

# 行政院及所屬各機關出國報告（出國類別：其他）

## 養禽場沙門氏菌流行病學監控技術研習

出國人員姓名/服務機關/單位/職稱/電話

蔡向榮/國立台灣大學/獸醫專業學院/教授/02-33663861

邱垂章/行政院農業委員會動植物防疫檢疫局/動物防疫組/組長  
/02-2343-4247

派赴國家：德國及荷蘭

出國期間：中華民國 98 年 10 月 31 日至 98 年 11 月 9 日

報告日期：中華民國 99 年 2 月

行政院及所屬各機關出國報告提要

出國報告名稱：養禽場沙門氏菌流行病學監控技術研習

頁數：95 含附件：是

出國計畫主辦機關/聯絡人/電話

農委會動植物防疫檢疫局/陸怡芬 /02-3343-2052

出國人員姓名/服務機關/單位/職稱/電話

蔡向榮/國立台灣大學/獸醫專業學院/教授/02-33663861

邱垂章/農委會動植物防疫檢疫局/動物防疫組/組長/02-2343-4247

出國類別： 1 考察 2 進修 3 研究 4 實習 5 其他

出國期間：98年10月31日 98年11月9日 出國地區：德國及荷蘭

報告日期：99年2月

分類號/目：F7/農產品檢疫及動物衛生

關鍵詞：家禽、沙門氏菌、監控

內容摘要：(二百至三百字)

沙氏桿菌症(salmonellosis)是一種重要的人畜共通傳染病。由於傳統的屠前與屠後檢查方法，無法檢出無明顯症狀的沙氏桿菌感染雞隻，因此發展了在整個生產過程（從養禽場、屠宰場、肉品加工廠、至超級市場等）的危害分析重點管制(HACCP)系統，在此系統中必須建立沙氏菌的偵測及監測方法，尤其是對個別菌株的表型(phenotype)及基因型(genotype)的區別技術，以供為對個別病原菌株在食物鏈中發生時的回溯追蹤的重要工具。世界各國的消費者對食品安全越來越重視，而在全球化的過程，家禽產品的進出口常以食品安全為主要的非關稅障礙，因此對食品安全的風險分析對保障國內消費者健康以及對爭取家禽產品的出口都十分重要，但我國尚未針對家禽場及其產品的沙氏桿菌做監控，有必要派員至先進國家學習並建立合作關係。本年度國立台灣大學獸醫專業學院蔡向榮教授與行政院農業委員會動植物防疫檢疫局邱垂章組長於10月31日至11月9日前往歐洲德國及荷蘭順利參訪大學、政府實驗室、私人實驗室、家禽團體、家禽場、家禽疫苗製造廠、甚至到超級市場實地了解其回溯追蹤系統，收穫豐碩，對於我國發展養禽場沙門氏菌監控措施至有助益。

## 壹、緣由及目的

沙氏桿菌症(salmonellosis)是一種重要的人畜共通傳染病。由於傳統的屠前與屠後檢查方法，無法檢出無明顯症狀的沙氏桿菌感染雞隻，因此發展了在整個生產過程（從養禽場、屠宰場、肉品加工廠、至超級市場等）的危害分析重點管制(HACCP)系統，在此系統中必須建立沙氏菌的偵測及監測方法，尤其是對個別菌株的表型(phenotype)及基因型(genotype)的區別技術，以供為對個別病原菌株在食物鏈中發生時的回溯追蹤的重要工具。世界各國的消費者對食品安全越來越重視，而在全球化的過程，家禽產品的進出口常以食品安全為主要的非關稅障礙，因此對食品安全的風險分析對保障國內消費者健康以及對爭取家禽產品的出口都十分重要，但我國尚未針對家禽場及其產品的沙氏桿菌做監控，有必要派員至先進國家學習並建立合作關係。本年度國立台灣大學獸醫專業學院蔡向榮教授與行政院農業委員會動植物防疫檢疫局邱垂章組長於 10 月 31 日至 11 月 11 日前往歐洲德國及荷蘭順利參訪大學、政府實驗室、私人實驗室、家禽團體、家禽場、家禽疫苗製造廠、甚至到超級市場實地了解其回溯追蹤系統，收穫豐碩，對於我國發展養禽場沙門氏菌監控措施至有助益。

## 貳、研習日程表

日期	研 習 內 容
10 月 31 日 (六)	於晚上 23:10 搭乘中華航空 CI 63 班機出發。
11 月 1 日(日)	於上午 06:30 抵達維也納 VIENNA SCHWECHAT 機場，其後 07:20 搭奧地利航空(OS 175 班機)出發，於 08:55 抵達漢堡 HAMBURG FUHLBUTTL 機場，晚上住宿於漢堡。
11 月 2 日(一)	上午由漢堡搭火車至 Cuxhaven，下午參訪德國羅曼國際動物保健公司(Lohmann Animal Health) 沙門氏菌減毒活疫苗(Avipro Salmonella Vac E)研發與製造部門，晚上住宿於 Bremen。
11 月 3 日(二)	上午由 Bremen 往漢若威 (Hannover)，下午 13:30 參訪德國漢若威獸醫大學(Veterinarian University Hannover)動物衛生研究所(Institute for Animal Hygiene, Welfare and Behaviour of

	Farm Animals)，由所長 Prof. Hartung (Member of EFSA)介紹 EFSA 與德國沙門氏菌之控制。下午 16:00 參訪漢若威獸醫大學 Ruthe 校區實驗農場。晚上住宿於漢若威。
11 月 4 日(三)	上午 9:30 參訪漢若威獸醫大學食品衛生與人畜共通傳染病研究所(Institute for Food Hygiene and Zoonosis) ，所長 Prof. Klein (Member of EFSA)親自介紹屠宰場沙門氏菌之控制，與該所之實驗室。下午 14:00 參訪私人獸醫實驗室(Vet Lab Ankum) ，研習有關商用雞群之沙門氏菌之控制，晚上住宿於 Rheine。
11 月 5 日(四)	前往荷蘭參訪位於 Deventer 之荷蘭動物保健服務中心 (GD Animal Health Service Deventer) ，晚上住宿於 Visbek。
11 月 6 日(五)	上午參訪位於 Oldenburg 的德國消費者保護及食品安全署下薩克遜州辦公室(LAVES, Lower Saxonian Office for Consumer Protection and Food Safety) 奧登堡獸醫所 (The Veterinary Institute Oldenburg) ，此為負責沙門氏菌檢測之官方實驗室。下午 14:00 參訪 PHW 集團中央實驗室 (PHW Central Laboratory) ，此為負責肉用家禽(包括肉雞、鴨與火雞)之沙門氏菌控制，晚上住宿於 Visbek。
11 月 7 日(六)	上午 PHW 集團中央實驗室(PHW Central Laboratory)由 Dr. Löhren 介紹沙門氏菌疫苗接種計畫，參訪地區超市雞肉與雞蛋產品之回溯追蹤系統(Traceability) 。晚上住宿於 Köln。
11 月 8 日(日)	上午由 Köln 搭火車至法蘭克福，邱垂章組長由法蘭克福 (FRA) FRANKFURT INTL 機場搭機返國，於 11 月 9 日抵達台灣桃園國際機場。蔡向榮教授則繼續訪英行程，於 12:25 搭德國漢莎航空(LH 4728 班機)出發，並於下午 13 :05 抵達倫敦希斯洛機場(LHR)。
11 月 9 日(一)	赴英國蛋業委員會 (British Egg Industry Council; BEIC) 參訪，由 Dr. Paul McMullin 介紹英國家禽場之沙門氏菌控制情形。
11 月 10 日(二)	上午 10:00 搭荷蘭航空(KL 1008 班機)由倫敦希斯洛機場 (LHR) 出發，並於下午 12:30 抵達阿姆斯特丹 (AMS) AMSTERDAM SCHIPHOL 機場，並於下午 14:25 轉搭中華航空(CI 66)返台。

11月11日(三)	於下午 13:00 抵達台灣桃園國際機場。
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## 參、內容摘要報告

### 一、德國部分

#### (一) 德國漢若瓦獸醫大學(Veterinarian University Hannover)

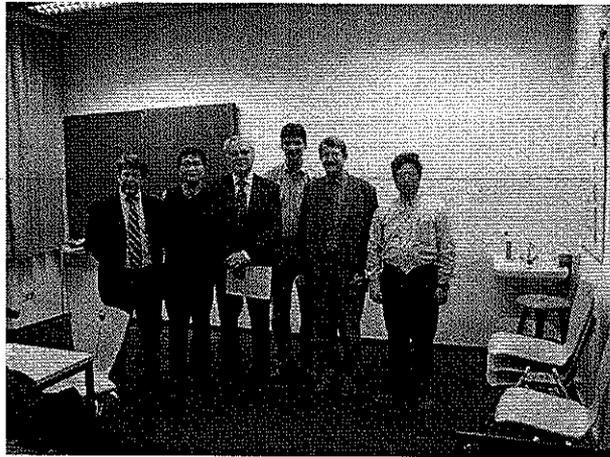
拜訪動物衛生研究所(Institute for Animal Hygiene , Welfare and Behaviour of Farm Animals)所長 Prof. Hartung (Member of EFSA)及食品衛生與人畜共通傳染病研究所(Institute for Food Hygiene and Zoonosis)所長 Prof. Klein (Member of EFSA)。德國漢諾威獸醫大學 (University of Veterinary Medicine Hannover)有四個校區包括 Campus Bischofsholer Damm(校總區)、Campus Bunteweg(動物醫院)、Ruthe 校區(Field and Research Station)、Bakum 校區(流行病學單位)，Bunteweg 校區有 20,000 m<sup>2</sup> 為大型複合式動物醫院，包括(1)小動物、(2)馬、(3)家畜、爬蟲類及家禽與野禽三大區塊。Ruthe 校區：有乳牛場、肉雞場、火雞場、鴨場、蛋雞場、學生宿舍，學生需在此的實驗牧場工作二週。漢諾威獸醫大學有悠久歷史，創立於 1778 年，為世界上第三個設立的獸醫學校(最早係 1762 年在法國里昂，第二個係 1766 年在阿爾弗(Alfort))，並在 1887 年升級成為大學，目前有 60 名教師，150 位研究助理及 500 名技術人員，包括 6 個臨床診所、13 個研究所、2 個中心，2008/2009 年學年度大學部學生 1,436 人，其中 90.8%為女生，碩士班 604 人，其中 79.5%為女生，博士班 97 人，其中 57.7%為女生。

Prof. Klein 另簡介歐洲食品安全局(European Food Safety Authority; EFSA)之職掌，該局係依據歐盟於 2002 年 1 月 28 日所公佈實施之規範 (Regulation (EC) No. 178/2002)所成立，其起因是一連串與食品有關的恐慌事件(例如狂牛病、載奧辛等)，使消費者對食物鏈之安全失去信心，同時也傷害到對主管當局的信任，因此需要加強歐盟的食品安全系統及政策。

EFSA 的三個目標：(1)改進歐盟的食品安全，(2)重建消費者對歐盟食品安全的信心，(3)重建歐盟貿易夥伴對歐盟供應食品之信心。因此 EFSA 被賦與之使命為：(1)提供歐盟在直接或間接影響食品和飼料安全各領域的立法或政策之科學諮詢及支持，(2)提供在這些領域內所有事務的獨立的資訊，(3)風險溝通。EFSA 成立後的主要改變為(1)風險分析與風險管理分開，(2)EFSA 提供歐盟建議，但並不隸屬於歐盟執行委員會 (EC)，(3) EFSA 有獨立的管理委員會，(4) EFSA 與國家政府當局密切合作，(5) EFSA 積極考慮並滿足相關利益者(包括消費者)的需求，(6)對科學小組(Scientific Panels)更好的支持和協調。EFSA 的任務來源包括(1) 歐盟執行委員會，(2)歐洲議會，(3)歐盟會員國，(4) EFSA 自己。EFSA 的任務如下：(1)提供歐盟立法及政策的科學建議、意見、資訊及技術支援，(2)收集及分析資料以使得能對風險定性及監測，(3)促進和協調發展一致的風險評估方法，(4)與 EFSA 使命有關的所有事件的風險溝通。

EFSA 並不負責食品安全之立法，也不負責食品安全及品質之控制、標示或其他此類之問題，也不作為國家當局的替代者。因此 EFSA 係歐盟有關食品及飼料安全風險評估的基石，經由與國家當局緊密合作以及與相關利益者的公開協商，EFSA 對既存在的和新興的風險提供獨立的科學諮詢意見和明確的溝通。

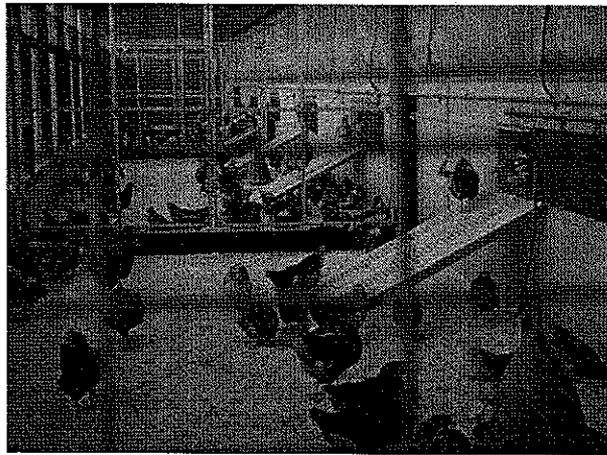
EFSA 總部於 2005 年 10 月從比利時的布魯塞爾搬到義大利的帕爾馬 (Parma)，目前有 400 名員工，在科學委員會及小組有 190 名以上之科學專家。EFSA 2007 年的預算為 5,160 萬歐元，2007 年共提出 185 個意見及報告。



與德國漢若瓦獸醫大學 (Veterinarian University Hannover) 動物衛生研究所 (Institute for Animal Hygiene, Welfare and Behaviour of Farm Animals ) 所長 Prof. Hartung 合影



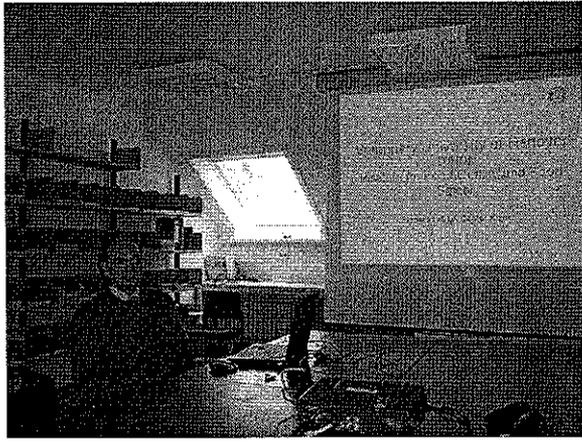
漢若瓦獸醫大學 Ruthe 校區的實驗農場人員  
簡介該場之設施



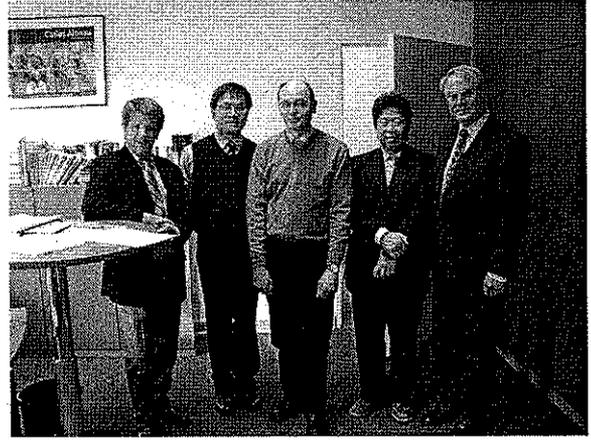
Ruthe 校區的實驗農場蛋雞舍運動場



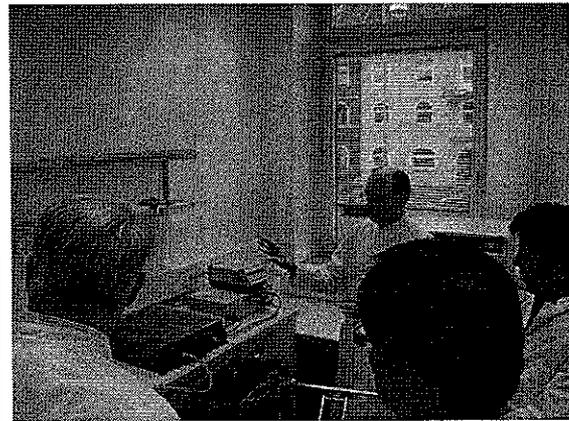
Ruthe 校區實驗農場介紹蛋品回溯追蹤系統之海報



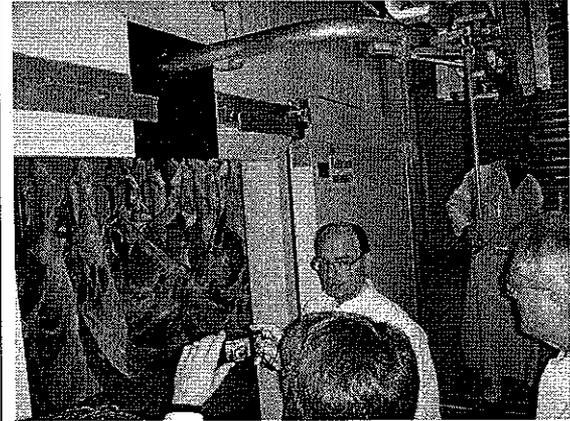
食品衛生與人畜共通傳染病研究所(Institute for Food Hygiene and Zoonosis)所長 Prof. Klein 親自進行簡報



