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A Biostatistical Insight into the As_2O_3 High Dilution Effects on the Rate and Variability of Wheat Seedling Growth

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Key Words
Homeopathy · Arsenic trioxide · Wheat seedlings · High dilutions · Variability

Summary
Background: Most criticism of homeopathy concerns the lack of scientific bases and theoretical models. Fundamental research could make important contributions to our understanding of the mechanisms of action of homeopathic treatments. Plant-based bioassays are suitable for basic research – lacking the placebo effect and ensuring large data samples for structured statistical analyses. **Objective:** The aim of this study was to reproduce a previous experiment on the effects of arsenic trioxide (As_2O_3) high dilutions on wheat seedling growth in order to verify whether the same significant results could be obtained working in a different place and with a different experimental team. A further goal was to investigate high dilution effects on variability. **Material and Methods:** A structured experiment was performed blind over 9 weeks, using wheat seeds previously stressed with a sublethal dose of As_2O_3. The seeds were then treated with either potentized As_2O_3 (5x, 15x, 25x, 35x, 45x), potentized water (equivalent potencies) or diluted As_2O_3 (10^{-5}, 10^{-15}, 10^{-25}, 10^{-35}, 10^{-45}). The working variable was the stem length, measured after 4, 5, 6 and 7 days. **Results:** Some potencies (As_2O_3 45x and H_2O 45x) induced a relevant increase in seedling growth and/or a variability decrease. Diluted As_2O_3 did not induce any significant results. **Conclusions:** Confirmation of a significant stimulating effect on seedling growth and a significant decrease of variability was obtained with ultra-high dilutions at the 45x potency. The model of wheat germination and growth has been confirmed to be a good tool for basic research in homeopathy.

Schlüsselwörter
Homöopathie · Arsentrioxide · Weizensprosse · Hohe Verdünnungen · Variabilität

Zusammenfassung
Hintergrund: Die meiste Kritik an der Homöopathie bezieht sich auf ihre schwache wissenschaftliche Basis und mangelnde theoretische Modelle. Die Grundlagenforschung könnte entscheidend zum Verständnis von Wirkmechanismen homöopathischer Behandlungen beitragen. Bioassays auf pflanzlicher Basis sind aufgrund des fehlenden Plazebo-Effekts und grosser Datenmengen für eine strukturierte statistische Analyse gut für die Grundlagenforschung geeignet. **Fragestellung:** Ziel dieser Studie war es, ein früheres Experiment zur Wirkung hoher Verdünnungen von Arsentrioxid (As_2O_3) auf das Wachstum von Weizensprossen zu reproduzieren, um zu überprüfen, ob trotz des unterschiedlichen Arbeitsumfeldes und Forscherteams gleich signifikante Ergebnisse erzielt werden können. Ein weiteres Ziel war die Untersuchung der Wirkung hoher Verdünnungen auf die Variabilität. **Material und Methoden:** Mit Weizensamen, die mit einer sublethalen Dosis As_2O_3 vorbehandelt waren, wurde ein strukturiertes Experiment blind über einen Zeitraum von 9 Wochen durchgeführt. Auf die Vorbehandlung folgte eine Behandlung der Samen mit entweder potenziertem As_2O_3 (5x, 15x, 25x, 35x, 45x), potenziertem Wasser (gleiche Potenzen) oder verdünntem As_2O_3 (10^{-5}, 10^{-15}, 10^{-25}, 10^{-35}, 10^{-45}). Als Arbeitsvariable diente die Sprosslänge, die nach 4, 5, 6 und 7 Tagen gemessen wurde. **Ergebnisse:** Einige Potenzen (As2O3 45x und H_2O 45x) führten zu einer relevanten Steigerung des Sprosswachstums und/oder einer Abnahme der Variabilität. Mit verdünntem As_2O_3 wurden keine signifikanten Ergebnisse erzielt. **Schlussfolgerung:** Eine signifikante stimulierende Wirkung auf das Sprosswachstum sowie Abnahme der Variabilität konnte für ultrahohe Verdünnungen (45er Potenz) bestätigt werden. Das Modell der Keimung und des Wachstums von Weizensprossen hat sich als ein gutes Instrument homöopathischer Grundlagenforschung erwiesen.

