OPEN ACCESS Freely available online

Journal of Clinical Toxicology

Mini Review

Abnormal Menstruation Following COVID-19 Vaccines: A Toxicologic Consideration

Deirdre Little*

Medical Director, Lily Rose Antenatal Clinic, Coffs Harbour, Australia

ABSTRACT

This paper responds to new onset irregular menstruation and post-menopausal bleeding occurring after COVID-19 vaccination. A causative mechanism is not understood. This paper therefore presents known effects of a vaccine component, polysorbate 80, that has evidenced toxicity in rats consistent with these co-occurring reports. The absence of available preclinical histology reports of COVID-19 vaccinated rat ovaries and uteri (and testes) and absence of clinical trial menstruation data precludes recognition of possible toxicological mechanisms that may be occurring. Other physiological aetiology has been postulated. The phenomenon observed before and after menopause, however, could suggest an unidentified oestrogenic effect. While no published research establishes these novel vaccines' long term gynaecological outcomes in humans, the delayed effects of polysorbate 80 observed in rat ovaries and uteri resemble those of diethylstilboestrol. It is unknown and unresearched whether the effect of a concentrated accumulation of COVID-19 vaccine nanoparticles measured in rat ovaries post vaccination could potentially be related to the abnormal uterine bleeding signalled in humans. While prior rat findings are sufficient to prompt human gynaecological research into this signal for possible toxicology causation, background observational studies and case series relative to other products using this chemical become relevant.

Keywords: Menstruation; Reproductive health; Excipients; COVID-19 vaccines; Polysorbate 80

INTRODUCTION

Reports to the UK Medicines and Healthcare Products Regulatory Agency (MHPRA) relating to new onset menstrual abnormalities and post-menopausal bleeding [1], following different COVID-19 vaccines have prompted discussion and research planning [2]. Reports exceeded 51,000 up to April 2022. No single pattern is evident. The Israel Ministry for Health recently announced its survey of third dose vaccine booster recipients, stating 'about 10% of women (under 54 years) reported other side effects such as menstrual irregularities' [3]. Some research suggests 50%-60% of reproductive-aged women experience cycle abnormalities after first vaccination and 60%-70% after second COVID-19 vaccination, irrespective of which vaccine was administered [4]. Sex and gender have not hitherto been taken into global consideration in research planning. Pharmaceutical studies have historically considered sexual health as the province of reproductive health research. Markers of sexual and reproductive health, therefore, have not been routinely sought or solicited in clinical trials, and reported abnormal bleeding occurring after COVID-19 vaccination caught the international community unawares and without existing clinical data. In response, the National Institutes of Health allocated $ 1.67 m post-marketing funding [5], to five institutions to determine if menstrual changes are linked to vaccinations; their duration; and 'to clarify the mechanisms underlying potential vaccine-related menstrual changes.' Some of these findings are now available. Potential causative mechanisms however are not clear. Pandemic stress has been postulated as a causative mechanism. Low platelet function has also been postulated. However, platelet counts would need to be severely impaired to cause spontaneous onset of bleeding and cannot account for delayed periods. Similarly, new onset post-menopausal bleeding cannot be attributed to stress. Other causative mechanisms need to be explored. Abnormal bleeding

Correspondence to: Deirdre Little, Medical Director, Lily Rose Antenatal Clinic, Coffs Harbour, Australia, E-mail: deirdrelittle2@gmail.com

Received: 15-Jul-2022, Manuscript No. JCT-22-17824; **Editor assigned:** 18-Jul-2022, PreQC No. JCT-22-17824 (PQ); **Reviewed:** 08-Aug-2022, QC No. JCT-22-17824; **Revised:** 19-Aug-2022, Manuscript No. JCT-22-17824 (R); **Published:** 26-Augl-2022, DOI: 10.35248/2161-0495.22.12.517

Citation: Little D (2022) Abnormal Menstruation Following COVID-19 Vaccines: A Toxicologic Consideration. J Clin Toxicol. 12:517.

