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COVID-19 vaccination, all-cause and non-COVID-19 mortality in an Italian Province. Data updated, re-presented biases

The SARS-CoV-2 pandemic has led to an unprecedented effort to generate real evidence on the safety and effectiveness of various treatments, mRNA vaccines included. We can now welcome with great interest the publication of a new article on mortality in the general population of the Italian province of Pescara, divided by vaccination status, with a follow-up of two years (1).

Already in articles published during the pandemic (2, 3) it has been argued that, in observational cohort studies, an incorrect management of follow-up times may introduce the so-called Immortal-Time Bias (ITB) in favour of the exposed group. Nevertheless, ITB still appears to be present in several cohort studies. As highlighted in an article (Intervention) recently published in Epidemiologia&Prevenzione (4), a study on the safety of COVID-19 vaccines in the population of an Italian province is no exception (5). A possible explanation that the ITB is still largely prevalent in such cohort studies may be that the structure of the ITB is still poorly understood (3). A new study on the effectiveness of COVID-19 vaccines (1) in the same Italian province as that examined in the article (5) we have previously considered (4) repeats the aforementioned bias, along with others, similarly common, that we have discussed before.

Reanalysis of the study on mortality by vaccination status in the province of Pescara

The cohort concerned is nearly the same as the one already considered (5), that is the entire population (≥10 years of age) in the province of Pescara, but the observation period is longer: about 2 years vs 18 months of the previous study. The construction of groups according to vaccination status follows the same criteria as the previous study:

1. "Not vaccinated", with the same previous meaning, that is "never vaccinated", followed-up for 750 days
2. "1 dose" (that is "vaccinated with 1 dose"), followed-up for 404 days
3. "2 doses" (that is "vaccinated with 2 doses only"), followed-up for 472 days
4. " 3 doses" ("vaccinated with ≥3 doses"), followed-up for 400 days

The outcomes sought for are now:

a. SARS-CoV-2 infection, diagnosed with a positive swab
b. severe COVID-19, with hospitalization
c. death related to COVID-19 (with or without hospitalization)
d. all-causes deaths.

The time zero for the start of the follow-up is not the same, but varies depending on the group:

1. January 16, 2021 for the "not vaccinated" group
2. 2 weeks after the first dose for the "1 dose" group
3. 2 weeks after the second dose for the "2doses" group
4. 2 weeks after the third dose for the "≥3 doses" group.

The end of the follow-up coincides either with the occurrence of any outcome, or with February 15, 2023 in the absence of outcomes.

With the above-mentioned criteria, it is evident that all the results of the article will be vitiated by the ITB already discussed in our previous contribution (Intervention) (4). Moreover, the univariate analysis no longer provides the average monthly incidence rates of each outcome, but only the proportions, expressed as a simple percentage of each outcome compared with the population of the group, without any reference to the time when the outcomes occurred. This introduces a further serious bias in the presentation of the results.
The first prerequisite to avoid the ITB would be to start the follow-up of all subjects at the same time zero, where they all are not exposed, i.e. not vaccinated, regardless of what will be the fate of any vaccinee. So, each subject would be placed in her/his own vaccination status moment by moment, allowing one to have the correct person-time for each vaccination status.
The study (5) does not meet this requirement. On the contrary, it makes the follow-up start at different times and considers the individual subcohorts in a static way, without taking into account their evolution over time.

The lack of individual exposure data over time prevents one from accurately reconstructing the correct person-time in each vaccination status, forcing one to make estimates based on average follow-up times. The average follow-up times are provided in Table 1 of the article (1) exclusively for the results "Deaths", so we will show, as an example, how the ITB substantially alters the results. We will present an approximate but realistic assessment of how they change when one takes into account the person-time correctly attributed to each vaccine category.
In detail, we first assumed that the vaccination with 2 doses was carried out one month (30 days) after the first dose, and that the one with 3 doses was carried out 6 months (180 days) after. Taking into account that:

