Environmental determinants of aplastic anemia in Pakistan: a case-control study

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Abstract
Aim Aplastic anemia (AA) affects the Asian population two to three fold more than people in other regions. Besides the host genetics and socioeconomic status, several other environmental exposures have been linked with an AA etiology. We aimed to examine the association of various environmental exposures with AA occurrence among Pakistani individuals.

Subjects and methods A case-control study was conducted in Karachi, Pakistan, where cases (diagnosed AA patients) were selected from the National Institute of Blood Disease and Bone Marrow Transplantation (NIBD), while for each case, a single control (who was free of AA and visited the outpatient department of the same hospital for the treatment of minor ailments) was selected matched by age and sex. A total of 428 participants were included in this study with equal proportions of cases and controls. Information related to disease characteristics, sociodemographics and exposure to chemicals was collected through a survey questionnaire, laboratory investigations and medical records. Descriptive results were reported as frequencies and proportions, adjusted odds ratios with 95% confidence intervals and population attributable risk (PAR) as percentage.

Results Among study participants (n = 428), AA was significantly associated with various environmental exposures. Participants residing in rural settings (OR = 2.29, 95% CI 1.12–4.67, p-value < 0.01) and those who reported exposure to pesticides (OR = 3.58, 95% CI 1.27–10.10, p-value 0.01; PAR = 18.16%) were significantly more likely to report AA. Participants with a formal education were significantly less likely to have AA (OR = 0.27, 95% CI 0.10–0.71, p-value < 0.01).

Conclusions This study observed a significant association of aplastic anemia with a lower socioeconomic profile, and certain environmental exposures among the Pakistani population. The evidence may be helpful in understanding the pathophysiology of aplastic anemia in the context of environmental exposures.

Keywords Aplastic anemia · Environmental exposures · Pesticides · Arsenic · Case-control study

Background
Aplastic anemia (AA) is defined as “bone marrow hypoplasia or aplasia resulting in pancytopenia”; it affects 2–7 million individuals globally (Shadduck 1995; Young and Kaufman 2008). Wide variation in the prevalence of AA has been observed (Young and Kaufman 2008). Individuals in Asian countries are affected two to three fold more than populations from other regions (Kojima 2002; Mary et al. 1990; Young and Kaufman 2008). Scientific evidence suggests that the wide variation in the prevalence of AA among different regions of the world could be due to variations in environmental exposures (Issaragrisil et al. 1997; Maluf et al. 2009; Young...
Population.

chemicals with AA disease occurrence in the Pakistani
explore the association of environmental exposure to various
2011; Shamsi et al. 2008; Zahra et al. 2015). We thus aim to
et al. 2009). Further, to the best of our knowledge, no such
AA prevalence is higher, is limited, but is called for (Maluf
study reports evidence related to environmental exposures
susceptibility and association with infectious agents such as
hepatitis (Adil et al. 2001; Niazia and Raziaq 2011; Rauff et al.
pesticides, arsenic and benzene have been found to be strongly
linked with an increased susceptibility of individuals to devel-
op AA (Beelte et al. 2009; Chatterjee et al. 2014; Fleming and
Timmeny 1993; Morton and Dunnette 1994; Peremarti et al.
2014; Prihartono et al. 2011).
Unfortunately, evidence from the developing world, where
AA prevalence is higher, is limited, but is called for (Maluf
et al. 2009). Further, to the best of our knowledge, no such
study reports evidence related to environmental exposures’
etiological link with AA among the Pakistani population.
The majority of the studies conducted in Pakistan report evi-
dence related to the clinical and pathological features, genetic
susceptibility and association with infectious agents such as
hepatitis (Adil et al. 2001; Niazia and Raziaq 2011; Rauff et al.
2011; Shamsi et al. 2008; Zahra et al. 2015). We thus aim to
explore the association of environmental exposure to various
chemicals with AA disease occurrence in the Pakistani
population.