與 Prof. Klein 合影



Prof. Klein 介紹該所實驗室

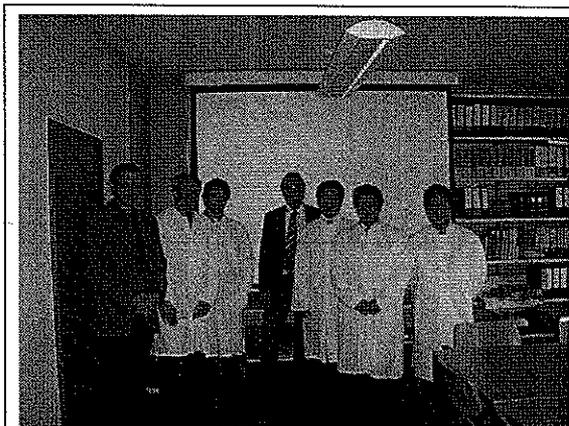


Prof. Klein 介紹該屠宰衛生實驗室

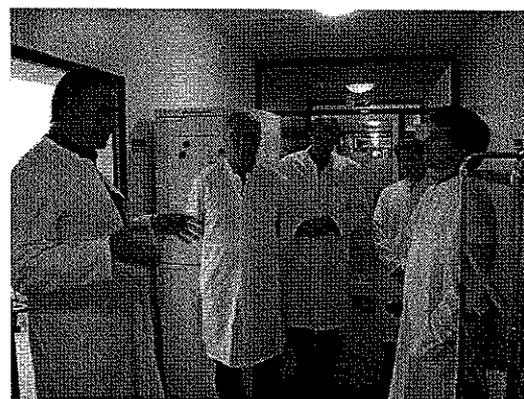
(二) 德國消費者保護及食品安全署下薩克遜州辦公室(LAVES, Lower Saxonian Office for Consumer Protection and Food Safety)的奧登堡獸醫所 (The Veterinary Institute Oldenburg)

由所長 Dr. Klarmann 介紹該所之業務，該所為沙門氏菌檢測之官方樣本以及農場自行採樣樣本中經由認證實驗室檢測之疑陽性樣本之再測試的官方檢測實驗室，下薩克遜州是德國最大的農業區，有 2700 萬頭牛，80 萬頭豬，7300 萬隻家禽 (其中 1200 個大型禽場飼養隻數佔了 90%，其他 10%屬於 20,000 個禽場)，農場數共計 60,000 戶，下薩克遜州的動物沙門氏

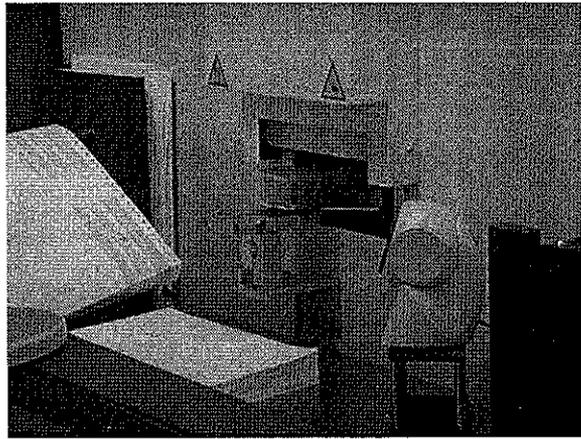
菌症之控制分由二個單位負責，西半部即為我們拜訪的奧登堡獸醫所，東半部則由漢諾瓦獸醫所(Veterinary Institute Hannover)負責，主要業務為依據下列法律規章進行調查：(1)動物疾病法及其他有關動物疾病之規章，包活歐盟指導綱要並著重於須通報之法定傳染病及人畜共通傳染病，(2)化製法及屠宰場廢棄物[VO(EG)1774/2002]，(3)動物保護法，(4)肉品衛生法、禽肉衛生法、乳蛋產品法[VO(EG)178/2002、882/2004、852-854/2004、2073/2005、RL2003/99/EG]，(5)確保前述要求，例如細胞培養、準備培養基、洗滌實驗室玻璃器皿等。以 2007 年為例，奧登堡獸醫所共執行傳播性海綿狀腦病(TSE)檢測 163,404 件，血清學檢測 745,108 件，病毒分子生物學檢測 87,602 件，細菌學及寄生蟲學檢測 12,139 件，病理學 5,220 件，藥劑殘留 14,718 件，肉品工業衛生管制 14,355 件，屠宰動物細菌學管制及抗生素殘留 50,070 件。



與德國消費者保護及食品安全署下薩克遜州辦公室(LAVES)的奧登堡獸醫所(The Veterinary Institute Oldenburg)所長 Dr. Klarmann 等合影



奧登堡獸醫所人員介紹實驗室



奧登堡獸醫所執行血清學檢測之自動化機器手臂



奧登堡獸醫所之外觀

(三) 德國羅曼國際動物保健公司(Lohmann Animal Health) 種雞部門與沙門氏菌減毒活疫苗(Avipro Salmonella Vac E)研發部門

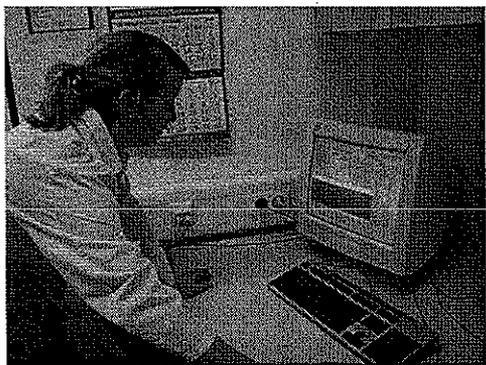
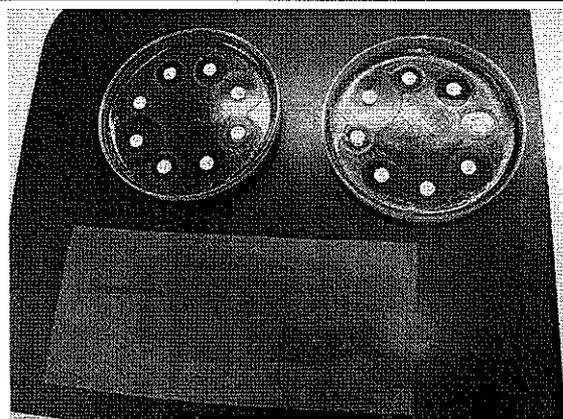
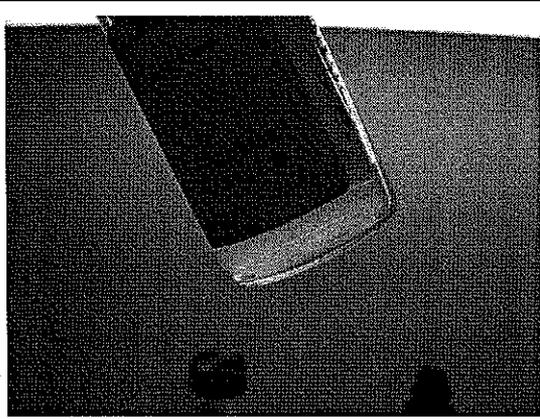
羅曼國際動物保健公司(Lohmann Animal Health)其總部在德國之集團，包括 40 家公司，其製藥部門所生產之沙門氏菌減毒活疫苗(Avipro Salmonella Vac E)係歐盟中英國等國家普遍用於家禽場之沙門氏菌控制計劃中，其雞白痢診斷抗原亦為美國 NPIP 計劃中之實驗室用於種雞測試，此外羅曼公司擁有海蘭(Hyline)、羅曼(Lohmann)、愛拔益加(AA)、羅絲(Ross)等蛋種雞及肉種雞原種雞場，其種雞場之沙門氏菌控制計劃非常成功值得借鏡，因此擬在本計畫中前往其研發部門及種雞部門參訪。Avipro 沙氏桿菌活菌苗有 SE 菌苗(Sm24/Rif12/Ssq 株)及 ST 菌苗(Nal2/RiF9/Rtt 株)二種，一劑量皆含  $1 \times 10^8 \sim 6 \times 10^8$  (cfu 菌量)，疫苗接種計畫在種雞及蛋雞皆為在 1 日齡、6-8 週齡，16-18 週齡三次飲水投與，肉雞可在 1 日齡飲水投與 1 次 ST 活菌苗。活菌菌苗株只會在雞隻臟器(肝)有短暫感染，在腸管可以早期定殖(Colonization)。

Avipro Plate，此係利用微量稀釋法進行抗藥性檢測(Microdilution Antimicrobial Susceptibility)來區別 Avipro 活疫苗菌株及野外菌株，Avipro Plate 為已加有不同濃度之 18 種抗生素的 96 孔盤，經由加入在肉湯培養液

中之定量沙氏桿菌後，在 35-37<sup>0</sup>C 培養 18-24 小時後判讀。

(四) PHW 集團 Wiesenhof 子公司的實驗室

Wiesenhof 旗下擁有 8 個孵化場，17 個屠宰加工廠，1 個實驗室，2 個香腸製造廠及 1 個產品開發中心，Dr. Ulrich Lohren 為負責人。此實驗室是德國的沙氏桿菌檢測認證實驗室之一。

	
PHW 集團中央實驗室	以菌株之生長能力區別沙氏桿菌疫苗株與野外株之方法
	
以菌株之抗藥性形式區別沙氏桿菌疫苗株與野外株之方法	培養後呈現混濁液體表示菌株在此特殊培養基具生長能力因此為野外株

(五) 位於 Delbruck 的私人獸醫實驗室(Vet Lab Ankum)，此係德國的沙門氏菌檢測認證實驗室之一。



與私人獸醫實驗室(Vet Lab Ankum)負責人 Dr. Manfred Poppel 等合影



實驗室人員示範沙氏桿菌採樣方法-鞋套法



以菌株之生長能力區別沙氏桿菌疫苗株與野外株之方法



以菌株之抗藥性形式區別沙氏桿菌疫苗株與野外株之方法

#### (六) 德國家禽沙門氏菌之監控計畫

德國需通報之人類傳染病包括沙氏桿菌症、彎曲桿菌症、輪狀病毒感染症及諾羅病毒(Norovirus)感染症。2006年德國有3,616件人沙門氏菌症病例，其中43.94%由SE所引起，24.81%由ST所引起，其他包括Salmonella Subspecies I 9.54%，S. Hadar 2.1%，S. Infantis 1.63%，S. Paratyphi B 1.27%，及其他>100種以上之血清型。2006-2007年由蛋雞所分離的沙氏桿菌中SE佔82.8%、ST佔2.4%，而肉雞分離株中SE佔3.4%、ST佔8.7%、S. paratyphi B佔19.5%(蛋雞為0%)，S. 4.12:D;-佔18.8%、S. Anatum佔12.1%、S. Infantis佔10.7%。

德國種雞群如果發現為沙氏桿菌陽性，其種蛋將不能再用於孵化，只能在低溫殺菌後出售，因此失去經濟價值，種雞場通常會將之除群。肉雞群如為陽性，需進行流行病學調查污染來源，糞便需在政府獸醫官監督下堆肥處理，目前如為 SE 或 ST 陽性可以在加工廠煮熟後出售，但在 2010 年 1 月後需在化製廠化製處理，目前其他血清型陽性場肉雞可以在陰性場肉雞屠宰後進行屠宰加工。

歐盟將在 2012 年 1 月 1 日起禁止以格子籠飼蛋雞(Battery Cages)，而需以豐富的籠架(Enriched Cages)取代，德國則在 2009 年底提前實施。在德國超市販售之雞蛋可以看見不同飼養系統的標示包括：有機(organic)、自由放牧(free range)、平飼(floor raised)、小群飼養(small colony keeping，每群 40-60 隻母雞)。

德國家禽沙門氏菌的盛行率及預期減少目標如下表：

動物別	生產別	群盛行率	目標	完成日期
種雞	雞群	0.3%	<1%	2007.1.1
蛋雞	雞群	29.3%	-30%	2008.1.2
		24.7%(SE,ST)		
肉雞	雞群	17.5%	≤1%	2011.12.31
		2.9%(SE,ST)		
肉雞	屠體	17.6%	?	?
		4.6%(SE,ST)		

德國家禽沙門氏菌之監控計畫如下表：

動物別	階層	自行採樣	官方採樣	規章 RL/VD(EG)
種雞	孵化場	每 6 週， S.Gal 每 2 週	每 16 週(當產蛋雞 未監控時)	90/539 1003/2005
	種雞	3 次(1 日齡、4 週齡、 14-16 週齡)	無	
	產蛋雞	1 次 Sal Gal, 每 2 週	生產週期中 3 次	
蛋雞	種雞	2 次(1 日齡、14-16 週)		1168/2006

		齡)		
	蛋雞場	每 15 週	1 次(如上次為沙氏桿菌陽性再加 1 次)	
肉雞	肉雞場	1 次(屠宰前 3 週)	1 次(10%肉雞群)	646/2007

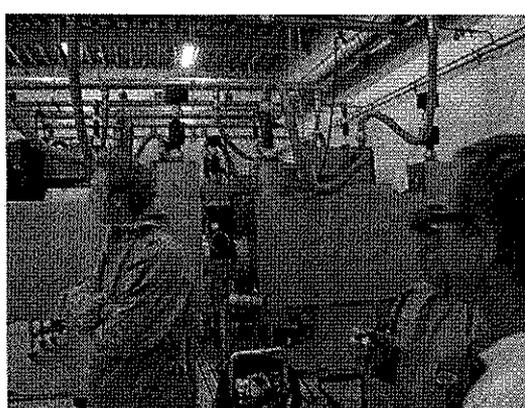
## 二、荷蘭 Deventer 動物保健服務中心 (GD Animal Health Service Deventer)

荷蘭的動物保健服務中心(Animal Health Service, AHS)位於荷蘭阿姆斯特丹東方約 90 公里處的 Deventer，由本來位於 Doorn 的家禽保健中心(Poultry Health Center, PHC)及位於 Gouda 的家禽保健中心合併成目前的動物保健服務中心。為提供動物疾病診斷、保健諮詢及疾病控制的服務中心。GD 在 2000 年轉型成爲一家以市場爲導向的私人公司，主要爲改善動物的健康和動物產品的安全。GD 的獸醫專家爲農民和執業獸醫提供對傳染病防治、畜牧業和動物福利各方面的協助和諮詢。GD 有一個大型診斷實驗室自 1993 年以來已被荷蘭認證委員會 (RVA) 基於 NEN-EN-ISO 17025 國際標準認證。GD 的員工約有 180 名員工，包括多方面領域的專家，包括病理學、微生物學、化學、和毒理學。