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Introduction

The present paper follows a recently developed field of investigation in which plant models are used for fundamental research in homeopathy [1]. Since it is very difficult to obtain reliable results in this field of study [2, 3], plant-based bioassays may represent a relatively simple model on a laboratory scale that allows a large number of experimental repetitions to be performed which is useful for studying the problem of irreproducibility so often reported in homeopathic literature. In this perspective, we attempted to reproduce a previous experiment which had yielded significant results based on the growth of wheat seedlings [4]. The in vitro germination and growth of wheat coleoptiles represented one of the first models adopted to investigate the effects of homeopathic potencies [5] and has become a classic test system for basic research in this field [6]. In the original experiment [4], we had observed a curative effect of arsenic trioxide (As_2O_3) at the 45 decimal potency on wheat seedling growth. This treatment – applied after having stressed the seeds with a sublethal dose of As_2O_3 – had induced a significant increase in stem length compared to stressed seedlings not treated homeopathically. Curative effects of different As_2O_3 homeopathic potencies have also been observed on other models based both on plants [7, 8] and animals [9–11].

The aim of the present study was to reproduce the original experiment in order to verify whether the same significant results could be obtained working in a different place and with a different experimental team. Other As_2O_3 potencies were also considered together with the corresponding As_2O_3 dilutions and H_2O potencies. In this way, we separately analyzed the effects of dilution and dynamization which are the main components of homeopathic treatment. Finally, we present an in-depth analysis of the sample variability, since it was previously shown in tobacco plants infected with tobacco mosaic virus that an essential target of homeopathic treatments could be the variability of the system under observation [7].

Materials and Methods

Biological Model

The present experiment was carried out over 9 weeks during the summer, using wheat seeds (*Triticum aestivum L.*) of the MEC variety, applying the same biological pattern as in a previous study [4]. The original experiment was performed at the Urania laboratory (Milano, Italy), whereas the present study was carried out at the Department of Agro-Environmental Science and Technology (University of Bologna, Italy) and performed by a different experimenter. A global comparison between the original trial and the present one is shown in table 1. According to the experimental protocol, each seed was fixed to a filter paper by a piece of clay. This paper was inserted into a transparent cellophane envelope (12 × 20 cm) which was placed in a larger cardboard envelope. That way, shoots and roots could develop in both natural light and darkness, controlled temperatures (25 ± 1 °C), a 16/8 hour day/night cycle, regular light intensity (1.87 W m^{-2}, with fluorescent Osram luminux tubes 58 W/ col. 84) and 75–80% relative humidity. The shoot length was manually measured on day 4, 5, 6 and 7 from seeding. Each week, 6 treatments were tested (10 seedlings per treatment) following a completely randomized design for the arrangement of the cardboard envelopes. Each treatment was repeated 3 times in different weeks, following a trial design built in order to have a control each week (table 2). The final sample size was 30 seeds per treatment.

Treatments

As in the original trial, wheat seeds were pre-treated by 30-min poisoning with a 0.1% arsenic trioxide watery solution (As_2O_3, Aldrich, St. Louis, MO, USA; H_2O p.a. Merck, Darmstadt, Germany), rinsed in tap water for 60 min, dried in ambient air until 12% humidity were attained and stored in darkness until use. This stress reduced the germination rate by 10–15% [8] and the shoot length on day 7 by 58% [4].

In the present study, only stressed seeds were considered, and in each experiment they were treated with a fixed quantity (3.2 ml) of the assigned treatment:

– C1, C2, C3 (untreated water p.a. Merck, control);
– WP (potentized water p.A. Merck) 5x, 15x, 25x, 35x, 45x;
– AD (diluted arsenic trioxide) 10^{-5}, 10^{-15}, 10^{-25}, 10^{-35}, 10^{-45};
– AP (potentized arsenic trioxide) 5x, 15x, 25x, 35x, 45x.