before and after menopause could also suggest an oestrogenic effect. In our present ignorance of a causative mechanism, previous research preclinical and clinical concerning COVID-19 vaccine components become highly relevant. While an immune response effect is currently being suggested [2,5], this theory ought not form the sole hypothesis. A toxicological hypothesis, however, is not yet being considered in published literature. Scientific research can only answer the questions we ask. The menstrual cycle is a vital indicator of women's health and wellbeing so a comprehensive scientific approach to this issue is imperative and includes excipient review. Inadequately informed hypotheses may be fruitless or falsely reassuring if not attentive to existing data.

LITERATURE REVIEW

Clinical data

Menstrual information was not elicited or extractable from COVID-19 vaccine trials. All participants were required to be using 'acceptable' contraception of which hormonal forms can affect or mask cycle disruption [6]. Furthermore, subsequent vaccination of placebo arm participants removed a medium-and long-term comparator, increasing our reliance on adverse event reports for our data on human ovarian and uterine health. While research that preceded vaccine availability found pandemic-induced stress was associated with menstrual cycle irregularities, a reduction in menstrual disorders during pandemic curfew is also described [7,8]. Conflicting pandemic effects on menstruation reduce generalizability of a stress effect to the vaccinated state. COVID-19 disease itself, as with other significant illness, can also temporarily affect cycles.

Research planning is now tasked with describing the signal and exploring any biologically plausible mechanisms common to both adenoviral vector and mRNA vaccines. Notified clinical presentations include [1,4,9], new onset amenorrhoea, irregular menstruation, post-menopausal bleeding, menorrhagia, dysmenorrhoea and oligomenorrhoea. Two published studies of the subsequently sought post-marketing research, with self-reporting data, found very small and transient, but statistically significant, cycle prolongation, and heavier than normal bleeding after first and second vaccine doses [10,11]. Though considered 'reassuring' these signals are unexplained [12]. One study excluded AstraZeneca vaccine recipients and post-menopausal women. It studied only cycle length, which was delayed by 0.64 days after first dose and 0.79 days after second dose, compared with controls. Most delay occurred when two vaccines were received in the same cycle. Authors suggested a 'robust immune response' temporarily affecting the hypothalamic-pituitary-ovarian axis as possible causation. This study did not track unexpected bleeding, quantity or quality of bleeding. It offers reassurance on the basis of a subsequent returned-to-normal cycle. The second study [11], in Norway, is incomplete but shows heavier periods after first and second vaccine doses, and a 'high incidence of various menstrual changes among menstruating women aged 18-30 years'. It states: 'data about the duration of disturbances after dose two are not yet available'. The paucity of menstrual research compels concomitant scrutiny of vaccine pre-clinical and component data. Formulation of potential causation hypotheses may be assisted by close review of histology of rat ovaries and uteri after COVID-19 vaccinations; animal research pertaining to vaccine constituents; previous research on menstruation following vaccines with the same or similar components; and nanoparticle biodistribution of novel vaccines.

Pre-clinical data

New onset abnormal menstruation and post-menopausal bleeding after vaccination urge close review of vaccinated rat organ histology. Reproductive organ microscopy reports referred to in COVID-19 vaccine reproductive toxicity studies [13], the Pfizer pre-submission briefing document to the Therapeutic Goods Administration (TGA) and in the Investigator's Brochure, however, are not therein presented. No histology report is accessible [14,15]. The assessment of the repeat-dose toxicity study (study number 38166) in the Investigator's Brochure (section 5.3.1 Table 9) does not include a description of reproductive tissue. The TGA has rejected Freedom of Information request (FOI 2565 and subsequent FOI Internal Review request) for rat gonad histology reports, even after refinement of the request to just three documents: two relating to Pfizer COVID-19 vaccine and one relating to AstraZeneca COVID-19 vaccine. The TGA's grounds were that these reports were too voluminous to process as histology reports were 'intermingled throughout the studies' and 'presented in such a way that it would be impractical to simply extract the relevant pages from the studies'. The TGA refusal to release rat gonad histology also stated 'both (Pfizer and AstraZeneca) have taken active steps to ensure the information contained within the documents is not disclosed to the general public'.

