period. This indicates a significant number of births among women in their mid to late 30s. In this study, the highest percentage of mothers (36.7%) was between the ages of 36-40 years (Table, 4). In addition, 66% of the mothers were aged ≥ 35 years with the mean age of the mother was 35.9 (Table, 3), which indicates that AMA may be a risk factor for DS in Libya. There is possible explanation for this result, that the birth rate for older mothers is also represent the general population in Libya. There are similarities in the mean ages of the DS mothers between the present study and those conducted by previous Libyan study on 150 DS cases [5]. They reported that the average age of DS mothers was 35.62 years with 82% of the DS mothers over 30 years old. Moreover, the mean age in the present study differed from that reported among mothers of children with DS in other populations. The possible clarifications for this result may be the percentage of AMA, which varies with the MA structure of a population or other different risk factors affecting the populations.

In our study, in all DS cases apart from the determination of Ch21 NDJ origin, the highest percentage of fathers ≥40 years old was 70% with the mean age was 42.3. Moreover, the highest percentage of fathers was 40.7% between the ages of (41-45) years (Table, 16), indicating the possible effect of paternal age in the etiology of DS. This is consistent with what has been reported in other populations [14], [16], [74], [75], [76]. On the other hand, there is a study in Switzerland found that younger men may be more at risk of Ch21 NDJ than older fathers [77]. Moreover, some studies did not find a possible link for increasing paternal age and the incidence of DS (Roth et al., 1983; Thompson, 2019).

This research revealed that a higher proportion of DS children were born in urban areas (84%) as opposed to rural areas (16%). It may be due to the fact that the majority of DS participants were from urban areas. Moreover, urban areas contain several environmental pollutants which may contribute to cause this syndrome. This is in contrast to previous studies in which the percentage of rural areas was higher than urban areas [12], [80]. They related that to several causes, including low socioeconomic status, environmental variables, and nutritional deficiencies.

Maternal and paternal education in this study showed the highest percentages for high school and above at 82% and 85.3%, respectively. This possibly that parents with higher levels of education are more concerned with their children's educational care through their participation in educational schools.

In present study, 52% of mothers were employees and 48% were housewives. Notably, the majority of the employed mothers (29.3%) are working in the education, training, and library occupation. These workplaces are surrounded with transmission towers, factories or oil fields. Therefore, maternal occupational exposure might be a risk factor for birth defects in their children [81], [82]. Maternal exposure to solvents at work may be a risk factor for Ch21 NDJ impacting the oocyte [22]. On the other hand, the majority of parental occupations (28.7%) were office and administrative support, mostly located around oil fields, factories or transmission towers. However, occupational characteristics present in modern workplaces, such as sitting for more than six hours or being exposed to mechanical vibrations, have been shown to increase sex chromosomal disomy [26], [27]

Maternal exposure to chemicals in this study were (2%) and paternal exposure to chemicals (4%). According to some researches, a buildup of environmental pollutants damaged meiotic machinery and reduced oocyte quality which may be increase the risk of NDJ [20], [21]. Moreover, a higher incidence of Ch21 disomy and sex chromosomal disomy was linked to paternal exposure to certain air pollutants, particulate matter (PM2.5), while exposure to PM10 was linked to Ch21 disomy (Jurewicz et al., 2014). A higher incidence of spermatozoa aneuploidy, including Ch21 disomy, was linked to paternal exposure to environmental pollutants such as pyrethroid pesticides [25]. However, the reproductive systems of both sexes (ovaries, testes) are affected by exposure to endocrine-disrupting chemicals, such as the common environmental contaminant bisphenol A (BPA) [23], [24].

An important finding in this study that only (2.7%) mothers have taken folic acid supplements during pregnancy. In this context, some studies have linked folate metabolism to the molecular markers of genomic instability observed in mothers of DS infants [49], [50]. However, lack of folic acid supplementation during pregnancy has been demonstrated to increase the incidence of maternal *MII* mistakes at AMA [51].

Numerous epidemiological studies examined the use of oral contraceptives prior to conception [53], [54], [55], [56], [57]. In present study, only one of the women in our study used oral contraceptives.

