normal increase in plasma corticosterone after NDV injection.

The range of values for plasma corticosterone concentrations Smith *et al.* reported for unstressed hypophysectomized mice is similar to what we observed. We do not know whether the NDV-induced increases in plasma corticosterone they observed in hypophysectomized mice were comparable with those observed in intact or sham-operated mice, because they did not present data from the latter. Nevertheless, the values they reported for plasma corticosterone after NDV injection in hypophysectomized mice are similar to those we found in intact or sham-operated mice. The major difference between the results of our respective studies is that we did not find an increase in the plasma corticosterone concentrations of hypophysectomized mice after injection with NDV under conditions where such an increase was observed in sham-operated mice. A small proportion of verified hypophysectomized CD-1 mice (4 of 47) did show relatively high plasma corticosterone concentrations after NDV administration, and it is these few mice that accounted for the small (not statistically significant) increases observed in this group (in two of the five experiments). Thus it is possible that a small proportion of mice can initiate an adrenocortical response by an extra-pituitary mechanism. However, because we injected NDV two or more days after the restraint test, it is just as likely that some recovery of pituitary corticotroph function had occurred in these few animals. Nevertheless, it is clear that an extra-pituitary mechanism cannot account quantitatively for the increase in plasma corticosterone normally observed after NDV injection in intact mice.

The major respect in which our experiments differ from that reported by Smith *et al.* is that we used male CD-1 mice from Charles River, whereas they used female Swiss Webster mice from Taconic Farms (*8*). In separate experiments on three batches of hypophysectomized female Swiss Webster mice from Taconic Farms, we found that a high proportion of the hypophysectomies appeared to be incomplete (25 of 57 mice did not pass our restraint test, as compared with 11 of 91 from Charles River). In these three batches some of the mice were not completely healthy; nevertheless the results after NDV injection were similar to those after injection of CD-1 mice.

We found that almost all of the hypophysectomized mice that we excluded because they showed increases in plasma corticosterone after restraint also responded to NDV (five of seven CD-1 mice, 13 of 16 Swiss Webster mice). Our observation that a high proportion of incompletely hypophysectomized mice were among those supplied by Taconic Farms suggests that the results of Smith *et al.* may have been due to inclusion of such mice in their experiments. We note that results from only five mice tested with cold water immersion were included in their report.

Our results call into question the proposed role of lymphocytes in initiating a pituitary-adrenal response after an immune challenge and suggest rather that the pituitary is indeed normally involved in the increase in plasma corticosterone that occurs after such a challenge. We believe the response to NDV more likely involves indirect activation of pituitary ACTH release by interleukin-1, as suggested by Besedovsky *et al.* (*9*), and may be secondary to the fever.

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4. Hypophysectomized CD-1 male mice or sham-operated controls were obtained from Charles River (Wilmington, MA). They were housed in individual cages on a 12:12 lighting cycle (lights on at 7 a.m.).
5. NDV of the mesogenic [N. J. Roakin-1946 (Daubney)] strain was grown in chick embryos according to the procedure of Henle and Hilleman [*Diagnostic Procedures for Viral and Rickettsial Infections*, E. H. Lennette and N. J. Schmidt, Eds. (American Public Health Association, New York, 1969), pp. 483–490]. A vehicle control was obtained from uninoculated embryos. The sample of virus used was infectious in BHK cells, but no live virus was obtained from the lungs or spleens of mice 8 hours after injection. We found it necessary to use 0.3 ml of NDV inoculum to obtain 750 hemagglutination units (close to the 800 hemagglutination units in 0.2 ml described by Smith *et al.*). In pilot experiments with intact mice we verified that peak concentrations of plasma corticosterone occurred approximately 8 hours after NDV injection.
6. Corticosterone concentrations in plasma were determined by radioimmunoassay of methylene chloride-extracted plasma. The antibody and procedure of A. Gwosdow-Cohen, C. L. Chen, and E. L. Besch [*Proc. Soc. Expl. Biol. Med.* **170**, 29 (1982)] were used.
7. We have performed five experiments in CD-1 mice involving a total of 172 mice (91 hypophysectomized). In one of these the verification of hypophysectomy was performed by injecting CRF; in this and in two other experiments, adrenocortical function was not verified, and NDV was injected 12 to 19 days after hypophysectomy. In no experiment did we observe a statistically significant increase in plasma corticosterone in hypophysectomized mice after NDV administration.
8. We have attempted to replicate the experiments of Smith *et al.* as closely as possible. Because Smith *et al.* performed the NDV injections 5 days after hypophysectomy, this time schedule was used in our last two experiments (Table 1).
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10. Supported by grants from the National Institute of Mental Health (MH25486) and the Office of Naval Research (N00014-85-K-0300). We thank Glenda Hall for growing the NDV.

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Response: It is now well established that leukocytes produce and secrete adrenocorticotropin (ACTH) either spontaneously or in response to immunostimulants, such as Newcastle disease virus (NDV) or bacterial endotoxin (*1*). Furthermore, they harbor the messenger RNA for proopiomelanocortin (POMC) (*2*) and have a POMC response to corticotropin-releasing factor (CRF) (*3*). We have previously shown that sufficient ACTH was produced in NDV-infected hypophysectomized mice to elicit a corticosterone response (*4*). When these findings are considered collectively, the reason for the inability of A. J. Dunn *et al.* to reproduce our in vivo results is especially puzzling. These authors suggest that this is due to incomplete hypophysectomy of the animals we employed. While this is a possibility, we continue to believe this is not the case, since our plasma corticosterone concentrations for unstressed hypophysectomized mice were similar to those observed by Dunn *et al.* Furthermore, we verified the completeness of the hypophysectomy by visual inspection of the sella tursica under a dissecting microscope and by functional testing the stress of cold-water immersion. Dunn *et al.* are correct that functional testing was performed on a separate group of mice. However, it would seem unlikely that complete hypophysectomies would have segregated to this group. One important difference between the studies which could account for the discrepancy is the omission of a crucial control in the present study. We showed that the spleens of NDV-infected animals in our study actually produced ACTH. In contrast, Dunn *et al.* have not verified the production of splenocyte ACTH under their experimental conditions. Perhaps the number of verified hypophysectomized CD-1 mice (4 of 47) that did show relatively high plasma corticosterone concentrations after NDV administration was small because those mice were the only ones that produced splenocyte ACTH under the experimental conditions of Dunn *et al.* This is a particularly important control, since they employed a mesogenic strain of virus in their studies, while we used a lentogenic strain. We do not know whether leukocytes consistently produce ACTH in response to a mesogenic strain of NDV, and Dunn *et al.* have not tested this idea.

Thus we believe that had Dunn *et al.* authors controlled their studies in the manner described above, they would have verified our results and shown an extra-pituitary adrenocortical response. This seems especially the case since such a response has now been demonstrated in humans by two laboratories. In one study, a case of "ectopic ACTH syndrome" was attributable to production of ACTH by normal leukocytes in an inflammatory mass (*5*). In the other, administration of CRF was shown to elicit both an ACTH and cortisol response in humans with a proved deficiency of pituitary ACTH (*6*). We do agree with Dunn *et al.* that the bulk of an ACTH response in NDV-infected intact animals probably involves an activation of pituitary ACTH release by IL-1, as shown by Woloski *et al.* (*7*) and by Besedovsky *et al.* (*8*). In contrast, however, this is not necessarily secondary to fever, since IL-1 can directly release ACTH from pituitary cells (*7, 9*).

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