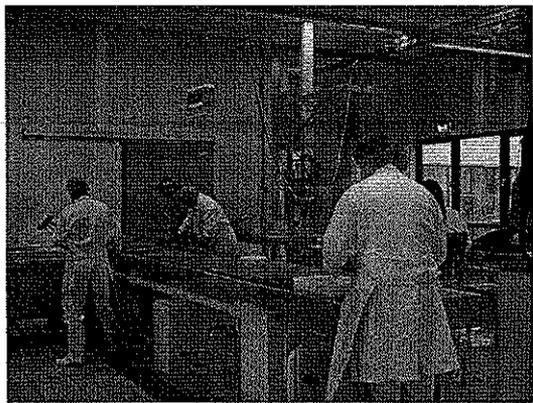
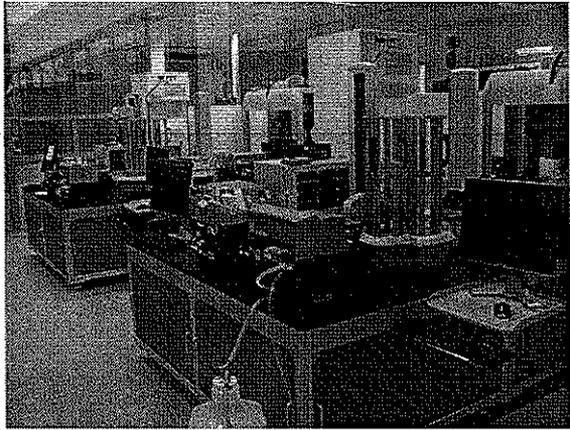
GD 成立之後業務逐漸擴大，至目前其服務範圍不只限於荷蘭全國，也遍及全世界其他 38 個國家，爲一頗具規模的國際性組織。其國際性的比重由其對國外的診斷工作佔其所有診斷工作的 40%可見。由國際送至機場的郵件，12 小時內就送達實驗室，對一般的檢驗，結果於 1 週內可傳真回答送檢者。



荷蘭動物保健服務中心(GD Animal Health Service Deventer)人員簡介該中心



參訪荷蘭動物保健服務中心飼養實驗雞隻之隔離箱設施

 <p>荷蘭動物保健服務中心人員進行送檢病例剖檢之情形</p>	 <p>荷蘭動物保健服務中心血清學檢測設施</p>
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### 荷蘭養雞場沙門氏桿菌監測計畫

家禽種類與飼養階段	蛋種雞及肉種雞育成階段	種雞之產蛋階段
執行產業及目標	蛋種雞場：針對歐洲最常見的五種血清型 (SE、ST、SH、SI、SV)。	蛋種雞場：針對歐洲最常見的五種血清型 (SE、ST、SH、SI、SV)。 肉種雞場：包含所有血清型之沙門氏桿菌。
採樣時程	(1) 1日齡時採取40片雞盒墊紙。 (2) 4週齡時採取糞便檢體，每兩個糞便混合成一份，共採30份檢體。 (3) 出雞前14天內採取糞便檢體，每6個糞便混合成一份，共採25份檢體。	每3週一次，自第24週齡開始採樣 (於2009年10月25日前為每2週一次)。
採樣方法	(1) chicken paper：每片大小為長5乘寬5公分的紙片，需可清楚看到有糞便污染情	(1) 籠飼型態雞場：以共泄腔拭子採取新鮮糞便檢體，每2個混合成一份檢體檢

	<p>形，放置於籠子底部。</p> <p>(2) 糞便檢體：下面 3 種採樣法擇一進行：</p> <p>a. 可以拭子採取籠子底部的新鮮糞便檢體。</p> <p>b. 採取共泄腔拭子。</p> <p>c. 於非籠飼雞場以鞋套採集糞便樣本，每 2 雙鞋套混合成 1 份檢體，共採集 5 份檢體。</p>	<p>測，共需 150 份檢體。</p> <p>(2) 非籠飼型態雞場：以鞋套採集糞便樣本，每 2 雙鞋套混合成一份檢體，共採集 5 份檢體進行檢測。</p>
採樣人	<p>(1) 需由雞場員工作為雛雞盒墊紙的採樣者。</p> <p>(2) 其餘檢體由 GD 或 GD 認證人員。</p>	<p>(1) 以上由雞場員工進行。</p> <p>(2) 另外官方樣本由 GD 或 GD 認證人員於每個生產週期中採樣三次，採樣時點為：</p> <p>a. 蛋中雞送達產蛋雞場的 4 週內。</p> <p>b. 雞群淘汰前八週。</p>

家禽種類與飼養階段	蛋雞之育成階段	白肉雞
執行產業及目標	針對 SE、ST。	包含所有血清型之沙門氏桿菌，對 <i>S. paratyphi B</i> var. Java 有特別的規定
採樣時程	運送至產蛋雞舍前 14 天 (即使是在同一雞場內)。	<p>(1) 1 日齡時採取 40 片雛雞盒墊紙、</p> <p>(2) 大於 3 週齡時採取 30 份糞便檢體 (如果在 34 日齡前屠宰，則可提前採樣，檢測結果必須能再運送至屠宰廠前 24 小時之前得到)。</p>
採樣方法	由農場主人擇一檢測： (1) 血液樣本：依照雞場	(1) 雛雞盒墊紙：每片大小為長 5 乘寬 5 公分

	飼養量決定樣本數，採檢總飼養數的 0.5% 雞隻；樣本數最少應達 24 隻，最多 60 隻。	的紙片，需可清楚看到有糞便污染情形，放置於籠子底部。
	(2) 糞便檢體：籠飼型態雞場至少須採兩份各 150 克重的糞便 (3) 非籠飼型態的雞場：2 雙鞋套混合成 1 份檢體，共採 2 份檢體。 糞便或鞋套檢體可於實驗室混合成一份進行檢測。	(2) 糞便檢體：下面 3 種採樣法擇一進行 a. 可以拭子採取籠子底部的新鮮糞便檢體。 b. 採取共泄腔拭子。 c. 於非籠飼雞場以鞋套採集糞便樣本，每 2 雙鞋套混合成 1 份檢體，共採集 5 份檢體。
採樣人	由 GD 或 GD 認證人員進行。	由雞場員工執行。

家禽種類與飼養階段	蛋雞之產蛋階段	陽性場如何處理
執行產業及目標	針對 SE、ST。	種雞：淘汰感染 SE 與 ST 的雞群，當感染 Virchow、Hardar、Infantis 這 3 種血清型時，由政府部門決定是否淘汰陽性雞群。
採樣時程	每 15 週一次，自第 22 週齡開始採樣。	
採樣方法	(1) 籠飼型態雞場：以共泄腔拭子採取新鮮糞便檢體，每 2 個混合成一份檢體檢測，共需 150 份檢體。 (2) 非籠飼型態雞場：以鞋套採集糞便樣本，每 2 雙鞋套混合成一份檢體，共採集 2 份檢體進行檢測。 所有檢體可混合成一份檢體進行檢測	商用蛋雞： 針對 SE 與 ST，產自污染場的蛋應先經處理以滅除沙門氏菌。  白肉雞：沙門氏菌陽性場雞群必需在陰性場屠宰後才可屠宰。
採樣人	主要由雞場員工進行，但是當以下情況時由 GD 或	其他尚包括：

	<p>GD 認證人員採取官方樣本：</p> <p>a. 當雞場飼養規模大於 1000 隻時，每個雞場需檢測 1 個雞群 1 次。</p> <p>b. 24 週齡時，若上批雞隻於此飼養階段時曾發生 SE 或 ST 陽性的情況。</p> <p>c. 當政府部門認為有任何理由可懷疑 SE 或 ST 污染時。</p>	<p>增加官方監控陽性場下批雞隻的情形、監控雞場之清潔消毒程度、強制追溯陽性場的污染源頭等；以上規則決定於感染的鳥種 (例如 repro 或白肉雞)，甚至感染的沙門氏菌血清型別等 (例如 S. Java 在白肉雞的污染有其特別的規範)。</p>
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### 三、英國雞場沙氏桿菌控制計畫

#### (一) 種雞場沙氏桿菌控制計畫

英國從 1993 年根據 Directive 1992/117 即開始執行控制計畫。此計畫已經成功地減少種雞感染 S. Enteritidis 與 S. Typhimurium 的程度。於 2004 年，以現行的檢測方法發現種雞感染 S. Enteritidis, S. Typhimurium, S. Hadar, S. Infantis, 與 S. Virchow 的比率低於 1.0%，已達到 2009/12/31 的預定目標。新的國家控制計畫將遵行 Regulation 2160/2003 實施，並且在 2007/1/1 以前達成。控制計畫內容包括：(1) 種雞場在養隻數達 250 羽以上或是孵化場孵化容量達 1000 顆以上，將必須向主管機關登記。(2)業者需要根據 EC Regulation 2160/2003 之 Annex IIB 實施採樣計畫。以育種用之一日齡小雞、四週齡及產蛋前兩週採取監測沙氏桿菌的樣本。供孵化之種蛋生產期間，業者需每兩週對種雞群採樣一次，以確定能夠達到成年種雞的監測標準。樣本需送到經主管機關授權，並且符合 EN/ISO 標準品質保證系統要求的實驗室。

#### (二) 蛋雞的沙門氏桿菌控制計畫

英國根據 Regulation (EC) No 2160/2003 以及 Regulation (EC) No 1168/2006 實施國家控制計畫。蛋雞的沙門氏桿菌國家控制計畫在 2008 年一月生效。控制計畫內容包括：(1)所有養殖蛋雞數目達 350 羽以上的禽場

將包括在國家控制計畫中。根據英格蘭的蛋雞註冊制度(Registration of Establishments (Laying Hens)(England) Regulations 2003)，這些禽場需要向主管機關註冊登記。而在威爾斯、蘇格蘭與北愛爾蘭亦需遵守同等法規。Regulation (EC) No 2160/2003 中，排除直接提供少量產品給終端顧客的業者。任何養殖少於 350 羽但未在 Regulation (EC) No 2160/2003 之 1.3 章中排除之業者將包括於國家控制計畫中。(2)英國爲了家禽疾病控制，特別是禽流感，已建立 GB 禽類登記系統(The GB Poultry Register)，包含管理飼養雞隻超過 50 羽之場所。GB 禽類登記系統包括英格蘭、威爾斯及蘇格蘭。北愛爾蘭有另一獨立之禽類登記系統。目前英國禽類登記系統除了禽流感以外，在獲得登記系統資料庫中之禽場的同意之前並不能用於其他疾病控制，未來該系統資料庫可能可以用於沙門氏桿菌之控制。(3)根據 EC 2160/2003 之 Annex IIB 條例中，要求業者實施採樣計畫。由雞群業者根據以下時間點採集檢體當做監測沙氏桿菌的樣本:以生產食用蛋之一日齡及產蛋前兩週，或是移入產蛋籠以前之蛋雞，隨後在 22-26 週時開始產蛋間期，每 15 週採樣一次。樣本需送到經主管機關授權，並且符合 EN/ISO 標準品質保證系統要求的實驗室。(4) 當有採集官方樣本時，可能可以取代由業者所需採集樣本。

### (三) 肉雞飼養場之沙門氏桿菌控制計畫

英國沙門氏桿菌國家控制計畫根據 Regulation (EC) No 2160/2003 and Regulation (EC) No 646/2007 執行。飼養肉雞之沙門氏桿菌國家控制計畫在 2009 年 1 月生效。所有肉雞飼養均包含在此國家控制計畫，除非是在 Regulation (EC) No 2160/2003 的 Article 1.3 中排除之飼養戶，例如雞群爲自家飼養食用，或是少量產品直接供應末端消費者或是提供當地零售商，而這零售商則是直接提供未加工產品給末端消費者。英國爲了家禽疾病控制，特別是禽流感，已建立 GB 禽類登記系統(The GB Poultry Register)，包含管理飼養雞隻超過 50 羽之場所。GB 禽類登記系統包括英格蘭、威爾斯

及蘇格蘭。北愛爾蘭有另一獨立之禽類登記系統。目前英國禽類登記系統僅應用於禽流感風險評估、預防與控制，然而經過公開協商，從 2009 年 8 月 1 日開始，此系統之應用擴展至其他禽病之管理，包括其他需通報的疾病及人畜共通傳染病。此系統提供大量飼養肉雞的場所資料庫資料。控制計畫內容包括：(1) 要求業者履行 EC Regulation 646/2007 之 Annex 中的採樣計畫，採樣流程於 Annex 2 中敘述。於出雞至屠宰場三週前期間，業者需採樣兩套鞋套或是兩個拭子樣本。如此在雞隻送至屠宰場前，有足夠的時間對這些樣本進行實驗室檢驗。在英國雞隻屠宰的年齡差異很大，一般大多數肉雞的屠宰雞齡是 35 日至 42 日齡，然而有機雞隻以及改良式雞舍系統(enhanced housing system)雞隻可能會較晚屠宰。偶爾有些雞隻會更早被移出(32 日齡)。在英國經常會由雞群移除一些雞隻，以減少日後雞群的數量。重點是在雞群第一批雞隻屠宰前，了解雞隻感染沙門氏桿菌的情況。從雞群採樣，至實驗室結果出爐，包含血清型鑑定，大約需耗時 2 週。於英國，根據過去經驗，大部分的肉雞在 2 到 4 週齡最可能成為沙門氏桿菌檢驗陽性。因此在第一批雞隻移出前，需要抽樣檢測沙門氏桿菌以了解雞群的感染情況(即使是要較晚屠宰的雞隻，最遲也要在進行屠宰前三週進行採樣)。(2) 樣本需送到經主管機關授權，並且符合 EN/ISO 標準品質保證系統要求的實驗室。(3) 主管機關或其授權人員每年將會隨機挑選 10%的飼養超過 5000 羽之禽場，每場至少一雞群，採取官方檢體(official sample)。此外，也會注意過去業者採樣之樣本為沙門氏桿菌陽性之禽場，特別是過去樣本曾分離出 S. Enteritidis 或 S. Typhimurium 之禽場。(4)採樣以檢測是否達到預定目標。由業者採集所需樣本，可以官方樣本採樣替代。(5)依照 Regulation (EC) No. 646/2007 Annex point 1 (C)，肉雞養殖場業者可向主管機關提出減少採樣申請，而非全場採樣。主管機關應根據下列條件評估減少採樣的申請，如均符合下列條件，主管機關可核准減少採樣。

1. 為一統進/統出的系統，

2. 所有雞群為相同的管理，
3. 所有雞群的飼料與飲水來源均相同，
4. 在一年內或 6 批雞隻飼養期間，曾對於根據標準監控計畫採樣並對沙門氏桿菌進行測試，且禽場所有雞群的樣本皆至少有一次由主管機關採檢，並於此加強測試期間的樣本未發現 SE 或 ST。

#### 肆、心得與建議

本年度赴歐參訪對歐洲家禽場之沙門氏菌控制已有整體之了解，基本上歐盟國家對沙門氏菌控制較美國積極與嚴格，經由設定各個階段的期程目標，促使沙門氏菌的盛行率逐步下降，此外德英等國由業者自行依監測計畫全面採樣送至主管機關授權的實驗室進行檢驗，在輔以由主管機關或其授權人員採取官方樣本在官方實驗室進行檢驗的抽檢方式，不但可以避免官方實驗室的過度負荷，又能達到全面檢驗的目標，基本上歐盟國家之家禽場沙門氏菌控制計畫的成效甚佳，頗值得我國借鏡處。

相較於美國僅對 SE 作規範，歐盟在沙氏桿菌之控制更為積極，除以 SE 及 ST 為初始控制目標外，更進一步擴及所謂 5 大血清型，此外由於沙門氏菌污染來源眾多，有時並不易完全清除，尤其是在同時有多種年齡之雞群同時存在之雞場，歐美皆允許疫苗的使用，但是美國要求疫苗接種雞群內必需有部份雞隻不打疫苗，以做為哨兵雞隻監控有無野外菌株之感染，歐盟則要求活菌減毒疫苗株必需能有與野外菌株簡易區別之方法，我國多年齡之蛋雞群在某些區域飼養十分密集，將來在改善種雞群與飼料等污染來源、以及蛋雞群本身之安全防護措施後，如仍未能將盛行率降到一定比例，例如 10% 時，亦應考慮開放以疫苗接種來協助沙氏桿菌之控制。

#### 伍、致謝

- 一、感謝防檢局長官給予此次出國機會，有幸瞭解德國及荷蘭家禽沙門氏菌監控計畫。
- 二、感謝國立台灣大學專業獸醫學院蔡向榮教授協助聯繫安排出國相關事宜及旅程上的指導。

## COMMISSION REGULATION (EC) No 1237/2007

of 23 October 2007

amending Regulation (EC) No 2160/2003 of the European Parliament and of the Council and Decision 2006/696/EC as regards the placing on the market of eggs from *Salmonella* infected flocks of laying hens

(Text with EEA relevance)

THE COMMISSION OF THE EUROPEAN COMMUNITIES,

down veterinary certification conditions for imports and transit through the Community of eggs and egg products.