Several studies investigated the lifestyle factors that may contribute to aneuploidy in spermatozoa, including smoking, caffeine, alcohol, paternal drug use and chemical exposure [12], [59], [60]. Significant result of the present study is that smoker fathers accounted for 38.7%.

Consanguineous marriage between DS parents has been identified in several studies as a risk factor for Ch21 NDJ [12], [30], [31]. In current findings, 22.7% of the affected children with DS had parents who were consanguineous. This finding agrees with previous Algerian and Egyptian studies where DS parents were blood relatives [38], [70].

In present study, there was family history of the child having DS in 31.3% of cases in different proportions from the mother's and father's side or both. There was a DS recurrence in the same family in 2% of cases. Numerous studies have estimated the chance of trisomy 21 recurrence risk for DS [28], [29], [83]. In addition, the absolute

risk of a second impacted pregnancy is always higher for pregnant women with a history of DS than for a mother's age-related risk [28], [29].

Spontaneous abortions were observed in 32.7% of mothers in this study. It was previously found that women who have experienced several spontaneous abortions are probably more susceptible to aneuploidy [36]. Furthermore, the relative risk of DS birth rises with the number of abortions and the mother's younger age as compared to women of the same age who never had an abortion [37]. In the present study, the ratio for parity was ≥5 at a rate of 64%. Mothers with higher parity are more likely to give birth to DS babies, according to numerous research [30], [38], [39], [40]. Higher statistically significant risk for DS birth for grand multiparas with ≥5 and statistically lower for primiparas [40]. In this result, there were four cases of DS born as twins with another normal child, which has also been reported in previous studies [41], [42], [43], [44]. Importantly, DS was discovered in one or both fetuses in 0.2% of twin pregnancies [42].

The current study indicated that 17.3% of parents had different diseases and health problems in preconception. In addition, it found that 45.3% of mothers with a BMI of ≥30Kg/m^2. This is consistent with what has been stated in many reports [34], [46], [47]. In same line, 63.3% of fathers with a BMI of ≥30Kg/m^2. This findings was consistent with research showing that spermatozoa's Ch21 NDJ has been linked to obesity (BMI 30–40 kg/m^2) [48].

A total of 150 family's participants were included in the factor analysis. The KMO measure of sampling adequacy was 0.708, indicating the dataset was suitable for factor analysis. Bartlett's test of sphericity was significant (χ^2 = 530.038, $p < .001$), confirming that correlations among variables were sufficient for factor analysis. A five-factor model was extracted using PCA, explaining 65.98% of the total variance (Table, 9). Due to conceptual similarity of some items, these items were combined into a single variable. For example, due to conceptual similarity between maternal exposure to X-rays and paternal exposure to X-rays, these items were combined into a single variable Parental X-rays exposure. This approach maintained measurement validity while reducing redundancy in the factor structure. After adjustments, items were excluded due to high cross-loadings or weak loadings (<0.30), ensuring a more distinct factor structure.

The findings emphasize the multifactorial nature of DS risk, with key contributions from distinction parental age and birth order, parental health and medication use, genetic and environmental exposure, reproductive health and parental obesity.

In conclusion, the most crucial risk factors associated with DS were parity and parental body mass index. However, low intake of folic acid was reported in mothers prior to pregnancy. Additionally, factor analysis has shown that the risk of DS might be affected by several factors, with significant contributions from variations in parental age and birth order of the child, health and medication use, genetic and environmental exposure, reproductive health and parental obesity. These findings highlight the multifactorial nature of the DS and emphasize the need for comprehensive prenatal screening, health education, and strengthened antenatal care services to reduce and prevent complications.

Compliance with ethical standards

Disclosure of conflict of interest

The author(s) declare that they have no conflict of interest.

References

[1] J. L. H. Down, "Observations on an ethnic classification of idiots," *Arch. Neurol.*, vol. 25, no. 1, pp. 89–90, 1866, doi: 10.1001/archneur.1971.00490010099014.

[2] J. Lejeune, M. Gautier, and R. Turpin, "[Study of somatic chromosomes from 9 mongoloid children].," *C. R. Hebd. Seances Acad. Sci.*, vol. 248, no. 11, pp. 1721–1722, Mar. 1959.