The untreated water p.a. Merck (control) was divided into 3 polyethylene bottles (C1/C2/C3 treatments) to prevent the identification of the control group which was present every week. Decimal potencies (5x, 15x etc.) were obtained by serial dilution (1:10) with distilled water (p.a. Merck) and succussions, starting from a 0.2% solution of As_2O_3 (i.e. 10 mM). The dynamization was performed with a specially designed succussion machine which vertically shakes 100 ml volumes (in polyethylene bottles filled to 90%) at 70 times min^{-1} with an oscillation amplitude of 24 cm. Each potency was succussed for 1 min. The potentized water was prepared by exactly the same method of serial dilution and succussion, the only difference being the absence of arsenic in the starting solution. The diluted arsenic trioxide was prepared by serial dilution with distilled water (Merck), starting from 0.2% solution of As_2O_3. At each dilution step, a gentle circular movement of 1 s was applied manually. These preparations were carried out simultaneously in the Urania laboratory (as in the original trial), and stored in the dark at 4 °C. All the experiments were performed blindly. The treatments had been coded by a person not participating in the research, so that all manipulations and measurements were carried out without knowledge of the treatment code, thus avoiding any subconscious or semi-subconscious influences by the experimenter.

Statistical Analysis

The working variable considered in the present paper is the stem length Y(d) of wheat seedlings measured after d days from seeding (d = 4, 5, 6, 7). We considered Y(7) as the main variable, i.e. the stem length on day 7. Some exploratory statistics (indices of location, dispersion, skewness) were calculated with these data (also including all Y(7) = 0 values, i.e. non-germinated seeds), revealing the differences between treatment and control groups which have also been presented graphically. We calculated the 'classic' parametric statistics (mean, M, standard deviation, SD), as well as the quantile-based statistics (median, Md, and mean absolute deviation, MAD). Since the distribution of Y(7) is notably skewed in all the experimental samples (checked by Pearson's index γ), we chose to use non-parametric methods for checking differences, specifically, the Mann-Whitney two-sample comparison and the Kruskal-Wallis non-parametric analysis of variance for independent samples [12]. The Kruskal-Wallis test, based on rank sums, was applied to check whether the control groups were homogeneous. The Mann-Whitney test, also based on ranks, was used to compare treatment and control groups. The multiple comparison of variability was done by the Levene test [13], and single comparisons have been checked with the Siegel-Tukey test [14]. Here, a graphical representation of stem length distribution is given by means of the graduation function which is the inverse of the cumulative distribution function and shows the values corresponding to different centiles. Having observed

Table 1. Comparison of the experimental setup of the original trial and the present trial

Parameters	Original trial	Present trial
Poison	Arsenic trioxide (As_2O_3)	Arsenic trioxide (As_2O_3)
Dilution medium	water p.a. Merck	water p.a. Merck
Plant species	*Triticum aestivum* (MEC)	*Triticum aestivum* (MEC)
Total number of seeds	360	540
Preliminary stress	0.1% As_2O_3*	0.1% As_2O_3
Classes of treatment (number of seeds)	control (150)	control (90)
	potentized As_2O_3 (150)	potentized As_2O_3 (150)
	–	diluted As_2O_3 (150)
	–	potentized water (150)
Potencies	45x	5x, 15x, 25x, 35x, 45x
Treatment per seed, ml	3.2	3.2
Working variables	length of shoot / primary and secondary roots	length of shoot
Randomization	yes, not reported	yes, reported
Blinding	yes	yes
Timing of measurement	days 4, 5, 6, 7	days 4, 5, 6, 7
Season	winter	summer
Location	Milan	Bologna

*In the original trial, seeds (n = 60) not submitted to preliminary stress were also considered as a further control.

