Quantitative marker of Immunoglobulin G (IgG) in foal

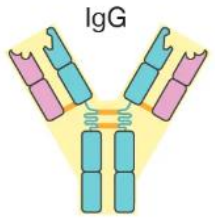
Vcheck Foal IgG

BIONOTE Marketing team

September 2023

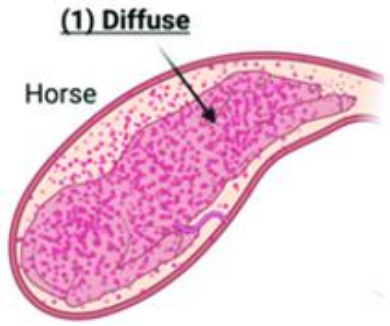
Immunoglobulins in Foals

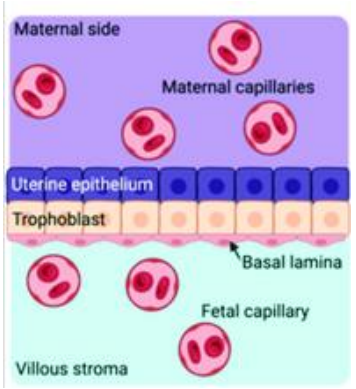
- **Immunoglobulins (Ig): Antibodies (IgM, IgD, IgA, IgE, and IgG)**
  - Protein used by the immune system to neutralize antigens (pathogenic bacteria and viruses)
  - **Immunoglobulin G (IgG)**: 75% of serum immunoglobulin



- **Foals have low concentrations of circulating immunoglobulins at birth**,
  because the diffuse epitheliochorial placenta does not allow antibody transfer during pregnancy[1].

- Foals begin to produce immunoglobulins immediately after they have been exposed to antigens,
  but protective concentrations of immunoglobulins may not be reached until they are 2 months old.





◀ The feature of equine placenta
(diffuse epitheliochorial placenta)

Reference: 1) J Vet Med Sci. 2012 Nov;74(11):1387-95.

Clinical utility of IgG in Foals

Immunoglobulin G in Foals

- **Transfer of maternal antibodies is critical for the well-being of foals.**
  - Foals are entirely dependent on maternal immunoglobulins (**primarily IgG**, lesser quantities of IgA and IgM) absorbed through ingestion of mare's colostrum **in the first 24 hours of life**[1)].
  - Immunoglobulin is not absorbed after 24–36 hours of age [2)].

- **Failure of passive transfer (FPT)** : failure to absorb colostral antibodies sufficient to achieve a IgG
  - **Healthy foals** consuming good-quality colostrum: IgG concentrations > 800 mg/dL
  - **Complete FPT**: IgG concentrations < 400 mg/dL after 24 hours of age
  - **Partial FPT**: serum IgG concentrations between 400 and 800 mg/dL

- **Incidence:** FPT (ranges from 3–24% in foals), partial FPT (ranges from 14–31%)[1].

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Reference: 1) J Vet Med Sci. 2012 Nov;74(11):1387-95. 2) J Vet Intern Med 2005;19:93–96

Clinical utility of IgG in Foals

Immunoglobulin G in Foals

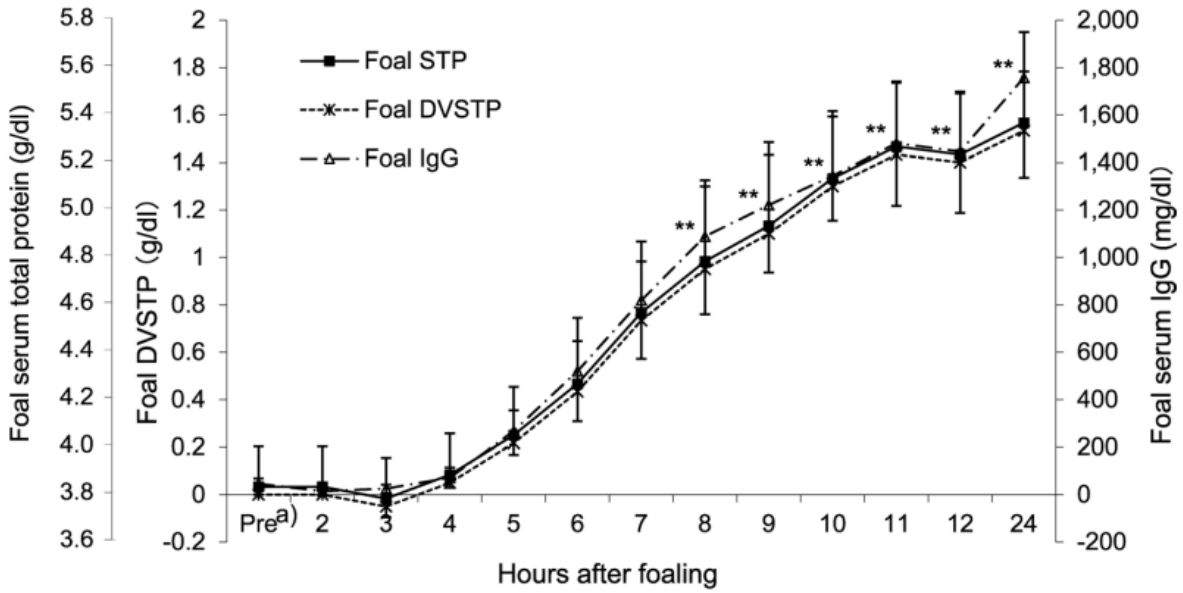
- Foals with insufficient serum immunoglobulin (IgG) levels are at increased risk of infectious diseases, such as septicemia, arthritis or respiratory tract infections.

