Research Needs

Risk assessments function not only to characterize factors leading to higher or lower risk, but also to identify where information is missing. Missing information identified during the modeling process targets the areas with priority for research. For the SE risk assessment, assumptions were stated throughout the text; these assumptions indicate additional places where data is needed. In this section, the most prominent research needs are summarized. Prioritization of these needs across modules was not attempted, as each module could significantly contribute to the public health outcomes depending on a given pathway or scenario.

Egg Production Module

The annual production of SE-positive eggs in the USA is estimated in the Production Module. Much of the data in the module needed to be adjusted for sampling size, the seasonal and temporal patterns of SE prevalence, and variability of SE prevalence associated with flock size strata (i.e. SE prevalence varies with the size of the flock). Research is needed to generate more data in these areas so that in the future, models can incorporate the actual observed data as opposed to the data generated by the algorithms currently in the module.

Egg production begins on the farm, yet little is known about farm environmental and/or management risk factors associated with SE-positive flocks or within-flock prevalences (i.e. proportion of SE-positive birds in SE-positive flocks). The effects of manure management and feeding practices may provide some examples of factors leading to an increase or decrease in SE-positive flocks and within-flock prevalence. Other examples of environmental and management risk factors include vaccination of flocks, rodent control in and around layer houses, cleaning/disinfection of layer houses, and the use of bacterial competitive exclusion techniques to prevent the colonization of the intestinal tract of hens with SE.

The size of the flock may also affect the probability that a flock is SE-positive. In the spent hen surveys (see page 32), the size of SE positive flocks was not documented. It is possible that flock size is correlated with whether or not a flock is SE-positive and with the within-flock prevalence of SE-positive hens. Future surveys should document and analyze the effect(s) of flock size.

A seasonal pattern to the proportion of flocks found to be SE-positive has been suggested (page 32 of Production Module). The number of positive flocks increases from March (11%), to April (29%), to May (39%) according to the 1991 spent hen survey. However, it is not known whether this increase is due to a true increase in SE-positive flocks or an increase in the prevalence within flocks associated with the season of the year. Studies should be conducted to confirm the seasonal pattern as well as to distinguish between the two possibilities. Likewise, longitudinal studies which document temporal and seasonal patterns of SE prevalence within positive flocks should be conducted.

Research Needs

The Production Module categorized flocks into either high or low prevalence SE-positive flocks by using SE contamination in the flock environment as an indicator of the status of the flock. An important question which should be addressed is whether or not these high prevalence flocks represent a constant proportion of SE-positive flocks (i.e. in other words, is there a seasonal or temporal change in the number of SE-positive flocks which are high prevalence?). This information would greatly enhance the value of survey data.

Other factors which should be investigated in future research projects include: (1) the association between severity of SE infection and specific strains of SE, (2) positive egg frequencies from geographically diverse SE-positive flocks, and (3) the efficacy of various molting strategies on SE infection.

Random surveys of eggs for the presence of SE and the number of SE bacteria in SE-positive eggs will be important in order to validate the numbers obtained from this module and future models. These surveys should occur on a national scale.

Shell Eggs Processing and Distribution Module

The Shell Eggs Processing and Distribution Module is the next module, following the Egg Production Module, in the SE farm-to-table risk assessment model. It calculates the likelihood of various numbers of SE bacteria in SE-positive shell eggs. Research needs which were identified from the Shell Eggs Processing and Distribution Module of the SE risk assessment model include the following:

Number of *Salmonella* Enteritidis in an infected egg

There are only two studies on the numbers of SE inside infected eggs at the time of laying. Both of these studies contain limited data. Gast and Beard (1992) used artificially infected hens which had been challenged with high numbers of SE. Humphrey (1994) describes an initial one log of growth in the number of SE before the pH of the egg increases and the albumen becomes inhibitory to growth. In addition, the two studies do not agree very well. These limitations and the conflicting results indicate that more research is needed to quantify the number of SE bacteria inside SE-positive eggs. It is preferably that these studies be conducted with naturally infected eggs.

