Research Article

Risk Factors of Parkinson's Disease: A Case-Control Study in Moroccan Patients

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Abstract

Background: Parkinson's disease (PD) remains a significant health concern in Morocco. Multivariate analysis was not used in any study to evaluate the role of various factors that contributed to the onset of the disease.

Objectives: This study investigates the role of family, environmental, and professional factors in PD development.

Methods: The present study is an age-matched case-control study with risk estimation based on odds ratios (OR) with a 95% confidence interval (CI). In total, 180 cases were matched with 360 controls.

Results: The average age of participants was 68.3 ± 11.2 years. Adjusted logistic regression analysis showed that the family history of PD (ORa = 7.19, CI 95% 3.41 - 15.13), male sex (ORa = 1.92, CI 95% 1.16 - 3.16), spring water consumption (ORa = 3.31, CI 95% 2.05 - 5.34), drug use (ORa = 2.12, CI 95% 1.33 - 3.38), a history of head injury (ORa = 3.38, CI 95% 1.16 - 9.83) and non - consumption of coffee (ORa = 3.04, CI 95% 1.56 - 5.90) were significantly associated with the onset of the disease. In a univariate analysis, well water consumption was observed as a significant risk factor but could not be shown to be significant in a multivariate analysis. Previous work on a farm (ORa = 0.30, CI 95% 0.16 - 0.54) and history of general anesthesia (ORa = 0.47, CI 95% 0.27 - 0.83) were inversely associated with PD risk. No statistical significance was observed in the data on occupational exposure and disease risk, although there was a 30% decrease in risk for the service occupations (ORa = 0.05, CI 95% 0.01 - 0.18).

Conclusions: As a result, further research is needed to determine additional risk factors.

Keywords: Parkinson's Disease, PD, Risk Factors, Conditional Logistic Regression, Morocco

1. Background

Parkinson's disease (PD) is a chronic, age-dependent neurodegenerative condition (1). The incidence of PD is increasing over time, and by 2030, the number of people with the disease will triple (2). There is evidence that the disease is present throughout the world, with studies in Africa producing hospital frequencies of 0.3 to 2.3% of neurological diseases (3).

Parkinson's disease is a multifactorial disease, but the complex interplay between the various factors is only just beginning to be understood. Parkinson's disease is primarily caused by aging (4, 5). Parkinson's disease is also associated with environmental exposures and genetic susceptibility. Parkinson's disease was associated with several environmental factors. Among these factors are pesticide exposure, head injuries, rural living, agricultural occupations, and drinking well water. Family history is associated with an increased risk of PD (6).

Despite the improvement of the Moroccan health system, there is limited epidemiological data on PD. The incidence and prevalence of this disease, however, increase with age. As life expectancy increases, projections indicate that the number of people over 60 will increase from 2.7 million in 2010 to 5.8 million in 2030 (7), possibly leading to an increase in the number of people with PD. According to a systematic analysis of epidemiological studies, PD prevalence increased by 39.7% in Morocco between 1990 and 2016 (8). Compared to other North African populations, Moroccans present a high genetic predisposition to PD due to the prevalence of mutations in the leucine-rich repeat kinase2 (LRRK2) and Parkine2 genes. These genes

correlate with PD risk (9, 10).

2. Objectives

Research like this will provide a better understanding of the etiology of PD, primarily since genetic causes represent only 5 - 10% of cases, while 75% are of uncertain origin, known as idiopathic PD. The results of this study will serve as a basis for identifying at-risk groups in the population in order to develop new prevention and treatment strategies.

3. Methods

This study is a case-control study aimed at identifying various characteristics of individuals associated with PD risk. Researchers examined risk factors for the disease, identified individuals at risk, and compared their characteristics with those of healthy control subjects.

3.1. Participants and Criteria of Inclusion

In this study, 180 patients (males and females), who had previously been diagnosed with PD, were examined cross-sectionally. By the recommendation of the Ethics Committees of the Faculty of Medicine and Pharmacy of Marrakech, all selected patients gave their written consent. The control subjects were recruited from departments other than the Department of Neurology without a prior diagnosis of neurodegenerative disease. Each case was compared with two age-matched controls per year to minimize the possibility of confounding factors.

3.2. Size of Sample

Open-epi Calculator determines the Size. Under this formula, the minimum sample size is 540 participants, distributed over 180 cases and 360 controls, for a power (% chance of detection) of 80%.

3.3. Data Collection

The study participants were informed of the objectives and protocol of the study. A physician conducted a neurological and clinical examination following the consent in writing. Clinicians completed a detailed clinical questionnaire during an interview with the patient to collect data.

3.4. Variables of Interest

The socio-demographic data and other clinical information. Additionally, the log sheet included questions regarding occupational experience and duration of employment. There was a list of all occupations in which participants had been employed for an extended period. The Dictionary of Occupational Titles (DOT) was used to categorize jobs.

In this survey, lifestyle habits were asked about past or present habits. The subjects were also surveyed regarding the medical and surgical comorbidities associated with PD.

3.5. Statistical Analysis

SPSS version 21 was used to process and analyze the data. The percentages of numbers were compared between groups using both Student's t-test and chi-square tests. The chi-square test was used for categorical variables and continuous variables. The Student's t-test was used. Because of the inequalities in sample sizes, the equality of variances premise was systematically verified. The Welsch test was used if there was a significant difference. An adaptation of the Student's t-test, the Welch t-test, is used for two samples with potentially unequal variances (11).