- **Several factors which can lead to FPT**
  - Poor colostrum quality (Not sufficient quantities IgG < 3,000 mg/dl)
  - Delayed suckling by the foal (mares reluctant to let foals suckle)
  - Ingestion of too little colostrum
  - Failure of the foal to absorb IgG from the colostrum

Reference: 1) J Vet Med Sci. 2012 Nov;74(11):1387-95.

Clinical utility of IgG in Foals

IgG concentrations in Foals



▲ Changes in foal serum IgG from before suckling to 24 hr after birth

- **From 4 hr after birth,**
  - ✓ The foal serum IgG: tendency to increase
  - ✓ The colostrum/milk IgG: tendency to decrease
- **6-12 hours**: maximum antibody transfer

Reference: 1) J Vet Med Sci. 2012 Nov;74(11):1387-95.

Result Interpretation

Interpretation of IgG results

- **Early testing for IgG concentration in newborn foals can identify potential cases of FPT.**
  (Treating foals with FPT may require hospitalization and costs can exceed $1500 per foal[1].)

- Test the foal's IgG concentrations after 12 hours after birth to assess passive transfer status.

- If the foal has partial or complete FPT, you must administer antibodies via fresh or frozen oral colostrum (within the first 12-18 hours of life), commercial products, or intravenous plasma (if 24 hours have passed).

REFERENCE RANGE

< 400 mg/dL	400 ~ 800 mg/dL	> 800 mg/dL
Failure of passive transfer in foal	Partial failure of passive transfer in foal	Successful passive transfer in foal

* Each laboratory should establish its own reference interval, as reference values may vary depending on the test population.
* The veterinarian in charge must conduct a clinical diagnosis along with the measured results of this reagent, clinical symptoms, and other test results.

◀ Reference range of Vcheck Foal IgG

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Reference: 1) http://www.seramune.com/faq.html

Diagnostic Algorithm

Algorithm to prevent Failure of Passive Transfer

Vaccinate the mare 4-6 weeks before foaling[1]

▼

Make sure the foal stands and nurses
Watch for common causes, such as early lactation, placental infections, premature birth, fescue toxicosis, and poor-quality colostrum

▼

Run an IgG test ideally after 12 hours of birth

▼

IgG < 400 mg/dL	IgG 400-800 mg/dL	IgG > 800 mg/dL
▼	▼	▼
Complete FPT (failure of passive transfer)	Partial FPT (failure of passive transfer)	Successful passive transfer in foal

▼

Administer antibodies
- (Within the first 12-18 hours of life) via fresh or frozen oral colostrum, commercial products
- (If 24 hours have passed) intravenous plasma

▶

After treatment, IgG concentrations should be rechecked in 8 to 12 hours.
to assess the adequacy of immunoglobulin therapy

Reference: 1) Vaccination and Passive Transfer, Reviewed by the AAEP Infectious Disease Committee in 2021 2) https://thehorse.com/197216/what-is-failure-of-passive-transfer-in-horses/ 3) Sally Vivrette, DVM, PhD, Colostrum and Oral Immunoglobulin Therapy in Newborn Foals

Case Study

- A foal born in September, 2021
- The foal was nursing from the mare within two to three hours after birth.
- When monitoring the mare carefully in the days prior to foaling, the mare drip milk for several days to weeks.
- IgG measurements were performed 12 hours after the foal was born. **The IgG result was 520 mg/dl**.
  → It was interpreted as "partial FPT".
- Oral immunoglobulin supplement (Seramune, Sera, Inc.) was immediately administrated at the manufacturer's recommended dosage.