Inhibition of *Salmonella* Enteritidis growth inside the egg

After the first day, the albumen is an excellent inhibitor of SE growth. This inhibition is maintained until the yolk membrane loses its ability to keep apart the SE in the albumen and the yolk contents. The time to yolk membrane breakdown depends upon the temperature storage: typical values are 17 days before yolk membrane breakdown when the egg is stored at 20° C and only 4 days before yolk membrane breakdown when the egg is stored at 35° C. This essential information comes from a single study. This single study needs to be validated by independent replication of the study.

Research Needs

Growth rates of *Salmonella* Enteritidis in eggs

Only two studies were found with significant information on the growth rates of SE in blended whole eggs at different temperatures (page 105). Blended whole eggs serve as an experimental approximation of the events following the breakdown of the yolk membrane and the utilization of yolk contents by SE in the albumen. Several experiments should be conducted on SE growth rates under various conditions in order to more accurately model the growth of SE in eggs.

Improved model for egg cooling

It would be very useful to have the ability to predict the temperature of an egg at a specified time given the initial temperature of the egg, the ambient air temperature, and the packaging characteristics . Only a few cooling curves have been published on the internal temperature of the egg over time, and no modeling or engineering studies are available. Studies are needed which correlate the internal egg temperature of an egg to the type of packaging material used, the position of the egg in a pallet of stacked cartons of eggs, and the ambient storage temperature.

Role of *Salmonella* contamination on the exterior of the shell

This model does not consider the role of SE or other *Salmonella* species found on the egg's exterior in producing human illness. Studies have shown that the bacteria on the exterior of the egg can potentially cross the shell and the two egg shell membranes, but the frequency or importance of this route of contamination for eggs in commercial channels has not been established. This route may not be significant in causing human illness; however, the question frequently arises as to the role and the significance of the exterior contamination of the egg shell in human illness. More research is needed to determine the role and the significance of exterior contamination of the egg shell in human illness .

Information about storage times and temperatures.

The sensitivity analyses indicate that storage times and temperatures are important risk factors (see page 108 of the Shell Eggs Processing and Distribution Module). More extensive surveys of industry practices (e.g. storage time after processing) should be conducted to streamline the distributions for these parameters. Industry and regulatory agencies should collaborate in these efforts.

As with the Production Module, validation of the Shell Eggs Processing and Distribution Module results is an important research need. Random egg surveys on the number of SE bacteria in SE-positive eggs should be conducted for eggs subjected to various processing, storage, and transportation scenarios.

Research Needs

Egg Products Processing Module

Sensitivity analysis of variables in the Egg Products Processing Module indicates that the number of SE bacteria in liquid egg before pasteurization is more highly correlated with the number of organisms remaining after pasteurization than any other variable tested. This relationship is true for whole egg, albumen, and yolk. This information has two implications. First, reduction in the number of bacteria in liquid egg prior to pasteurization will result in a reduction in the number of bacteria post pasteurization. Plant sanitation including washing and sanitizing of incoming eggs, and prevention of cross contamination from breaking machinery, machine operators, airborne *Salmonella*, and the surface of the shell in the breaking process are the most promising means of reducing *Salmonella* in the final product. Second, refining our estimate of the number of organisms before pasteurization would allow us to predict with greater certainty the number remaining after pasteurization.

Our sources of data for the number of organisms in liquid egg before pasteurization have significant shortcomings. The 1991 and 1995 liquid egg surveys (Ebel, 1991 and Hogue, 1997) provide no information on the number of organisms because samples were only analyzed for the presence or absence of SE and other *Salmonella*. The 1969 survey (Garibaldi, 1969) includes the number of *Salmonella* organisms present but the information is nearly thirty years old and is not specific to *Salmonella* Enteritidis. Significant changes have occurred in the egg products industry over the last 30 years. A current survey of the level of *Salmonella* in liquid egg prior to pasteurization including the variation in *Salmonella* across breaker plants nationwide would provide information useful in predicting the number of organisms likely to remain after pasteurization.