3.6. Analysis of Factors Associated with Parkinson's Disease

The factors associated with the disease were analyzed using a conditional logistic regression model in which the variable to be explained was the presence or absence of disease. For the present study, that model was developed through the following steps: choosing the independent variables, exploring data and verifying associations (analysis of variable distribution), and conducting univariate and multivariate analyses. The odds ratio (OR) was used to quantify the strong association between the variables. A multivariate analysis was conducted using conditional logistic regression to estimate the adjusted odds ratio with confidence intervals. Variables inserted in a model were selected according to their clinical significance and statistical association with a dependent variable at a 20% significance level in the univariate analysis (P value of Wald's test).

3.7. Modeling Strategy

In the analysis, all explanatory variables that appeared significant in the interpretation of a dependent variable were retained in a final model. The regression procedure is based on a stepwise approach.

3.8. Model Selection Criteria

In the end, all variables that were not significant on the 5% significant level were excluded from the model. Akaike Criterion was used to estimate the quality of the model. We used the Nagelkerk R2 to examine the model summary and verify its association strength.

3.9. Ethical Consideration

According to the Declaration of Helsinki and the requirements of the Medical Ethics Committee of the Faculty of Medicine and Pharmacy in Marrakech, the study group adhered to the ethical principles of biomedical research. The Ethics Committee also approved a protocol for the study of the Faculty of Medicine and Pharmacy of Marrakech.

4. Results

4.1. Comparison of Socio-demographic Variables Between Cases and Controls

Five hundred forty eligible participants (180 cases and 360 controls) were invited to participate in this study. Table 1 compares the general demographic characteristics of cases and controls. Cases and controls did not differ in age, with controls averaging 68.09 years (SD 11.3) (t = -0.278, P > 0.05). Due to the matching, the age distribution among controls was comparable. Males' average age was 68.81 years (SD 10.84), and females' average age was 67.67 years (SD 11.71). In total, 54.07% of the participants (n = 292) were males. There were significant gender differences between the case and control groups (P < 10 - 3). Cases and controls have similar education levels.

4.2. Comparison of Cases and Controls by Principal Occupation

This study included a professional story. The dictionary of occupational titles (DOT) was used to code jobs. The DOT classified jobs into nine categories based on their similarity to other occupations. Both cases and controls in this study worked in the category of "service occupations" (cases 39.4%, controls 28.3%). Professionals in this group include military personnel, homemakers, housekeepers, janitors, and security guards. Table 2 shows that there was a significant difference between cases and controls by occupational and industrial category, except for benchwork occupations and agricultural, fishery, forestry, and related occupations (see Table 2). Comparisons were made between the patient and control groups. (Tables 1 and 2) present these comparisons.

Table 1. Comparison of Socio-demographic Variables Between Cases and Controls [a]

Characteristics	Cases	Controls	P Value
Number of participants	180	360	
Age (y)	68.09 ± 11.3	68.38 ± 11.24	0.78 [b, c]
Age (y)			0.42 [d]
$\leq$ 50	13 (7.2)	23 (6.4)	
> 50	167 (92.8)	337 (93.6)	
Body mass index (kg/m^2)	25.3 ± 3.9	25.7 ± 3.2	0.2 [b]
Gender			
Female	63 (35)	185 (51.4)	
Male	117 (65)	175 (48.6)	0.0001 [d]
Area of residence			0.53 [e]
Urban	108 (60)	226 (62.7)	
Rural	72 (40)	134 (37.3)	
Level of education			0.27 [e]
None	92 (51.1)	198 (55)	
Primary	44 (24.4)	92 (25.5)	
Secondary	28 (15.6)	53 (14.7)	
University	16 (8.9)	17 (4.8)	

[a] Values are expressed as mean ± SD or No. (%).
[b] Student's t-test
[c] Age matching
[d] Fisher's test
[e] Chi-square test

Table 2. Comparison of Cases and Controls by Major Occupation (Dictionary of Occupational Titles) [a]

Occupation (Classification Dictionary of Occupational Titles #)	Cas	Témoins	P Value*
(0/1) Professional, technical, and managerial occupations	14 (7.7)	09 (2.5)	0.004
(2) Clerical and sales occupations	14 (7.7)	12 (3.3)	0.02
(3) Service occupations	71 (39.4)	102 (28.3)	0.009
(4) Agricultural, fishery, forestry, and related occupations	51 (28.3)	77 (21.4)	0.07
(5) Processing occupations	5 (2.7)	79 (21.9)	0.001
(6) Machine trades occupations	5 (2.7)	02 (0.5)	0.03
(7) Benchwork occupations	3 (1.6)	11 (3)	0.4
(8) Structural work occupations	11 (6.1)	61 (16.9)	0.001
(9) Miscellaneous occupations	10 (5.5)	09 (2.5)	0.08

[a] Values are expressed as No. (%).
[b] Pearson chi-square test

4.3. Univariate Analysis

The univariate Logistic regression analysis revealed that the distribution of gender among both populations was different: there was a preponderance of males in cases.

There was a 1.96-fold (CI 95% 1.35 - 2.84) increase in the risk of the disease in males compared to females. Cases (n = 43, 23.9%) had more family history of PD as controls (n = 18, 5%). Family history of PD correlated with significantly increased risks of PD (OR = 5.96, (CI 95% 3.32 - 10.70)). Also, a previous head injury is a risk factor with an OR of 3.41 (CI 95% 1.51-7.68). Additionally, a history of diabetes is associated with an Odds Ratio of 1.12 (CI 95% 0.76 - 1.66) but is not statistically significant. The use of medications for a condition other than PD was less frequent in the group with PD than in the controls. The use of these medications was associated with a decreased risk of PD (OR = 0.55, (CI 95% 0.38 - 0.79)).