[3] D. Mutton, E. Alberman, and E. B. Hook, "Cytogenetic and epidemiological findings in Down syndrome: England and Wales 1989-1993," *Am. J. Med. Genet.*, vol. 33, no. 5, pp. 387–394, 1996, doi: 10.1002/ajmg.a.35248.

[4] WHO, "World Down Syndrome Day 21 March," United Nations. Accessed: Jan. 28, 2025. [Online]. Available: https://www.un.org/en/observances/down-syndrome-day

[5] I. C. Verma, A. R. Mathews, A. Faquih, A. A. El-Zouki, G. R. Malik, and F. Mohammed, "Cytogenetic Analysis of Down Syndrome in Libya," *Indian J. Pediatr.*, vol. 57, no. 2, pp. 245–248, 1990, doi: 10.1007/BF02722095.

[6] J. Vranekovic *et al.*, "Down syndrome: parental origin, recombination, and maternal age," *Genet. Test. Mol. Biomarkers*, vol. 16, no. 1, pp. 70–73, 2012, doi: 10.1089/gtmb.2011.0066.

[7] E. G. Allen *et al.*, "Maternal age and risk for trisomy 21 assessed by the origin of chromosome nondisjunction: a report from the Atlanta and National Down Syndrome Projects," *Hum. Genet.*, vol. 125, no. 1, pp. 41–52, 2009, doi: 10.1007/s00439-008-0603-8.Maternal.

[8] P. W. Yoon *et al.*, "Advanced Maternal Age and the Risk of Down Syndrome Characterized by the Meiotic Stage of Chromosomal Error: A Population-Based Study," *Am. J. Hum. Genatics*, vol. 58, no. 3, pp. 628–633, 1996.