Methods

Study settings and participants

A case-control study was conducted in the largest metropoli-
tan city, Karachi, in Pakistan. The participants included pa-
tients accessing NIBD for healthcare from January 2014 to
December 2014.

Cases

Cases included all patients aged 12 and above with a con-
firmed diagnosis of aplastic anemia as per the Camitta
Criteria (Camitta et al. 1975). Eligible cases included patients
who met at least two of the following three criteria: (1) white
cells <3.5 × 10^9/l; (2) platelets <50 × 10^9/l; (3) hemoglobin
<10.0 g/dl or hematocrit <30 %. Further following the criteria
set by E. Maluf et al., hypocellularity was shown on bone
marrow biopsy, without leukemic, lymphomatous or carcino-
matous infiltration or fibrosis (Maluf et al. 2009). All patients
were residing in Pakistan at the time of the study. Exclusion
criteria included presence of hypocellular myelodysplasia,
other severe hematologic diseases such as other neoplasias,
near tube defects, megaloblastic anemia, neutropenia or pan-
cytopenia associated systemic diseases, previous organ trans-
plantation, chemotherapy, radiotherapy or immunosuppres-
sive therapy, Felty’s syndrome or Kostmann’s syndrome.
Profiles of all cases were finally validated by consultant he-
matologists who had access to patients’ files containing the
complete clinical history, blood reports and biopsy results.

Controls

The controls were selected by matching for age and sex. For
each case of aplastic anemia, a single control was selected.
They consisted of walk-in patients who attended the outpa-
ten department during the study period for ailments other
than blood disorders, including minor ailments such as sore
throat and diarrhea.

Data collection tool

Participants’ information was collected through a pretested,
structured questionnaire. The same questionnaire was used
for both cases and controls. The questionnaire consisted of
modules related to (1) sociodemographic characteristics in-
cluding sex, age, ethnicity, urban/rural type of residence, mar-
ital status and education level; (2) eating habits and drinking
water source; (3) environmental exposures related to pesti-
cides and arsenic (a pictorial album was used by the interview-
er to facilitate participants’ recall of exposures related to pes-
ticides and arsenic). Further, information related to the hema-
tological profile of the patients was retrieved from their med-
ical records. Data were collected by trained clinical research
officers.

Statistical analysis

Data were analyzed using STATA SE version 11.1. Chi-square
test was used to determine the significance of association of
categorical variables between cases and controls. Univariate
and multivariate conditional logistic regression analyses were
used, and odds ratios with 95 % confidence intervals were
reported to determine an association between exposures and
disease occurrence (AA). First, an association between the
different forms of environmental exposures and AA was re-
ported in model 0. Subsequently, model 1 reported the results
after adjusting for sociodemographic characteristics including
type of residence, education level, ethnicity and marital status.
Third, model 2 reports the results after adjusting for the type of
residence, education level, ethnicity, marital status, water
source and source of milk intake. Finally, model 3 reports
the results after adjusting for all covariates with the environ-
mental exposures to arsenic and pesticides.
Further, the population attributable risk (PAR) percentage was also calculated for the exposures that were found significantly associated with AA disease occurrence. This may be defined as “the proportional reduction in population disease or mortality that would occur if exposure to a risk factor were reduced to an alternative ideal exposure scenario” [Bruzzi et al. 1985; Health statistics and information systems. Metrics: population-attributable fraction (PAF)].

A subgroup analysis was also performed by matching for additional factors such as marital status, education level and type of residence. Both univariate and multivariate analyses were repeated on this subgroup of the sample to validate the findings obtained from age- and sex-matched analysis.