Table 2. Experimental pattern: each week (9 weeks in total) 6 treatments were tested (10 seeds per treatment)

	Number of week								
	1	2	3	4	5	6	7	8	9
C 1		•					•		•
C 2	•				•			•	
C3			•	•		•			
WP 5x			•			•			•
WP 15x	•					•		•	
WP 25x		•		•					•
WP 35x	•		•				•		
WP 45x		•			•		•		
AD 10^{-5}		•		•				•	
AD 10^{-15}	•						•	•	
AD 10^{-25}		•		•					•
AD 10^{-35}	•				•		•		
AD 10^{-45}			•	•					•
AP 5x					•	•		•	
AP 15x	•				•			•	
AP 25x			•	•		•			
AP 35x			•		•		•		
AP 45x		•				•			•

C = control, WP = potentized water, AD = diluted As_2O_3, AP = potentized As_2O_3.

Table 3. Exploratory statistics of stem length data (cm) after 7 days for controls (C), potentized water (WP), diluted As_2O_3 (AD) and potentized As_2O_3 (AP) groups

Treatment	n	M	SD	Md	MAD	γ
C1	30	5.89	3.86	7.05	3.12	–0.41
C2	30	6.36	3.50	7.28	2.79	–0.43
C3	30	6.28	3.70	7.35	2.84	–0.46
WP 5x	30	6.56	2.86	6.60	2.24	–0.31
WP 15x	30	5.04	3.43	6.32	2.80	–0.27
WP 25x	30	7.32	3.78	8.90	2.98	–0.78
WP 35x	30	5.91	4.32	6.78	3.92	–0.20
WP 45x	30	7.22	3.07	8.08	2.04	–1.27
AD 10^{-5}	30	6.62	3.15	7.20	1.85	–0.41
AD 10^{-15}	30	5.97	2.81	6.70	1.40	–0.73
AD 10^{-25}	30	7.08	3.86	8.03	2.60	–0.66
AD 10^{-35}	30	6.32	3.37	5.80	2.45	–0.38
AD 10^{-45}	30	6.84	4.37	8.35	2.60	–0.55
AP 5x	30	7.02	4.09	7.78	3.17	–0.58
AP 15x	30	5.85	3.15	6.18	2.53	–0.04
AP 25x	30	6.86	2.77	7.22	1.98	–1.23
AP 35x	30	6.54	3.29	7.25	2.42	–0.78
AP 45x	30	7.51	2.57	8.00	1.79	–1.42

that the most relevant differences were related to smaller centiles (left side of the graduation function), we decided to follow an ad hoc procedure, computing some exploratory indices and performing a simple non-parametric inference to the 'left-half' of each sample which is the set of values less or equal to the median.

Furthermore, between the observation days 4 to 7, we analyzed the day-to-day growth by comparing each treatment with the control group by common exploratory statistics. Finally, the results related to AP 45x in the present trial have been compared with the original trial results, using the same test applied there, which was the t-test.

Table 4. Non-parametric inference for the 45x potency/dilution groups vs. controls

	Mann-Whitney U-test	Siegel-Tukey W-Test
WP 45x	not significant	not significant
AD 10^{-45}	not significant	not significant
AP 45x	$p = 0.042$	$p = 0.034$

Results

In order to check the regularity and stability of the experimental setting, we compared the three control groups (C1, C2, C3). Some exploratory statistics are shown in table 3. The variable Y(7), i.e. the stem length after 7 days of observation, shows similar distribution in the 3 groups as regards location (M and Md), variability (SD and MAD) and skewness (γ). Since the samples are negatively skewed ($\gamma < 0$) and are not to be considered as normal, we chose to follow a non-parametric approach by applying the Kruskal-Wallis non-parametric test which showed no significance whatsoever.