The uncertainty in our estimates of the logs of bacteria reduced in liquid egg pasteurized according to FSIS regulations is large (Table 7). The 95% confidence interval for whole egg and yolk extends from about five to more than 20 and albumen is about one to 16. The reason for this large uncertainty is that our estimates were made by regression of data from all current experimental pasteurization studies on egg products. Variation within studies is generally low but variation between studies is large (see regression charts for whole egg [page 130], yolk [page 136], and albumen [page 132]). Minor differences in methods between studies do occur but no single variable has been identified as responsible for the lack of repeatability of pasteurization studies between laboratories. Egg may, by its composition (high fat and globular), provide less repeatable results, or the conditions under which bacteria are grown prior to inoculation into the egg may influence experimental results. In any case, a better understanding of the reasons for the lack of repeatability in experimental pasteurization studies on liquid egg is essential in accurately predicting the number of bacteria remaining in liquid egg after pasteurization.

Table 7. Estimates of the reduction in *Salmonella* Enteritidis from pasteurization

Product	FSIS minimum pasteurization requirements	Expected logs of SE reduced	95% confidence interval
whole egg	60°C for 3.5 minutes	8.1	5.2-17.8
yolk	60°C for 6.2 minutes	7.8	4.7-19.6
	61.1°C for 3.5 minutes	8.2	5.4-22.5
albumen (pH=8.3)	55.6°C for 6.2 minutes	3.6	1.1-15.7
	56.7°C for 3.5 minutes	3.7	1.1-16.0

pH is inversely correlated with the number of bacteria in albumen after pasteurization (i.e. the higher the pH of albumen, the greater the reduction in the number of SE during pasteurization). The pH of egg albumen increases from about 7.8 to 9.3 over the first three or four days after lay. FSIS requirements for pasteurization are based on a pH for albumen of about nine. FSIS minimum time and temperature requirements for egg white pasteurization were adequate in the late 1960's when the regulation was enacted because eggs generally took more than four days to reach breaker plants. The industry in 1998 includes in-line operations where eggs generally reach the breaker plant in less than 24 hours and thus have a lower initial pH. Also, restricted eggs currently make up a smaller proportion of the eggs broken than in 1969 because of the growth of the egg products industry. Since restricted eggs are sorted during grading and diverted to pasteurization, they are generally older when they arrive at breakers.

Information used in this model about the pH of albumen in breaker plants across the U.S. comes largely from the opinion of industry experts and experimental studies on the change in pH of egg white over time. More accurate survey information about the pH of albumen in breaker plants and how pH varies across breaker plants nationwide will reduce the uncertainty in our estimate of the number of SE in egg white after pasteurization.

Preparation and Consumption Module

The purpose of the Preparation and Consumption Module is to determine the number of egg containing servings which are contaminated with SE and the extent of this contamination. Due to a lack of sufficient data, the Preparation and Consumption Module estimated several preparation and consumption variables in both the home setting and in the institutional settings. Storage times and temperatures, probability of eggs being pooled or non-pooled, thoroughness of cooking, and the number of eggs consumed as “eggs” versus “eggs as ingredients” were estimated from the results of limited surveys or by members of the core group of the SE risk assessment team. Much research is needed in these areas, as preparation and consumption variables have significant effects on public health outcomes.

Research Needs

Specific examples of data needs are as follows:

--Retail storage times and temperatures before cooking
--Home storage times and temperatures before cooking
--Institutional storage times and temperatures before cooking
--Pooling factors in home and institutional settings
--Thoroughness of cooking for pooled vs. non-pooled eggs
--Thoroughness of cooking in home and institutional settings
--Home and institutional storage times and temperatures after pooling
--Home and institutional storage times and temperatures after cooking
--Consumption patterns for institutional and home settings--pooled vs. non-pooled
--Consumption patterns for institutional and home settings--cooked vs. undercooked
--Consumption patterns for institutional and home settings--"eggs" vs "eggs as ingredients"

This information could be obtained through cooperative studies amongst retailers, institutions, and regulatory agencies. Information about home practices could be obtained through surveys conducted by regulatory agencies, retailers, and/or consumer groups.