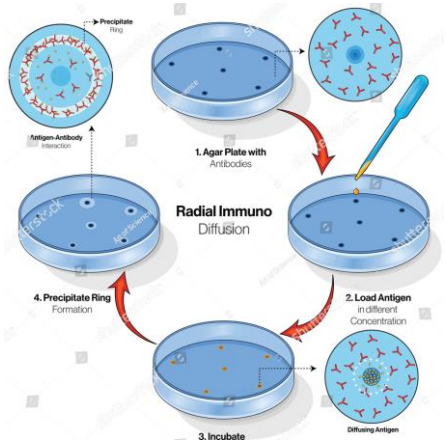
example photos

Reference: 1) https://thehorse.com/130821/foal-igg-antibody/ 2) Photo: Anne M. Eberhardt/The Horse, https://equine-reproduction.com/articles/mares/fpt

Gold standard method

Gold standard method for the IgG test

- Several screening tests have been developed over the years for evaluating the IgG concentration in foals.
- **The single radial immunodiffusion (SRID) test**
  : considered to be the most accurate test for quantitative measurement of IgG concentration in foals
- **However, results of SRID test are generally not available for 24 hr.**
  The 24-hr delay in obtaining test results decreases the usefulness of this test for routine use on a breeding farm, in a field situation or at a veterinary clinic because rapid results are required for therapeutic intervention in FPT cases in foals.



▲ Radial immunodiffusion (RID) test

Reference: 1) J Vet Med Sci. 2012 Nov;74(11):1387-95. 2) Photo: shutterstock, edvotek

Performance

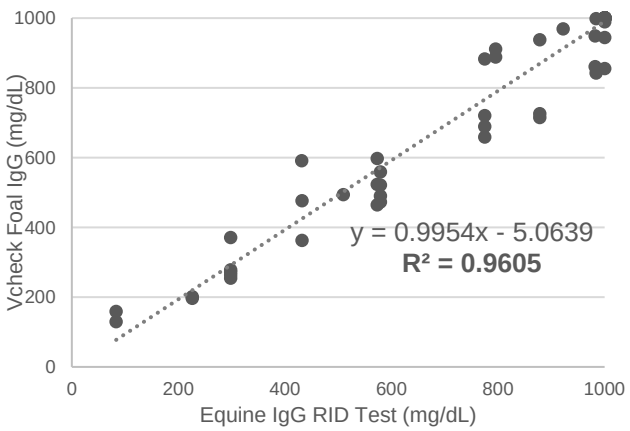
Strong correlation with a gold standard method

▲ Triple J Farms IgG

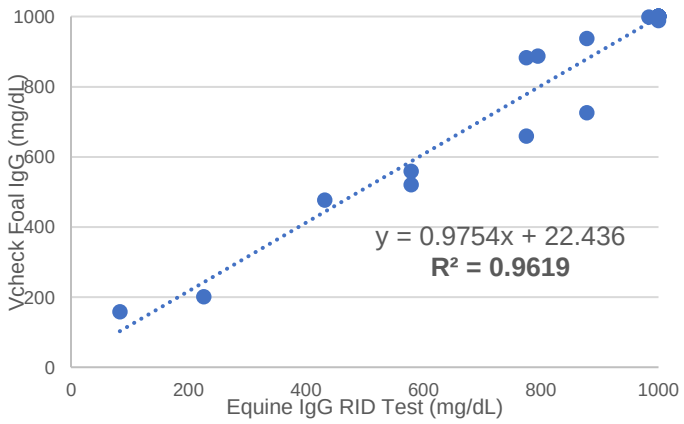
- **The reference method of Vcheck Foal IgG**: Triple J Farms IgG* (RID assay)

  *Validated assay[1]

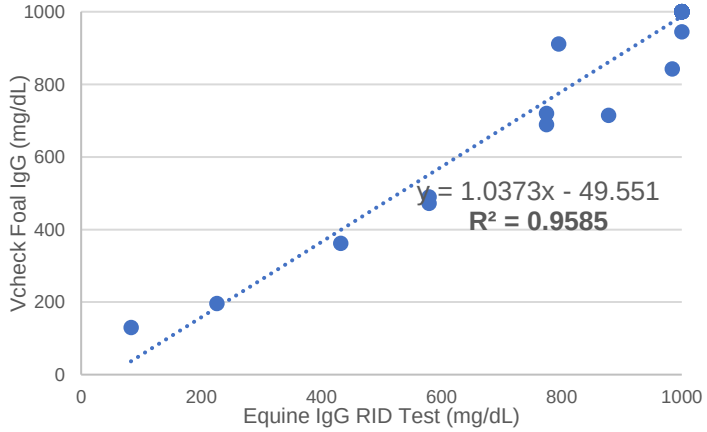
- There is a strong correlation (slope 0.995, **R^2=0.96**) was found between the two test methods when analyzing 102 whole blood, plasma, and serum samples[2].