**Results**

A total of 428 individuals were included in the study. Of these, 214 patients had confirmed diagnoses of AA, and 214 were taken as matched controls. Age and sex distributions of cases and controls were similar because of matching (chi-square test $p$-value = 1.00). Among cases of AA, 11.3 % ($n = 24$) were exposed to arsenic, and 25.2 % ($n = 54$) were exposed to pesticides, as compared to only 4.7 % ($n = 10$), 3.7 % ($n = 8$) and 23.3 % ($n = 50$) among controls, respectively. Regarding education status and type of residence, 30.2 % of AA patients ($n = 65$) had no education, and 39.5 % ($n = 85$) of them resided in rural environments, as compared to only 8.9 % ($n = 19$) and 15.3 % ($n = 33$) among controls, respectively. Among AA cases, 84.2 % ($n = 181$) used fresh milk, and only 6 % ($n = 13$) used tetra-pack milk, whereas use of fresh milk was lower among controls, i.e., 74.4 % ($n = 160$) and 13.5 % ($n = 29$) of them used tetra-pack milk. The source of household water supply among 68.4 % ($n = 147$) of the cases was tap water, as compared to 78.6 % ($n = 169$) among controls (Table 1).

**Exposure to pesticides** Univariate logistic regression (model 0) showed that exposure to pesticides was significantly associated with AA cases ($OR = 7.57$, $95 \% CI$ 3.44–16.65, $p$-value < 0.01). After adjusting for important sociodemographic variables such as type of residence, education level, ethnicity and marital status in model 1, exposure to pesticides remained significantly associated with AA cases ($OR = 3.73$, $95 \% CI$ 1.47–9.42, $p$-value < 0.01). In model 2, when we adjusted for the type of milk consumed and source of drinking water, pesticide exposure remained significantly associated with AA ($OR = 3.12$, $95 \% CI$ 1.14–8.53, $p$-value 0.02). At the model 3 level, we adjusted for all covariates in the study and found that pesticide exposure remained a significant risk factor for developing AA ($OR = 3.42$, $95 \% CI$ 1.24–9.47, $p$-value 0.01) (Table 2). The PAR was calculated as 17.83 %.

**Exposure to arsenic** Univariate logistic regression (model 0) showed that exposure to arsenic was significantly associated with AA cases ($OR = 2.74$, $95 \% CI$ 1.22–6.17, $p$-value 0.01). After adjusting for important sociodemographic variables such as the type of residence, education level, ethnicity and marital status in model 1, exposure to arsenic remained positively associated with AA cases ($OR = 2.05$, $95 \% CI$ 0.69–6.09, $p$-value 0.19). In model 2, we adjusted for the type of milk consumed and source of drinking water, and arsenic

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**Table 1** Distribution of cases of aplastic anemia (AA) and controls according to exposure to pesticides and arsenic with sociodemosographics ($n = 428$)

| Characteristics                              | Cases $n$ (%) | Controls $n$ (%) | $p$-value* |
|----------------------------------------------|--------------|-----------------|------------|
| Exposure to pesticides (organophosphates/DTT/insecticides/mosquito repellent) |              |                 |            |
| No                                           | 160 (74.8)   | 206 (96.3)      | <0.01      |
| Yes                                          | 54 (25.2)    | 8 (3.7)         |            |
| Exposure to arsenic                          |              |                 |            |
| No                                           | 190 (88.7)   | 204 (95.3)      | 0.01       |
| Yes                                          | 24 (11.3)    | 10 (4.7)        |            |
| Source of milk intake                        |              |                 |            |
| Fresh milk                                   | 179 (84.2)   | 160 (74.4)      | 0.04       |
| Tetra-pack                                   | 14 (6.0)     | 28 (13.5)       |            |
| Powdered milk                                | 17 (7.9)     | 21 (9.8)        |            |
| Mixed source                                 | 4 (1.9)      | 5 (2.3)         |            |
| Water source                                 |              |                 |            |
| Tap water                                    | 146 (68.4)   | 168 (78.6)      | <0.01      |
| Hand pump                                    | 44 (20.5)    | 13 (6.1)        |            |
| Mineral/filter                               | 16 (7.4)     | 31 (14.4)       |            |
| River                                        | 8 (3.7)      | 2 (0.9)         |            |
| Type of residence                            |              |                 |            |
| Urban                                        | 130 (60.5)   | 181 (84.7)      | <0.01      |
| Rural                                        | 84 (39.5)    | 33 (15.3)       |            |
| Education level                              |              |                 |            |
| No education                                 | 64 (30.2)    | 20 (8.9)        | <0.01      |
| Primary                                      | 59 (27.4)    | 70 (33.2)       |            |
| Secondary                                    | 63 (29.3)    | 73 (34.1)       |            |
| Higher                                       | 28 (13.1)    | 51 (23.8)       |            |
| Marital status                               |              |                 |            |
| Unmarried                                    | 152 (70.9)   | 159 (74.4)      | 0.44       |
| Married                                      | 62 (29.1)    | 55 (25.6)       |            |
| Ethnicity                                    |              |                 |            |
| Urdu speaking                                | 67 (30.8)    | 81 (38.1)       | <0.01      |
| Sindhi speaking                              | 44 (20.6)    | 54 (25.1)       |            |
| Punjabi speaking                             | 38 (17.8)    | 38 (17.7)       |            |
| Pashtun speaking                             | 44 (21.0)    | 13 (6.1)        |            |
| Balochi speaking                             | 16 (7.5)     | 19 (8.8)        |            |
| Others                                       | 5 (2.3)      | 9 (4.2)         |            |