1. the group of vaccinated with ≥3 doses has the same numerical consistency (and the same duration of follow-up) as reported in Table 1 of the study;
2. the group of vaccinated with 2 doses consists of two subgroups:
   a. vaccinated with only 2 doses (with number of people and follow-up time as shown in Table 1)
   b. those who have continued vaccination with at least 3 doses (with the number of people equal to that of vaccinated with ≥3 doses and follow-up time of 180 days);
3. the group of vaccinated with one dose consists of two subgroups:
   c. vaccinated with only 1 dose (with number of people and follow-up time as shown in Table 1)
   d. those who have continued vaccination with at least 2 doses (with the number of persons equal to the sum of subgroup a. and subgroup b. and a 30-day follow-up time);
4. the unvaccinated group consists of four subgroups:
   e. those who have never been vaccinated (with number of people and time to follow-up as shown in Table 1. of the study);
   f. those who continued vaccinations until the third dose and beyond (with the number of people equal to group 1.);
   g. those who stopped at the second dose (with the number of people as reported in Table 1. of the study);
   h. those who did not go beyond the first dose (with number of people as per Table 1. of the study).

With regard to the average follow-up times as unvaccinated of sub-groups f., g. and h., not having the average start time of vaccinations with the first dose of such subgroups, theoretically the possible assumptions about their mean times are infinite. However, all of them are contained within a finite interval. Let us examine the three subgroups individually:

1. **Vaccinated with 3 doses**. First, let us keep in mind that the average time between one dose and the other is 6 months (180 days) between the second and the third, and one month (30 days) between the first and the second. The hypothesis that we consider most plausible, at least as a first approximation, is to make the end of the average follow-up of this subgroup coincide with that of the never vaccinated, similarly to what we have done in our previous contribution (Intervention) in this Journal (4). The average follow-up time as unvaccinated of the members of this subgroup can thus be obtained from the difference between the follow-

up time of the never vaccinated (750 days) and that of the vaccinated with 3 doses (400 days) increased by 210 days (=610 days), that is 140 days.

2. **Vaccinated with 2 doses**. In this case, as well as in the following one, having no information about the average time when the first dose was administered, we will assume two hypotheses, placing the average follow-up at the two extremes of the possible range of variability:

   1st - assuming that the follow-up time as unvaccinated of this subgroup, as well as that referred to in point h., coincides with that of the vaccinated with ≥3 doses, that is 140 days (more conservative)

   2nd - assuming that the end of the follow-up of the vaccinated with 2 doses coincides with that of the never vaccinated, so that the average follow-up as unvaccinated of this subgroup can be obtained from the difference between the follow-up time of the never vaccinated (750 days) and that of the vaccinated with only 2 doses (472 days) increased by 30 (=502 days), i.e. 248 days (less conservative).

3. **Vaccinate with one dose**. Also in this case the hypotheses are those referred to in point g., so that the average time of follow-up as unvaccinated will be:

   1st - equal to that of f., that is 140 days

   2nd- equal to the difference between the average time of the never vaccinated (750 days) and that of the vaccinated with only one dose (404 days), that is 346 days

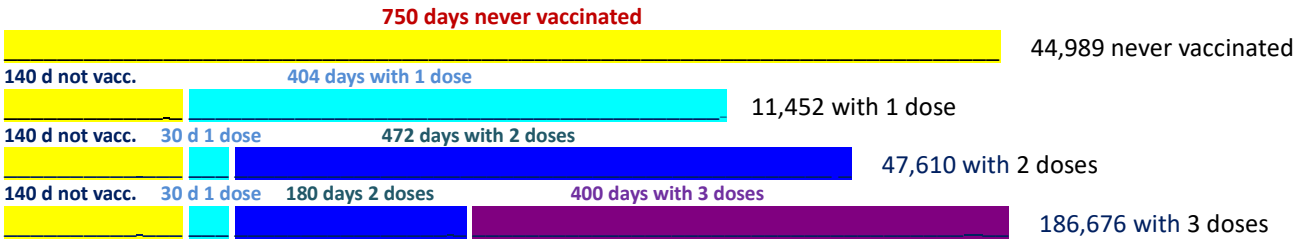
The person-times of each subgroup are obtained by multiplying the number of persons of the subgroup by the average time of follow-up of the subgroup itself; those of each group by adding together the person-times of the subgroups into which the group is divided. As the person-times thus obtained are expressed in person-days, for the purposes of calculating the annual mortality rates we will convert them into person-years, dividing the results obtained by 365.