The risk of the disease is increased with lower levels of education. Compared with university-level instruction, people without any level of schooling have an odds ratio of 2.02 (CI 95% 0.98 - 4.78). The consumption of well - water constitutes a PD risk factor with odds ratios of OR = 1.93, (CI 95% 1.34 - 2.78). On the contrary, source drinking was significantly and negatively associated with the risk of disease (OR = 0.29, (CI 95% 0.20 - 0.43)). Bivariate analysis with definite smoking status in three classes indicated an inverse association between tobacco use and disease risk (OR = 0.69, (CI 95% 0.26 - 1.79)), but not significantly compared to non - smokers. We observed a stronger association for former smokers (OR = 1.65, (CI 95% 1.04 - 2.64)). A significant inverse association was reported with coffee consumption (OR = 0.46, 95% CI 0.32 - 0.66). Such association was mainly detected for consumption of more than 21 years (OR = 0.3, CI 95% 0.12 - 0.71) (Table 3).

There were also higher odds ratios for technical and management occupations (OR = 4.25, (CI 95% 1.68 - 10.73)), clerical and sales occupations (OR = 2.95; (CI 95% 1.28 - 6.78)) and service occupations (OR = 1.53, (CI 95% 1.05 - 2.24)) in the case/control occupations analysis. Processors were found to have a lower risk of PD (OR = 0.07; (CI 95% 0.02 - 0.22). The protective associations ranged from 0.07; (CI 95% 0.02 - 0.22) for professionals in processing occupations to 0.31; (CI 95% 0.16 - 0.61) for those in structural occupations.

The risk estimation was not statistically significant for agriculture - related activities and metalworking occupations. OR = 1.22; (CI 95% 0.79 - 1.88) and OR = 1.08; (CI 95% 0.423 - 2.765), respectively. In contrast, homemakers had a high risk of contracting the disease (OR = 1.74, CI 95% 1.11 - 2.72). Previous work on a farm significantly reduced the risk (OR = 0.42; (CI 95% 0.27 - 0.63)). In Table 4, we present the odds ratios associated with specific occupations among both groups.

4.4. Multivariate Analysis

In the multivariate analysis, we used a descending stepwise logistic model with Wald's test probabilities of no

more than 20%. With the addition of the area of residence (forced factor), we included only the 16 variables significantly associated with PD risk in univariate analysis. The logistic regression model considered all variables for which the P value was less than 0.05.

The interaction effects between the variables were detected. A final model is composed of the nine non-interactive variables at the end of the step-by-step descending procedure. Multivariate analysis revealed significant associations of several predictors with PD in this population when independent variables were introduced descendingly. Spring water consumption exposes a person to a risk of 3. In contrast, familial PD is associated with a positive association. There is a seven-fold increase in disease risk. In this study, the use of drugs contributed to the risk for PD (ORa = 2.12, (CI 95% 1.33 - 3.38)), whereas surgery under general anesthesia was significantly negatively associated with PD (ORa = 0.47, (CI 95% 0.27 - 0.83)). There was a 3-point (P × 0.05) higher risk of PD for non-coffee drinkers after adjusting for age and smoking. The results indicate a significant inverse association between past farm work and depression (ORa = 0.30 with a 95% confidence interval (CI 95% 0.16 - 0.54)). It appears that working in processing occupations ORa = 0.05, (CI 95% 0.01 - 0.18) is protective of the occurrence of the disease. Based on the baseline data, the coefficient of adjustment measures the fit of the model. Cox & Snell have an R-square of 33%, Nagelkerke has an R^2 of 48%, and McFadden has an R^2 of 30% for this study. Those values indicate a positive correlation between the dependent and independent variables. According to this model, 48% of the variance of the studied variable can be explained by this model.

5. Discussion

Multifactorial pathogenesis of PD requires the identification of the population at risk. Many epidemiological studies (12) support a hypothesis of an increased risk of development of PD in the male population. Our findings are consistent with those of these earlier investigations. In the present research, when the gender factor was separately analyzed or adjusted (by age and area of residence), a significantly enhanced risk of the disease was observed for the males (crude OR = 1.96, (CI 95% 1.35 - 2.84), and (ORa= 1.92, (CI 95% 1.16 - 3.16)), both respectively.

Several studies have also reported an association between education and PD. The lowest level of education in this study was associated with a positive risk (crude OR = 2.02, (CI 95% 0.980 - 4.781)). The results of (13), therefore, are confirmed.

There is a link between PD and rural living, well and spring water use, and pesticide exposure (14). It examined

Table 4. Bivariate Analysis of Work-Related Factors Using Conditional Logistic Regression

Occupation	Cases [a]	Controls [a]	OR, CI 95%	P
Occupation (classification DOT)				
Occupation (classification DOT)				
(0/1) Professional, Technical, and Managerial Occupations	14 (7.8)	07 (1.9)	4.25, 1.68 - 10.73	0.002 [b]
(2) Clerical and Sales Occupations	14 (7.8)	10 (2.8)	2.95, 1.28 - 6.78	0.01 [b]
(3) Service Occupations	68 (37.8)	102 (28.3)	1.53, 1.05 - 2.24	0.02 [b]
(4) Agricultural, Fishery, Forestry, and Related Occupations	51 (28.3)	77 (21.4)	1.45, 0.96 - 2.19	0.07
(5) Processing Occupations	4 (2.3)	80 (22.2)	0.07, 0.02 - 0.22	0.001 [b]
(6) Machine Trades Occupations	5 (2.8)	02 (0.5)	5.11, 0.98 - 26.62	0.05
(7) Benchwork Occupations	3 (1.7)	11 (3)	0.53, 0.148 - 1.95	0.3
(8) Structural Work Occupations	11 (6.1)	62 (17.2)	0.31, 0.16 - 0.61	0.001 [b]
(9) Miscellaneous OccupationsOccupation	10 (5.4)	09 (2.7)	Reference	
Farmer and related activities				
Yes	41 (22.7)	70 (19.4)	1.22, 0.79 - 1.88	0.9
No	139 (77.3)	290 (80.6)	Reference	
Housewife				
Yes	43 (23.9)	55 (15.3)	1.74, 1.11 - 2.72	0.01 [b]
No	137 (76.1)	305 (84.7)	Reference	
Metalworking				
Yes	07 (3.9)	13 (3.6)	1.08, 0.42 - 2.75	0.87
No	173 (96.1)	347 (96.4)	Reference	
Previous work on a farm				
Yes	45 (25)	115 (31.9)	0.42, 0.27 - 0.63	0.001 [b]
No	135 (75)	145 (68.1)	Reference	

Abbreviation: DOT, major divisions of dictionary of occupational titles.