*p-value calculated using the chi-square test.
exposure remained positively associated with AA cases (OR = 1.75, 95% CI 0.55–5.57, p-value 0.34). At the model 3 level, we adjusted for all covariates in the study and found that the odds of having AA increased (OR = 2.18, 95% CI 0.64–7.43, p-value 0.21) (Table 2).

After adjusting for all covariates in model 3, we plotted the random effect of each variable against AA through forest plots. We found that rural residents were significantly associated with AA cases as compared to urban residents (OR = 2.68, 95% CI 1.33–5.38, p-value 0.01). Regarding education status, having primary education (OR = 0.29, 95% CI 0.11–0.74, p-value < 0.01), secondary education (OR = 0.24, 95% CI 0.09–0.63, p-value < 0.01) and higher education (OR = 0.16, 95% CI 0.05–0.46, p-value < 0.01) was found to be significantly protective against AA (see Fig. 1).

Furthermore, a subgroup analysis was also performed after matching for age, gender, type of residence, education level and marital status. The basic demographics of this subgroup analysis with additional matching are shown in Table 3. A total of 194 individuals were included in the subanalysis, 97 patients had confirmed diagnoses of AA, and 97 were taken as matched

**Table 2** Risk estimates of aplastic anemia (AA) with exposure to environmental factors (n=428)

| Characteristics | Model 0 |  | Model 1 |  | Model 2 |  | Model 3 |  |
|-----------------|---------|---|---------|---|---------|---|---------|---|
| Exposure to pesticides (organophosphates/DDT/insecticides/mosquito repellent) | OR (95% CI) | p-value | OR (95% CI) | p-value | OR (95% CI) | p-value | OR (95% CI) | p-value |
| No | 1 |  | 1 |  | 1 |  | 1 |  |
| Yes | 7.57 (3.44–16.65) | <0.01 | 3.73 (1.47–9.42) | <0.01 | 3.12 (1.14–8.53) | 0.02 | 3.42 (1.24–9.47) | 0.01 |
| Exposure to arsenic | OR (95% CI) | p-value | OR (95% CI) | p-value | OR (95% CI) | p-value | OR (95% CI) | p-value |
| No | 1 |  | 1 |  | 1 |  | 1 |  |
| Yes | 2.74 (1.22–6.17) | 0.01 | 2.05 (0.69–6.09) | 0.19 | 1.75 (0.55–5.57) | 0.34 | 2.18 (0.64–7.43) | 0.21 |

Model 0 = univariate analysis
Model 1 = model 0 + sociodemographics (type of residence, education level, marital status, ethnicity)
Model 2 = model 1 + source of milk intake + water source
Model 3 = model 0 + model 1 + model 2

**Fig. 1** Forest plot showing the random effect of each variable against aplastic anemia (AA) after adjusting for all covariates