All sample types (n=102)



Serum (n=42)



Whole blood (n=40)

Reference: 1) J Vet Intern Med 2017;31:1170–1177 2) Bionote study

Performance

Better performance than IDEXX SNAP KIT

- When compared to the RID test (cut-off 800 mg/dl)
  - ✓ **Vcheck:** sensitivity of 97% (64/66), specificity of 81.3% (13/16)
  - ✓ **IDEXX SNAP**: sensitivity of 92.4% (61/66), specificity of 75% (12/16)

▲ IDEXX SNAP Foal IgG

Results are interpreted as '<400 mg/dl', '400 mg/dl', '400-800 mg/dl', '800 mg/dl', or '>800 mg/dl'.

- Based on our comparative analysis, the in-clinic Vcheck test demonstrated superior performance compared to the SNAP kit when evaluated against the reference RID test for measuring foal IgG levels.

Vcheck (vs. RID)

IgG		RID			Total
		<400	400-800	>800	
Vcheck (mg/dl)	<400	4	1	0	**5**
	400-800	0	8	2	**10**
	>800	0	3	64	**67**
Total		**4**	**12**	**66**	**82**

Concordance rate 92.7% (76/82)

Sensitivity 97.0% (64/66, cut-off 800 mg/dl)

Specificity 81.3% (13/16, cut-off 800 mg/dl)

IDEXX SNAP (vs. RID)

IgG		RID (mg/dl)			Total
		<400	400-800	>800	
SNAP (mg/dl)	<400	4	1	0	**5**
	400-800	0	7	5	**12**
	>800	0	4	61	**65**
Total		**4**	**12**	**66**	**82**

Concordance rate 87.8% (72/82)

Sensitivity 92.4% (61/66, cut-off 800 mg/dl)

Specificity 75.0% (12/16, cut-off 800 mg/dl)

Reference: 1) Bionote study

FAQ

Is the ‘colostrum IgG test’ useful?

- Serum IgG concentration in a newborn foal correlates well with the IgG concentration in its dam’s colostrum[1].
- However, one report has indicated that **foals that fail to nurse due to neonatal weakness or delayed development of the nursing reflex have low IgG concentrations** despite adequate equine colostrum quality[1].

Is the ‘total protein (TP) test’ useful?

- One of the most rapid and inexpensive tests (by refractometry)
- However, several reports have indicated that this test is an unreliable indicator of FPT in foals because unlike in calves, **there is a wide range of total protein concentrations in newborn foals**[1].

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Reference: 1) J Vet Med Sci. 2012 Nov;74(11):1387-95.

Thank you for your time!

Any questions?

Key points

- ✓ Transfer of maternal antibodies is critical for the well-being of foals to achieve a IgG.
- ✓ Early testing for IgG concentration in newborn foals can identify potential cases of FPT.
- ✓ The Vcheck had superior performance compared to the SNAP kit when evaluated against the reference RID test for measuring foal IgG levels

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Quantitative marker of Foal IgG

Vcheck Foal IgG

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Vchck Foal IgG

Vcheck Foal IgG

Quantitative marker of Immunoglobulin G in foal

✓ Simple and useful method of assessment of neonatal foals' IgG levels.

- **Species :** Horse
- **Sample :** Whole blood (EDTA or heparin)10 µl
  or Serum, Plasma (EDTA or heparin) 5 µl
- **Testing time :** 5 minutes
- **Measurement Range :** 100 ~ 1,000 mg/dl
- **Storage Condition :** 2 ~ 30 ºC

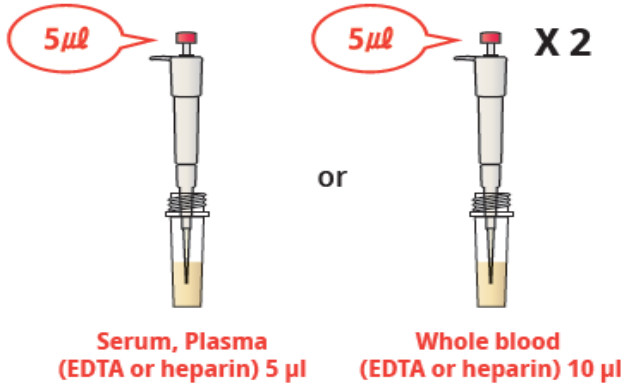
Vchck Foal IgG

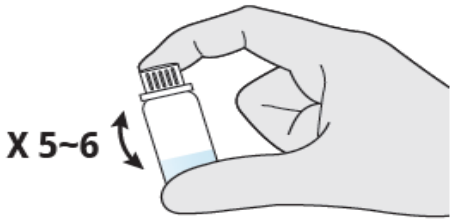
Vcheck Foal IgG

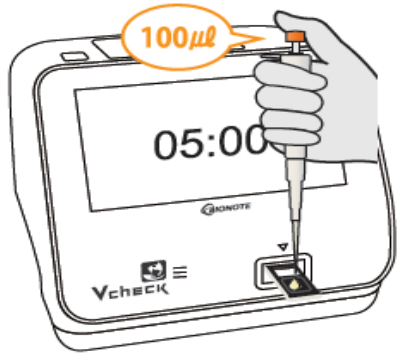
Quantitative marker of Immunoglobulin G in foal

✓ Simple and useful method of assessment of neonatal foals' IgG levels.