[a] Values are expressed as No. (%).

[b] Significant at 0.05.

the exposure to environmental factors, including the location of residence, the source of drinking water, and the exposure to pesticides and various chemicals at home and work. Several environmental factors related to the disease have been linked to the place of residence in previous studies (15, 16). The unadjusted analyses, independently of the other factors, disclosed no significant association between residential exposures (crude OR = 1.49, (CI 95% 1.00 - 2.20)) and PD. It is consistent with Hancock et al. (2008), who reported that residence or working on a farm was not significantly associated with an increased risk of PD (17). While matching was only done according to age, that result may be explained by the previous population immigrating to the urban region. Several epidemiological studies have linked well water to an increased risk of PD (18, 19), but this has not been confirmed in other populations (20-22). According to the findings of this study, drinking well water is

associated with the disease in univariate, unadjusted analysis (crude OR = 1.93, (CI 95% 1.34 - 2.78)). It is suspected that healthy water may act as a vector for toxicants, possibly causing PD. Studies have also found positive univariate associations between source drinking water use and PD (23). Spring water use was negatively associated with PD in the current study (crude OR = 0.29, (CI 95% 0.20 - 0.43)). In stratified analysis, the consumption of well-water was excluded from the multivariate analysis as a risk factor unrelated to PD. Healthy water consumption probably interacted with rural residency and pesticide exposure to create confusion. The association between spring water use and PD persisted in multivariate analysis, with an ORa of 3.31 (CI 95% 2.05 - 5.34) in favor of a positive correlation. Although healthy and spring water composition may differ, the reason for the difference was unclear. Several studies have found that working in agriculture increases the risk

of PD ([18], [24]). In univariate conditional logistic regression analysis, work in agriculture and agriculture-related activities was associated with more significant risks (raw OR = 1.22, (CI 95% 0.79 - 1.88)) but not significantly. The same results have been reported in other studies ([13]). After adjusting for other predictors (e.g., pesticide use, well water consumption, living area), the risk associated with these activities disappeared in the multivariate analysis. In order to further explore the question of work-related exposures as a contributing factor to the development of diseases in the population under study, we examined both agricultural activities and occupations involving pesticides and other chemicals.

It has been reported that pesticides are primarily responsible for PD development ([13], [25]). Numerous epidemiological studies have shown that pesticide exposure at work is associated with the disease. Pesticide use was associated with an increased risk (crude OR = 1.51, (CI 95% 0.88 - 2.60)), but not significantly. That study did not cover some information on pesticide use (use of equipment, protection, duration, and type of pesticide. Because of the relatively lower prevalence of pesticide manipulators in this study, we were also unable to establish the association of pesticides with PD, as reported earlier in a Washington study ([26]). Consequently, it may help investigate additional occupational exposures associated with PD, such as endotoxin (lipopolysaccharide) bacteria, common in various occupational and environmental settings, including agriculture ([27], [28]).

In the environmental risk factors, long-term experiences of living, such as occupation, can be critical. In the present study, consideration has been given to elucidating other occupational exposures while also considering the major confounding factors (cigarette smoking). The occupational activities employed during the most extended period were coded using the DOT and stratified into nine major groups. It showed that high crude risk was associated with working in agriculture, fishing, forestry, and related occupations, but this result was not statistically significant. An earlier study ([29]) confirms this observation. There may be an explanation for this by the fact that a large proportion of the study population is located primarily in urban and peri-urban areas that have little agriculture, which may have the effect of masking the association. Based on the results of this study, increased risks were observed for the occupation category of "clerical and sales occupations" and for the occupation category of "service occupations". According to an unadjusted analysis, the personnel employed in service occupations (i.e., military officers, officials, medical and nursing professionals, and homemakers) were 1.53 times more likely to develop this disease than those employed in other occupations. It is only partially

consistent with the results of the literature ([26]). There may be an explanation for such an outcome in that PD was recognized and acknowledged earlier in the educational and healthcare sectors ([30]).

To examine any confounding or interaction among the exposure variables, a multivariate analysis ([Table 5]) was conducted, followed by a logistic regression procedure. As a result of adjusting for potential confounders (gender, smoking), the results indicate that there is a decreased risk of PD in processing occupations (aluminum, mining, and ironworkers), which is consistent with several other studies ([26]), and not consistent with other studies that have shown an increased risk of the disease. Despite the difficulty in interpreting these results, classifications using the DOT cover a wide range of work activities and exposure potentials.

Medical and tobacco smoking histories, as well as information regarding family history, were also examined in this case-control study. Numerous epidemiological studies have demonstrated that individuals with a history of head injuries have an increased risk of developing PD ([31-33]). Additionally, we found a significant association between individuals with a history of head injury and the disease (crude OR = 3.41, (CI 95% 1.51 - 7.68)). Consistency of the evidence from multiple studies and different populations supports the conclusion that previous head injury is an etiological factor in PD.