**Aplastic Anemia**

| Independent Variables | OR (95% CI) |
|-----------------------|-------------|
| Exposure to Pesticides | 3.42 (1.24, 9.47) |
| Exposure to Arsenic | 2.18 (0.64, 7.43) |
| Tetra-pack vs fresh milk | 0.66 (0.29, 1.51) |
| Powder milk vs fresh milk | 1.15 (0.46, 2.88) |
| Mixed source vs fresh milk | 0.97 (0.18, 5.18) |
| Hand pump vs tap water | 1.30 (0.47, 3.60) |
| Mineral/Filter vs tap water | 0.54 (0.23, 1.26) |
| River vs tap water | 0.75 (0.09, 6.25) |
| Rural vs Urban | 2.68 (1.33, 5.38) |
| Primary education vs no education | 0.29 (0.11, 0.74) |
| Secondary education vs no education | 0.24 (0.09, 0.63) |
| Higher education vs no education | 0.16 (0.05, 0.46) |
| Unmarried vs married | 1.29 (0.51, 3.25) |
| Sindhi speaking vs urdu speaking | 0.40 (0.19, 0.84) |
| Punjabi speaking vs urdu speaking | 0.76 (0.37, 1.57) |
| Pashtun speaking vs urdu speaking | 1.58 (0.66, 3.76) |
| Balochi speaking vs urdu speaking | 0.57 (0.21, 1.59) |
| Others vs urdu speaking | 0.17 (0.03, 0.81) |

Odds ratio

[Springer]
Table 3 Distribution of cases of aplastic anemia (AA) and controls according to exposure to pesticides and arsenic with sociodemographics (n = 194)

| Characteristics                                | Cases n (%) | Controls n (%) | p-value* |
|------------------------------------------------|-------------|----------------|----------|
| Exposure to pesticides (organophosphates/DDT/insecticides/mosquito repellent) |             |                |          |
| No                                             | 80 (82.5)   | 92 (94.8)      | <0.01    |
| Yes                                            | 17 (17.5)   | 5 (5.2)        |          |
| Exposure to arsenic                            |             |                |          |
| No                                             | 85 (88.5)   | 90 (92.8)      | 0.31     |
| Yes                                            | 11 (11.5)   | 7 (7.2)        |          |
| Source of milk intake                          |             |                |          |
| Fresh milk                                     | 84 (86.7)   | 74 (76.3)      | 0.15     |
| Tetra-pack                                      | 8 (8.2)     | 16 (16.5)      |          |
| Powdered milk                                  | 1 (1.0)     | 4 (4.1)        |          |
| Mixed source                                   | 4 (4.1)     | 3 (3.1)        |          |
| Water source                                   |             |                |          |
| Tap water                                      | 64 (66.0)   | 71 (73.3)      | 0.06     |
| Hand pump                                      | 20 (20.6)   | 11 (11.3)      |          |
| Mineral/filter                                  | 8 (8.2)     | 13 (13.4)      |          |
| River                                          | 5 (5.2)     | 2 (2.0)        |          |
| Gender                                         |             |                |          |
| Male                                           | 67 (69.1)   | 67 (69.1)      | 1.00     |
| Female                                         | 30 (30.9)   | 30 (30.9)      |          |
| Age group                                      |             |                |          |
| <16                                            | 46 (47.4)   | 46 (47.4)      | 1.00     |
| 16–29                                          | 40 (41.3)   | 40 (41.3)      |          |
| ≥30                                            | 11 (11.3)   | 11 (11.3)      |          |
| Type of residence                              |             |                |          |
| Urban                                          | 77 (79.4)   | 77 (79.4)      | 1.00     |
| Rural                                          | 20 (20.6)   | 20 (20.6)      |          |
| Education level                                |             |                |          |
| No education                                   | 19 (19.6)   | 19 (19.6)      | 1.00     |
| Primary                                        | 40 (41.3)   | 40 (41.3)      |          |
| Secondary                                      | 24 (24.7)   | 24 (24.7)      |          |
| Higher                                         | 14 (14.4)   | 14 (14.4)      |          |
| Marital status                                 |             |                |          |
| Unmarried                                      | 72 (74.2)   | 72 (74.2)      | 1.00     |
| Married                                        | 25 (25.8)   | 25 (25.8)      |          |
| Ethnicity                                      |             |                |          |
| Urdu speaking                                  | 41 (42.3)   | 27 (27.8)      | 0.04     |
| Sindhi speaking                                | 13 (13.4)   | 27 (27.8)      |          |
| Punjabi speaking                               | 20 (20.5)   | 22 (22.7)      |          |
| Pashtun speaking                               | 15 (15.5)   | 8 (8.2)        |          |
| Balochi speaking                               | 6 (6.2)     | 9 (9.3)        |          |
| Others                                         | 2 (2.1)     | 4 (4.2)        |          |