Add 5 µl of serum, plasma (EDTA or heparin) or 10 µl of whole blood to the assay diluent bottle	Close the bottle cap and shake for 5-6 times to mix thoroughly	Add 100 µl of the mixed sample into the test device





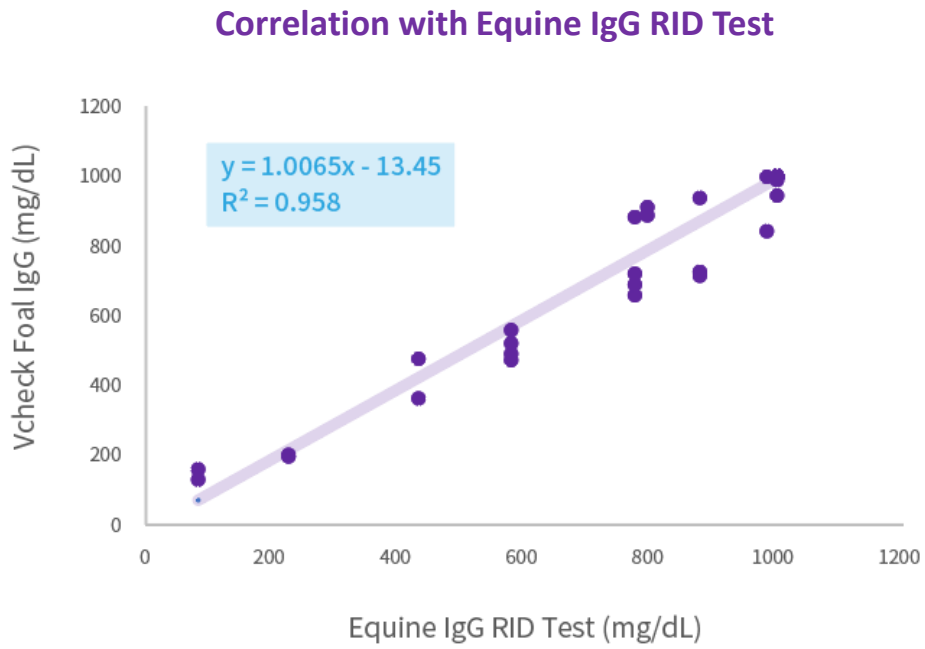


< 400 mg/dL	400 ~ 800 mg/dL	> 800 mg/dL
Failure of passive transfer in foal	Partial failure of passive transfer in foal	Successful passive transfer in foal

Performance

Correlation

Vcheck Foal IgG has a strong correlation (y=1.0065x - 13.45, **R2=0.958**) with the reference method (Equine IgG RID), which has been used in reference laboratories



Vchck Foal IgG

Vcheck Foal IgG

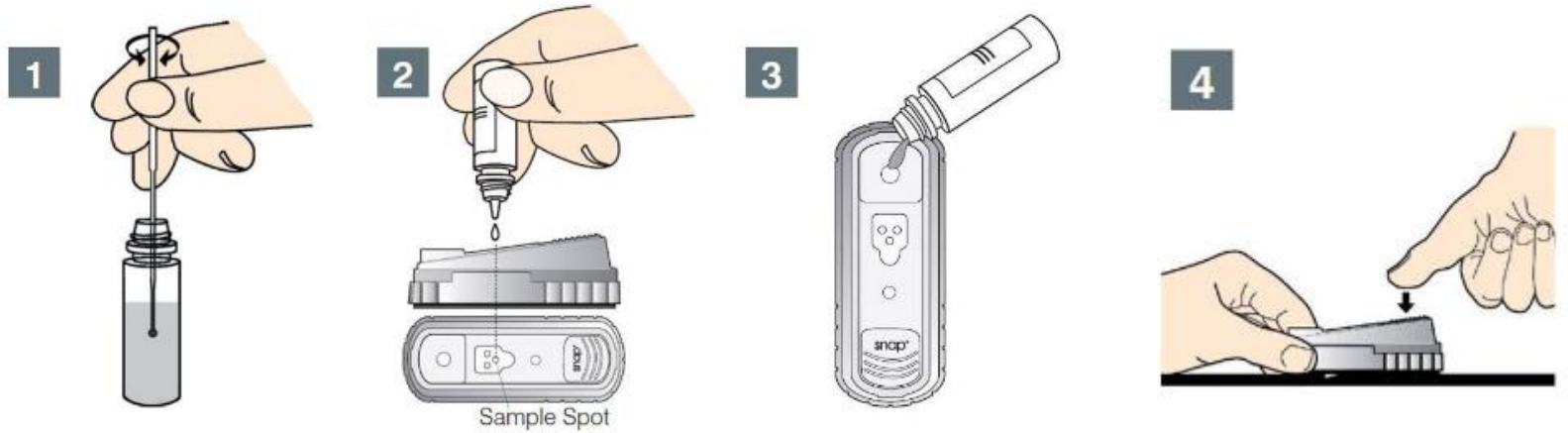
Quantitative marker of Immunoglobulin G in foal