The 4 experimental groups (C, WP i, AD i, AP i ; i = 5, 15, 25, 35, 45) were compared simultaneously, with group C including only the corresponding control observations (table 2). The multiple comparison proved to be significant ($p < 0.05$) for i = 45. After this overall test, we focused our analysis on particularly the 45x potency/dilution group, showing also the exploratory results of even the other groups (table 3). If we consider the average values, it is noticeable that AP 45x shows the largest value of all. Compared to the corresponding controls, the average stem length is about 25% larger. The analogous comparison on Md, although going in the same direction, was less clear. However, it must be remembered that Md value is based on only the 2 central observations and is much more sensitive to random oscillation. Furthermore, we compared each treatment of the 45x potency/dilution group versus the corresponding controls by the Mann-Whitney rank sum test (U-statistic). The stimulating effect of AP 45x proved to be significant ($p < 0.05$), unlike all the other groups (table 4). WP 45x and AD 10^{-45} treatment showed no significant results compared to the controls, and neither did the comparisons AP 45x versus WP 45x and AP 45x versus AD 10^{-45}.

We also noticed that there are relevant effects on variability, expressed here by means of SD and MAD (table 3). There seems to be a general tendency towards a reduction of variability with respect to controls. We performed multiple comparisons by means of the Levene test, taking into consideration 4 experimental groups (C, WP i, AD i, AP i; i = 5, 15, 25, 35, 45). Once more, for i = 45 the multiple comparison resulted to be significant ($p < 0.01$). To the significant group we then applied the non-parametric Siegel-Tukey test (W-test) to check the significance of the observed differences. The AP 45x group shows a significant decrease at $p < 0.05$ (two-tailed test), as shown in table 4. It is important to point out that the AP 45x treatment is the only one showing both a stimulating effect on growth and a significant decrease in variability.

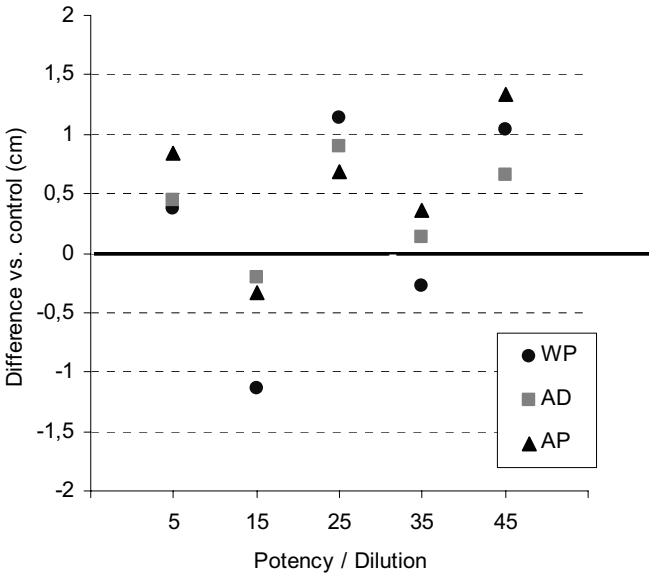

Fig. 1. Graphical representation of the stem length difference (cm) between treated and control groups considering each kind of treatment (WP = potentized water, AD = diluted arsenic trioxide, AP = potentized arsenic trioxide) and each level of potency/dilution.

Looking at the graphical representation of the average effect of treatments compared with controls (fig. 1), it is apparent that every potency/dilution group of treatments goes into the same direction (except group 35 which shows no relevant difference to the control). Moreover, there is a patent oscillatory trend, since groups 5, 25 and 45 induce a stimulating effect (average difference above zero in fig. 1), whereas groups 15 and 35 do not (average difference below or near zero).

In order to visualize the time trend and the global distribution of the variable Y(d) (d = 4, 5, 6, 7), we presented the graduation function for the 25x and 45x treatment groups (fig. 2) which induced the most relevant stimulating results, as seen in figure 1. It is evident that in AP 25x treatment and – even more so – in AP 45x, the lower centiles show higher values compared with the controls. In other words, the treatment has its main effect on the left side of the distribution. This trend becomes more evident day by day, until it reaches the final result after 7 days. The WP 45x treatment, although less evidently than AP 45x, shows a similar daily behavior, while this effect was not seen in the WP 25x treatment.