Based on this study, diabetes and high blood pressure do not increase the risk of PD; crude OR = 0.97, (CI 95% 0.64 - 1.45) for diabetes and crude OR = 0.91, (CI 95% 0.61 - 1.34) for high blood pressure. Other studies have found an inverse association between hypertension and diabetes ([34]); however, other studies have found a positive association between diabetes and hypertension ([35], [36]). Differences in the population studied and the protocol used may explain these variances. In both univariate and multivariate analyses, drug use (crude OR = 0.55, CI 95% 0.38 - 0.79) and general anaesthesia history (crude OR = 0.57, CI 95% 0.36 - 0.89) were significantly associated with PD. Medication use was a significant risk factor in the logistic regression model with other PD risk factors (ORa = 2.12, (CI 95% 1.33 - 3.38)). Parkinson's disease is well documented to have a genetic etiology ([21], [37]). Parkinson's disease is more likely to occur in people with a family history. An unadjusted odds ratio of 5.96 was observed in this study, with a 95% confidence interval of 3.32 - 10.70. Statistically significant associations with PD persisted in the multivariate analysis (ORa = 7.19 (CI 95% 3.41 - 15.13)), suggesting that a positive family history of PD is an independent risk factor. Therefore, genetic factors may also be involved. Personal lifestyle habits, such as smoking and drinking, have been studied concerning PD risk. Based on the univariate analysis, coffee consump-

Table 5. Multivariate Analysis Using Conditional Logistic Regression for Risk Factors for Parkinson's Disease

Factors Selected	Beta	AIC	Error	aOR [a]	CI 95% [a]	Seuil P
Previous work on a farm	-1.19	517.63	0.33	0.30	0.16-0.54	0.003
Use of drugs	0.75	510.92	0. 23	2.12	1.33-3.38	0.02
History of head injury	1.21	505.73	0.54	3.38	1.16-9.83	0.02
No consumption of coffee	1.11	521.67	0.34	3.04	1.56-5.90	0.001
Processing occupations	-2.85	562.59	0.85	0.05	0.01-0.18	0.001
Operation under general anesthesia	-0.74	507.79	0.28	0.47	0.27-0.83	0.009
Consumption of spring water	1.19	525.92	0.23	3.31	2.05-5.34	0.001
The notion of familial Parkinson's	1.97	530.86	0.36	7.19	3.41-15.13	0.001
Male gender	0.65	507.27	0.25	1.92	1.16-3.16	0.01
Constant				-2.23		

Abbreviations: aOR, adjusted odds-ratio; CI, confidence interval; AIC, Akaike information criterion

[a] Calculated by logistic regression analysis for the age difference ~ 5 years between cases and controls

tion was found to have a low protective effect against PD (crude OR = 0.46, (CI 95% 0.32 - 0.66)), most notably over 30 years (crude OR = 0.22, (CI 95% 0.09 - 0.56)). Accordingly, the results are consistent with the results of previous studies ([14], [38]-[40]), which indicated that coffee consumption is a negative predictor of PD risk.

In a stratified analysis controlling for other potential risk factors for PD, such as family history, history of severe head injury, area of residence, and age, non-consumption of coffee was found to be a significant risk factor for PD development (ORa = 3.04, (CI 95% 1.56 - 5.90)). The results of the present study support some hypotheses about how coffee drinking may benefit PD. Dopaminergic neurons of substantia nigra are protected from excitotoxic lesions by caffeine's adenosine 2A receptor antagonist ([41]).

A final model (Table 5) identified family PD as the most significant predictor of PD risk, followed by previous head injuries and later spring water consumption. In the adjusted multivariate analysis, medication use was a significant risk factor for PD. The odds of contracting the disease were higher for men (ORa = 1.92, CI 95% 1.16 - 3.16) than for women. The risk of having disease following surgery with general anesthesia, as well as among those in the treatment professions, appeared to be reduced after adjustment for other risk factors. In our study, previous work on a farm reduced PD risk by 30%.

5.1. Conclusions

A serious public health issue of PD in Morocco has significant health, social, and economic repercussions.

The final PD model of risk suggested that the environmental and possibly genetic factors played an essential role in the etiology of the disease in this series. According to the present case-control study, positive family his-

tory is the primary nonenvironmental risk factor for PD, indicating the importance of expanding understanding of the genetic underpinnings of this disease. There is also an association between drinking spring water and PD. In the study sample, the male gender was also confirmed as a factor associated with the disease and had high statistical significance. However, considered classical risk factors for the disease, agricultural work, healthy water consumption, and residence in rural areas were not significantly different between cases and controls.

A single risk factor, therefore, is likely to pose a low risk and will likely differ depending on the specific characteristics of the population. Future research will need to identify additional risk factors.

Footnotes

Authors' Contribution: A. A.: Conceived and designed the evaluation and drafted the manuscript; A. A.: Participated in designing the evaluation, performed parts of the statistical analysis, and helped to draft the manuscript; S. A.: Re-evaluated the clinical data, revised the manuscript and performed the statistical analysis, and revised the manuscript; H. S.: Collected the clinical data, interpreted them, and revised the manuscript. ; A. K.: Statistical analysis; Y. B.: Re-analyzed the clinical and statistical data and

revised the manuscript. All authors read and approved the final manuscript.; A. B.: Re-analyzed the clinical and statistical data and revised the manuscript. All authors read and approved the final manuscript.; M. N.: Re-analyzed the clinical and statistical data and revised the manuscript. All authors read and approved the final manuscript.

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Informed Consent: All the selected patients gave their written consent.

