4rd FAO-APHCA/OIE/DLD Regional Workshop on Brucellosis Diagnosis and Control in Asia-Pacific Region "Proficiency Test and Way Forward" 19-21 Mar, 2014.





Brucellosis- Indian Scenario



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INDIA

- ✓ India is a country of sub-continental proportions
- ✓ Total area 3.29 million sq. kms.
 - (2.4% area of world holding 16.7% human population)
- ✓ 29 States and 7 Union territories
- ✓ Population = 1,270,272,105 persons.
- ✓ Sex ratio = 940 females Vs 1000 males



Livestock Wealth of INDIA

Total Livestock population522.7 million

- ✓ Largest buffalo population in the world (105.34 million- 57.3%)
- ✓ 2nd largest cattle population
- ✓ 2nd largest goat population
- ✓ 3rd largest sheep population
- ✓ 5th largest Poultry population
- ✓ 6th largest camel population
- ✓ Canine population

- (199.08 million- 14.7%)
- (140.54 million- 16.7%)
- (71.56 million- 12.4%)
- (648.88 million- 24.7%)
- (0.52 million- 29.31%)
- (19.9 million)



Contribution to Animal Products

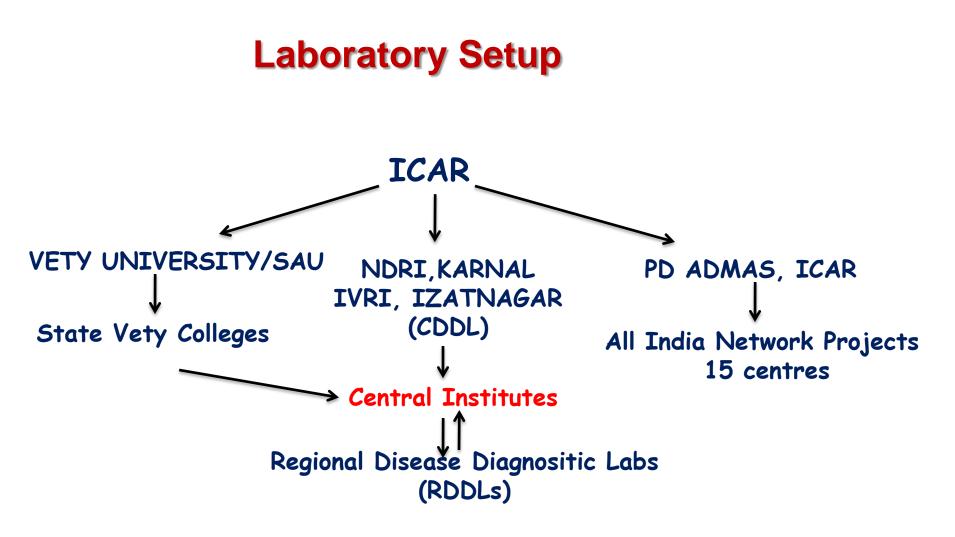
- ✓ Largest producer of milk in the world (121.8 million tons)
- ✓ 3rd largest producer of Chicken eggs (66 billion)
- ✓4th highest poultry meat producer (3.4 million tons)



Veterinary Services in India (Federal Government)

- At the Federal (Central) level, a Central Cabinet Minister heads Ministry of Agriculture having control of three Departments :
- Department of Animal Husbandry, Dairying and Fisheries (DADF)
- Department of Agriculture and Cooperation (DOAC)
- Department of Agricultural Research & Education (DARE)







Location of Regional Disease Diagnostic Laboratories in India





Brucellosis Situation in Animals

- Seroprevience of brucellosis in goats and humans in Bikaner distt of Rajasthan reveals 11.45% prevalence in milch goats and 2.97% in humans.(Kapoor *et al* 1985).
- Serological survey of brucellosis was performed in 23 states of India. A total of 30,437 bovine samples were screened with RBPT and STAT which reveals 1.9% prevalence in cattle and 1.8% in buffaloes .(Isloor et al 1998).
- PD-ADMAS conducted long-term serological studies which indicated 5% of cattle and 3% of buffaloes are infected with brucellosis.(Rajasekhar et al 2002).
- Sero-prevalence of brucellosis in Mithun (*Bos frontalis*) in Nagaland state by ELISA, STAT and RBPT is 34%, 20% and 11% respectively (Rajkhowa *et al* 2004).
- Serological study of *B.melitensis* with RBPT, STAT and ELISA reveals incidence of 13.85%,9.96% and 20.35% in Tamil Nadu State (Maher Sulima *et al* 2006).
- Prevalence rate for brucellosis was 8.58% in cattle, 8.85% in goat and 7.08% in sheep from the states of Rajasthan and Bihar.(Singh, 2007).
- Prevalence was 40.4% in bovines by AB-ELISA in Maharashtra State. (Lodhe *et al* 2011).