Taking all this into account, we decided to add a statistical comparison between the left-half of each experimental set of data on day 7. We have computed the left-half mean and the corresponding Md which coincides with the lower quartile of the whole set of data. As before, we have performed a multiple comparison by means of the Kruskal-Wallis non-parametric test, which proved to be significant for i = 45 ($p < 0.05$). Therefore we applied – to the only significant group – the Mann-Whitney U-test versus the corresponding controls. Re-

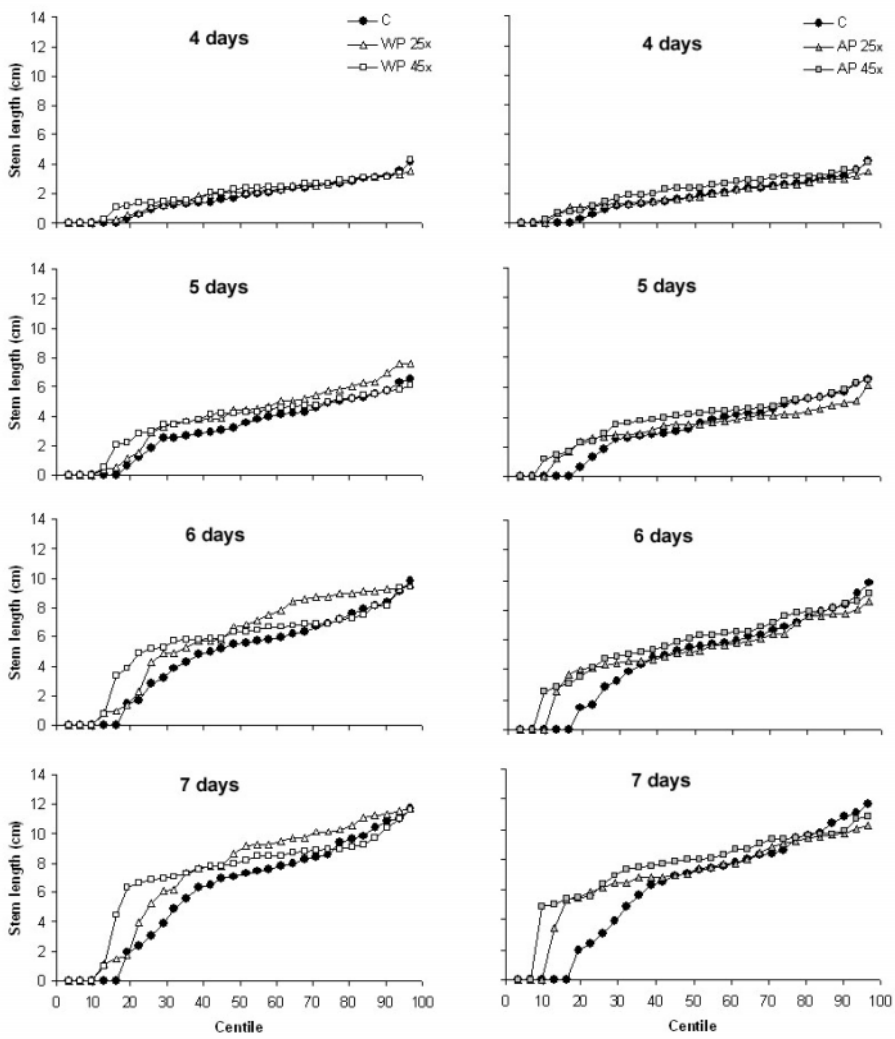

Fig. 2. Graduation function of stem length on day 4, 5, 6, 7 for controls (circles), 25x (triangles) and 45x (squares) treatment groups. Controls (C) are represented by black symbols, potentized water (WP) and potentized arsenic trioxide (AP) by white and gray symbols, respectively.

sults are shown in table 5. In this analysis the differences between treatments and controls are much more evident. In particular, the average stem length of the AP 45x group is about 90% larger than of the corresponding controls, and in the WP 45x it is about 70% larger. These differences are even stronger when comparing the Md values. Checking the statistical significance with the U-test, we realized that both WP 45x and AP 45x are highly significant ($p < 0.01$). Once again, the AD group does not yield any significant result.