Having regard to the Treaty establishing the European Community,

Having regard to Regulation (EC) No 2160/2003 of the European Parliament and of the Council of 17 November 2003 on the control of *Salmonella* and other specified food-borne zoonotic agents<sup>(1)</sup> and, in particular Article 5(6) thereof,

Having regard to Regulation (EC) No 853/2004 of the European Parliament and of the Council of 29 April 2004 laying down specific hygiene rules for food of animal origin<sup>(2)</sup>, and in particular Article 9 thereof,

Whereas:

- (1) Regulation (EC) No 2160/2003 lays down rules to ensure that proper and effective measures are taken to detect and control *Salmonella* and other zoonotic agents at all relevant stages of production, processing and distribution, particularly at the level of primary production, in order to reduce their prevalence and the risk they pose to public health.
- (2) Pursuant to Annex II to Regulation (EC) No 2160/2003, with effect from 72 months after the date of entry into force of that Regulation, eggs may only be used for direct human consumption as table eggs if they originate from a commercial flock of laying hens subject to a national control programme and not under official restriction.
- (3) Commission Decision 2006/696/EC of 28 August 2006 laying down a list of third countries from which poultry, hatching eggs, day-old chicks, meat of poultry, ratites and wild game-birds, eggs and egg products and specified pathogen-free eggs may be imported into and transit through the Community and the applicable veterinary certification conditions, and amending Decisions 93/342/EEC, 2000/585/EC and 2003/812/EC<sup>(3)</sup> lays

- (4) A high prevalence of *Salmonella* Enteritidis and *Salmonella* Typhimurium was detected in flocks of laying hens in Member States during a study carried out in accordance with Commission Decision 2004/665/EC of 22 September 2004 concerning a baseline study on the prevalence of *Salmonella* in laying flocks of *Gallus gallus*<sup>(4)</sup>.
- (5) According to the Report of the European Food Safety Authority on Trends and Sources of Zoonoses, Zoonotic agents and Antimicrobial Resistance and Foodborne Outbreaks in the European Union in 2005<sup>(5)</sup> eggs and egg products are the most important source of known foodborne outbreaks of salmonellosis in humans. In addition, according to that report, *Salmonella* Enteritidis and *Salmonella* Typhimurium were responsible for 88 % of the outbreaks where the serovar was demonstrated.
- (6) In view of the high prevalence of *Salmonella* Enteritidis and *Salmonella* Typhimurium in certain Member States, its public health impact and the reluctance of food business operators to trade table eggs from infected flocks, the date on which restrictions on the consumption of table eggs are to apply, should be brought forward but should still allow food business operators sufficient time to comply with the new requirements without causing any disturbance to markets.
- (7) However, where a flock of laying hens has been indicated as the source of infection in a food borne outbreak as a result of the epidemiological investigation of food-borne outbreaks in accordance with Directive 2003/99/EC of the European Parliament and the Council of 17 November 2003 on the monitoring of zoonoses and zoonotic agents, amending Council Decision 90/424/EEC and repealing Council Directive 92/117/EEC<sup>(6)</sup>, the restrictions on the use of table eggs set out in Annex II to Regulation (EC) No 2160/2003 should apply without delay.

<sup>(1)</sup> OJ L 325, 12.12.2003, p. 1. Regulation as last amended by Council Regulation (EC) No 1791/2006 (OJ L 363, 20.12.2006, p. 1).

<sup>(2)</sup> OJ L 139, 30.4.2004, p. 55, as corrected by OJ L 226, 25.6.2004, p. 22. Regulation as last amended by Regulation (EC) No 1791/2006.

<sup>(3)</sup> OJ L 295, 25.10.2006, p. 1.

<sup>(4)</sup> OJ L 303, 30.9.2004, p. 30.

<sup>(5)</sup> The EFSA Journal (2006), 96.

<sup>(6)</sup> OJ L 325, 12.12.2003, p. 31. Directive as amended by Council Directive 2006/104/EC (OJ L 363, 20.12.2006, p. 352).

- (8) Taking account of the public health risk of eggs infected with *Salmonella*, rules should be laid down on the marking of eggs to guarantee that eggs from flocks which are subject to restrictions within the framework of a *Salmonella* control programme provided for in Regulation (EC) No 2160/2003, are marked in a way which easily distinguishes them from table eggs before being placed on the market.
- (9) In order to exclude false-positive initial results, the competent authority should be allowed to lift the restrictions laid down in Paragraph 2 of Part D to Annex II of that Regulation if the *Salmonella* infection is not confirmed in the flocks of laying hens using a strict protocol.
- (10) Third countries, from which Member States are authorised to import eggs, should provide guarantees, equivalent to the requirements within the Community and the model certificate for eggs in Decision 2006/696/EC should be amended accordingly.
- (11) Regulation (EC) No 178/2002 of the European Parliament and of the Council of 28 January 2002 laying down the general principles and requirements of food law, establishing the European Food Safety Authority and laying down procedures in matters of food safety<sup>(1)</sup>, in particular Article 12 thereof, lays down rules concerning food and feed exported or re-exported from the Community for placing on the market in a third country. Those provisions apply to table eggs. Accordingly, it is not necessary to lay down specific provisions with regard to the export of such eggs in this Regulation.
- (12) To avoid any disturbance of trade, the use of certificates issued in accordance with the current model certificates set out in Decision 2006/696/EC should be allowed for a period of 60 days following the date of application of this Regulation.
- (13) Regulation (EC) No 2160/2003 should therefore be amended accordingly.
- (14) The measures provided for in this Regulation are in accordance with the opinion of the Standing Committee on the Food Chain and Animal health,

HAS ADOPTED THIS REGULATION:

Article 1

Annex II to Regulation (EC) No 2160/2003 is amended in accordance with Annex I to this Regulation.

Article 2

Annex II to Decision 2006/696/EC is amended in accordance with Annex II to this Regulation.

Article 3

Consignments of eggs for which certificates have been issued in accordance with Decision 2006/696/EC in its version before 1 November 2007 may be imported into the Community for a period of 60 days following that date.

Article 4

This Regulation shall enter into force on the third day following its publication in the *Official Journal of the European Union*.

It shall apply from:

- 1 November 2007 where *Salmonella* ssp. are identified in the flock of laying hens as the source of infection for humans by the consumption of eggs or egg products as a result of the epidemiological investigation of food-borne outbreaks in accordance with Article 8 of Directive 2003/99/EC,
- 1 January 2009 at the latest to all other flocks of laying hens.

This Regulation shall be binding in its entirety and directly applicable in all Member States.

Done at Brussels, 23 October 2007.

For the Commission  
Markos KYPRIANOU  
Member of the Commission

<sup>(1)</sup> OJ L 31, 1.2.2002, p. 1. Regulation as last amended by Commission Regulation (EC) No 575/2006 (OJ L 100, 8.4.2006, p. 3).

## ANNEX I

In Annex II to Regulation (EC) No 2160/2003, Part D is replaced by the following:

**D. Specific requirements concerning flocks of laying hens**

1. Eggs shall not be used for direct human consumption as table eggs unless they originate from a commercial flock of laying hens subject to a national control programme established under Article 5 and not under official restriction.
2. Eggs originating from flocks with unknown health status, that are suspected of being infected or that are infected with *Salmonella* serotypes for which a target for reduction has been set or which were identified as the source of infection in a specific human foodborne outbreak, may be used for human consumption only if treated in a manner that guarantees the destruction of all *Salmonella* serotypes with public health significance in accordance with Community legislation on food hygiene.

Eggs originating from flocks with unknown health status, that are suspected of being infected or that are infected with *Salmonella* serotypes for which a target for reduction has been set or which were identified as the source of infection in a specific human foodborne outbreak, shall be:

- (a) considered as Class B eggs as defined in Article 2(4) of Commission Regulation (EC) No 557/2007 laying down detailed rules for implementing Council Regulation (EC) No 1028/2006 on marketing standards for eggs<sup>(1)</sup>;
  - (b) marked with the indication referred to in Article 10 of Commission Regulation (EC) No 557/2007 which clearly distinguishes them from Class A eggs prior to being placed on the market;
  - (c) prohibited access to packaging centres unless the competent authority is satisfied with the measures to prevent possible cross-contamination of eggs from other flocks.
3. When birds from infected flocks are slaughtered or destroyed, steps must be taken to reduce the risk of spreading zoonoses as far as possible. Slaughtering shall be carried out in accordance with Community legislation on food hygiene. Products derived from such birds may be placed on the market for human consumption in accordance with Community legislation on food hygiene and, once applicable, part E. If not destined for human consumption, such products must be used or disposed of in accordance with Regulation (EC) No 1774/2002.
  4. In order to exclude false-positive initial results, the competent authority may lift the restrictions laid down in point 2 of this Part:
    - (a) when the flock of layers is not the source of infection for humans by the consumption of eggs or egg products as a result of the epidemiological investigation of food-borne outbreaks in accordance with Article 8 of Directive 2003/99/EC; and
    - (b) where the flock is subjected to a national control programme established under Article 5 and *Salmonella* serotypes for which a target for reduction has been set, is not confirmed by the following sampling protocol carried out by the competent authority:
      - (i) the technical specifications referred to in Article 5 of Commission Decision 2004/665/EC (seven samples); however, a sub-sample of 25 grams must be collected of each faecal material and dust sample for analysis; all samples must be analysed separately;or
      - (ii) bacteriological investigation of the caeca and oviducts of 300 birds;or
      - (iii) bacteriological investigation of the shell and the content of 4 000 eggs of each flock in pools of maximum 40 eggs.

In addition to the sampling in point (b), the competent authority shall verify the absence of the use of antimicrobials, potentially affecting the result of the analyses of the sampling.

<sup>(1)</sup> OJ L 132, 24.5.2007, p. 5.

## ANNEX II

In Part 2 of Annex II to Decision 2006/696/EC, the model veterinary certificate for eggs (E) is replaced by the following:

**Model veterinary certificate for eggs (E)**

COUNTRY		Veterinary certificate to EU		
Part I : Details of dispatched consignment	I.1. Consignor Name Address Tel. No		I.2. Certificate reference number I.2.a. <input type="checkbox"/>	
			I.3. Central Competent Authority	
			I.4. Local Competent Authority	
	I.5. Consignee Name Address Postal code Tel. No		I.6. <input type="checkbox"/>	
	I.7. Country of origin	ISO code	I.8. Region of origin	Code
	I.9. Country of destination	ISO code	I.10. Region of destination	Code
	I.11. Place of origin Name Address Approval number		I.12. <input type="checkbox"/>	
	I.13. Place of loading		I.14. Date of departure	
	I.15. Means of transport Aeroplane <input type="checkbox"/> Ship <input type="checkbox"/> Railway wagon <input type="checkbox"/> Road vehicle <input type="checkbox"/> Other <input type="checkbox"/> Identification: Documentary references:		I.16. Entry BIP in EU	
			I.17. <input type="checkbox"/>	
	I.18. Description of commodity		I.19. Commodity code (HS code) 04.07	
			I.20. Quantity	
	I.21. Temperature of product Ambient <input type="checkbox"/> Chilled <input type="checkbox"/> Frozen <input type="checkbox"/>		I.22. Number of packages	
	I.23. Identification of container/Seal number		I.24. Type of packaging	
I.25. Commodities certified for: Human consumption <input type="checkbox"/>				
I.26. <input type="checkbox"/>		I.27. For Import or admission into EU <input type="checkbox"/>		
I.28. Identification of the commodities Species (Scientific name)      Manufacturing plant      Approval number of establishments Cold store      Number of packages      Net weight				

## E (Eggs)

	II. Health information	II.a. Certificate reference number	II.b.
Part II: Certification	<b>II.1. Health attestation</b>		
	<p>I, the undersigned official veterinarian/official inspector, declare that I am aware of the relevant provisions of Regulations (EC) Nos 178/2002, 852/2004, 853/2004 and 2160/2003 and hereby certify that the eggs described in this certificate have been obtained in accordance with those requirements, and in particular that:</p> <p>II.1.1. they come from (an) establishments(s) implementing a programme based on the HACCP principles in accordance with Regulation (EC) No 852/2004;</p> <p>II.1.2. they have been kept, stored, transported and delivered in accordance with the relevant conditions laid down in Section X, Chapter I of Annex III to Regulation (EC) No 853/2004;</p> <p>(1) II.1.3. they fulfil the requirements of Commission Regulation (EC) No 1688/2005 of 14 October 2005 implementing Regulation (EC) No 853/2004 of the European Parliament and of the Council as regards special guarantees concerning <i>Salmonella</i> for consignments to Finland and Sweden of certain meat and eggs;</p> <p>II.1.4. the guarantees covering live animals and products thereof provided by the residue plans submitted in accordance with Directive 96/23/EC, and in particular Article 29 thereof, are fulfilled.</p> <p>(2) II.1.5. they fulfil the requirements in Article 10(6) of Regulation (EC) No 2160/2003 of the European Parliament and of the Council of 17 November 2003 on the control of <i>Salmonella</i> and other specified food-borne zoonotic agents. In particular:</p> <p>Eggs shall not be imported from flocks of laying hens in which <i>Salmonella</i> ssp. has been detected a result of the epidemiological investigation of a food-borne outbreak or if no equivalent guarantees have been provided.</p> <p>[From 1 January 2009 on, eggs shall also not be imported from flocks of laying hens with unknown health status, that are suspected of being infected or from flocks infected by <i>Salmonella</i> ssp. for which a target for reduction has been set in Community legislation (3) and on which monitoring equivalent to the monitoring laid down in the provisions in the Annex of Regulation (EC) No 1168/2006 is not applied, or if no equivalent guarantees have been provided.]</p> <p><b>Notes</b></p> <p><b>Part I:</b></p> <p>Box I.8: provide the code for the region of origin, if necessary, as defined under code of territory in column 2, Part 1 of Annex II of Decision 2006/696/EC [as last amended].</p> <p>Box I.11: Name, address and approval number of establishment of dispatch.</p> <p>Box I.15: Indicate the registration number(s) of railway wagons and lorries, the names of ships and, if known, the flight numbers of aircraft. In the case of transport in containers or boxes, the total number of these and their registration and seal numbers, where applicable, should be indicated in box I.23.</p> <p><b>Part II:</b></p> <p>(1) Delete if the consignment is not intended for export to Sweden or Finland.</p> <p>(2) Only applicable in case of import of eggs, Class A in accordance with Article 3 of Regulation (EC) No 1028/2006. Delete as appropriate.</p> <p>(3) <i>Salmonella Enteritidis</i> and <i>Salmonella Typhimurium</i></p>		
	<p>Official veterinarian or official inspector</p> <p>Name (in capitals): _____ Qualification and title: _____</p> <p>Local competent authority: _____ Signature: _____</p> <p>Date: _____</p> <p>Stamp: _____</p>		

COMMISSION REGULATION (EC) No 1168/2006  
of 31 July 2006

implementing Regulation (EC) No 2160/2003 as regards a Community target for the reduction of the prevalence of certain salmonella serotypes in laying hens of *Gallus gallus* and amending Regulation (EC) No 1003/2005

(Text with EEA relevance)

THE COMMISSION OF THE EUROPEAN COMMUNITIES,

Having regard to the Treaty establishing the European Community,

Having regard to Regulation (EC) No 2160/2003 of the European Parliament and of the Council of 17 November 2003 on the control of salmonella and other specified food-borne zoonotic agents<sup>(1)</sup> and, in particular Article 4(1) and Article 13 thereof,

Whereas:

(1) The purpose of Regulation (EC) No 2160/2003 is to ensure that proper and effective measures are taken to detect and control salmonella and other zoonotic agents at all relevant stages of production, processing and distribution, particularly at the level of primary production, in order to reduce their prevalence and the risk they pose to public health.

(2) Regulation (EC) No 2160/2003 provides for a Community target is to be established for the reduction of the prevalence of all salmonella serotypes with public health significance in laying hens of *Gallus gallus* at the level of primary production. Such reduction is important in view of the strict measures which are to apply to infected flocks in accordance with that Regulation (EC) No 2160/2003 from December 2009 on. In particular, eggs originating from flocks with unknown salmonella status, that are suspected of being infected or from infected flocks may be used for human consumption only if treated in a manner that guarantees the elimination of salmonella serotypes with public health significance in accordance with Community legislation on food hygiene.

(3) Regulation (EC) No 2160/2003 provides that the Community target is to include a numerical expression of the maximum percentage of epidemiological units remaining positive and/or the minimum percentage of reduction in the number of epidemiological units

remaining positive, the maximum time limit within which the target must be achieved and the definition of the testing schemes necessary to verify achievement of the target. It is also to include a definition, where relevant, of serotypes with public health significance.

(4) In order to set the Community target, comparable data on the prevalence of the concerned salmonella serotypes in laying hens of *Gallus gallus* in Member States have been collected in accordance with Commission Decision 2004/665/EC of 22 September 2004 concerning a baseline study on the prevalence of salmonella in laying flocks of *Gallus gallus*<sup>(2)</sup>.