Prior vaccine constituent research

This difficulty directs attention to previous research of rat ovaries and uteri for information following the injection of the same or related chemicals present in COVID-19 nanoparticle vaccines. While these vaccines are new, research is available on some nanoparticle constituents. Concerns relating to polyoxyethylene sorbitan monooleate [16], present in the AstraZeneca vaccine and also known as polysorbate 80 or 'Tween 80', stimulated earlier rat research by Gajdova, et al. [17]. Polysorbate 80 is a combination of polyethylene glycol plus an oleic acid. Polyethylene glycol is present in Pfizer vaccine nanoparticles. Gajdova, et al. [17], rat data showed significant ovarian toxicity, with cystic changes and decreased ovarian weight, at all injected doses of polysorbate 80 tested (1 milligram, 5 milligram and 10 milligram). There was no dose-response relationship to this effect. Rats also displayed a significantly prolonged oestrous cycle and induction of persistent vaginal oestrous. The effect on rat ovaries and uteri resembled that of the potent oestrogenic endocrine disruptor, diethylstilboestrol, injected as a positive control to a rat cohort in the same study. The diethylstilboestrol arm had been included in the study to explore previously identified 'oestrogenous properties' observed in substances containing the polysorbate 80 emulsifier. A safe threshold dose

of injected polysorbate 80 and related compounds for rat ovaries has not yet been identified.

Menstrual information from a related vaccine constituent

Analysis of disproportionate reports of new onset irregular menstruation, Premature Ovarian Insufficiency (POI) and amenorrhoea has been published regarding a separate vaccine, Human Papillomavirus Quadrivalent Vaccine (HPV4) [18]. Authors flagged a signal from statistical analysis (a positive result by both Bayesian Confidence Propagation Neural Network method and Multi-item Gamma Poisson Shrinker method) of the Vaccine Adverse Event Reporting System (VAERS) notifications. Researchers reported signals for POI and related events of amenorrhoea, raised Follicle Stimulating Hormone level and irregular menstruation as having a stable and strong association on time scan, with a steady upward trend and 95% Confidence Interval. HPV4 vaccine injects three serial 50 microgram doses of polysorbate 80. No signal was detected for the bivalent HPV vaccine, which does not contain polysorbate 80. Only one signal was detected for the nonavalent HPV vaccination, which injects only two such doses of polysorbate 80. Authors concluded the HPV4 correlation strongly supports further causality investigation. An earlier population based retrospective cohort study of POI following HPV4 found no correlation, but rested on ten identified research deficiencies, errors and incorrect claims which detracted from a valid conclusion [19,20]. Disturbed menstruation has also been recorded after serial injections of HPV4 in observational studies, and in case series, with subsequent diagnosis of POI [21-24]. Rat ovary histology after HPV4 vaccination was unavailable (FOI request 001-1112). Rat testicular histology was normal. While present in some foods and cosmetics, polysorbate 80's injected bioavailability and biodistribution effects for ovary and uterus may have been overlooked. The TGA argues Gajdova's young rats received from 20 times the human 50 microgram excipient dose. Although vaccinated children received 150 micrograms in total, the rat dose is still significantly larger. More research may be needed to establish the threshold dose with evidenced certainty to avoid future controversies.

Polysorbate 80 was included within all the placebos of all controlled phase two and phase three trials of HPV4 as per 2016 amended Australian Product Information [25]. Furthermore, the unresearched menstrual conundrum also places interest on the 10 milligram polysorbate 80 excipient dose used in the vital vitamin K injections ('Konakion®') needed by newborns. Excipients have no active therapeutic role.