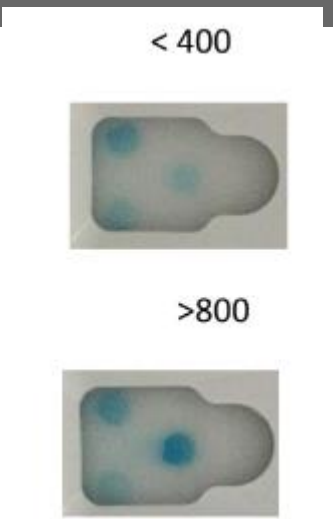

Equine IgG	Vcheck F	IDEXX (USA)	TargetVet (USA)	VMRD (USA)
Product Name	Equine Foal IgG	SNAP Foal IgG	Immuno-Chek G	Foal IgG
Analyzer	V200, V2400	x	x	VMRD reader
Assay	Fluorescent immunoassay (FIA)	ELISA	Lateral Flow Tech	Lateral Flow Tech
Specimen	Serum, Plasma(EDTA, heparin), whole blood (EDTA, heparin)	Whole boold, Serum, Plasma(heparin, EDTA)	Serum, whole blood (Heparin, EDTA)	Serum, whole blood (EDTA)
Measurement Range	**100 mg/dL~1,000 mg/dL**	100-800 mg/dL	10-2700 mg/dL	200-1000 mg/dL
Test Time	**5mins**	7mins	10mins	10 mins
Storage	**2-30°C**	2~8°C	Room temperature	Room temperature
Reference Range	< 400 mg/dL Failure of passive transfer in foal 400 ~ 800 mg/dL Partial failure of passive transfer in foal > 800 mg/dL Successful passive transfer in foal	IgG > 800 mg/dL → adequate passive transfer (good/normal immune protection) IgG between 400 – 800 mg/dL → partial failure of passive transfer (partial protection) IgG < 400 mg/dL → complete failure of passive transfer (very susceptible to infection)	>800 Very Good Transfer 400 Partial Transfer 200 Total Failure	0-400 mg/dL: Very low IgG 400-800 mg/dL: Low IgG >800 mg/dL: Normal IgG
Package unit	5 Tests/Kit	5 or 10 Tests/Kit	5 or 10 Tests/Kit	15Tests/Kit
Note	**•27 Items available •3 equine panel**	semi-quantitative test	semi-quantitative test	Quantitative

Foal IgG

IDEXX –Snap Foal IgG

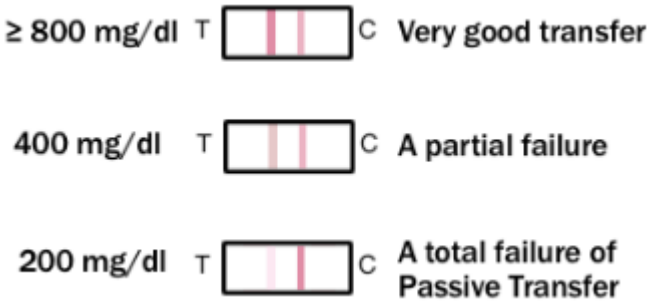


- Sample: Plasma(heparin), Serum, Whole blood
- Testint time: 7 mins
- Assay: ELISA
- Video: https://www.youtube.com/watch?v=BuJMPcSsqNg
- Sensitivity: 94.7%
- Result: : 100-800 mg/dL
- Package unit: 5T/kit