(5) Regulation (EC) No 2160/2003 provides that for a transitional period of three years, the Community target for laying hens of *Gallus gallus* is to cover *Salmonella enteritidis* and *Salmonella typhimurium*.

(6) In order to verify achievement of the Community target, it is necessary to organise repeated sampling of flocks.

(7) In accordance with Article 15 of Regulation (EC) No 2160/2003, the European Food Safety Authority (EFSA) was consulted on the setting of the Community target for laying hens of *Gallus gallus*.

(8) Since the adoption of Commission Regulation (EC) No 1003/2005 of 30 June 2005 implementing Regulation (EC) No 2160/2003 as regards a Community target for the reduction of the prevalence of certain salmonella serotypes in breeding flocks of *Gallus gallus* and amending Regulation (EC) No 2160/2003, alternative analysis methods have been developed and validated. In addition salmonella strains detected in breeding flocks should be stored for future phage typing and antimicrobial susceptibility testing. Therefore Regulation (EC) No 1003/2005 should be amended accordingly.

(9) The measures provided for in this Regulation are in accordance with the opinion of the Standing Committee on the Food Chain and Animal Health,

<sup>(1)</sup> OJ L 325, 12.12.2003, p. 1. Regulation as amended by Commission Regulation (EC) No 1003/2005 (OJ L 170, 1.7.2005, p. 12).

<sup>(2)</sup> OJ L 303, 30.9.2004, p. 30.

HAS ADOPTED THIS REGULATION:

*Article 1*

**Community target**

1. The Community target referred to in Article 4(1) of Regulation (EC) No 2160/2003 for the reduction of *Salmonella enteritidis* and *Salmonella typhimurium* in adult laying hens of *Gallus gallus* (Community target) shall be as follows:

- (a) An annual minimum percentage of reduction of positive flocks of adult laying hens equal to at least:
- (i) 10 % if the prevalence in the preceding year was less than 10 %;
  - (ii) 20 % if the prevalence in the preceding year was between 10 and 19 %;
  - (iii) 30 % if the prevalence in the preceding year was between 20 and 39 %;
  - (iv) 40 % if the prevalence in the preceding year was 40 % or more;
- or;
- (b) a reduction of the maximum percentage to 2 % or less; however, for Member States with less than 50 flocks of adult laying hens, not more than one adult flock may remain positive.

The first target should be achieved in 2008 based on the monitoring starting in the beginning of that year. With regard to the target in 2008, the results of the baseline study as carried out pursuant to Article 1(1) of Decision 2004/665/EC shall be used as reference referred to in this Article.

2. The testing scheme to verify the progress on the achievement of the Community target is set out in the Annex.

The achievement shall be evaluated taking into account the results of three consecutive years.

When not described in the Annex, the technical specifications referred to in Article 5 of Commission Decision 2004/665/EC

shall be considered as recommendations for the implementation of this point in the national control programmes.

3. The Commission shall consider a review of the testing scheme in the Annex based on the experience gained during the first year of the control programme as referred to in Article 5(1) of Regulation (EC) No 2160/2003 (the national control programme).

*Article 2*

**Amendment to Regulation (EC) No 1003/2005**

In the Annex to Regulation (EC) No 1003/2005, the following points 3.4 and 3.5 are inserted:

*3.4. Alternative methods*

With regard to samples taken at the initiative of the operator, the methods of analysis provided for in Article 11 of Regulation (EC) No 882/2004 (\*), may be used instead of the methods for the preparation of samples, detection methods and serotyping provided for in point 3 of this Annex, if validated in accordance with EN/ISO 16140/2003.

*3.5. Storage of strains*

At least the strains isolated as part of the official controls, shall be stored for future phage typing or anti-microbial susceptibility testing, using the normal methods for culture collection, which must ensure the integrity of the strains for a minimum period of two years.

(\*) OJ L 191, 28.5.2004, p. 1.

*Article 3*

**Entry into force**

This Regulation shall enter into force on the third day following its publication in the *Official Journal of the European Union*.

It shall apply from 1 August 2006.

This Regulation shall be binding in its entirety and directly applicable in all Member States.

Done at Brussels, 31 July 2006.

For the Commission  
Markos KYPRIANOU  
Member of the Commission

## ANNEX

**Testing scheme necessary to verify the achievement of the Community target for the reduction of *Salmonella enteritidis* and *Salmonella typhimurium* in adult laying hens of *Gallus gallus*, as referred to in Article 1(2)****1. SAMPLING FRAME**

The sampling frame shall cover all flocks of adult laying hens of *Gallus gallus* (laying flocks) referred to in Article 1 of Regulation (EC) No 2160/2003.

**2. MONITORING IN LAYING FLOCKS****2.1. Frequency and status of sampling**

Laying flocks shall be sampled at the initiative of the food business operator (operator) and by the competent authority.

Sampling at the initiative of the operator shall take place at least every fifteen weeks. The first sampling shall take place at the age of  $24 \pm 2$  weeks.

Sampling by the competent authority shall take place at least:

- (a) in one flock per year per holding comprising at least 1 000 birds;
- (b) at the age of  $24 \pm 2$  weeks in laying flocks housed in buildings where salmonella was detected in the preceding flock;
- (c) in any case of suspicion of *Salmonella enteritidis* or *Salmonella typhimurium* infection, as a result of the epidemiological investigation of food-borne outbreaks in accordance with Article 8 of Directive 2003/99/EC of the European Parliament and of the Council<sup>(1)</sup>;
- (d) in all other laying flocks on the holding in case *Salmonella enteritidis* or *Salmonella typhimurium* are detected in one laying flock on the holding;
- (e) in cases where the competent authority considers it appropriate.

A sampling carried out by the competent authority may replace one sampling at the initiative of the operator.

**2.2. Sampling protocol**

In order to maximise sensitivity of sampling, both faecal material and the environment shall be sampled at least as provided for in (a) and (b):

- (a) In cage flocks,  $2 \times 150$  grams of naturally pooled faeces shall be taken from all belts or scrapers in the house after running the manure removal system; however, in the case of step cage houses without scrapers or belts  $2 \times 150$  grams of mixed fresh faeces must be collected from 60 different places beneath the cages in the dropping pits.
- (b) In barn or free-range houses, two pairs of boot swabs or socks be taken, without changing overboots between boot swabs.

In the case of sampling by the competent authority, 250 ml containing at least 100 gram of dust shall be collected from prolific sources of dust throughout the house. If there is not sufficient dust, an additional sample of 150 grams naturally pooled faeces or an additional pair of boot swabs or socks shall be taken.

<sup>(1)</sup> OJ L 325, 12.12.2003, p. 31.

In the case of sampling referred to in point 2.1(b), (c) and (d), the competent authority shall satisfy itself by conduction further tests as appropriate that the results of examinations for salmonella in birds are not affected by the use of antimicrobials in the flocks.

Where the presence of *Salmonella enteritidis* and *Salmonella typhimurium* is not detected but antimicrobials or bacterial growth inhibitory effect are it shall be accounted for as an infected laying flock for the purpose of the Community target referred to in Article 1(2).

### 3. EXAMINATION OF THE SAMPLES

#### 3.1. Transport and preparation of the samples

Samples shall be sent by express mail or courier to the laboratories referred to in Article 11 of Regulation (EC) No 2160/2003, on the day of collection. At the laboratory, samples shall be kept refrigerated until examination, which shall be carried out within 48 hours following receipt.

##### 3.1.1. Boot swab samples

- (a) The two pairs of boot swabs ('or socks') shall be carefully unpacked to avoid dislodging adherent faecal material, pooled and placed in 225 ml Buffered Peptone Water (BPW) which has been pre-warmed to room temperature;
- (b) The sample shall be swirled to fully saturate it and culture shall be continued by using the detection method in 3.2.

##### 3.1.2. Other faecal material and dust samples

- (a) The faeces samples shall be pooled and thoroughly mixed and a 25 gram sub-sample shall be collected for culture.
- (b) The 25 gram sub-sample shall be added to 225 ml of BPW which has been pre-warmed to room temperature.
- (c) Culture of the sample shall be continued by using the detection method in 3.2.

If ISO standards on the preparation of faeces for the detection of salmonella are agreed on, they shall be applied and replace the above provisions on sampling preparation.

#### 3.2. Detection method

The method recommended by the Community Reference Laboratory (CRL) for Salmonella in Bilthoven, the Netherlands, for detection shall be used. This method is described in the current version of draft Annex D of ISO 6579 (2002): 'Detection of *Salmonella* spp. in animal faeces and in samples of the primary production stage'. In this method, a semi-solid medium (modified semi-solid Rappaport-Vassiladis medium, MSRV) is used as the single selective enrichment medium.

#### 3.3. Serotyping

At least one isolate from each positive sample shall be serotyped, following the Kaufmann-White scheme.

#### 3.4. Alternative methods

With regard to samples taken at the initiative of the operator, the methods of analysis provided for in Article 11 of Regulation (EC) No 882/2004<sup>(1)</sup>, may be used instead of the methods for the preparation of samples, detection methods and serotyping provided for in point 3 of this Annex, if validated in accordance with EN/ISO 16140/2003.

#### 3.5. Storage of strains

At least the strains isolated from samples collected by the competent authority, shall be stored for future phage typing or anti-microbial susceptibility testing, using the normal methods for culture collection, which must ensure integrity of the strains for a minimum of two years.

<sup>(1)</sup> OJ L 191, 28.5.2004, p. 1.

#### 4. RESULTS AND REPORTING

A laying flock shall be considered positive for the purpose of verifying the achievement of the Community target, where the presence of *Salmonella enteritidis* and *Salmonella typhimurium* (other than vaccine strains) was detected in one or more samples in the laying flock. Positive laying flocks shall be counted only once, irrespective of the number of sampling and testing operations and only be reported in the first year of detection.

Reporting shall include:

- (a) the total number of flocks of laying hens tested and the number of laying flocks tested for each status of sampling referred to in point 2.1;
- (b) the total number of infected flocks and the results of the testing for each status of sampling referred to in point 2.1;
- (c) explanations on the results, in particular concerning exceptional cases.

The results referred to in this point and any additional relevant information shall be reported as part of the report on trends and sources provided for in Article 9(1) of Directive 2003/99/EC.

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THE UNIVERSITY OF CHICAGO

MEMORANDUM

TO : [Name]

FROM : [Name]

SUBJECT : [Topic]

- [Text]

[Main body of text, possibly a summary or detailed notes]

[Closing text, possibly a signature line or date]

GD - Animal Health Service Denter



## Zoonotic Salmonella in Dutch officiërs



[www.gdoverloos.com](http://www.gdoverloos.com)

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## Programme

- Part I: The root of all evil.
- Part II: The role of GD

### TOUR OF THE LAB

- Part III: Monitoring of zoonotic Salmonella

[www.gdoverloos.com](http://www.gdoverloos.com)

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## PART I: THE ROOT OF ALL EVIL



[www.gdoverloos.com](http://www.gdoverloos.com)

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# PART I: THE ROOT OF ALL EVIL

## Salmonella

"There are probably more people making a living out of Salmonella, than people dying because of it."

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# YOPI



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# Salmonella

- **>2500 types**
  - some host specific (human, animal)
  - many non-specific (potentially zoonotic)
- **Human disease**
  - in NL 50,000 / year
  - average 1 week

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### Clinical significance

**Mature birds:** seldom diseased, S.E. sometimes peritonitis, mortality

**Chicks:** dull, locomotion problems, sepsis, enlarged edematous gall bladder, poly-arthritis, omphalitis

**Salmonella strain differences**  
\* O-Ag variations  
\* plasmids



**Host-sensitivity differences**  
\* moulting, transportation  
\* coccidiosis  
\* Gumboro, CAV, ...

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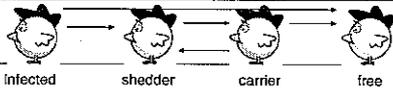
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### Salmonella

- **Vertical:** transovarial, contaminated eggs
- **Horizontal:**
  - \* the front door: men, equipment, other animals
  - \* the back door: rodents, insects, birds, other vermin
  - \* the side door: feed, water, air (dust/feces/feathers)



- Continuous re-infection of sensitive birds in flock,
- limited (low) percentage shedders

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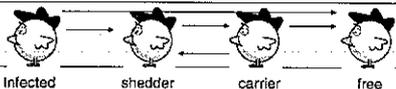
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### Salmonella




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### CASE-CONTROL STUDY OF SE RISK FACTORS

- Occurrence was explained for 30% by factors concerning:
  - preventive hygiene
  - surroundings of the farm
  - farm itself
- Other factors not included in the study may be important:
  - imported poultry introducing new infections
  - quality of intestinal flora, varies from farm to farm
  - all in all not important (majority of farms operate this system, therefore not relevant in this study)

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### Seven major risk factors associated with SE in broiler breeders

1. Breed A (selective breeding)
2. Flock size (stress favouring colonization)
3. Poultry house visits
4. Feed silo cleaning and disinfection
5. Farmyard disinfection
6. Industry related to animal products in surroundings (migration rodents, insects, by air, passing trucks, visits, etc.)
7. Animal farms in close proximity = 1 km

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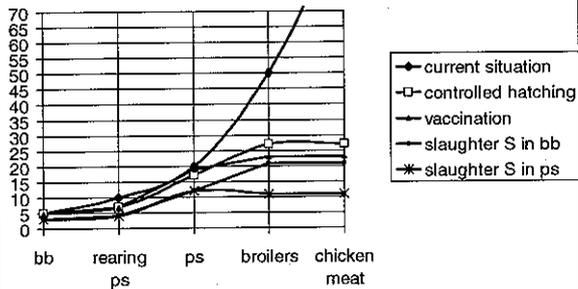
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with controlled hatching and vaccination (phase 1), controlled slaughtering (phase 2) and elimination from production (phase 3)




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### EU Order 2073/2005 (microbiological criteria on food)

Permanent: S. Enteritidis, S. Typhimurium,  
Variable: S. Hadar, S. Infantis, S. Virchow

- 2007 reproduction chicken
- 2008 commercial layers
- 2009 broilers
- 2010 turkeys

- Monitoring based on bacteriology only
- Implementation in NL: Action Plan Salmonella PPE

[www.gdseventer.com](http://www.gdseventer.com)

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## PARTNERHOOP OF GD



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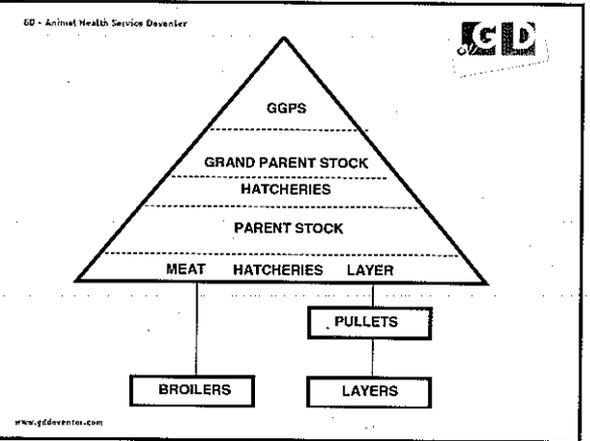
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### Dutch poultry industry

- Broiler breeders : 280 farms
- Broilers: 800 farms
- Layer breeders: 50 farms
- Layers 1300 farms
- Breeder turkeys 3 farms
- Meat turkeys 60 farms
- Duck farms 75 farms
- All private farmers!