[9] A. Jyothy *et al.*, "Parental age and the origin of extra chromosome 21 in Down syndrome," *J. Hum. Genet.*, vol. 46, no. 6, pp. 347–350, 2001, doi: 10.1007/s100380170071.
[10] J. Blanco *et al.*, "Chromosome 21 disomy in the spermatozoa of the fathers of children with trisomy 21, in a population with a high prevalence of dow syndrome: Increased incidence in cases of paternal origin," *Am. J. Hum. Genet.*, vol. 63, no. 4, pp. 1067–1072, 1998, doi: 10.1086/302058.
[11] M. P. Kava, M. S. Tullu, M. N. Muranjan, and K. M. Girisha, "Down Syndrome : Clinical Profile From India," *Arch. Med. Res.*, vol. 35, no. 1, pp. 31–35, 2004, doi: 10.1016/j.arcmed.2003.06.005.
[12] H. M. A. Shalaby, "A study of new potential risk factors for Down syndrome in Upper Egypt," *Egypt. J. Med. Hum. Genet.*, vol. 12, no. 1, pp. 15–19, 2011, doi: 10.1016/j.ejmhg.2011.02.009.
[13] Q. Yang, S. W. Wen, A. Leader, X. K. Chen, J. Lipson, and M. Walker, "Paternal age and birth defects: How strong is the association ?," *Hum. Reprod.*, vol. 22, no. 3, pp. 696–701, 2007, doi: 10.1093/humrep/dem134.
[14] H. Fisch, G. Hyun, R. Golden, T. W. Hensle, C. A. Olsson, and G. L. Liberson, "The influence of paternal age on Down syndrome," *J. Urol.*, vol. 169, no. 6, pp. 2275–2278, 2003, doi: 10.1097/01.ju.0000067958.36077.d8.
[15] E. M. Sartorelli, L. F. Mazzucatto, and J. M. de Pina-Neto, "Effect of paternal age on human sperm chromosomes," *Fertil. Steril.*, vol. 76, no. 6, pp. 1119–1123, 2001, doi: 10.1016/s0015-0282(01)02894-1.
[16] J. Stene, E. Stene, S. Stengel-Rutkowski, and J. D. Murken, "Paternal age and Down's syndrome data from prenatal diagnoses (DFG)," *Hum. Genet.*, vol. 59, no. 2, pp. 119–124, 1981, doi: 10.1007/BF00293059.
[17] J. A. Thompson, "Disentangling the roles of maternal and paternal age on birth prevalence of down syndrome and other chromosomal disorders using a Bayesian modeling approach," *BMC Med. Res. Methodol.*, vol. 19, no. 1, p. 82, 2019, doi: 10.1186/s12874-019-0720-1.
[18] E. B. Hook and R. R. Regal, "A search for a paternal-age effect upon cases of 47,+21 in which the extra chromosome is of paternal origin," *Am. J. Hum. Genet.*, vol. 36, no. 2, pp. 413–421, 1984.
[19] T. R. Oliver, A. Bhise, E. Feingold, S. Tinker, N. Masse, and S. L. Sherman, "Investigation of Factors Associated With Paternal Nondisjunction of Chromosome 21," *Am. J. Med. Genet. Part A*, vol. 149A, no. 8, pp. 1685–1690, 2009, doi: 10.1002/ajmg.a.32942.
[20] S. I. Nagaoka, T. J. Hassold, and P. A. Hunt, "Human aneuploidy: mechanisms and new insights into an age- old problem," *Nat. Rev. Genet.*, vol. 13, no. 7, pp. 493–504, 2012, doi: 10.1038/nrg3245.Human.