All-cause deaths based on vaccination status (S1)

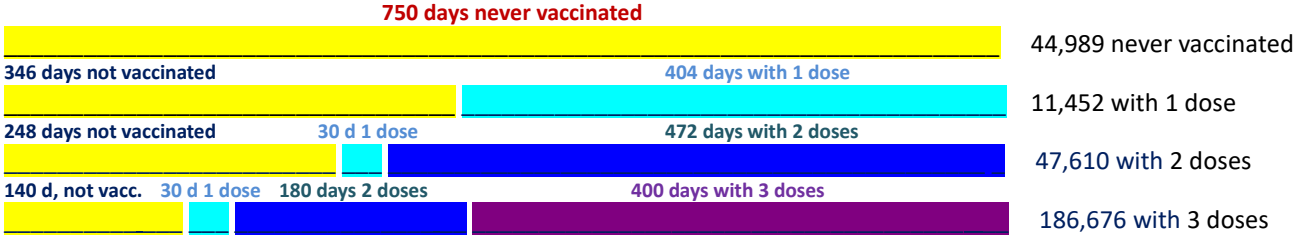
The following figure shows the correct denominators, namely the duration of exposure (in days) without vaccine (yellow), with one dose (light blue), with two doses (dark blue) and with three doses (purple).

Observation times for each category of vaccinated / unvaccinated:

1. according to the 1st hypothesis



2. according to the 2nd hypothesis



Over the period of just more than two years of observation 1686 deaths were recorded among the never vaccinated, 325 among the vaccinated with a single dose, 1964 among the vaccinated with two doses and 2546 among the vaccinated with three doses.

In the less conservative hypothesis that the end of the follow-ups of each group of vaccinated coincides with that of the never vaccinated, the annual incidence rate for 1000 people per year will therefore be:

- 9.58/1000 for the unvaccinated
- 10.18/1000 for the vaccinated with one dose
- 12.78/1000 for the vaccinated with almost three doses

The relative crude risk of death of the vaccinated (with confidence intervals of 95%) compared to the unvaccinated will therefore be:

for 1 dose 10.18 /9.58 =1.06 (CI_{95}: 0.95 – 1.19); for 2 doses 12.78 /958 = 1.33 (CI_{95}: 1.25.1,42); for 3 doses 12.44 /9.58 = 1.30 (CI_{95}: 1.23-1.38)

In the more conservative hypothesis that the non-vaccinated period before any vaccine was the same for all, i.e. 140 days, the following RR would be obtained: 0.96 (CI_{95}: 0.85-1.08) for vaccinated with one dose, 1.20 (CI_{95}: 1.10-1.28) with two doses and 1.17 (CI_{95}: 1.10-1.24) with three doses.

These results cancel the excessive gap between the rates of the vaccinated with 1 and 2 doses as compared to vaccinated with ≥3 or more doses and make disappear the inexplicable and unlikely very high protection from all-cause deaths (around 80%) of vaccination with ≥3 or more doses, which would imply implausible vaccine benefits even for deaths unrelated to COVID-19. Relative Risks, albeit unadjusted (*), appear much more plausible than "nonsensical finding", i.e. "the higher risk of all-cause deaths observed among the subjects that received one or two vaccine doses, as opposed to the much lower mortality of those who received three or more doses", as admitted by the Authors of the debated study (1).
Our results prove able to question the entire structure of the study and the validity of its results.
Note that we have focused exclusively on the ITB, but also in this study (1) there are other biases that we have already discussed (4). An example is the calendar-time bias, caused by the fact that the follow-up of the different vaccination statuses begins in calendar periods characterized by different risks for the outcome "all-cause deaths". In particular, this gives rise to an imbalance in favour of the vaccinated versus the unvaccinated, whose follow-up begins in the winter period (with higher mortality from cooling diseases) and during the second pandemic wave, characterized by high COVID-19 related mortality.