(Source: Kwantitatief 1st Q 2009)

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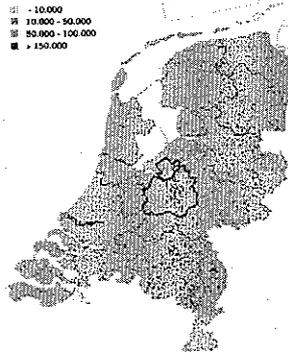
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### Dutch poultry industry

Total annual shots plumbeo per beest

- < 10.000
- 10.000 - 50.000
- 50.000 - 100.000
- > 150.000




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### Organizations

- EU REF LAB
- Min.ANF CVI
- PPE AHS (POULTRY DEP)
- INDUSTRY PRACTICIONERS

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## Organizations

- **Ministry of Agriculture, Nature and Food quality (MANF)**
  - Legal regulations (notifiable diseases)
    - National
    - EU
- **Product board for Poultry and Eggs (PPE)**
  - Promotion of Dutch poultry and eggs and -products
  - Private law organization
  - Identification and registration
  - License to produce
  - Self regulating according to the needs
  - Total surveillance of production

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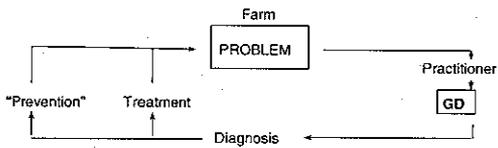
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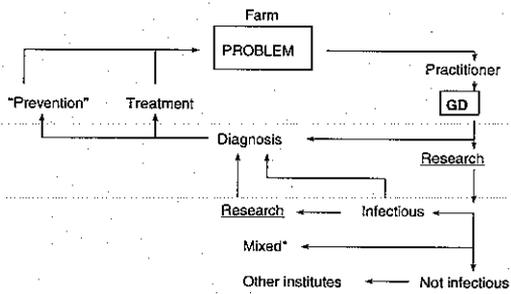
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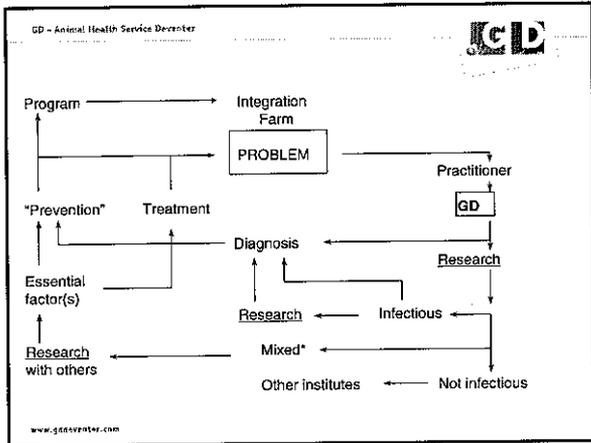
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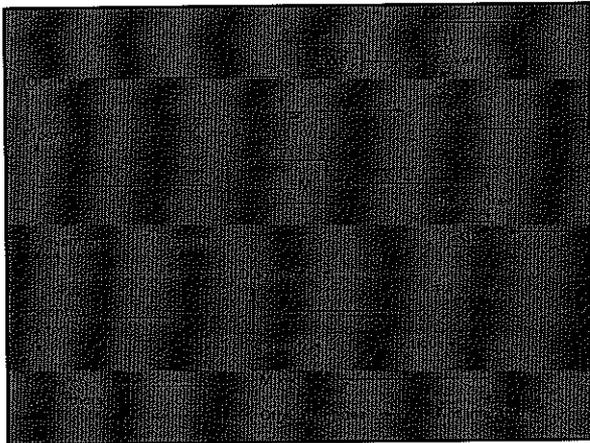
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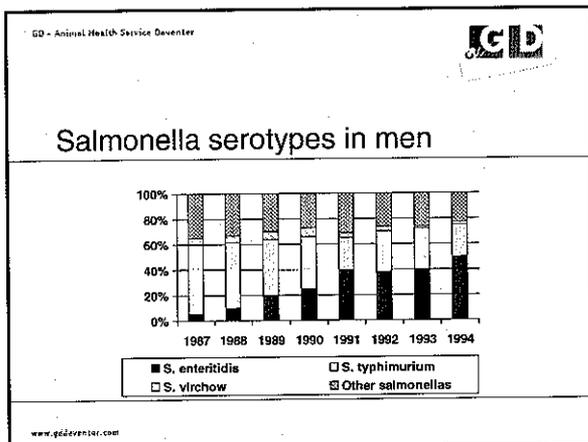
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### EU baselinstudy – commercial layers

Europe	30,7	20,3
Netherlands	15,8	7,9

[www.gdcentr.com](http://www.gdcentr.com)

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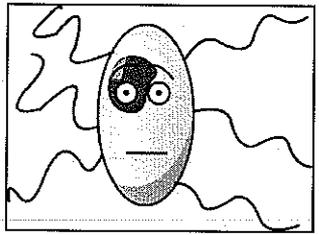
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### Questions?



[www.gdcentr.com](http://www.gdcentr.com)

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### PRATIEN VOOR NIEUW EN NIET-ENKEL



[www.gdcentr.com](http://www.gdcentr.com)

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2 parts:

- GD involvement
- The monitoring scheme

*Program*

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### GD involvement

Specific monitoring programs

1. Preparation
2. Execution
3. Surveillance, control
4. Adaptation

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### 1. Preparation

**Advices, textual suggestions**

- a. EU regulations
- b. NL regulations (MANF, PPE)
- c. GD planning and monitoring
- d. GD veterinary know-how

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### 1a. EU regulations

- The essential meaning
- **Unambiguity**
- Similarities and differences with other regulations
- Detailed SOP's or broadly outlined
- **Application in NL** (vaccination)

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### 1b. NL regulation (MANF, PPE)

- "Translation" (definitions, apprehensions)  
- genetically valuable or valuable genetic material
- Connection with other NL regulations
- Working details, SOP's
- **Combinations (in sampling)**

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### 1c. GD planning and monitoring

- Regulation per flock, house, farm
- Relation with type of animal/housing
- Planning on age/calendar/production stage
- Combination of program's in 1 scheme per animal type / farm / visit

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### 1d. GD veterinary know-how

- Which test(s) (specificity, sensitivity)
- Kind of samples (immun., bact., PCR)
- **Number of samples** (representativity)
- Frequency of testing
- Screening (routine)/verification (suspicion)
- Consequences result, black/white or gradual

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### Number of samples

Shedder prevalence	Confidence level		
	99%	95%	90%
0.5%	840	564	
1%	440	290	
2%	225	147	90
5%	92	59	45
10%	46	29	22

- Flocks > 1000: 95% confidence level, at 1% prevalence 300 samples

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### 2. Execution, planning

- Flock / farm (based on "KIP")
- Partition may be relevant, but (yet) unknown
- Not every removal gives a (whole) new flock
- Planning at age / period / otherwise
- Samples taken by owner / VP / GD
- Addressing (instructions, test results, bill)
- Relation with (previous) test results

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## 2. Execution, laboratory

- Examination feces / blood
- Judgment of result by GD vet, before sending report, a.o. regarding
  - vaccination
  - verification
  - consequences
- Communication of result, if verification also directly to PPE

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## 3. Surveillance

- Reporting
  - general / exceptions
- Actions on "errors"
  - criteria: what is an error, in relation with animal type (ND), housing (AI)
  - when to check (end laying period)

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## 4. Adaptation

- In consultation with responsible authority
- Based on experiences with execution, test results, (re)actions at errors

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### The monitoring scheme

- But first some considerations:
  - Vaccination
  - Sampling methods

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### SALMONELLA VACCINATION

- Vaccines, specific use in NL
- (G)GPS : no vaccination, regarding claims of "reduction" only
- Parents : killed SE/ST, specific? sero-pos, maternal protection
- Layers : live SE/ST, group-specific? <50% sero-pos  
: live SG, group-specific SE/ST sero-neg (test based on g,m/l)

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### Sampling methods

- Purpose defines which detection method to follow
- Detection method defines what kind of samples to examine
- Kind of samples defines sampling method

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### Purpose of testing

What is the reason for the examination?

Should all Salmonella's be detected  
or only specific serotypes  
or antibodies due to infection

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### Detection methods

#### Bacteriological examination

##### Advantages

- direct information about presence of Salmonella
- applicable for many types of sample material

##### Disadvantages

- only samples from inside the birds give reliable information about actual infection
- excretion in feces is irregular, percentage positive samples is rather low

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### Detection methods

#### Serological examination :

##### Advantages

- sensitive and specific
- quick result
- continuously positive: less samples needed

##### Disadvantages

- for invasive strains (SE and ST) only, others do not stimulate antibody production
- indirect information:
  - “false” positive; old inf, “false” negative; early inf
- Y**- may be positive after vaccination
- not for samples from outside the birds

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## Sample material

### Bacteriological examination

- Feces (swabs, caecal droppings, overshoes)
- Cloaca swabs (individual, not environment)
- Hatch material (fluff, dead-in-shells, non vital chicks)

### Serological examination

- Blood serum

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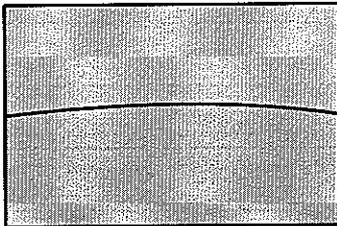
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## The environment



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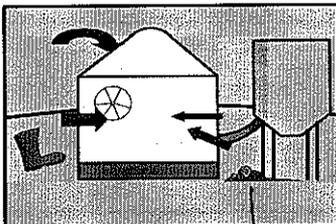
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## ..and into the barn



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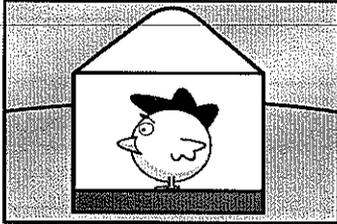
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So what happens now?



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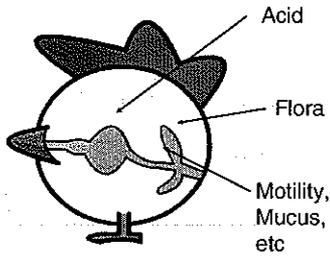
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The chicken



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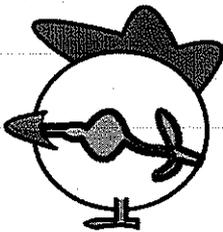
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The chicken



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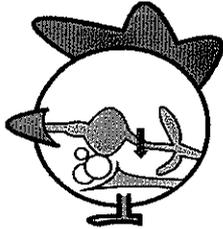
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### The chicken, laying hens

From gut to egg:

- Intracellular, bloodstream
- Ascending, cloaca
- Feces, eggshell




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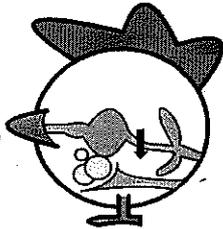
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### The chicken, laying hens

Favorite place for SE  
In the repro tract:  
Isthmus

But can be found in  
all locations along the  
reproductive tract




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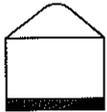
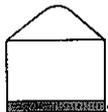
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### Possible outcomes




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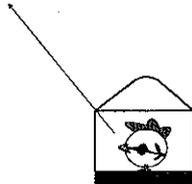
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### Possible outcomes

- Highest risk for vertical transmission



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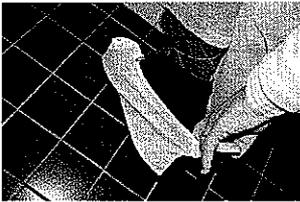
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### Current situation

- Basic monitoring based on overshoe



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### Current situation

- Monitoring based on overshoe and direct sampling of caecal feces

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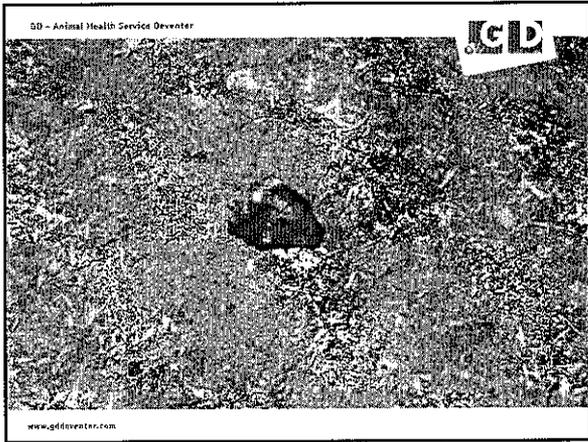
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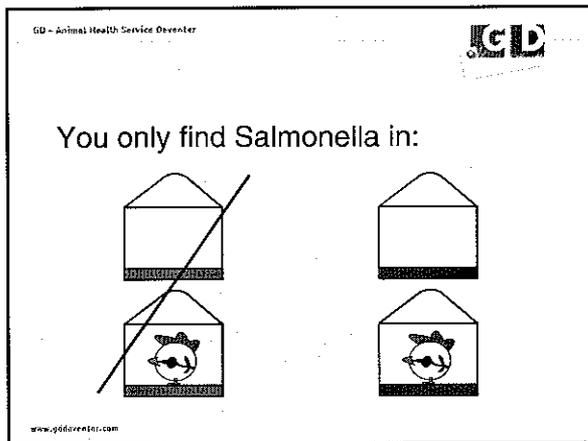
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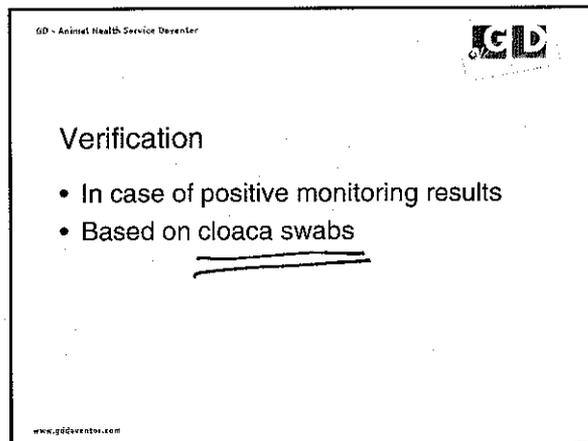
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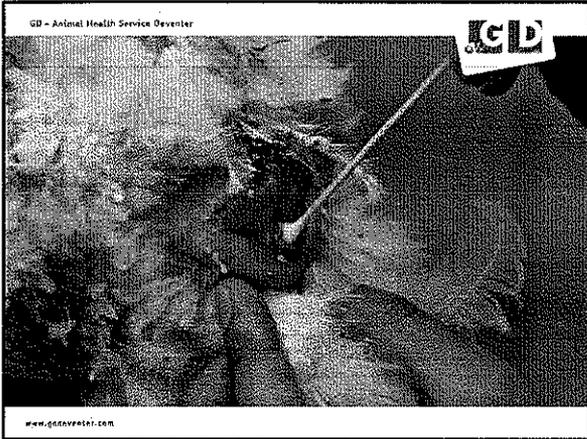
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GD - Animal Health Service Deventer



Combining these two methods,  
you only find Salmonella in:



www.gdvet.nl

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GD - Animal Health Service Deventer



The monitoring scheme

- See little computer program on your DVD

www.gdvet.nl

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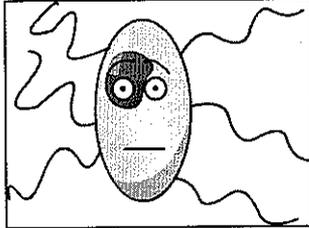
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Questions?



[www.g4e.nl](http://www.g4e.nl)

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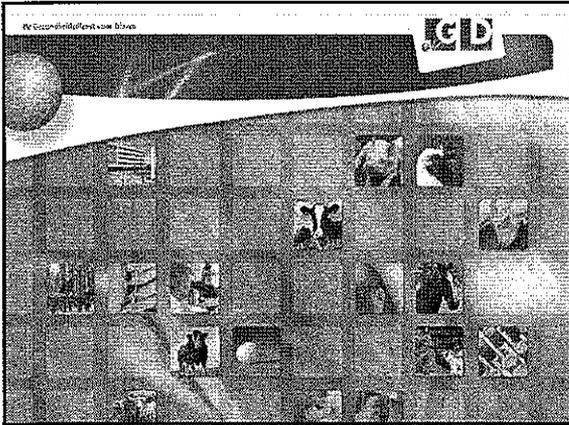
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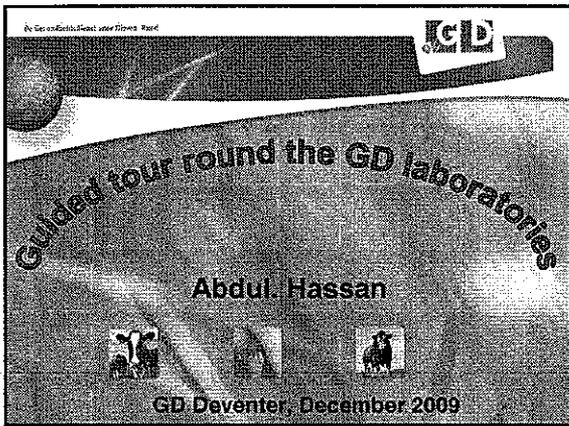
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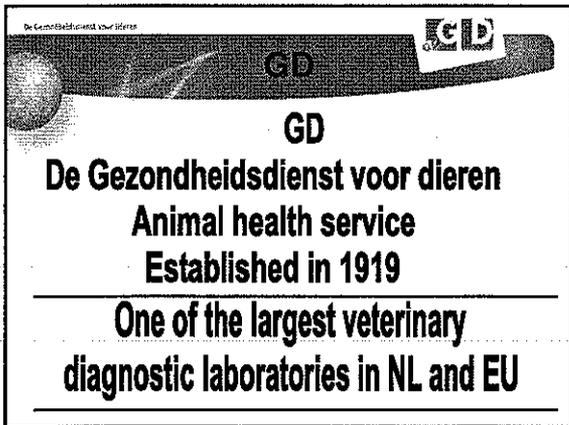
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De Gezondheidsdienst voor Dieren

**Quality system in GD** 

**GD certified according to quality system  
ISO 9001 as well as ISO 17025**

**Accreditation**  
**GD laboratories are accredited by  
The Dutch Accreditation Council (RVA)  
since 1994**

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De Gezondheidsdienst voor Dieren

**General informations** 

- **400 employees (130 employees in Lab., 40 specialised veterinarians, 20 researchers)**
- **>250 different tests**
- **3.5 - 4 million samples per year**
- **15.000-20.000 samples investigate daily**

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De Gezondheidsdienst voor Dieren

**GD Cooperations** 

**Cooperated with other organizations in NL**

- **Central Veterinary Institute (CVI)**
- **Utrecht University**
- **The National Institute for Public Health and the Environment (RIVM)**

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GD Cooperations

- Cooperation with other countries  
Russia, Indonesia, France, Germany
- Cooperation with other Laboratories
  - Qlip (mastitis)
  - van Haeringen (PCR and Molecular)
  - KBBL (food microbiology)
  - Böse Laboratory, Germany (special laboratory for horses)

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GD in practice

Service provider

Researcher

Partner

Advice/consultant

Provide creative/innovative solution

Entrepreneur

Developer

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GD Organization

Animals Dep.