[21] F. Pacchierotti and U. Eichenlaub-Ritter, "Environmental hazard in the aetiology of somatic and germ cell aneuploidy," *Cytogenet. Genome Res.*, vol. 133, no. 2–4, pp. 254–268, 2011, doi: 10.1159/000323284.
[22] C. Keen, J. E. Hunter, E. G. Allen, C. Rocheleau, M. Waters, and S. L. Sherman, "The association between maternal occupation and down syndrome: A report from the national Down syndrome project," *Int. J. Hyg. Environ. Health*, vol. 223, no. 1, pp. 207–213, 2020, doi: 10.1016/j.ijheh.2019.09.001.
[23] C. V Sartain and P. A. Hunt, "An old culprit but a new story: bisphenol A and 'NextGen' bisphenols," *Fertil. Steril.*, vol. 106, no. 4, pp. 820–826, 2016, doi: 10.1016/j.fertnstert.2016.07.1114.An.
[24] T. S. Horan *et al.*, "Replacement Bisphenols Adversely Affect Mouse Gametogenesis with Consequences for Subsequent Generations," *Curr. Biol.*, vol. 28, no. 18, pp. 2948–2954, 2018, doi: 10.1016/j.cub.2018.06.070.
[25] M. Radwan *et al.*, "The association between environmental exposure to pyrethroids and sperm aneuploidy," *Chemosphere*, vol. 128, pp. 42–48, 2015, doi: 10.1016/j.chemosphere.2014.12.077.
[26] M. Radwan, J. Jurewicz, P. Radwan, A. Ulańska, L. Jakubowski, and W. Hanke, "Occupational risk factors and frequency of sex chromosome disomy," *Hum. Fertil.*, vol. 18, no. 3, pp. 200–207, 2015, doi: 10.3109/14647273.2015.1006693.
[27] A. F. Olshan, P. A. Baird, and K. Teschke, "Paternal occupational exposures and the risk of Down syndrome," *Am. J. Hum. Genet.*, vol. 44, no. 5, pp. 646–651, 1989.
[28] J. K. Morris, D. E. Mutton, and E. Alberman, "Recurrences of free trisomy 21: Analysis of data from the National Down Syndrome Cytogenetic Register," *Prenat. Diagn.*, vol. 25, no. 12, pp. 1120–1128, 2005, doi: 10.1002/pd.1292.
[29] E. DeSouza, J. Halliday, A. Chan, C. Bower, and J. K. Morris, "Recurrence risks for trisomies 13, 18, and 21," *Am. J. Med. Genet. Part A*, vol. 149, no. 12, pp. 2716–2722, 2009, doi: 10.1002/ajmg.a.33099.
[30] T. I. Farag and A. S. Teebi, "Possible evidence for genetic predisposition to nondisjunction in man," *J. Med. Genet.*, vol. 25, no. 2, pp. 136–137, 1988, doi: 10.1136/jmg.25.2.136-a.
[31] O. S. Alfi, R. Chang, and P. Azen, "Evidence for Genetic Control of Nondisjunction in Man," *Am. J. Hum. Genatics*, vol. 32, no. 4, pp. 477–483, 1980, [Online]. Available: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC1686121/pdf/ajhg00190-0003.pdf
[32] A. A. Rezayat *et al.*, "Down syndrome and consanguinity," *J. Res. Med. Sci.*, vol. 18, no. 11, pp. 995–