Brucellosis Situation in Animals....

In Punjab State

Cattle and Buffaloes

- The Avidin biotin ELISA in sera samples from 52 villages of Punjab state revealed prevalence in Buffaloes was 13.4% and in Cattle was 9.9% (Dhand et al 2005).
- Milk-ELISA in 345 animals from 32 villages, 18.26% of animals were found positive (Aulakh *et al* 2008).

Caprines

Dot-ELISA and STAT, 4.0% were found positive (Singh et al 2000).



Sero-prevalence of Brucellosis in Punjab State GADVASU, Ludhiana

Year	Suspected Sample Tested	Positive By RBPT	% Positive
2007-08	1603	420	26.20
2008-09	2961	829	27.90
2009-10	2588	829	32.03
2010-11	2689	775	28.82
2011-12	1064	191	17.80
2012-13	662	95	14.32

Annual reports-ADRC



Sero-prevalence of Brucellosis in Northern States

Year	Random Sample Tested	Positive	%
2003-04	19692	1615	8.20
2004-05	14968	564	3.70
2005-06	16806	122	1.10
2006-07	13906	431	3.09
2008-09	1861	163	8.76

(Annual reports-NRDDL)



Brucellosis in Western States

State	Sample tested	Positive	% Positive
Gujarat	260	82	31.54
Maharastra	941	328	34.86
Rajasthan	39	5	12.82
Madhya Pradesh	2795	575	20.57
Uttar Pradesh	712	294	41.29

Brucellosis in Eastern States					
Manipur	700	2	0.28%		
Assam	395	0	-		
Mizoram	676	38	5.62%		

Brucellosis in Southern States						
Andhra Pradesh28431224.3%						
Kerla	1361	109	8.0%			
Karnataka 10767 402 3.8%						

Gajendragad,2007



Brucellosis Situation in Human Population

- Sero-prevalence of 5.8 to 14.3% in veterinary workers and of 8.5% in general population of Belgaum. (Bhat *et al* in 1997).
- In Kashmir area (1992-1997) a total of 3532 patients of FUO were screened by Wright's tube agglutination test, out of which 28 (0.8%) were found positive. (Kadri *et al* 2000).
- Chronic brucellosis was noticed in 28 (6.8%) patients with fever of unknown origin out of 414 studied. (Sen *et al* 2002).
- In 2003, an outbreak of brucellosis was studied in HP state of North India and Serological study revealed 10% of contact animal handlers.(Chahota *et al* 2003).
- 292 Blood samples of blood donors were collected in Postgraduate Institute of Medical Sciences, Chandigarh, Punjab for sero-monitoring of brucellosis. One case found positive and one was doubtful.(Vaishnavi et al 2007).



HUMAN BRUCELLOSIS IN PUNJAB STATE

Year	Total Samples	Positive Samples	% Positive
Upto 2002	2430	383	15.8%
2003	139	40	28.7%
2004	905	95	10.5%
2005	430	44	10.2%
2006	297	44	14.8%
2007	175	47	26.8%
2008	241	64	26.6%

School of Veterinary Public Health



Human Brucellosis.....

Occupation	RBPT	STAT	Incidence on basis of RBPT
Veterinarians (N=118)	13	13	11.01 %
Veterinary paraclinical staff (N= 87)	22	20	25.28 %
Dairy farmers (N=62)	10	10	16.10%
Total (267)	45	43	16.85%

School of Veterinary Public Health-2012-13



Canine Brucellosis in Punjab State

Total no of canine serum samples analyzed	112
Serologically positive samples	9.8%
Positive among showing clinical symptoms (Abortion, orchitis, anorexia, persistent Temperature, itching etc.)	32.6%