*p-value calculated by using the chi-square test

Exposure to pesticides Univariate logistic regression (model 0) showed that exposure to pesticides was significantly associated with AA cases (OR = 3.91, 95 % CI 1.38–11.07, p-value 0.01). In model 1, when we adjusted for the type of milk consumed and source of drinking water, the pesticide exposure was statistically significantly associated with AA (OR = 3.64, 95 % CI 1.03–12.81, p-value 0.04). In model 2, we adjusted for all covariates including exposure to arsenic and found that pesticide exposure remained a significant risk factor for developing AA (OR = 3.66, 95 % CI 1.04–12.88, p-value 0.04) (Table 4). The PAR was calculated as 12.72 %.

Exposure to arsenic Univariate logistic regression (model 0) showed that exposure to arsenic was not significantly associated with AA cases (OR = 1.64, 95 % CI 0.60–4.43, p-value 0.76). Similarly, after adjusting for other variables, i.e., ethnicity, type of milk consumed, source of drinking water and exposure to pesticides, no association was found between exposure to arsenic and AA cases (OR = 0.88, 95 % CI 0.26–2.97, p-value 0.84) (Table 4).

Discussion

The findings of this study indicate that AA is associated with a lower socioeconomic profile and environmental exposure to several toxic substances among the Pakistani population. Individuals who were exposed to pesticides were significantly more likely to be diagnosed with AA. Our study results are suggestive of the fact that besides host genetics, several other hemotoxic factors may contribute to an environmental etiology of AA (Montané et al. 2008).

We found that literacy, which is the attainment of formal education, was significantly protective against aplastic anemia, and the odds of reporting AA decreased significantly with increasing levels of education. In other words, the illiterate remained at higher risk of acquiring aplastic anemia. Further, rural residents were also found more likely to report AA compared with their urban counterparts. This finding further adds to the international evidence that a lower socioeconomic profile is a risk factor for AA (Issaragrisil et al. 1995; Malhotra et al. 2015). As noted by S. Issaragrisil et al., a lower socioeconomic profile may very well be acting as a surrogate measure for several of the environmental exposures that may have an etiological role in the development of AA (Issaragrisil et al. 1995). The illiterate and rural residents of the country may be exposed to several toxic substances, pathogenic agents or medications that may play a role in the development of AA. For instance, evidence related to a higher association of hepatitis infection with AA continues to pour in (Rauff et al. 2011; Shah et al. 2011, 2012). Knowing that such
infections are the diseases of poverty may further add to the importance of recognizing the poor as a high-risk subpopulation for AA (Awofeso 2001; Engels and Savioli 2006). Thus, this calls for a deeper understanding of the specific characteristics of illiterate and rural Pakistanis to provide evidence related to the risk factors for AA.