References

1. Aroonvilairat S, Kespichayawattana W, Sornprachum T, Chaisuriya P, Siwadune T, Ratanabanangkoon K. Effect of pesticide exposure on immunological, hematological and biochemical parameters in thai orchid farmers- a cross-sectional study. *Int J Environ Res Public Health.* 2015;**12**(6):5846–61. doi: 10.3390/ijerph120605846. [PubMed: 26024358]. [PubMed Central: PMC4483675].

2. Langston JW, Wiley JC, Tagliati M. Optimizing Parkinson's disease diagnosis: the role of a dual nuclear imaging algorithm. *NPJ Parkinsons Dis.* 2018;**4**:5. doi: 10.1038/s41531-018-0041-9. [PubMed: 29507872]. [PubMed Central: PMC5824845].

3. Okubadejo NU, Ojo OO, Oshinaike OO. Clinical profile of parkinsonism and Parkinson's disease in Lagos, Southwestern Nigeria. *BMC Neurol.* 2010;**10**:1. doi: 10.1186/1471-2377-10-1. [PubMed: 20051133]. [PubMed Central: PMC2806862].

4. Driver JA, Logroscino G, Gaziano JM, Kurth T. Incidence and remaining lifetime risk of Parkinson disease in advanced age. *Neurology.* 2009;**72**(5):432–8. doi: 10.1212/01.wnl.0000341769.50075.bb. [PubMed: 19188574]. [PubMed Central: PMC2676726].

5. Pringsheim T, Jette N, Frolkis A, Steeves T. The prevalence of Parkinson's disease: A systematic review and meta-analysis. *Mov Disord.* 2014;**29**(13):1583–90. doi: 10.1002/mds.25945.

6. Noyce AJ, Bestwick JP, Silveira-Moriyama L, Hawkes CH, Giovannoni G, Lees AJ, et al. Meta-analysis of early nonmotor features and risk factors for Parkinson disease. *Ann Neurol.* 2012;**72**(6):893–901. doi: 10.1002/ana.23687. [PubMed: 23071076]. [PubMed Central: PMC3556649].

7. Minato T, Maeda T, Fujisawa Y, Tsuji H, Nomoto K, Ohno K, et al. Progression of Parkinson's disease is associated with gut dysbiosis: Two-year follow-up study. *PLoS One.* 2017;**12**(11). e0187307. doi: 10.1371/journal.pone.0187307. [PubMed: 29091972]. [PubMed Central: PMC5665539].

8. G. B. D. Parkinson's Disease Collaborators. Global, regional, and national burden of Parkinson's disease, 1990-2016: a systematic analysis for the Global Burden of Disease Study 2016. *Lancet Neurol.* 2018;**17**(11):939–53. doi: 10.1016/S1474-4422(18)30295-3. [PubMed: 30287051]. [PubMed Central: PMC6191528].

9. De A, Bose R, Kumar A, Mozumdar S. *Targeted delivery of pesticides using biodegradable polymeric nanoparticles.* New York, USA: Springer; 2014.

10. Saad S, Nomani AZ, Badshah M, Afzal A. Frequency of non-motor symptoms in Parkinson disease: experience from Pakistan. *Pakistan Journal of Neurological Sciences (PJNS).* 2017;**12**(1):8–15.

11. Ahad NA, Yahaya S. Sensitivity analysis of Welch's t-test. *21st National Symposium on Mathematical Sciences.* Penang, Malaysia. AIP Publishing LLC; 2014. p. 888–93.

12. Baldereschi M, Di Carlo A, Vanni P, Ghetti A, Carbonin P, Amaducci L, et al. Lifestyle-related risk factors for Parkinson's disease: a population-based study. *Acta Neurol Scand.* 2003;**108**(4):239–44. doi: 10.1034/j.1600-0404.2003.00128.x. [PubMed: 12956856].

13. Pouchieu C, Piel C, Carles C, Gruber A, Helmer C, Tual S, et al. Pesticide use in agriculture and Parkinson's disease in the AGRICAN cohort study. *Int J Epidemiol.* 2018;**47**(1):299–310. doi: 10.1093/ije/dyx225. [PubMed: 29136149].

14. Breckenridge CB, Berry C, Chang ET, Sielken RL, Mandel JS. Association between Parkinson's Disease and Cigarette Smoking, Rural Living, Well-Water Consumption, Farming and Pesticide Use: Systematic Review and Meta-Analysis. *PLoS One.* 2016;**11**(4). e0151841. doi: 10.1371/journal.pone.0151841. [PubMed: 27055126]. [PubMed Central: PMC4824443].

15. Wright Willis A, Evanoff BA, Lian M, Criswell SR, Racette BA. Geographic and ethnic variation in Parkinson disease: a population-based study of US Medicare beneficiaries. *Neuroepidemiology.* 2010;**34**(3):143–51. doi: 10.1159/000275491. [PubMed: 20090375]. [PubMed Central: PMC2865395].

16. Priyadarshi A, Khuder SA, Schaub EA, Priyadarshi SS. Environmental risk factors and Parkinson's disease: a metaanalysis. *Environ Res.* 2001;**86**(2):122–7. doi: 10.1006/enrs.2001.4264. [PubMed: 11437458].

17. Hancock DB, Martin ER, Mayhew GM, Stajich JM, Jewett R, Stacy MA, et al. Pesticide exposure and risk of Parkinson's disease: a family-based case-control study. *BMC Neurol.* 2008;**8**:6. doi: 10.1186/1471-2377-8-6. [PubMed: 18373838]. [PubMed Central: PMC2323015].

18. Martino R, Candundo H, Lieshout PV, Shin S, Crispo JAG, Barakat-Haddad C. Onset and progression factors in Parkinson's disease: A systematic review. *Neurotoxicology.* 2017;**61**:132–41. doi: 10.1016/j.neuro.2016.04.003. [PubMed: 27058967].