Foal IgG

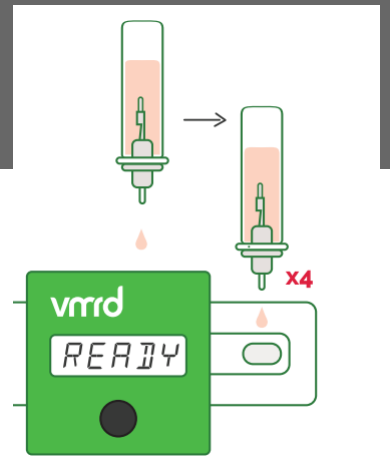
Target vet- Immuno Check G

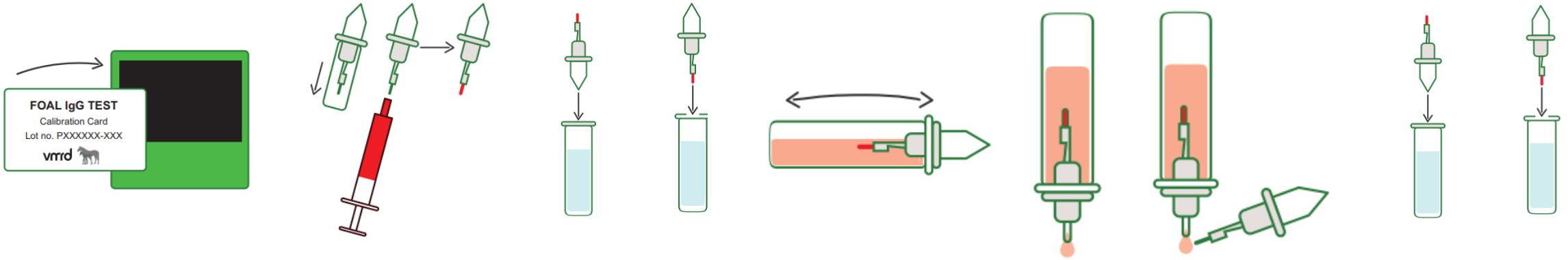


- Sample: Serum, Whole blood
- Testint time: 10 mins
- Assay: Lateral flow tech
- Video: https://www.youtube.com/watch?v=2qySoaU9Xi4
- Sensitivity: 100%
- Result: : 10-2700 mg/dL
- Package unit: 5T/kit

Foal IgG

VMRD Foal IgG



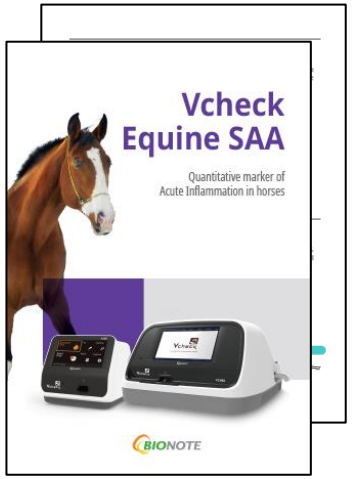


- Sample: Plasma(EDTA), Whole blood
- Testint time: 10 mins
- Assay: Lateral flow tech
- Video: https://www.youtube.com/watch?v=Y9V6hIZ6jpM
- Result: : 10-2700 mg/dL
- Package unit: 15T/kit

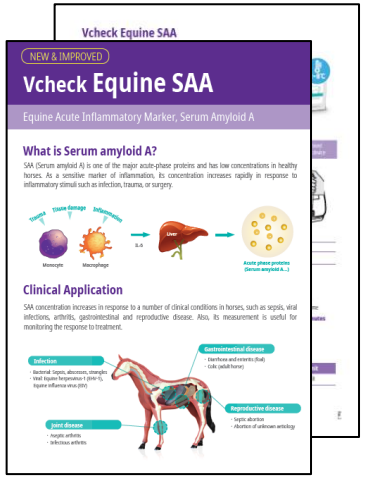
Marketing materials

✓ Brochures/leaflets

- Brochure: Detailed resource, showcases product values, used by vets to explain benefits and encourage tests.
- Leaflet/Pamphlet: Summarized content, highlights key product advantages, helps vets communicate benefits to patients.