997, 2013.
[33] C. P. Torfs and R. E. Christianson, “Socioeconomic effects on the risk of having a recognized pregnancy with Down syndrome,” *Birth Defects Res. Part A - Clin. Mol. Teratol.*, vol. 67, no. 7, pp. 522–528, 2003, doi: 10.1002/bdra.10071.
[34] J. E. Hunter *et al.*, “The association of low socioeconomic status and the risk of having a child with Down syndrome : a report from the National Down Syndrome Project,” *Genet. Med.*, vol. 15, no. 9, pp. 698–705, 2013, doi: 10.1038/gim.2013.34.
[35] M. S. Rana, M. M. Parvin, and M. T. Islam, “The Multifactorial Causes of Down Syndrome During Pregnancy: A Narrative Review of Genetic, Environmental, and Maternal Influences,” *Heal. Sci. Reports*, vol. 8, no. 12, 2025, doi: 10.1002/hsr2.71615.
[36] K. Bianco, A. B. Caughey, B. L. Shaffer, R. Davis, and M. E. Norton, “History of Miscarriage and Increased Incidence of Fetal Aneuploidy in Subsequent Pregnancy,” *Obstet. Gynecol.*, vol. 107, no. 5, pp. 1098–1102, 2006, doi: 10.1097/01.AOG.0000215560.86673.22.
[37] E. B. Hook and P. K. Cross, “Spontaneous abortion and subsequent Down syndrome livebirth,” *Hum. Genet.*, vol. 64, no. 3, pp. 267–270, 1983, doi: 10.1007/BF00279407.
[38] F. Belmokhtar, R. Belmokhtar, and A. Kerfouf, “Cytogenetic study of down syndrome in Algeria: Report and review,” *J. Med. Sci.*, vol. 36, no. 2, pp. 46–52, 2016, doi: 10.4103/1011-4564.181526 ORIGINAL.
[39] V. P. Doria-Rose, H. S. Kim, E. T. J. Augustine, and K. L. Edwards, “Parity and the Risk of Down’s Syndrome,” *Am. J. Epidemiol.*, vol. 158, no. 6, pp. 503–508, 2003, doi: 10.1176/appi.psychotherapy.1970.24.4.545.
[40] K. Källén, “Parity and Down syndrome,” *Am. J. Med. Genet.*, vol. 70, no. 2, pp. 196–201, May 1997.
[41] B. Boyle *et al.*, “Prevalence and risk of Down syndrome in monozygotic and dizygotic multiple pregnancies in Europe: Implications for prenatal screening,” *BJOG An Int. J. Obstet. Gynaecol.*, vol. 121, no. 7, pp. 809–820, 2014, doi: 10.1111/1471-0528.12574.
[42] T. N. Sparks, M. E. Norton, M. Flessel, S. Goldman, and R. J. Currier, “Observed Rate of Down Syndrome in Twin Pregnancies,” *Obstet. Gynecol.*, vol. 128, no. 5, pp. 1127–1133, 2016, doi: 10.1097/AOG.0000000000001690.
[43] A. Garchet-Beaudron, S. Dreux, N. Leporrier, E.-F. Oury, and F. Muller, “Second-trimester Down syndrome maternal serum marker screening: a prospective study of 11 040 twin pregnancies,” *Prenat. Diagn.*, vol. 28, no. 12, pp. 1105–1109, 2008, doi: 10.1002/pd.
[44] P. E. Doyle, V. Beral, B. Botting, and C. J. Wale, “Congenital malformations in twins in England and Wales,” *J. Epidemiol. Community Health*, vol. 45, no. 1, pp. 43–48, 1991, doi: 10.1136/jech.45.1.43.
[45] E. Hildebrand, B. Källén, A. Josefsson, T. Gottvall, and M. Blomberg, “Maternal obesity and risk of Down syndrome in the offspring,” *Prenat. Diagn.*, vol. 34, no. 4, pp. 310–315, 2014, doi: 10.1002/pd.4294.
[46] S. Ghosh, P. Ghosh, and S. K. Dey, “Altered incidence of meiotic errors and Down syndrome birth under extreme low socioeconomic exposure in the Sundarban area of India,” *J. Community Genet.*, vol. 5, no. 2, pp. 119–124, 2014, doi: 10.1007/s12687-013-0159-8.
[47] J. H. Park, B. E. Lee, H. S. Park, E. H. Ha, S. W. Lee, and Y. J. Kim, “Association between pre-pregnancy body mass index and socioeconomic status and impact on pregnancy outcomes in Korea,” *J. Obstet. Gynaecol. Res.*, vol. 37, no. 2, pp. 138–145, 2010, doi: 10.1111/j.1447-0756.2010.01332.x.
[48] J. Jurewicz *et al.*, “The Relationship Between Exposure to Air Pollution and Sperm Disomy,” *Environ. Mol. Mutagen.*, vol. 56, no. 1, pp. 50–59, 2014, doi: 10.1002/em.
[49] X. Wang, P. Thomas, J. Xue, and M. Fenech, “Folate deficiency induces aneuploidy in human lymphocytes in vitro - Evidence using cytokinesis-blocked cells and probes specific for chromosomes 17 and 21,” *Mutat. Res. - Fundam. Mol. Mech. Mutagen.*, vol. 551, no. 1–2, pp. 167–180, 2004, doi: 10.1016/j.mrfmmm.2004.03.008.
[50] S. Beetstra, P. Thomas, C. Salisbury, J. Turner, and M. Fenech, “Folic acid deficiency increases chromosomal instability, chromosome 21 aneuploidy and sensitivity to radiation-induced micronuclei,” *Mutat. Res. - Fundam. Mol. Mech. Mutagen.*, vol. 578, no. 1–2, pp. 317–326, 2005, doi: 10.1016/j.mrfmmm.2005.05.012.
[51] N. D. Hollis *et al.*, “Preconception Folic Acid Supplementation and Risk for Chromosome 21 Nondisjunction : A Report From the National Down Syndrome Project,” *Am. J. Med. Genet. - Part A*, vol. 161A, no. 3, pp. 438–444, 2013, doi: 10.1002/ajmg.a.35796.
[52] S. S. Young, B. Eskenazi, F. M. Marchetti, G. Block, and A. J. Wyrobek, “The association of folate, zinc and antioxidant intake with sperm aneuploidy in healthy non-smoking men,” *Hum. Reprod.*, vol. 23, no. 5, pp. 1014–1022, 2008, doi: 10.1093/humrep/den036.
[53] A. Ray *et al.*, “Maternal Telomere Length and Risk of Down Syndrome: Epidemiological Impact of Smokeless Chewing Tobacco and Oral Contraceptive on Segregation of Chromosome 21,” *Public*