We then refined our time trend analysis of the growth pattern. Table 6 shows some of the results for the 25x and 45x potencies, also presented in figure 2, which seem to have exerted the most interesting effects. For easier reading, we let the corresponding controls be equal to 100. We indicate here only 1 mean value (M) and 1 measure of variability (SD), but the Md and MAD do have quite a similar pattern. The stimulating effect (difference in average) seems already present after 4 days, both for WP and AP, and does not seem to increase with time. The decreasing trend of variability (difference in SD) seems to be progressive, which is more evident for AP 25x and (even more) for AP 45x whose variability was higher than that of the controls (+6.1%) after 4 days but markedly lower than the control (–29.7%) after 7 days.

The homeopathic potency showing the most evident stimulating effects is AP 45x, confirming our previous results on germination [8, 15] and stem length [4]. When comparing the average increase observed for AP 45x in the original stem length experiment (+24.0%, $p < 0.05$) with that observed in the present one (+24.7%, $p < 0.05$), the results prove to be very close (fig. 3), although the different growth capacity was related to the season (winter and summer for the original and the present trial, respectively).

Discussion

The adopted experimental setup allows us to obtain a set of data large enough to perform a detailed statistical analysis. With the control groups being internally homogeneous, we may attribute the results essentially to treatment effect. This

Table 5. Exploratory statistics of left-half stem length data (cm) after 7 days: average, median and Mann-Whitney U-test results for the 45x potency/dilution groups

Treatment	n	M	Md[1]	U-Test
C1	15	2.77	2.10	–
C2	15	3.57	3.90	–
C3	15	3.44	2.40	–
WP 45x	15	5.18	6.85	p = 0.006
AD 10^{-45}	15	3.24	2.25	not significant
AP 45x	15	5.72	6.35	p = 0.002

[1]The left-half Md is equivalent to the lower quartile estimated from the whole set of data.

Table 6. Daily growth trend of the 25x and 45x potency groups compared with controls (controls = 100)

Day	WP 25x		WP 45x		AP 25x		AP 45x	
	M	SD	M	SD	M	SD	M	SD
4	120.3	88.4	109.5	87.6	109.4	82.3	149.5	106.1
5	129.2	93.1	110.6	88.7	105.1	72.5	129.8	82.2
6	133.2	92.5	106.2	89.3	109.9	73.5	128.7	79.6
7	130.2	86.1	106.3	86.6	113.0	72.3	128.9	70.3

gives us confirmation that the present model, as already proposed in previous papers [4, 16], may be considered suitable for evaluating the efficacy of homeopathic treatments. Moreover, the simplicity of the model makes it possible to easily perform the same experiment in different laboratories, which is a very important requirement for validation of studies on high dilutions. Actually, the experimental model reported in the present paper has been adopted, with minor modifications, by another research group in Switzerland [17]. As regards the results, in the present study we obtained confirmation of the significant stimulating effect we had previously observed with the 45x potency, both on germination and growth of wheat seeds. In particular, the As_2O_3 45x treatment of stressed seeds, which had induced an increase in germination of 6.4% and 12.1% in two successive experiments [8] and a stimulating effect on seedling stem growth of 24.0% [4], once again showed its efficacy, enhancing shoot growth by 24.7% (which is impressively close to 24.0%). The range of the observed effects (about 6–25%) is consistent with that reported in literature for a similar experimental setup based on seedling growth of wheat [6, 18] or barley [19]. Effects of the same magnitude have been observed with other plant-based models, such as cowpea infected by root-knot nematodes [20] or tobacco infected with tobacco mosaic virus [7]. The greatest effects (40–60%) were obtained with ultra-high dilutions of hormones or other chemical mediators [21–24]. Recently, in a study based on a luminescent bacterium as test organism [3], even very small effects (1.5%) proved statistically significant.