DISCUSSION

Biodistribution

Nanoparticle biodistribution studies of 50 micrograms mRNA vaccine in rats show concentration of nanoparticles in the ovary, liver and spleen at ten times that of other organs at 48 hours [26]. It is not known when this concentration subsequently plateaus or reduces. The toxicity assessment in the nonclinical evaluation report [26], for the Pfizer COVID-19 vaccine also advises that the BNT162b2 (mRNA) COVID-19 vaccine excipients 'have long elimination half-lives. Repeat dose toxicity studies with a dosing interval of 2 or 3 weeks would be more appropriate for investigating the potential toxicity of the vaccine'. Pfizer vaccine reproductive toxicity studies [13], found a higher percentage of pre-implantation losses in the vaccinated rat group compared with the control group ($p<0.5$). Authors attributed this to a higher ovulation rate in the vaccinated cohort, considered to be within the historical control data range, though not seen in study controls. The potential effect of lipid nanoparticle constituents such as polyethylene glycol, related to potentially ovary-toxic polysorbate 80, concentrating in ovaries is unknown. It bears further investigation, however, since cycle delay was found most marked in those receiving double COVID-19 vaccinations within the same cycle of ovarian follicular development. Although it has been argued that 50 micrograms is a large rat dose, double vaccinated persons receive 60 micrograms, and vaccine development research 'Guidance for Industry' has stated "where possible we recommend that you administer the maximum human dose e.g., 1 human dose=1 rabbit dose regardless of body weight' [27].

CONCLUSION

In the context of the widespread reports of abnormal uterine bleeding of unknown cause in reproductive aged women and beyond, following COVID-19 vaccination, all previously identified, reported and signalled problems relating to vaccine components become relevant. Working hypotheses would best not exclude the scope of toxicology. After signal identification, formulation of causative hypotheses in evidence-based medicine should consider all available, relevant research. Where clinical data is not adequate, consideration of previous research and pre-clinical data becomes pertinent. It is unfortunate that collection of menstrual data is not incorporated into clinical safety trials of new medications and particularly those of population vaccines in the 21st century. Resulting ignorance affects signal detection and clarification of aetiology and leaves a temporary gap in vaccine knowledge which could undermine public vaccine confidence unless fully addressed. If we next overlook the wealth of pharmacological knowledge gathered by others before us we risk continuing the habit of underrating reproductive health information. Clarifying 'the mechanisms underlying potential vaccine-related menstrual changes' as sought by the NIH research grants requires rigorism. Research narrowed to presumptive hypotheses not informed by existing data may be less accurate. We have not yet established the research courses to pursue, but the resolve to do so is at the core of scientific method and principles, as we build on the work of fellow researchers. The review of past toxicology data is a good place to start since there is a history that remains insufficiently addressed.

REFERENCES

1. Medicines and healthcare products regulatory agency. Coronavirus vaccine-weekly summary of Yellow Card reporting. 2022.
2. Male V. Menstrual changes after COVID-19 vaccination. BMJ. 2021;374:n2211.