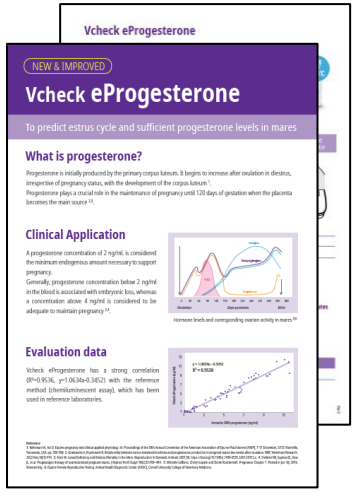


▲ E.SAA brochure



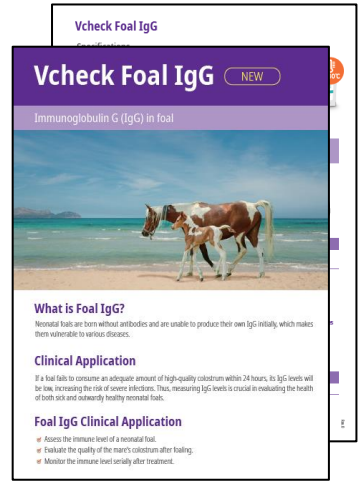
▲ E.SAA Leaflet

▲ ePRG brochure



▲ ePRG Leaflet

▲ Foal IgG brochure

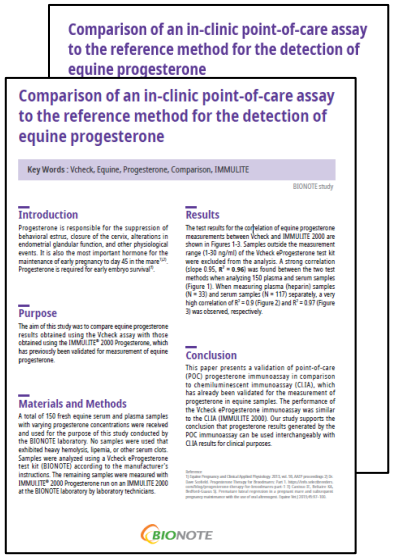


▲ Foal IgG Leaflet

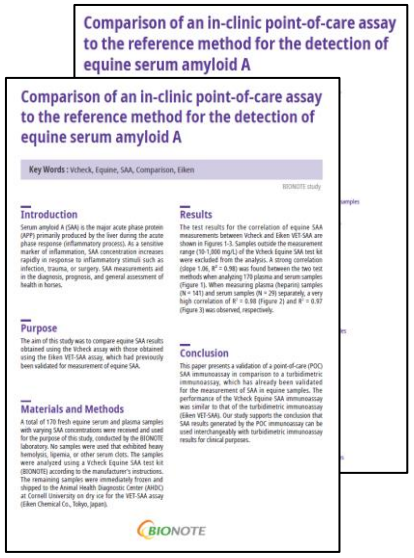
Marketing materials

✓ Evaluation

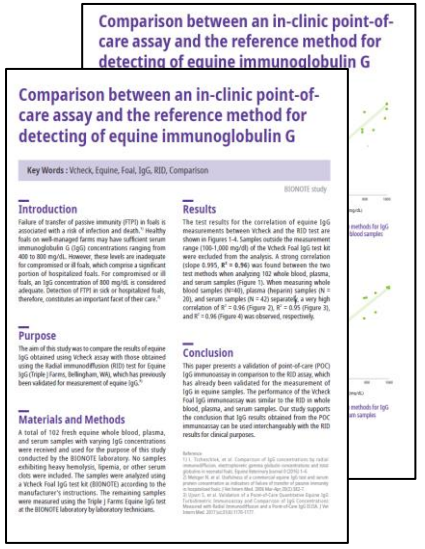
- **eProgesterone**: Comparative assessment with over 150 samples against the IMMULITE 2000 equipment demonstrated excellent correlation (R2=0.96), based on data from the BN Research Division.
- **E.SAA**: Using over 170 samples, a comparative assessment against the Eiken VET-SAA equipment demonstrated exceptional correlation (R2=0.98), based on data from the BN Research Division.
- **Foal IgG**: A strong correlation (slope 0.995, R2 = 0.96) was found between Vcheck and the RID test when analyzing 102 whole blood, plasma, and serum samples



▲ ePRG



▲ E.SAA

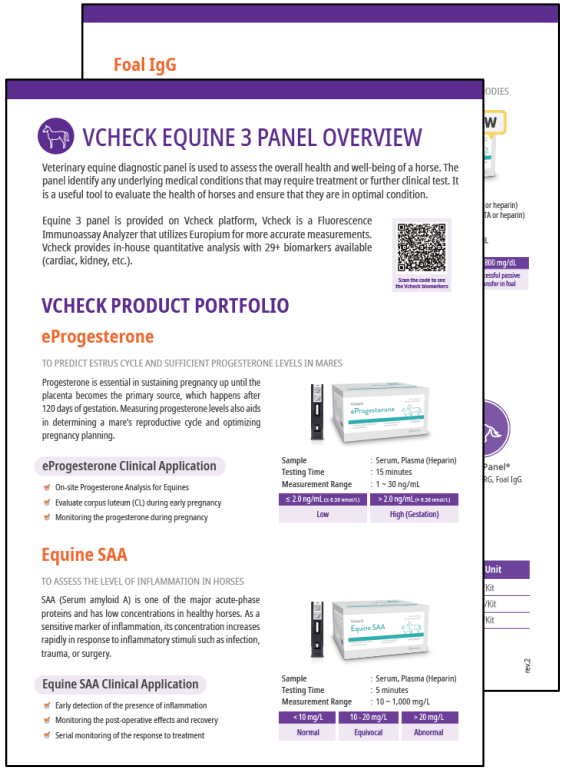


▲ Foal IgG

Marketing materials

✓ Fact sheet

- Quick product overview in one page.
- Showcases product range and benefits.
- Aids veterinarians in explaining options to customers.



▲ Fact sheet

Thank you

BIONOTE Marketing team

September 2023