19. Gatto NM, Cockburn M, Bronstein J, Manthripragada AD, Ritz B. Well-water consumption and Parkinson's disease in rural California. *Environ Health Perspect.* 2009;**117**(12):1912–8. doi: 10.1289/ehp.0900852. [PubMed: 20049211]. [PubMed Central: PMC2799466].

20. Silver M, Nielsen SS, Dube U, Racette B. Well-water and risk of Parkinson Disease: a case-control study of US Medicare beneficiaries (P4. 321). *Neurology.* 2017;**88**.

21. Beghdadli B, Ghomari O, Hamimed MEA, Azza A, Edjekouane I, Ider M, et al. [Parkinson's disease and occupational and environmental risk factors: a case-control study in western Algeria]. *Arch des Mal Prof et de l'Environnement.* 2016;**77**(1):21–6. French. doi: 10.1016/j.admp.2015.08.010.

22. Gorell JM, Johnson CC, Rybicki BA, Peterson EL, Richardson RJ. The risk of Parkinson's disease with exposure to pesticides, farming, well water, and rural living. *Neurology.* 1998;**50**(5):1346–50. doi: 10.1212/wnl.50.5.1346. [PubMed: 9595985].

23. Vlajinac HD, Sipetic SB, Maksimovic JM, Marinkovic JM, Dzoljic ED, Ratkov IS, et al. Environmental factors and Parkinson's disease: a case-control study in Belgrade, Serbia. *Int J Neurosci.* 2010;**120**(5):361–7. doi: 10.3109/00207451003668374. [PubMed: 20402575].

24. Sallmén M, Warden M, Uuksulainen S, Racette B, Sainio M, Searles Nielsen S. Risk of Parkinson disease in agricultural occupations in Finland. *Rev Epidemiol Sante Publique.* 2018;**66**. doi: 10.1016/j.respe.2018.05.077.

25. Pezzoli G, Cereda E. Exposure to pesticides or solvents and risk of Parkinson disease. *Neurology.* 2013;**80**(22):2035–41. doi: 10.1212/WNL.0b013e318294b3c8. [PubMed: 23713084].

26. Firestone JA, Lundin JI, Powers KM, Smith-Weller T, Franklin GM, Swanson PD, et al. Occupational factors and risk of Parkinson's disease: A population-based case-control study. *Am J Ind Med.* 2010;**53**(3):217–23. doi: 10.1002/ajim.20788. [PubMed: 20025075]. [PubMed Central: PMC3299410].

27. Niehaus I, Lange JH. Endotoxin: is it an environmental factor in the cause of Parkinson's disease? *Occup Environ Med*. 2003;**60**(5):378. doi: 10.1136/oem.60.5.378. [PubMed: 12709528]. [PubMed Central: PMC1740529].

28. Li AA, Mink PJ, McIntosh LJ, Teta MJ, Finley B. Reply: Endotoxin, a possible agent in the causation of Parkinson's disease. *J Occup Environ Med*. 2006;**48**(7):655. author reply 655-6. doi: 10.1097/01.jom.0000214446.94586.62. [PubMed: 16832216].

29. Kirkey KL, Johnson CC, Rybicki BA, Peterson EL, Kortsha GX, Gorell JM. Occupational categories at risk for Parkinson's disease. *Am J Ind Med*. 2001;**39**(6):564–71. doi: 10.1002/ajim.1055. [PubMed: 11385640].

30. Frigerio R, Elbaz A, Sanft KR, Peterson BJ, Bower JH, Ahlskog JE, et al. Education and occupations preceding Parkinson disease: a population-based case-control study. *Neurology*. 2005;**65**(10):1575–83. doi: 10.1212/01.wnl.0000184520.21744.a2. [PubMed: 16301484].

31. Goldman SM, Kamel F, Ross GW, Jewell SA, Bhudhikanok GS, Umbach D, et al. Head injury, alpha-synuclein Rep1, and Parkinson's disease. *Ann Neurol*. 2012;**71**(1):40–8. doi: 10.1002/ana.22499. [PubMed: 22275250]. [PubMed Central: PMC3270814].

32. Goldman SM, Tanner CM, Oakes D, Bhudhikanok GS, Gupta A, Langston JW. Head injury and Parkinson's disease risk in twins. *Ann Neurol*. 2006;**60**(1):65–72. doi: 10.1002/ana.20882. [PubMed: 16718702].

33. Fang F, Chen H, Feldman AL, Kamel F, Ye W, Wirdefeldt K. Head injury and Parkinson's disease: a population-based study. *Mov Disord*. 2012;**27**(13):1632–5. doi: 10.1002/mds.25143. [PubMed: 23143933].

34. Miyake Y, Tanaka K, Fukushima W, Sasaki S, Kiyohara C, Tsuboi Y, et al. Case-control study of risk of Parkinson's disease in relation to hypertension, hypercholesterolemia, and diabetes in Japan. *J Neurol Sci*. 2010;**293**(1-2):82–6. doi: 10.1016/j.jns.2010.03.002. [PubMed: 20347450].

35. Pressley JC, Louis ED, Tang MX, Cote L, Cohen PD, Glied S, et al. The impact of comorbid disease and injuries on resource use and expenditures in parkinsonism. *Neurology*. 2003;**60**(1):87–93. doi: 10.1212/wnl.60.1.87. [PubMed: 12525724].

36. Pagano G, Ferrara N, Brooks DJ, Pavese N. Age at onset and Parkinson disease phenotype. *Neurology*. 2016;**86**(15):1400–7. doi: 10.1212/WNL.0000000000002461. [PubMed: 26865518]. [PubMed Central: PMC4831034].