Marketing New Business

Manag. offices

Operation

Facilities service

Planning Strategie

Info. & Info. Technol.

DRE

CLA and Labs

Quality & Enviro

LIMS and COS

Immunology

Molecular biology/Virology

Vet. Chemistry/Toxicology/fertility

Bacteriology and Parasitology

Pathology/Histology

Pigs & poultry

Humans

Per animals

Horses

Wages office

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De Gezondheidsdienst voor Dieren

**GLD**



**CLA and Laboratories**

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De Gezondheidsdienst voor Dieren

**GLD**

**Central Laboratory Administration (CLA)**

- working more than 15 employees
- This section arrival all the samples from farmers or veterinarians
- 20.000 samples daily, 3.5-4 million samples annually
- Most of these samples (3 millions) for serological test screening of important diseases in NL
- All the samples obtaining the code/ number using the LIMS program
- Blood samples centrifuged for serological test and moved forward to the immunology lab
- Others samples sendt to different labs according to the type of examinations

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De Gezondheidsdienst voor Dieren

**GLD**

**Immunology Laboratory**

- About 3 million samples determined annually
- Large health screening and monitoring program for NL farms
- Give health certificate to these farms

**Important tests**

- Rapid plate agglutination test (e.g. Mycoplasma)
- Agar gel Immuno diffusion test
- Haemagglutination test
- Enzyme Linked Immuno Sorbent Assay (ELISA)

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De Gezondheidsdienst voor Dieren

**Important diseases determined in Immunology Lab**

**Cattle:**

- BVD (Bovine Virus Diarrhea)
- IBR (Infectious bovine rhinotracheitis)
- Salmonella
- Paratb

**Pigs:**

- AD (Aujeszky's disease)
- Salmonella
- SVD (Swine Vesicular Disease)

**Poultry:**

- Mycoplasma
- NCD
- Avian influenza
- Gumburo disease

**Horses:**

- Equine influenza
- Equine herpesvirus




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De Gezondheidsdienst voor Dieren

**Molecular Biology and Virology Laboratory**

- Using traditional PCR and Real-Time-PCR Techniques
- Examination about 170.000 PCR samples as individual and pooled tests
- Examination about 3500 viral samples using tissue cultural method, IPMA (Immunoperoxidase Monolayer Assay), VNT (Virus Neutralization Test) and SNT (Serum Neutralization Test)




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De Gezondheidsdienst voor Dieren

**Important diseases determined in Molecular Biology/Virology Laboratory**

- Screening of paratb (Johne's disease)
- BVD (Bovine Virus Diarrhea)
- PRRS (Porcine reproductive and respiratory syndrome)
- AR (Atrophic rhinitis)
- Mycoplasma gallisepticum; Mycoplasma synoviae
- Avian influenza




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De Gezondheidsdienst voor Dieren 

**Poultry section**

**Bacteriological examination of the PM samples**  
(*Salmonella Gallinarum*, *Enterococcus spp.*, *E coli*,  
..... etc)

**Poultry projects e.g. Enterococcus species**  
(*E. Cecorum*, *E. durans* and *E. hirae* ....etc)

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De Gezondheidsdienst voor Dieren 

**Pathology Laboratory**

- National courier service
- Pathological examination (PM of 5000 animals annually)
- Histological investigation
- Tumor diagnostic (5000-6000 samples annually)
- Poultry examinations (2000 cases annually)

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De Gezondheidsdienst voor Dieren 

**Veterinary Chemistry and Toxicology Laboratory**

- Clinical chemistry and hematological tests (15,000 clinical samples annually)
- Fertility examination (horses)
- water examination (7000 samples annually)
- Determination of free fatty acid patterns (omega-3) in milk samples (4000-5000 samples annually)

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De Landbouwkundige Dienst

**Veterinary Chemistry  
and Toxicology Laboratory**



- **Residue determination: analysis of prohibited substances such as increase hormones and  $\beta$ -agonists in feed, urine and water**
- **GD is the only laboratory in the Netherlands for Vet. Toxicology analysed the heavy metal, pesticides and toxic plants**

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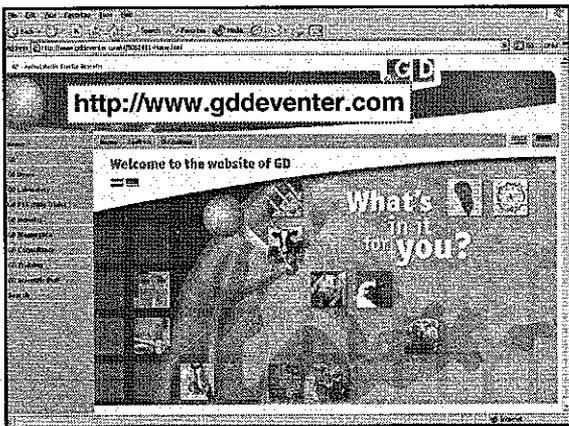
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http://www.gddeventer.com

Welcome to the website of GD

What's that for you?

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De Landbouwkundige Dienst



**THANK YOU FOR ATTENTION**

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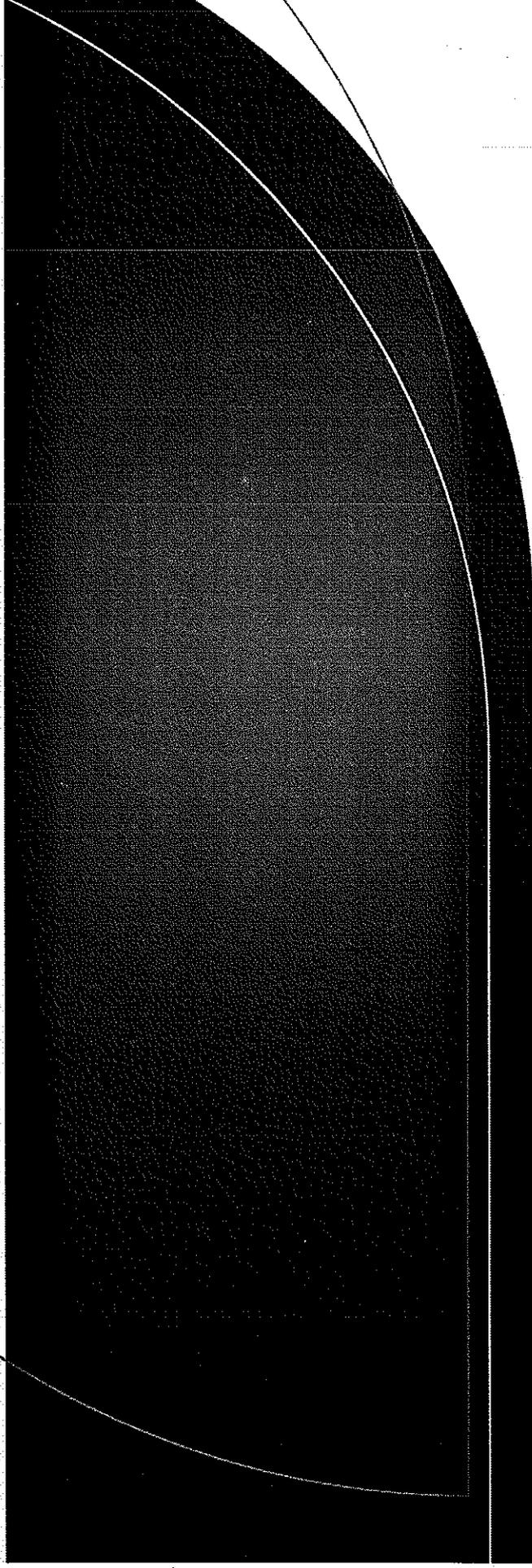
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# **Salmonella live vaccines**

**Dr. Sarah Schriek**

# 1. Salmonella – the problem



## Taxonomy

Group: gram-negative bacteria

Family: Enterobacteriaceae

Genus: *Salmonella*

Species: *Salmonella enterica*

Subspecies: *Salmonella enterica* spp. *enterica*

Serovar: more than 1400 known

## **Salmonella in chickens – zoonotic agents**

- *Salmonella* Typhimurium
  - rare cases of problems in young chicks
  - But impact as zoonotic agent –  
mainly meat related transmission but also eggs
- *Salmonella* Enteritidis
  - Rare cases of disease in young chicks
  - But major impact as zoonotic agent
  - Meat and egg related transmission
    - SE readily colonises and persists in ovaries;
    - shedded *in* and *on* eggs

## **Salmonella as zoonosis in man (1)**

- Infection through consumption / ingestion of contaminated food
- Clinically from asymptomatic to severe diarrhoea and nausea
- Confined to gut as gastro-enteritis
- Focal infection in any organ of the body
- Cases of death

## **Salmonella as zoonosis in man (2)**

- Severity depending on dose ingested and on constitution/susceptibility of individual person:
    - ~ 1 mio bacteria can cause disease in healthy person
    - ~ 1000 bacteria in sensitive person
- Salmonella control is a numbers game...

## **A PROBLEM IS:**

- food cannot always be absolutely "STERILE"
- improper handling of food / poor kitchen hygiene
- interruption of cooling chain
- rapid multiplication at room temperature
- Initially low number of bacteria may increase to a number that poses a health risk

## **THE AIM MUST BE:**

- To prevent colonisation in the first place as early and as effectively as possible!
- To subsequently reduce shedding and spreading
- To consolidate Salmonella free steps in food production and processing
- To reduce initial Salmonella load at any step of food production and processing



## 2. Salmonella – why vaccinate?

## Why vaccinate?

- Monitoring and culling of infected flocks
- adequate for *S. Gallinarum* – host specific / unless endemic
- adequate (?) for valuable genetic breeding stock
- not adequate for *S. Enteritidis* and *S. Thyphimurium* - broad host range/reservoir
- not adequate for layers – compensation???
- complex strategy needed to reduce overall *S.*-input

## **Why vaccinate?**

Treatment of infected animals with antibiotics

- expensive
- recurrence of disease after end of treatment
- withdrawal period
- residues in meat and eggs
- promotion of new resistances

## **Live Salmonella Vaccines**

### **Mode of action**

- early colonisation of the intestine – after hatch
  - development of intestinal immunity – IgA
  - transient infection of organs (liver)
  - development of cell mediated immunity
  - in case of challenge/natural infection
    - reduced colonisation density -> 2 log 10
    - reduced shedding period of field strain
    - reduced mortality in young chickens
- ⇒ **LESS** animals shed
- ⇒ **LESS** bacteria for a
- ⇒ **SHORTER** period of time

## **The European Food Safety Agency states:**

“Beside the microbiological colonisation-inhibition effect, the presence in the intestine of large numbers of live *Salmonella*-derived vaccine bacteria early post-hatch can induce infiltration of immune cells into the intestinal mucosa and sub-mucosa, which confers resistance to invasion and systemic spread by virulent *Salmonella* strains and, in mammals, gastro-enteritis (Van Immerseel *et al.*, 2002b).

This opens new perspectives for vaccine usage in broilers, layers and breeders.”

*The EFSA Journal* (2004)  
114 1-74, The use of  
vaccines for the control of  
*Salmonella* in poultry.

## **Why go live?**

“The development of efficacious live vaccines has been acknowledged by the WHO as one part of an overall control strategy to contain Salmonellosis in food animals.” Cooper et al Infect and Imm vol 62 no.11 pages 4747-4754.

But why is this the WHO view?

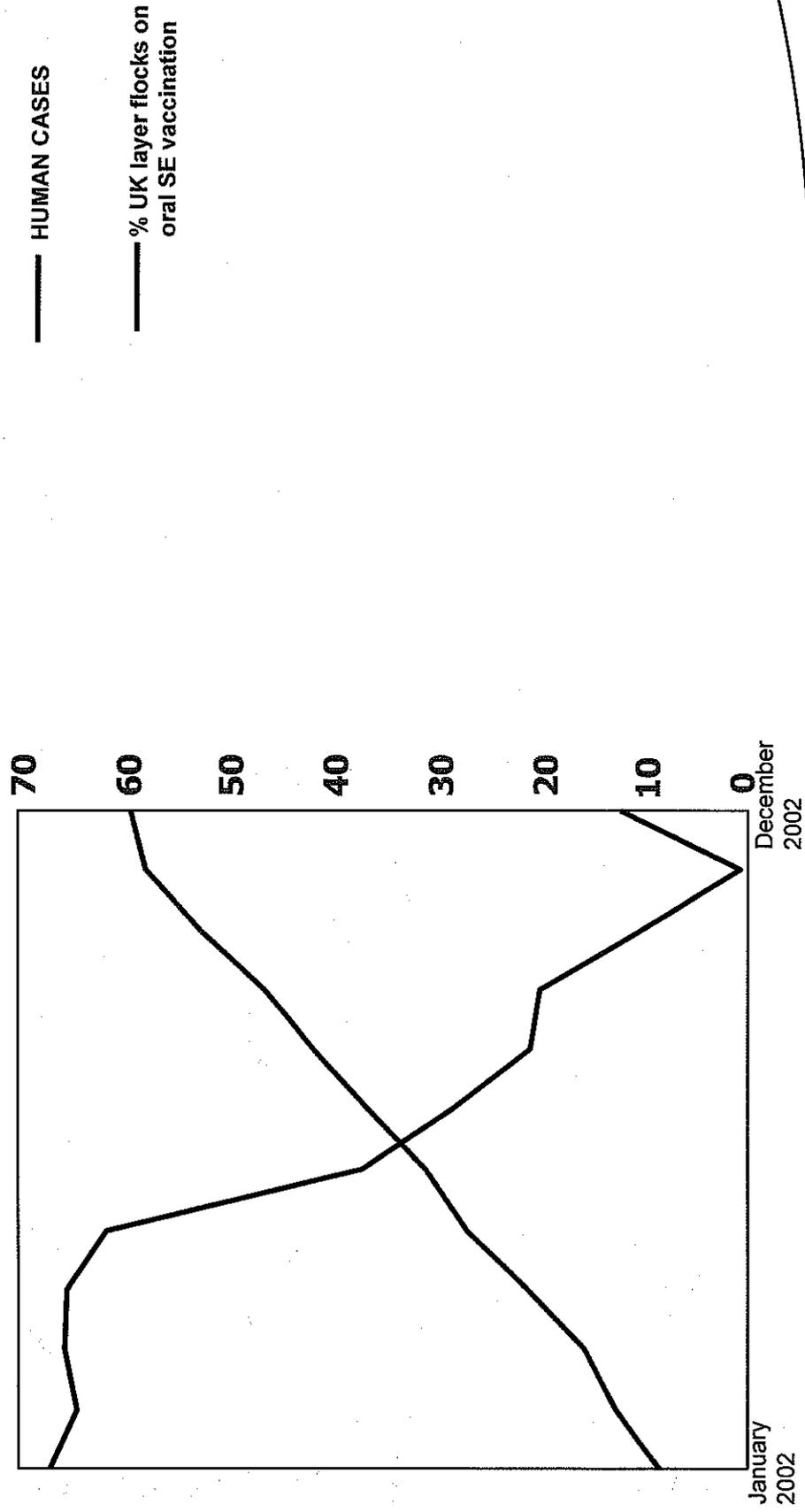
## **For more than half a century..**

live vaccines have been shown to be more effective.