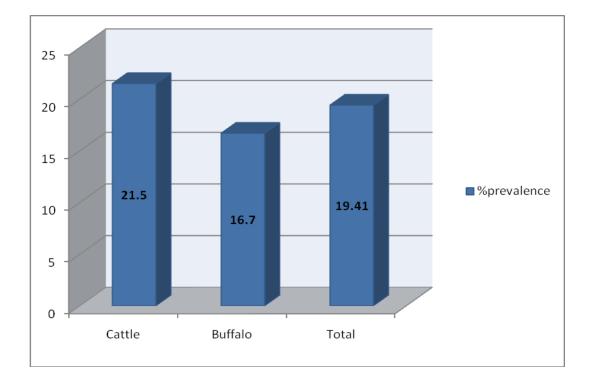


Zone wise sero-prevalence of brucellosis in Punjab State



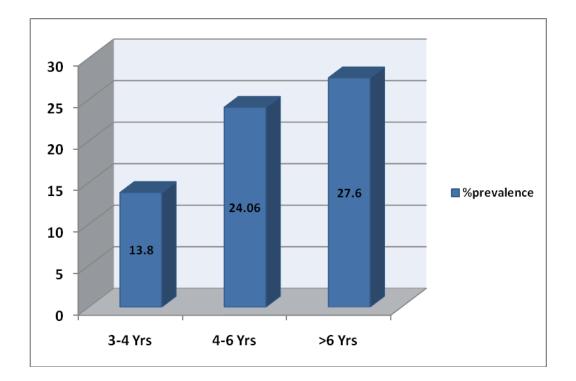
7		Cattle			Buffalo)	Overell
Zone	Tested	Positive	% prevalenc e	Tested	Positive	% prevalence	Overall prevalence (%)
I Sub- mountaneou s Zone	28	3	10.71	34	8	23.52	17.74
II Central Zone	188	50	26.59	148	27	18.24	22.91
III Arid Irrigated Zone	48	4	8.33	33	1	3.03	6.17





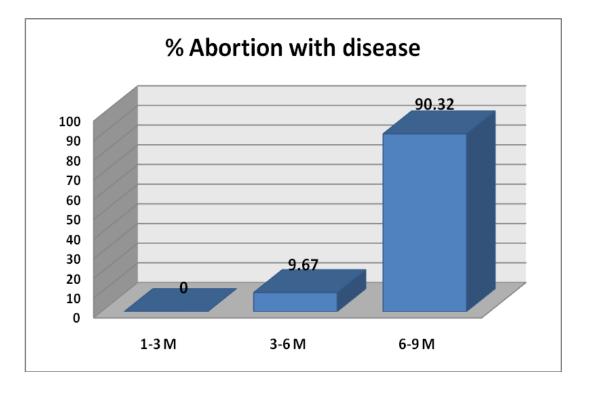
Species wise sero-prevalence of brucellosis in Punjab State





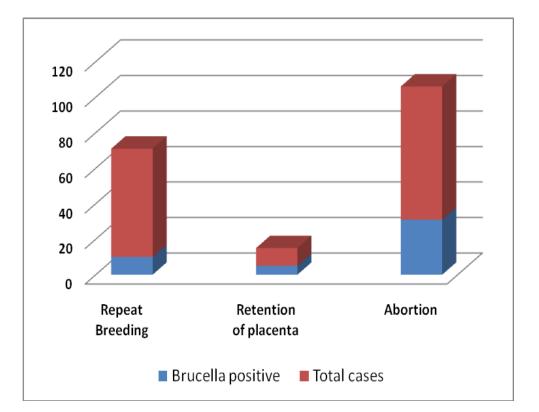
Age wise Sero-prevalence of brucellosis in Punjab State





Relationship between month of gestation and abortion in cows and buffaloes affected with brucellosis in Punjab State





Relationship of abortion, Retention of Placenta and Repeat Breeding with brucellosis



Surveillance Program and Diagnostic Strategy

- For sero-surveillance, Milk Ring Test is applied on bulk tank samples.
- The suspected herds are screened with Rose Bengal Plate Test and positive reactors are confirmed with Tube Agglutination Test and indirect ELISA.
- Isolation, identification and molecular characterization of the organism.
- Segregation of positive animals and no slaughter of positive animals.



Work under progress

- Sero-surveillance using RBPT, STAT, ELISA
- To compare the polymerase chain reaction (PCR) based techniques with conventional bacteriological techniques employed for diagnosis of brucellosis.
- Bruce-ladder multiplex PCR to differentiate various Brucella species.
- Real time PCR and LAMP standardised for detection of Brucella sp.
- Fluorescent polarisation assay is under going standardisation process.
- Cloning, expression and purification of different OMPs.



Asia Pacific Bovine Brucellosis Serum Proficiency Test 2013

RBPT Test performed on 27.12.13 Antigen batch no RBA 0513 Expiry date Sep 2015			IELISA Test performed on 28.12.13 Kit- Idexx lab, Batch no 220-z751, Expiry date 30.6.14			
Sample No	Tube No	Result	Interpretation	OD	Calculating Index	Interpretation (Positive >80%)
1	92	++++	Р	0.8052	120.46	Р
2	312	++++	Р	0.8434	126.65	Р
3	267	++++	Р	0.83105	124.65	Р
4	173	++++	Р	0.84915	127.59	Р
5	446	0	N	0.68575	101.11	Р
6	509	++	Р	0.65725	96.50	Р
7	611	0	N	0.6657	97.86	Р
8	794	++	Р	0.82665	123.94	Р
9	827	0	N	0.0778	2.624	N
10	1132	++	Р	0.7944	118.72	Р
11	1208	++++	Р	0.81075	121.36	Р
12	1370	++++	Р	0.8215	123.11	Р
13	918	0	N	0.2218	25.95	N
14	1569	0	N	0.0751	2.18	N
15	1477	0	N	0.17475	18.33	N
16	1603	++	Р	0.81515	122.08	Р
17	1037	++	Р	0.81085	121.38	Р



THANKS