

ERYTHROCYTIC ISOIMMUNIZATIONS

by

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Stormont (~1956, pers. comm.) knew that Aberdeen-Angus were poor responders to isoimmunizations to cattle red cells in comparison with other cattle breeds. Similar experiments with cattle in Iowa belonging to the Animal Science Department of Iowa State University have supported this observation.

The Holstein-Friesian breed (purebred and grade cattle) has been most frequently used for the earlier isoimmunizations in the United States. Jersey, Hereford and Angus have been used perhaps next in frequency. A sprinkling of Ayrshire, Brown Swiss, Gernsey, Shorthorns, and grade cattle with an occasional rare breed individual or two complete the list of recipients.

The immunization procedure known to work well in such breeds generally is to inject 50 ml of 20-50% washed red cells intravenously weekly for 3 or 4 weeks. Weekly samples of antiserum are titer-tested for a rise in antibody response. The test is a hemolytic one (see Miller, 1966) although saline agglutinating antibodies can be noted if they occur. The infrequent agglutination response invariably has been weaker than the blocking antibodies or hemolytic response.

Aberdeen-Angus (or Hereford) donor blood differed in a total 94 combinations of factors of 8 blood group systems that were absent in the recipient (table 1). Holstein-Friesians especially were known to be potentially able to elicit such desired antibodies with the exception of R_2 . Only one of the 27 (~4%) immunizations succeeded (C36). The combination $O_1/O_x T_1 Y_1 A' E_3' G'$ as a donor into $O_x T_1 A' E_3' G'/I'$ produced a weak O_1 reagent not regarded as strong enough to use in a standard test. The other 26 immunizations were complete failures. They are listed in Table 1 by the blood group system on the left and the factors involved on the right. The number of immunizations involving the factors follows.

Since the preponderant majority of donor cells in Angus recipients were also Angus, Angus donor cells used in other breeds of recipients needs comparison with the Angus recipients. Only 17 such isoimmunizations were made at ISU, but the response was obviously better among the 17 recipients of other breeds, 12 Holstein-Friesian, four Shorthorn, and one Hereford. There were 43 factors of 6 systems that could have stimulated antibodies,

Of course, not all the donor factors or phenogroups were the same. There were 14 factors (successful) in common in the two groups of isoimmunizations, and 14 others not successful. More important, some factors (A_1 , Y, R_1 , S, and Z) with 50% frequency of success or greater in isoimmunizations failed in Angus.

In table 3 are details of 111 additional isoimmunizations of other breeds as donors and recipients. There was 28% overall success. The most antigenic systems are F-V, S and Z systems. The A and Z' factors are also excellent antigens in isoimmunizations, although that is not evident from this particular data, since no Z' isoimmunizations were included. It should be remembered that the combinations are not random. Those factors less frequently antigenic are, perforce, repeated more frequently in immunization attempts.

Ideal controlled comparisons are not economically feasible in this material, so the results require considerable qualification or reservation. Nevertheless, the evidence favors an opinion that Aberdeen Angus are poor responders in erythrocytic isoimmunizations.

In the modern parlance, Angus would lack the appropriate type of immune response (IR) gene. The purveyors of this term seldom mention that many genes are usually responsible for a genetic character, and that the first "IR genes", were evident in the cattle (and human) isoimmunizations in years past in the form of subtypes. This point was made clear to W.H. Stone and myself when Stormont described the O_x reagent. We immediately improved our success in producing O_3 reagent by using non O_x recipients, and better absorbing cells. Therefore, the lower subtypes, such as A_2 are non-immune response genes to the higher antigenic types, such as A_1 and the lack of subtypes in a recipient are controlled by "IR" genes. This principle has exceptions, but the generality is well known to cattle blood groupers.

A possible next step is to compare the success of factors as antigens within particular phenogroups. In the B and C systems it is well known that some factors at least are more antigenic in particular phenogroups than others. I have found, for example, that T_1 in the phenogroup $O_1 T_1 E_3 F' K'$ is seldom antigenic and even then only weakly so, while $T_1 E_3 F'$ is a good phenogroup for producing strong anti-T. More exact details and further data need documentation. Perhaps comparisons between laboratories would further this endeavor.