Further, discussing the identified environmental toxic exposures, pesticides were found to be strongly associated with AA disease occurrence among participants, which echoes results from other studies (Ahamed et al. 2006; Prihartono et al. 2011). Most of the individuals in Pakistan are exposed to pesticides in either the drinking water or vegetables, fruits and other edible items with various concentrations above the WHO/FAO permissible limits. Being an agricultural country, a 1169 % increase has been recorded with the use of different types of pesticides in the last 2 decades, and an almost similar rise in the burden of diabetes (Azizullah et al. 2011; Tariq et al. 2007). Studies report that occupational exposure to pesticides among farmers is strongly associated with all hematopoietic cancers (Merhi et al. 2007). Additionally, even the general population living in areas with extensive agricultural operations have high exposure to pesticides (Tahir and Anwar 2012). It is important to note here that our study results indicate that rural residents had double the risk of acquiring AA as compared to their urban counterparts. Although pesticide contamination of food and water and domestic use of pesticides were the main exposures assessed in our study, occupational exposures among the predominantly agrarian rural population in Pakistan cannot be ruled out. We therefore call for further evidence from the Pakistani context in this regard.

Arsenic exposure caused by soil and ground water contamination has remained a serious health concern for populations globally and has been related to various cancers and genetic and metabolic dysfunctions in humans (Shankar et al. 2014). The situation is a major public health concern in Pakistan as well, where contamination of drinking water in affected areas exceeds the WHO permissible limits (Bahadar et al. 2014; Rahman et al. 2009). Although we found this to be significantly associated with AA disease occurrence in the univariate analysis, after careful adjustment with important covariates, the significance of an association between arsenic exposure and AA eventually diminished. Nevertheless, arsenic substantially affects large population subgroups, and further evidence is warranted to explore its toxic effects on the etiology of blood disorders (Subhani et al. 2015). On subgroup analysis, the association of exposure to arsenic with AA was not statistically significant; this may perhaps have been due to the smaller number of exposed individuals in the subgroup analysis.

The findings of our study may provide useful information regarding environmental exposures to certain chemicals among patients with aplastic anemia. The study was conducted at the NIBD, which may be regarded as paramount in its expertise, and it caters to a large number of patients with blood disorders. Further, the sample selection of cases and controls was finely matched according to age and gender, also adding to the strengths of the study findings. Nevertheless, there are several limitations. First, exposure data were collected retrospectively, so there a chance of recall and information bias may remain. Second, despite the fact that there is a significant association of aplastic anemia with environmental exposures, the case-control nature of study limits the ability to establish a temporal association. However, despite the above-mentioned limitations, this study has provided useful information regarding the sociodemographic- along with lifestyle-related environmental exposure in acquired aplastic anemia. The

### Table 4

| Characteristics | Model 0 | Model 1 | Model 2 |
|-----------------|---------|---------|---------|
|                 | OR (95% CI) | p-value | OR (95% CI) | p-value | OR (95% CI) | p-value |
| Exposure to pesticides (organophosphates/DDT/insecticides/mosquito repellent) | | | | | |
| No              | 1       | 1       | 1       |
| Yes             | 3.91 (1.38–11.07) | 0.01 | 3.64 (1.03–12.81) | 0.04 | 3.66 (1.04–12.88) | 0.04 |
| Exposure to arsenic | | | | | |
| No              | 1       | 1       | 1       |
| Yes             | 1.64 (0.60–4.43) | 0.76 | 0.95 (0.29–3.06) | 0.94 | 0.88 (0.26–2.97) | 0.84 |

Model 0 = univariate analysis
Model 1 = model 0 + water source + source of milk intake
Model 2 = model 0 + model 1
information may be helpful in building evidence related to environmental risk factors for AA.

**Conclusion**

This study observed a significant association of aplastic anemia with a lower socioeconomic profile and certain environmental exposures. The evidence may be helpful in understanding the pathophysiology of aplastic anemia in the context of environmental exposures.

**Compliance with ethical standards**  The study was approved by the NIBD review board. Participants were enrolled after receiving written informed consent. Participants aged less than 12 years were interviewed after obtaining consent from their parents/guardians. The parents/guardians were also present at the time of interview for facilitation regarding information recall.

All procedure followed were in accordance with the ethical standards of the responsible committee on human experimentation (institutional and national) and with the Helsinki Declaration of 1975, as revised in 2008.

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**Conflict of interest**  The authors declare that they have no conflict of interest.

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