In addition to preparation and consumption variables, bacterial death rates with respect to cooking times and preparation of various egg dishes have not been well-studied. Results used in the Preparation and Consumption Module are taken largely from one study (Humphrey et al., 1989). Log reduction of bacteria in this study is enumerated for total cooking time on inoculated eggs that are cooked as "eggs", not ingredients. No data exist for bacterial death rates with "eggs as ingredients". Future studies should (1) monitor the log reduction of bacteria for each minute given each style of cooking (e.g. log reduction in bacteria after boiling a shell egg for one minute, after boiling a shell egg for two minutes, after boiling a shell egg for three minutes, and after boiling a shell egg for four minutes--total time 4 minutes); 2) focus on the death rates with respect to "eggs as ingredients", and 3) incorporate naturally infected eggs into the studies.

Results of the Preparation and Consumption data should be validated by surveying homes and institutions for preparation and consumption practices and subsequently sampling egg-containing dishes from the surveyed groups for the presence and enumeration of SE.

Research Needs

Public Health Outcomes Module

The Public Health Outcomes Module links exposure to SE-contaminated foods with adverse health outcomes. Limitations of this module were summarized in the text of the module (see page 252). These limitations are indicators of specific research needs.

Proportion susceptible

Data on the types and numbers of people more susceptible to foodborne pathogens are limited. Currently, the data does not account for differences within the susceptible and "normal" sub-populations, but rather dichotomizes the human population into these two groups. The probability of a certain clinical outcome may be very similar between the most "sensitive" of the "normal" sub-population and the most "robust" of the susceptible sub-population. There is most likely a continuum of susceptibilities in the human population. Future research should focus on further stratification of the susceptible and "normal" sub-populations (i.e. defining subcategories of each of the two groups).

Dose-Response Parameters (probability of illness)

Much of the dose-response data is based upon animal models or human feeding trials using strains other than SE. The human feeding trials are also of limited use because of the small sample sizes, the repeated use of subjects, and the use of a quantitatively large dose as the minimum doses. Given the differences between animals and humans, the variation in human susceptibility, and bacterial strain/species differences, there is a great deal of uncertainty in dose-response calculations, especially in the low dose ranges. Because human studies should not be conducted with pathogenic organisms, research is needed to determine how to deal with this uncertainty. Better quantitative methods (e.g. better functional forms) for modeling the probability of illness are needed.

Dose-response data can be obtained from outbreaks. Epidemiologic investigations which trace the food vehicle and enumerate the number of disease causing organisms in this vehicle should be expanded. Current surveillance efforts (e.g. FoodNet USDA/FDA/CDC) should enhance efforts to track the causative food source.

Probability of clinical outcomes when ill

Important parameters describing probabilities for clinical outcomes of disease often were not described explicitly due to a lack of data. Research is needed to determine these outcome probabilities, particularly with respect to the susceptible sub-population. More extensive surveys of the general population and medical professionals are needed to determine these values.

The number of ingested organisms may be related to the severity of illness due to SE and to the incubation period for a particular illness. Evidence suggests that there is an increase in the probability of treatment by a physician and in the probability of hospitalization when higher doses are ingested. Likewise, with higher doses, the

incubation periods (i.e. the time between infection and the onset of clinical signs) of SE illnesses appear to be shorter. However, because of the lack of data, these relationships were not incorporated into the computation of clinical outcomes. Research is needed to collect more data concerning the relationship between the number of ingested organisms and clinical health outcomes.

Probability of long-term sequelae to illness

Only one long-term sequelae to illness, reactive arthritis, was estimated in the current Public Health Outcomes Module. Several other long-term sequelae of SE infection have been identified; however, information is limited with respect to the proportion of these long-term sequelae that can be attributed to SE. Epidemiologic studies should be expanded to enumerate the long-term sequelae due to foodborne illness and to investigate the food vehicles involved.

End