Health Genomics, vol. 19, no. 1, pp. 11–18, 2016, doi: 10.1159/000439245.
[54] B. Klllgn, "Maternal use of oral contraceptives and Down syndrome," *Contraception*, vol. 39, no. 5, pp. 90105-, 1989.
[55] G. R. Nagy, B. Gyorffy, B. Nagy, and J. Rigó, "Lower risk for Down syndrome associated with longer oral contraceptive use: A case-control study of women of advanced maternal age presenting for prenatal diagnosis," *Contraception*, vol. 87, no. 4, pp. 455–458, 2013, doi: 10.1016/j.contraception.2012.08.040.
[56] Q. Yang *et al.*, "Risk factors for trisomy 21: maternal cigarette smoking and oral contraceptive use in a population-based case-control study," *Genet. Med.*, vol. 1, no. 3, pp. 80–88, 1999, doi: 10.1097/00125817-199903000-00004.
[57] M. L. Martínez-Frías, E. Bermejo, E. Rodríguez-Pinilla, and L. Prieto, "Periconceptional exposure to contraceptive pills and risk for Down syndrome," *J. Preinatology*, vol. 21, no. 5, pp. 288–292, 2001, doi: 10.1038/sj.jp.7210538.
[58] S. Ghosh *et al.*, "Epidemiology of Down Syndrome : New Insight Into the Multidimensional Interactions Among Genetic and Environmental Risk Factors in the Oocyte," *Am. J. Epidemiol.*, vol. 174, no. 9, pp. 1009–1016, 2011, doi: 10.1093/aje/kwr240.
[59] A. Kaur and A. Kaur, "Assessment of risk factor associated with down syndrome," *J. Pediatr. Assoc. India*, vol. 9, no. 1, p. 24, 2020, doi: 10.4103/jpai.jpai_3_20.
[60] C. Templado, F. Vidal, and A. Estop, "Aneuploidy in human spermatozoa," *Cytogenet. Genome Res.*, vol. 133, no. 2–4, pp. 91–99, 2011, doi: 10.1159/000323795.
[61] K. Sperling, H. Neitzel, and H. Scherb, "Evidence for an Increase in Trisomy 21 (Down Syndrome) in Europe after the Chernobyl Reactor Accident," *Genet. Epidemiol.*, vol. 36, no. 1, pp. 48–55, 2012, doi: 10.1002/gepi.20662.
[62] E. Zevanya, W. Indrarto, D. Lestari, and T. M. M. Widagdo, "Maternal Age Increases the Risk of Down Syndrome: a Case-Control Study in Yogyakarta, Indonesia," *Berk. Ilm. Kedokt. Duta Wacana*, vol. 9, no. 1, pp. 4–9, 2024, doi: 10.21460/bikdw.v9i1.636.
[63] A. A. Wahab, A. Bener, A. L. Sandridge, and G. F. Hoffmann, "The Pattern of Down Syndrome among Children in Qatar : A Population-Based Study," *Birth Defects Res. Part A*, vol. 76, no. 8, pp. 609–612, 2006, doi: 10.1002/bdra.20290.
[64] W. Amayreh, K. Al Qa'qa, A. Al Hawamdeh, and I. Khashashneh, "Clinical and Cytogenetic Profile of Down Syndrome at King Hussein Medical Centre," *J. R. Med. Serv.*, vol. 19, no. 3, pp. 14–18, 2012.
[65] S. K. Jaiswal, A. Kumar, and A. K. Rai, "Molecular Cytogenetic Classification of Down Syndrome and Screening of Somatic Aneuploidy in Mothers," *Cytogenet. Genome Res.*, vol. 161, no. 8–9, pp. 397–405, 2021, doi: 10.1159/000519624.
[66] D. K. Griffin, M. A. Abruzzo, E. A. Millie, E. Feingold, and T. J. Hassold, "Sex ratio in normal and disomic sperm: Evidence that the extra chromosome 21 preferentially segregates with the Y chromosome," *Am. J. Hum. Genet.*, vol. 59, no. 5, pp. 1108–1113, 1996.
[67] Z. Elmagrpy, A. Rayani, A. Shah, E. Habas, and E. H. Aburawi, "Down syndrome and congenital heart disease : why the regional difference as observed in the Libyan experience ?," *Cardiovasc. J. Afr.*, vol. 22, no. 6, pp. 306–309, 2011, doi: 10.5830/CVJA-2010-072.
[68] M. Aloui *et al.*, "Fetopathological examination for the fetuses with Down syndrome in Tunisia: Epidemiological study and associated malformations," *Pathol. Res. Pract.*, vol. 213, no. 9, pp. 1200–1206, 2017, doi: 10.1016/j.prp.2017.05.001.
[69] A.-H. El-Gilany, S. Yahia, and Y. Wahba, "Prevalence of congenital heart diseases in children with Down syndrome in Mansoura, Egypt: a retrospective descriptive study," *Ann. Saudi Med.*, vol. 37, no. 5, pp. 386–392, 2017.
[70] A. H. El-Gilany, S. Yahia, M. Shoker, and F. El-Dahtory, "Cytogenetic and comorbidity profile of down syndrome in Mansoura university children's hospital, Egypt," *Indian J. Hum. Genet.*, vol. 17, no. 3, pp. 157–163, 2011, doi: 10.4103/0971-6866.92092.
[71] K. Barclay and M. Myrskyla, "Advanced Maternal Age and Offspring Outcomes: Reproductive Aging and Counterbalancing Period Trends," *Adv. Matern. Age Offspring Outcomes*, vol. 42, no. 1, pp. 69–94, 2016.
[72] M. C. Cusimano *et al.*, "Delay of Pregnancy Among Physicians vs Nonphysicians," *JAMA Intern. Med.*, vol. 181, no. 7, pp. 905–912, Jul. 2021, doi: 10.1001/jamainternmed.2021.1635.
[73] S. Galal, "Number of births in Libya between 2015 and 2020, by age of mother," Statista. Accessed: Feb. 21, 2025. [Online]. Available: https://www.statista.com/statistics/1261074/number-of-births-in-libya-by-age-of-mother/
[74] J. Stene, G. Fischer, E. Stene, M. Mikkelsen, and E. Petersen, "Paternal age effect in Down's syndrome," *Ann. Hum. Genet.*, vol. 40, no. 3, pp. 299–306, 1977, doi: 10.1111/j.1469-1809.1977.tb00194.x.
[75] J. D. Erickson and T. O. Bjerkedal, "Down syndrome associated with father's age in Norway," *J. Med.*