37. Rybicki BA, Johnson CC, Peterson EL, Kortsha GX, Gorell JM. A family history of Parkinson's disease and its effect on other PD risk factors. *Neuroepidemiology*. 1999;**18**(5):270–8. doi: 10.1159/000026222. [PubMed: 10461053].

38. Kalia LV, Lang AE. Parkinson's disease. *Lancet*. 2015;**386**(9996):896–912. doi: 10.1016/S0140-6736(14)61393-3. [PubMed: 25904081].

39. Ventura-Chávez R, Reyna-Gil AI, García S. [Factores asociados a la enfermedad de Parkinson en pacientes de la Comarca Lagunera, México]. *Rev Mex de Neurocienc*. 2019;**20**(4). Spanish. doi: 10.24875/rmn.M19000067.

40. Wirdefeldt K, Adami HO, Cole P, Trichopoulos D, Mandel J. Epidemiology and etiology of Parkinson's disease: a review of the evidence. *Eur J Epidemiol*. 2011;**26 Suppl 1**:S1–58. doi: 10.1007/s10654-011-9581-6. [PubMed: 21626386].

41. Munoz DG, Fujioka S. Caffeine and Parkinson disease: A possible diagnostic and pathogenic breakthrough. *Neurology*. 2018;**90**(5):205–6. doi: 10.1212/WNL.0000000000004898. [PubMed: 29298853].

Table 3. Bivariate Analysis of Socio-demographic and Environmental Factors Using Conditional Logistic Regression of Predictors of Parkinson's Disease

	Cases (n = 180) [a]	Controls (n = 360) [a]	Univariate Analysis (OR (IC à 95%))	Seuil P
Gender				
Female	63 (35)	185 (51.4)	Reference	
Male	117 (65)	175 (48.6)	1.96 (1.357 - 2.841)	0.0001
Origin				
Urban	108 (60)	226 (62.8)	Reference	——
Rural	72 (40)	134 (37.2)	1.12 (0.77 - 1.62)	0.5
Level of education				
None	92 (51.1)	198 (55)	2.02 (0.980 - 4.781)	0.05
Primary	44 (24.4)	92 (25.5)	1.96 (0.910 - 4.256)	0.08
Secondary	28 (15.5)	53 (14.7)	1.78 (0.783 - 4.053)	0.16
University	16 (9)	17 (4.8)	Reference	——
Body mass index (kg/m^2)				
< 18.5	1 (0.6)	2 (0.6)	Référence	——
18.5 au 24.99	87 (48.3)	148 (41.1)	1.34 (0.93 - 1.92)	0.11
$\geq$ 25 à < 30	80 (44.4)	176 (48.9)	0.83 (0.58 - 1.19)	0.32
$\geq$ 30	12 (6.7)	34 (9.4)	0.68 (0.34 - 1.35)	0.27
Residence near an agricultural area				
Yes	59 (32.7)	93 (25.8)	1.39 (0.94 - 2.06)	0.09
No	121 (67.3)	267 (74.2)	Reference	——
Water consumption				
Well water				
Yes	104 (57.8)	149 (41.4)	1.93 (1.34 - 2.78)	0.001
Never	76 (42.2)	211 (58.6)	Référence	——
Spring water				
Yes	54 (33.9)	218 (60.6)	0.29 (0.20 - 0.43)	0.001
Never	126 (66.1)	142 (39.4)	Reference	——
Use of pesticides				
Yes	26 (14.4)	36 (85.6)	1.51 (0.88 - 2.60)	0.12
No	154 (42.7)	324 (57.3)	Reference	——
Family history of PD				
Positive	43 (23.9)	18 (5)	5.96 (3.32 - 10.70)	0.001
Negative	137 (76.1)	342 (95)	Référence	——
Use of drugs				
Yes	69 (38.3)	191 (53)	0.55 (0.38 - 0.79)	0.001
No	111 (61.7)	169 (47)	Reference	——
Medical and surgical history				
History of head injury				
Yes	16 (8.9)	10 (2.8)	3.41 (1.51 - 7.68)	0.003
No	164 (91.1)	350 (97.2)	Reference	——
High blood pressure				
Yes	47 (26.1)	96 (26.7)	0.97 (0.64 - 1.45)	0.89
No	133 (73.9)	264 (73.3)	Reference	——
Diabetes				
Yes	55 (30.5)	101 (28)	1.12 (0.76 - 1.66)	0.54
No	16 (69.5)	8 (72)	Reference	——
Operation under general anesthesia				
Yes	31 (17.2)	96 (26.6)	0.57 (0.36 - 0.89)	0.01
No	147 (82.8)	263 (73.4)	Reference	——

Lifestyle

Smoking status				
Never	136 (75.5)	239 (66.4)	Reference	—
Smoker	6 (3.4)	17 (4.7)	0.69 (0.26 - 1.79)	0.45
Ex-smoker	38 (21.1)	50 (28.9)	1.65 (1.04 - 2.64)	0.03
Coffee consumption				
Never	99 (55)	130 (36.1)	Reference	—
Yes	81 (45)	229 (63.9)	0.46 (0.32 - 0.66)	0.0001
Duration of coffee consumption (y)				
Never	99 (55)	130 (36.1)	Reference	—
1 - 10	35 (19.4)	95 (26.4)	0.27 (0.16 - 0.47)	0.001
11 - 20	12 (6.7)	17 (4.7)	0.57 (0.31 - 1.04)	0.07
21 - 30	12 (6.7)	13 (3.6)	0.3 (0.12 - 0.71)	0.007
> 30	22 (12.7)	104 (29.2)	0.22 (0.09 - 0.56)	0.001

[a] Values are expressed as No. (%).