3. Israel Ministry of Health. Reported adverse events after receiving Pfizer's third (booster) dose of the vaccine for COVID-19. 2022.
4. Laganà AS, Veronesi G, Ghezzi F, Ferrario MM, Cromi A, Bizzarri M, et al. Evaluation of menstrual irregularities after COVID-19 vaccination: Results of the MECOVAC survey. Open Med (Wars). 2022;17(1):475-484.
5. Eunice Kennedy Shriver National Institute of Child Health and Human Development. Item of interest: NIH funds studies to assess potential effects of COVID-19 vaccination on menstruation. 2021.
6. Pfizer B. A phase 1/2/3, placebo-controlled, randomized, observer-blind, dose-finding study to evaluate the safety, tolerability, immunogenicity, and efficacy of SARS-CoV-2 RNA vaccine candidates against COVID-19 in healthy individuals. 2020.
7. Takmaz T, Gundogmus I, Okten SB, Gunduz A. The impact of COVID-19-related mental health issues on menstrual cycle characteristics of female healthcare providers. J Obstet Gynaecol Res. 2021;47(9):3241-3249.
8. Aolymat I. A cross-sectional study of the impact of COVID-19 on domestic violence, menstruation, genital tract health, and contraception use among women in Jordan. Am J Trop Med Hyg. 2021;104(2):519.
9. Muhaidat N, Alshrouf MA, Azzam MI, Karam AM, Al-Nazer MW, Al-Ani A. Menstrual symptoms after COVID-19 vaccine: A cross-sectional investigation in the MENA region. Int J Womens Health. 2022;14:395-404.
10. Edelman A, Boniface ER, Benhar E, Han L, Matteson KA, Favaro C, et al. Association between menstrual cycle length and coronavirus disease 2019 (COVID-19) vaccination: A US Cohort. Obstet Gynecol. 2022;139(4):481-489.
11. Norwegian Institute of Public Health. Increased incidence of menstrual changes among women after coronavirus vaccination. 2021.
12. Male V. Menstruation and COVID-19 vaccination. BMJ. 2022;376:o142.
13. Bowman CJ, Bouressam M, Campion SN, Cappon GD, Catlin NR, Cutler MW, et al. Lack of effects on female fertility and prenatal and postnatal offspring development in rats with BNT162b2, a mRNA-based COVID-19 vaccine. Reprod Toxicol. 2021;103:28-35.
14. Pfizer-biontech COVID-19 vaccine (bnt162, pf-07302048) vaccines and related biological products advisory committee briefing document. 2020.
15. Investigator's brochure BNT162/PF-07302048. 2020.
16. Gajdová M, Vargová M, Jakubovskỳ J, Grunt J, Války J, Galbavỳ Š. Estrogenic effects of fosmet on neonatal rat uterus. Bratisl Lek Listy. 1988;89:843-847.
17. Gajdová M, Jakubovsky J, Války J. Delayed effects of neonatal exposure to Tween 80 on female reproductive organs in rats. Food Chem Toxicol. 1993;31(3):183-190.
18. Gong L, Ji HH, Tang XW, Pan LY, Chen X, Jia YT. Human papillomavirus vaccine-associated premature ovarian insufficiency and related adverse events: Data mining of vaccine adverse event reporting system. Sci Rep. 2020;10(1):10762.
19. Naleway AL, Mittendorf KF, Irving SA, Henninger ML, Crane B, Smith N, et al. Primary ovarian insufficiency and adolescent vaccination. Pediatrics. 2018;142(3):e20180943.
20. Little DT, Ward HR. Ongoing inadequacy of quadrivalent HPV vaccine safety studies. BMJ Evid Based Med. 2020;25(2):44-45.
21. Brinth L, Theibel AC, Pors K, Mehlsen J. Suspected side effects to the quadrivalent human papilloma vaccine. Dan Med J. 2015;62(4):A5064.
22. Ozawa K, Hineno A, Kinoshita T, Ishihara S, Ikeda SI. Suspected adverse effects after human papillomavirus vaccination: A temporal relationship between vaccine administration and the appearance of symptoms in Japan. Drug Saf. 2017;40(12):1219-1229.
23. Colafrancesco S, Perricone C, Tomljenovic L, Shoenfeld Y. Human papilloma virus vaccine and primary ovarian failure: Another facet of the autoimmune/inflammatory syndrome induced by adjuvants. Am J Reprod Immunol. 2013;70(4):309-316.
24. Little DT, Ward HR. Adolescent premature ovarian insufficiency following human papillomavirus vaccination: A case series seen in general practice. J Investig Med High Impact Case Rep. 2014;2(4): 2324709614556129.
25. Product information Gardasil®. 2011.
26. Australian government department of health therapeutic goods administration. 'Nonclinical evaluation report BNT162b2 (mRNA) COVID-19 vaccine (ComirnatyTM). 2021.
27. US department of health and human services. Guidance for industry: Considerations for developmental toxicity studies for prevention and therapeutic vaccines for infectious disease indications. 2006.