Genet., vol. 18, no. 1, pp. 22–28, 1981, doi: 10.1136/jmg.18.1.22.
[76] E. Matsunaga, A. Tonomura, H. Oishi, and Y. Kikuchi, "Reexamination of paternal age effect in Down's syndrome," *Hum. Genet.*, vol. 40, no. 3, pp. 259–268, 1978, doi: 10.1007/BF00291534.
[77] B. Steiner *et al.*, "An unexpected finding: Younger fathers have a higher risk for offspring with chromosomal aneuploidies," *Eur. J. Hum. Genet.*, vol. 23, no. 4, pp. 466–472, 2014, doi: 10.1038/ejhg.2014.122.
[78] M. P. Roth, J. Feingold, A. Baumgarten, P. Bigel, and C. Stoll, "Reexamination of paternal age effect in Down's syndrome," *Hum. Genet.*, vol. 63, no. 2, pp. 149–152, 1983, doi: 10.1007/BF00291534.
[79] G. O. Roecker and C. A. Huether, "An Analysis for Paternal-Age Effect in Ohio's Down Syndrome Births, 1970-1980," *Am. J. Hum. Genet.*, vol. 35, no. 6, pp. 1297–1306, 1983.
[80] L. Verma, F. Macdonald, P. Leedham, M. McConachie, S. Dhanjal, and M. Hultén, "Rapid and simple prenatal DNA diagnosis of Down's syndrome," *Lancet*, vol. 352, no. 9121, pp. 9–12, 1998, doi: 10.1016/S0140-6736(97)11090-X.
[81] S. Lin, M. L. Herdt-Losavio, B. R. Chapman, J. P. Munsie, A. F. Olshan, and C. M. Druschel, "Maternal occupation and the risk of major birth defects: A follow-up analysis from the National Birth Defects Prevention Study," *Int. J. Hyg. Environ. Health*, vol. 216, no. 3, pp. 317–323, 2013, doi: 10.1016/j.ijheh.2012.05.006.
[82] M. L. Herdt-Losavio *et al.*, "Maternal occupation and the risk of birth defects: An overview from the National Birth Defects Prevention Study," *Occup. Environ. Med.*, vol. 67, no. 1, pp. 58–66, 2009, doi: 10.1136/oem.2009.048256.
[83] S. Arbuzova, H. Cuckle, R. Mueller, and I. Sehmi, "Familial Down syndrome: Evidence supporting cytoplasmic inheritance," *Clin. Genet.*, vol. 60, no. 6, pp. 456–462, 2001, doi: 10.1034/j.1399-0004.2001.600609.x.