COVID-19 related and unrelated deaths, depending on vaccination status (S2)

By applying the same corrections to the relative risk estimation of vaccinated versus unvaccinated for COVID-19 related deaths, we obtain the following results:
RR = 0.72 (CI_{95}: 0.56 - 0.93) for vaccinated with one dose
RR = 0.54 (CI_{95}: 0.47 – 0.63) for vaccinated with two doses
RR = 1.14 (CI_{95}: 1.02 – 1.28) for vaccinated with three doses.

The results suggest that the booster does not reduce COVID-19 related mortality. It is clear that different confounders can affect the result, not least the fact that the boosters started much later and therefore with variants much more contagious even if less lethal. Moreover, the follow-up of the booster status took place mostly in the Omicron era. However, these results appear to be consistent with what was published on 16 March 2023 in a Technical Report of the European Centre for Disease Prevention and Control (ECDC) (6), showing that in the 11 participating European countries the vaccination effectiveness (VE) of the booster on severe acute respiratory infections (SARI) by SARS-CoV-2 in adults and the elderly is close to nil between 120-179 days (4-6 months) compared to those who completed the primary vaccination series. Continuing the follow-up from 180 days forward, the effectiveness of booster vaccination has fallen to a -34%, of borderline significance, compared to those who completed the primary series.
In any case, in the light of these results, the conclusions of the article (1) should be drastically reviewed, in particular where it states that there are "remarkably higher levels of protection among the subjects who received one or more booster doses".

In the article under discussion deaths for causes other than COVID-19 are not taken into account. However, through the analysis of the latter it is possible to bring out more clearly the contradictions found with deaths for all causes, in particular those "meaningless" results admitted by the same Authors (1):

RR = 1.20 (CI_{95}: 1.05 – 1.37) for vaccinated with one dose

RR = 1.66 (CI_{95}: 1.55 – 1.78) for vaccinated with two doses

RR = 1.36 (CI_{95}: 1.27 – 1.46) for vaccinated with three doses.

Conclusion

The results obtained with a correction, although approximate, taking into account the ITB, suggest that non-vaccinated people do not run that higher risk of all-cause deaths as compared with vaccinated people that would result from the study under question (1).

To draw more certain conclusions about the relationship between the different vaccine statuses and mortality in the province of Pescara it would be necessary to have access to the raw data, on which to carry out multivariate analyses. Given the importance of the possible implications for national health policies, the Authors of the study (1) are invited to make the suggested corrections, and in any case to make available the set of data that meritoriously they have obtained, to allow other working groups (including ours) to carry out further independent analyses.

(*) We are aware that the adjustments that can be achieved with a multivariate analysis could still significantly change the results compared to what appears now. Therefore, in addition to inviting the Authors to implement the necessary corrections, we ask that they make public the information in their possession and the procedures adopted, allowing also other research groups to replicate such important analyses.

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Keywords: all-cause mortality in province of Pescara; COVID-19 vaccination status and all-cause mortality; COVID-19 vaccination status and non-COVID-19 mortality; immortal-time bias; calendar bias

Author Contributions: Conceptualization, F.B., A.D., M.A. and G.M.; methodology, F.B., A.D., M.A. and G.M.; validation, F.B., A.D., M.A. and G.M.; formal analysis, F.B., A.D., M.A. and G.M.; resources, A.D. and G.M.; data curation, F.B., A.D., M.A. and G.M.; writing—original draft preparation F.B., A.D. and G.M.; writing—review and editing, F.B., A.D., M.A. and G.M.; supervision, F.B., A.D., M.A. and G.M. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Conflict of interests: The authors declare no conflict of interests.

Disclaimer: The statements, opinions and data contained in this Intervention are those of the individual authors.