Table 1 - Eight systems and 28 factors in 94 possible immunological sensitizations in 27 Angus cattle.

System	Factors and number of times involved	System	Factors and number of times involved
A	A - 1	C	C ₁ - 1
	H - 1		*E - 10
			R ₁ - 2
B	B - 2		*R ₂ - 5
	I ₂ - 4		W - 1
	O ₁ - 2		X ₂ - 3
	(O ₁) - 4		
	O _x - 1	F - V	F - 1
	P - 2		
	Y ₂ - 1	M	M ₁ - 4
	A' - 4		
	E' ₁ - 3	S	S - 4
	(E' ₁) - 2		H' - 5
	E' ₃ - 2	Z	Z ₁ - 5
			Z ₂ - 1
	G' - 2	R' - S'	R' - 6
	I' - 5		S' - 3
	O' - 5		

() O₁ and E'₁ in parenthesis refer to a lower subtype O_x and E'₃ in the recipient which lowers the chances of response, although as in the one (C36) that succeeded, the response is not obviated.

*Known to be poor or rarely antigenic in other breeds (H.F.) also.

Table 2 - Six systems and 20 factors in 43 possible immunological sensitizations by Angus cells in other breeds of recipients.

<u>System</u>	<u>Factor</u>	<u>Antibody Stimulation</u>		<u>System</u>	<u>Factor</u>	<u>Antibody Stimulation</u>	
		<u>Successful</u>	<u>Failed</u>			<u>Successful</u>	<u>Failed</u>
B	B	1	1	C	C ₁	1	1
	G	2			E	2	2
	I ₂	1	1		R ₂	5	5
	O ₁		1				
	T	3	1	F-V	F	1	
	Y		2		V		1
	A'	1	1				
	D'	1		M	M ₁	1	4
	E'		3				
	G'		1	L	L	1	
	O'		2				
	Y'		2	S	S	1	

12/43 = .387 = ~40% succeeded.

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TABLE 3 - A summary of 111 additional ISU isoimmunizations in Holstein-Friesian, Shorthorn, Hereford, Brown Swiss, Jersey, and grade cattle. nine systems and 53 different factors in 559 possible immunological sensitizations (28% successful).

System	Factor	Antibody Stimulation		System	Factor	Antibody Stimulation	
		Successful	Failure			Successful	Failure
A 35% Successful	A ₁	6	1	B	O'	3	11
	A ₂	1	-	-	Y'	-	4
	D	-	6	-	7	2***	16
	H	2	10				
B 27% Successful	B	5	14	C	C ₁	6	9
	G	7	14		C ₂	-	1
	K	3	10	18% Successful	E	-	13
	I	-	2	R ₁	9****	8	
	I ₂	4	18	R ₂	-	24	
	O ₁	5*	7	W	1	10	
	O ₂	-	1	X ₁	4	3	
	O ₃	-	3	X ₂	-	22	
	O _x	-	6	X ₃	1***	-	
	P	1	3	L ₁	1	7	
	Q	5	4	F-V 47% Successful	F	3	1
	Y ₁	5	6	V	5	8	
	Y ₂	8	8	L 15%	L	2	11
	A'	4	23	M 33%	M ₁	-	-
	B'	-	1	M ₂	2	3	
	D'	8	15	S	S ^R _(-/-)	2	1
E' ₁	6	7	55%	S ^R _(H')	9	5	
E' ₂	1	4	H'	3	4		
E' ₃	3**	3	U ₂	3	4		
F'	2	14	Z	Z ₁	6	5	
G'	8	10	41%	Z ₂	-	4	
I'	5	8	Z ₁ → Z ₂	1	1		
J'	1	8	R'-S'	R'	1	6	
K'	1	11	25%	S'	2	3	
					157	402/559	

* O₁ → O_x Successful 3 times

** 1 anti - E₃ → E₂' reagent!

*** "Successful" does not always mean strong enough to use in a Standard test

**** One R₁ → R₂ Successful.