“The Salmonella vaccine strain protected against challenge. These results offered a striking contrast to the failure of killed vaccines to protect, even against a very low challenge dose (Robson et al 1972).”

“Live attenuated vaccines against Salmonellosis have been shown many times to be more effective than killed bacterins in limiting mortality and faecal excretion”(Smith 1956, Collins 1972, Robertson 1983, Barrow 1990,1991).

### Comparison: Human salmonella cases (2002 vs 2001) with the up take of oral SE vaccination of layers in UK



### 3. Salmonella – The vaccines

## AViPro® SALMONELLA VAC E - Ingredients

- *Salmonella* Enteritidis vaccine, live, lyophilized

### Active pharmaceutical ingredient

*Salmonella* Enteritidis       $1 \times 10^8$  to  $6 \times 10^8$   
Sm24/Rif12/Ssq              cfu/dose

### Other ingredients

Peptone

Sucrose

Gelatine

HEPES-buffer



## AVIPro® SALMONELLA VAC E – Application and benefit

- utilization in layer and breeder chicken from first day of age
- application via the drinking water
- active immunization of chicken against *Salmonella* Enteritidis infection
- reduction of the number of birds shedding *Salmonella* Enteritidis-field strains after infection



### Vaccination scheme

- |                |                      |
|----------------|----------------------|
| 1. Vaccination | 1. Day of life       |
| 2. Vaccination | 6.-8. Week of life   |
| 3. Vaccination | 16.-18. Week of life |

## AViPro® SALMONELLA VAC T - Ingredients

- *Salmonella* Typhimurium vaccine, live, lyophilized

### Active pharmaceutical ingredient

*Salmonella* Typhimurium  
Na12/Rif9/Rtt      1 x 10<sup>8</sup> to 6 x 10<sup>8</sup>  
cfu/dose

### Sonstige Bestandteile

Milk powder

Sucrose

Gelatine

HEPES buffer



## AVIPro® SALMONELLA VAC T – Application and benefit

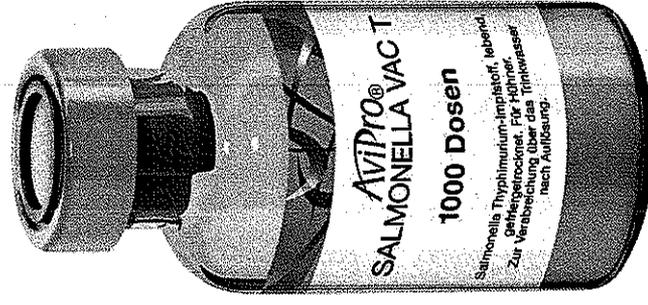
- for utilization in chicken (breeder, layer, broiler) from first day of life
- application via the drinking water
- active immunization of chicken
- protection against mortality induced by infection with *Salmonella* Typhimurium and reduction of colonization and shedding of *Salmonella* Typhimurium field strains
- immunity lasts for at least 6 weeks in broilers and for at least 50 weeks after third vaccination in breeders and layers

### Vaccination scheme breeder and layer

- |                |                  |
|----------------|------------------|
| 1. Vaccination | 1. Day of life   |
| 2. Vaccination | 7. Week of life  |
| 3. Vaccination | 16. Week of life |

### Vaccination scheme broiler

- |        |                           |
|--------|---------------------------|
| 1 dose | From first day of life on |
|--------|---------------------------|



## Presentations

**AviPro SALMONELLA VAC E    AviPro SALMONELLA VAC T**

1 x 1000 Doses

1 x 2000 Doses

1 x 2000 Doses

1 x 5000 Doses

10 x 1000 Doses

10 x 2000 Doses

10 x 2000 Doses

10 x 5000 Doses

## Efficacy of AviPro® Salmonella Vac E

- Layers (2 groups, 36 birds per group)
- Challenge: 52 weeks of life ( $1,0 \times 10^{10}$  CFU/bird, orally)

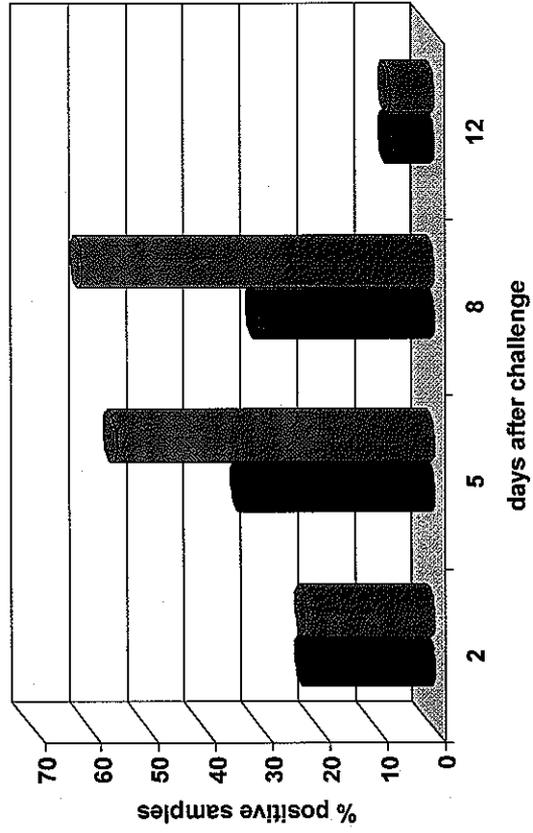
	Group I	Group II
	vaccinated	control
1. day of life	AviPro® Salmonella Vac E	
7. week of life	AviPro® Salmonella Vac E	
16. week of life	AviPro® Salmonella Vac E	

Trial 250/93

## Duration of immunity

■ Vaccinated ■ Control

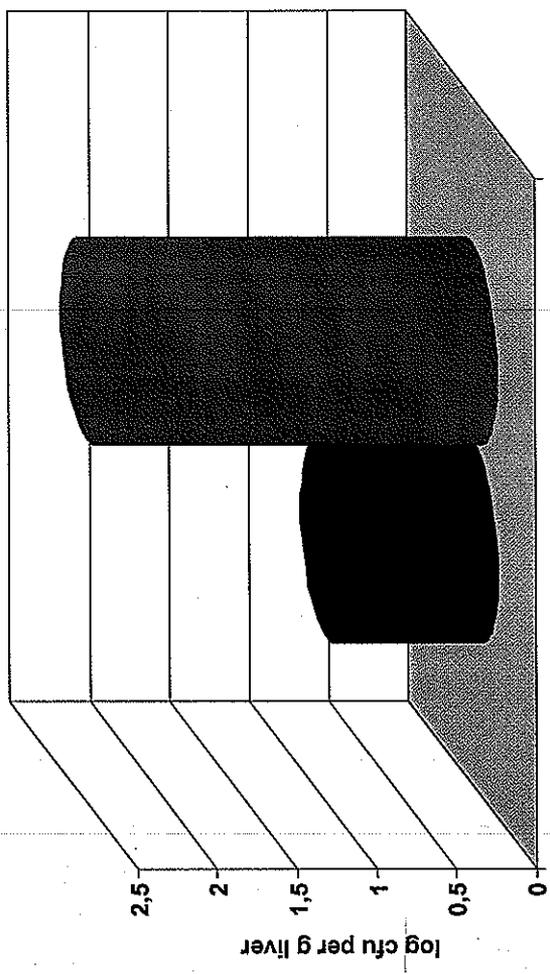
Excretion of challenge strain via  
cloacal swabs at 2, 5, 8, 12 day p.c.



## Duration of immunity

■ Vaccinated ■ Control

Persistence of the challenge strain  
in the liver 5 days post challenge



## Summary

- Vaccination with AviPro<sup>®</sup> Salmonella Vac E leads to a strong reduction in shedding of the challenge strain
- 5 days past challenge infection a significant reduction in organ colonization by the challenge strain in vaccinated birds as compared to unvaccinated birds can be seen

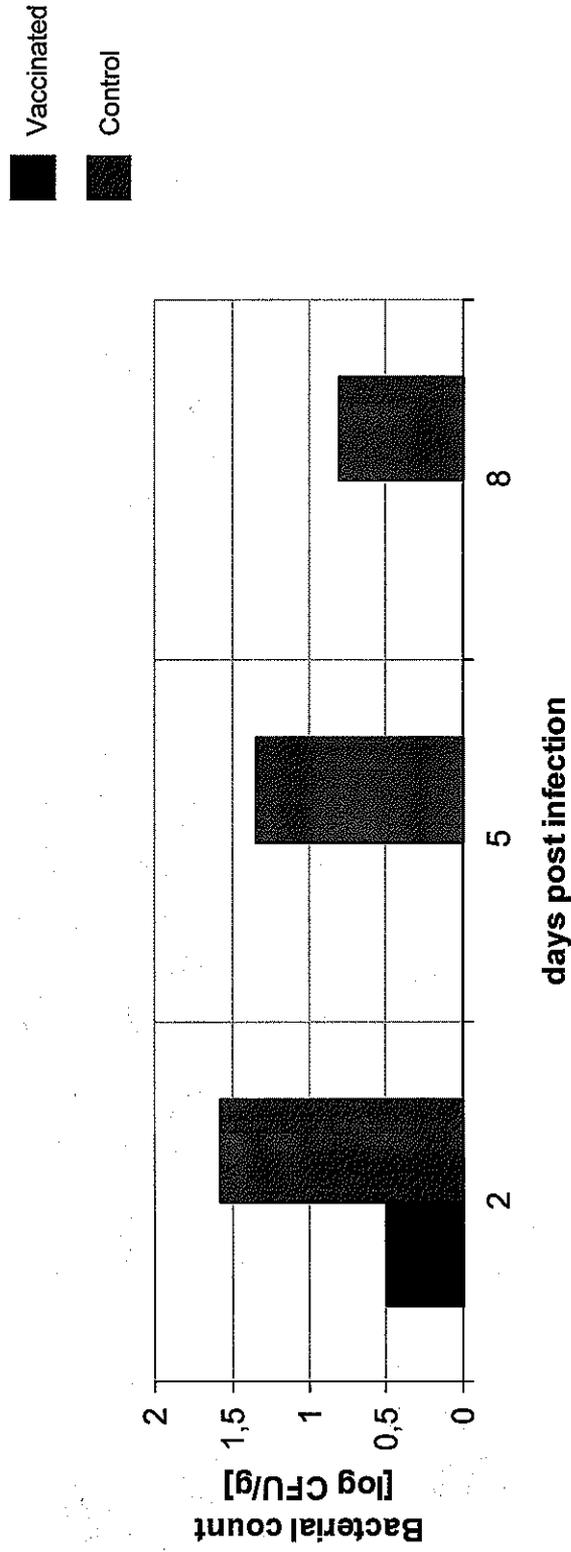
## Efficacy of AviPro® Salmonella Vac T

- Layers (two groups, 24 birds each)
- Challenge: 50 weeks of life (1,8 x 10<sup>10</sup> CFU/bird, orally)

	Group I	Group II
	vaccinated	control
1. day of life	AviPro® Salmonella Vac T	
7. week of life	AviPro® Salmonella Vac T	
16. week of life	AviPro® Salmonella Vac T	

Trial 250/93

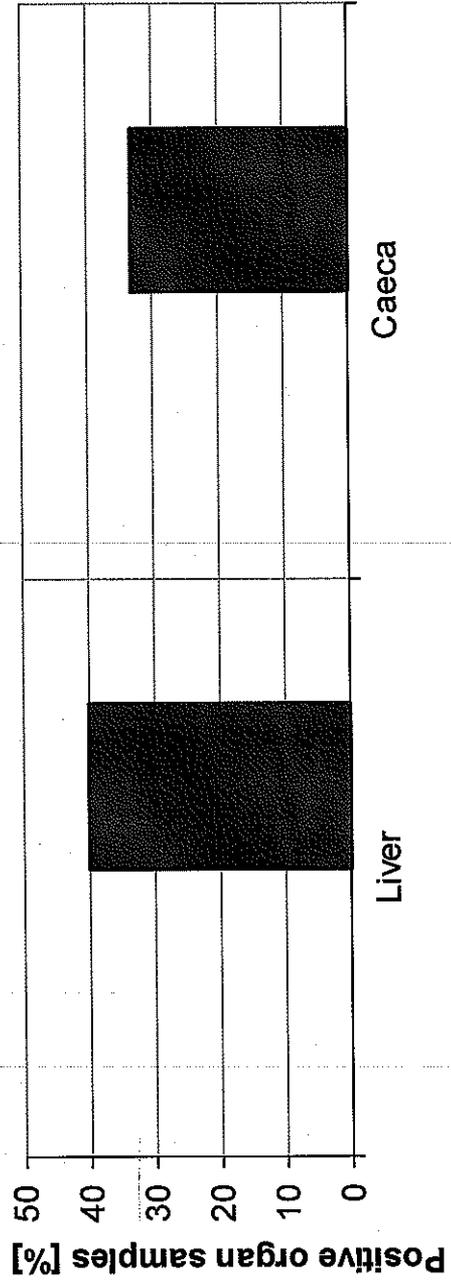
## Efficacy of AviPro® Salmonella Vac T Persistence of the challenge strain in the liver



Trial 250/93

## Efficacy of AviPro® Salmonella Vac T Reisolation of the challenge strain in organs 14 days post infection

■ Vaccinated  
■ Control



Trial 250/93

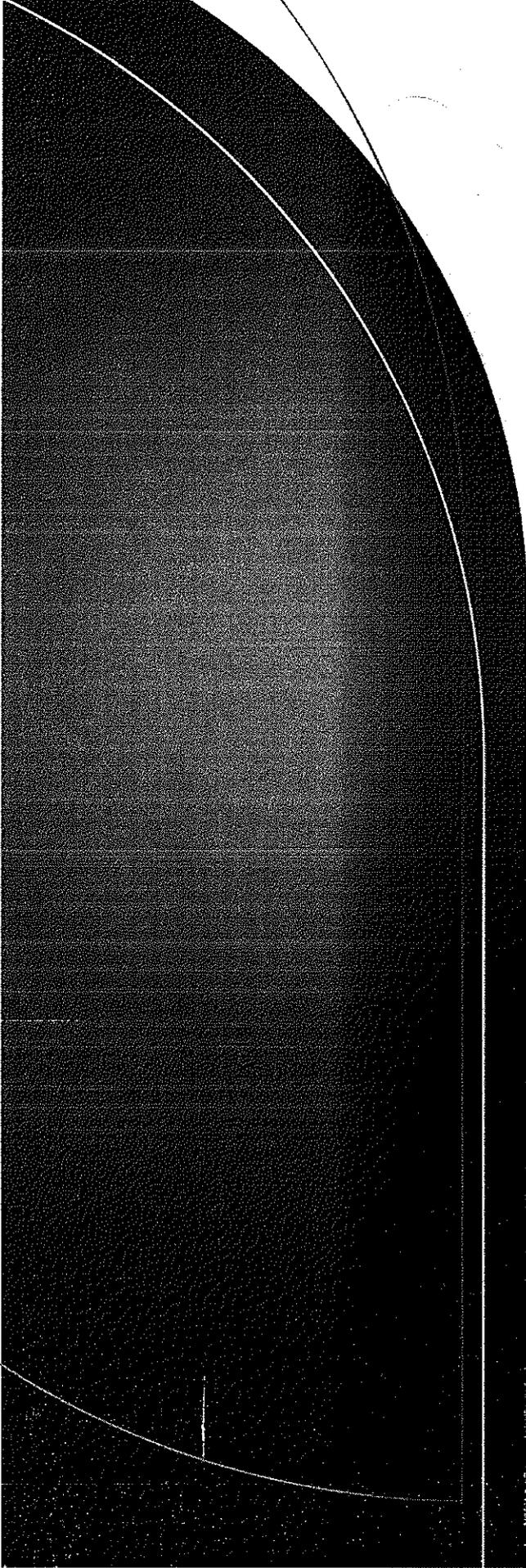
## Summary

- Vaccination with AviPro® Salmonella Vac T leads to a strong reduction in shedding of the challenge strain
- 14 days post infection liver and caeca of vaccinated birds are free of the Salmonella challenge strain, while the internal organs of unvaccinated birds are still colonized by the challenge strain

## Facts

- more than 15 years of experience in producing Salmonella live vaccines
- registrations in more than 45 countries around the world
- more than 3.5 billion doses sold so far
- vaccine strain was never found in humans





**Thank you very much for your attention!**