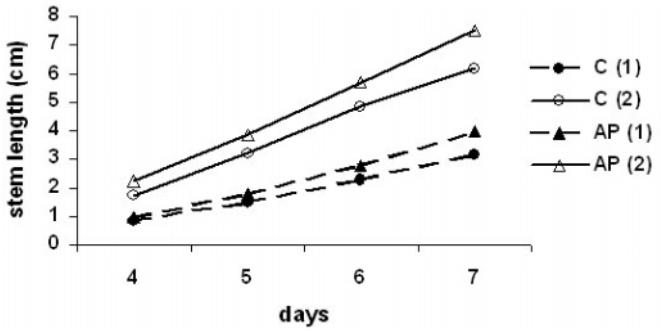

Fig. 3. Time trend of the effect on the stem length of potentized arsenic trioxide (AP) 45x treatment vs. control (C): comparison between the original trial (1, dotted line and black symbols) and present trial (2, continuous line and white symbols). Control is represented by circles and AP by triangles.

Similar small effects, albeit statistically significant, had been observed in our experiments on germination of non-stressed wheat seeds [8, 15]. Therefore, for all successive experiments, we adopted a preliminary stress (sublethal dose of As_2O_3) in order to enhance the seed response to As_2O_3 homeopathic treatments. There is much evidence that high doses of a toxicant do sensitize the bio-object to a subsequent recovery induced by low doses of the same toxic substance, suggesting that this homologous or isopathic stimulation (isotherapy) may represent a general principle [25–27].

As far as the other AP (potentized As_2O_3) treatments are concerned, no significant effect on stem growth was observed, either with lower (5x and 15x) or higher dilutions (25x and 35x). This confirms that diverse potencies of the same substance are to be considered as an inhomogeneous group of effective and ineffective samples [3], with its effectiveness being related to the adopted model. In fact, the treatment AP 5x that induced no significant results on wheat germination model, had proved significant on a tobacco/tobacco mosaic virus model [7].

We recently pointed out that an essential target of homeopathic treatments could be the variability of the system under observation [7]. This assumption seems to be confirmed by the results obtained in the present study, where repeated and significant reduction in variability (i.e. a greater homogeneity between the seedlings belonging to the same treatment group) has been observed for AP 45x. This treatment, which gave the most relevant results on stem growth, was also significant as regards variability. This decrease in variability may be explained by analyzing the treatment effect on distribution of seedling population. The growth stimulation becomes much more evident when considering the left half of experimental samples, suggesting the presence of a particular curative effect on wheat seedlings which had mostly been weakened by the preliminary stress (or intrinsically weak). When studying the time growth process, we noted that the difference in variability seems to become progressively more evident day by day, which is particularly pronounced for AP 45x. On the

other hand, the treatment effect on growth is generally completely developed after 4 days of observation.

As regards the effectiveness of potentized H_2O (WP) treatments, the results suggest that solvent dynamization alone is able to induce some effects, as also previously evidenced by chemico-physical methods [28, 29]. These effects prove to be significant for WP 45x when analyzing the left side of data distribution. This may be indicative for a 'tuned' energetic support to an intrinsic physiological homeostatic mechanism [30]. On the other hand, the mere dilution (AD treatments) did not show any significant result on stem growth rate, evidence confirmed in our previous findings [8] and consistent with other experimental studies in biological [3, 31, 32] and chemico-physical models [33]. The particular process of dynamization thus appears to be as fundamental in the activation of the different behavior of the high and ultra-high dilutions, as also recently demonstrated by experiments on thermoluminescence of different substances at ultra-high dilutions [34].

Finally, it is noteworthy that the effects of every potency/dilution group go to the same direction with respect to the controls. Moreover, the oscillatory trend of the effects, observed in figure 1, which yields a W-shaped representation, corresponding to a fixed pattern with 10 potency/dilution steps between 2 consecutive groups (5, 15, 25 etc.), recalls the V-shaped pattern detected by other authors [5, 6, 18] when considering directly consecutive potency groups.

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